

**UNIVERSITY OF MODENA AND REGGIO EMILIA**

**Ph.D. School in: Agri-Food Sciences, Technologies and Biotechnologies**

---

**XXXV Cycle**

**A MULTIDISCIPLINARY APPROACH TO IMPROVE THE  
STABILITY OF VINEGAR-BASED CONDIMENTS**

**Ph.D. Candidate: Andrea Goldoni**

**Supervisor: Dr. Maria Gullo**

**Dean of the PhD School Professor Alessandro Ulrici**

---



# Index

<i>Thesis outline</i>	<i>page 7</i>
<i>Abstract of the thesis</i>	<i>page 9</i>
<i>Riassunto</i>	<i>page 11</i>
<i>Aims, objectives and strategies</i>	<i>page 13</i>

## **Chapter 1 - Introduction** *page 14*

<i>1.1 Food condiments</i>	<i>page 14</i>
<i>1.2 Vinegars towards condiments</i>	<i>page 15</i>
<i>1.3 The glaze, in particular the Glaze with Aceto Balsamico di Modena PGI</i>	<i>page 15</i>
<i>1.4 Ingredients of the Glaze with Aceto Balsamico di Modena PGI</i>	<i>page 17</i>
<i>1.5 The stability and safety of the Glaze with Aceto Balsamico di Modena PGI</i>	<i>page 23</i>
<i>1.6 Microbiological aspects affecting glazes stability</i>	<i>page 25</i>
<i>1.7 Company description</i>	<i>page 28</i>
<b>REFERENCES</b>	<i>page 33</i>

## **Chapter 2 - First insight into glazes stability** *page 36*

<i>2.1 Introduction</i>	<i>page 36</i>
<i>2.2 Materials and method</i>	<i>page 36</i>
<i>2.2.1 Glaze with Aceto Balsamico di Modena PGI - sample</i>	<i>page 36</i>
<i>2.2.2 Analytical determinations (pH, titratable acidity and refractometric degree)</i>	<i>page 36</i>
<i>2.2.3 Enzymatic determinations (Ethanol, acetic acid, sucrose, D-fructose and D glucose)</i>	<i>page 37</i>
<i>2.2.4 Microbiological analyses</i>	<i>page 37</i>

2.2.4.1	<i>Total Mesophilic Bacterial Load</i>	page 37
2.2.4.2	<i>Acetic Acid Bacteria growth</i>	page 37
2.2.4.3	<i>Yeasts growth</i>	page 38
2.2.5	<i>Bacterial strains used in this study</i>	page 38
2.2.5.1	<i>Strain preservation and revitalization</i>	page 39
2.2.6	<i>Challenge tests</i>	page 40
2.2.7	<i>Bacterial cellulose qualitative test</i>	page 41
2.2.8	<i>Quantitative bacterial cellulose test</i>	page 42
2.2.9	<i>Optical microscopy analysis</i>	page 42
2.3	<i>Results and discussion</i>	page 42
2.3.1	<i>Characterization of the Glaze with Aceto Balsamico di Modena PGI</i>	page 42
2.3.2	<i>Evaluation of AAB growth in glaze at different dilutions and storage temperature by challenge tests</i>	page 44
2.3.2.1	<i>A. pasteurianus AB0220</i>	page 46
2.3.2.2	<i>G. oxydans, ATCC 621H</i>	page 48
2.3.2.3	<i>Ga. johannae, DSM 13595<sup>T</sup></i>	page 49
2.3.2.4	<i>Komagataeibacter sp., K1A18</i>	page 49
2.3.3	<i>Evaluation of bacterial cellulose production</i>	page 51
2.3.4	<i>K. xylinus K2G30 as a case study in glaze with Aceto Balsamico di Modena PGI</i>	page 52
2.4	<i>Conclusion and perspectives</i>	page 58
	<i>REFERENCES</i>	page 59

<b>Chapter 3 - Microbiological and chemical study of Glaze with Aceto Balsamico di Modena PGI</b>	<b>page 61</b>
3.1 Introduction	page 61
3.2 Materials and method	page 62
3.2.1 Samples description and experimental set up	page 62
3.2.2 Yeast strain and culture conditions	page 63
3.2.3 Microbiological analysis	page 64
3.2.4 Physico-chemical analysis	page 64
3.3 Results and discussion	page 65
3.3.1 Microbial spoilage	page 65
3.3.2 Total mesophilic count	page 65
3.3.3 Yeasts count	page 66
3.3.4 Molds, lactic acid bacteria and acetic acid bacteria count	page 68
3.3.5 Changes in physico-chemical parameter caused by microbial spoilage	page 69
3.3.5.1 <i>a</i> W	page 69
3.3.5.2 pH	page 71
3.3.5.3 Total acidity in acetic acid	page 72
3.3.5.4 Relative density at 20°C	page 74
3.3.5.5 Alcoholic degree	page 77
3.3.5.6 Ochratoxin A	page 77
3.3.5.7 Sugars content and profile	page 78
3.3.5.7.1 Reducing sugars	page 78
3.3.5.7.2 Galactose, Sucrose, Lactose and Maltose content	page 80
3.3.5.7.3 Glucose content	page 82
3.3.5.7.4 Fructose content	page 84

3.3.5.8	<i>Organic acids profile</i>	<i>page 86</i>
3.3.5.8.1	<i>Citric acid</i>	<i>page 86</i>
3.3.5.8.2	<i>Lactic acid</i>	<i>page 88</i>
3.3.5.8.3	<i>Malic acid</i>	<i>page 90</i>
3.3.5.8.4	<i>Succinic acid</i>	<i>page 92</i>
3.3.5.8.5	<i>Tartaric acid</i>	<i>page 94</i>
3.3.5.9	<i>Brookfield rotational viscosity</i>	<i>page 96</i>
3.4	<i>Concluding remarks</i>	<i>page 100</i>
	<i>REFERENCES</i>	<i>page 101</i>
	<i>Acknowledgements</i>	<i>page 104</i>
	<i>Ringraziamenti</i>	<i>page 105</i>
	<i>Appendix A:</i>	
	<i>Neutron SPA analysis certificates</i>	<i>page 106</i>

## Thesis outline

This PhD project was developed in the frame of a collaboration between Unimore Microbial Culture Collection (UMCC) laboratory and the *Acetificio Carandini Emilio S.p.A* company. The thesis manuscript covers the research activity performed over the three years, focusing on the role of viscosity value in glazes with Aceto Balsamico di Modena PGI. The goal was to assess the potentiality of viscosity values as control parameter for product stability by contrasting microbial growth.

Glazes with Aceto Balsamico di Modena PGI, considered in this study, are a pool of particular foodstuff product made by mixing Aceto Balsamico di Modena PGI, concentrated or cooked grape must, eventually wine vinegar, and modified starch. Based on previous knowledge and acquired one by intensively studying glaze composition and contamination, both acetic acid bacteria strain and yeasts strains were chosen as model microorganisms to conduct challenge tests for setting a useful viscosity value able to avoid microorganisms spoilage.

A detailed outline of the thesis is reported below.

**Chapter 1** introduces vinegar, and glazes with Aceto Balsamico di Modena PGI. In addition, a brief introduction of *Acetificio Carandini Emilio S.p.A* is given. This chapter gives an overview on glazes with Aceto Balsamico di Modena PGI composition and focusing on stability factors, such as physico-chemical characteristics. However, first, food condiment category and vinegar-based condiment are discussed to contextualize market and legal aspects of glazes with Aceto Balsamico di Modena PGI. Then, physico-chemical composition is deeply elucidated for targeting potential spoilage microorganisms and pointing out critical raw materials and steps of production process.

In **chapter 2** a first study assessing viscosity potential role against microorganisms' spoilage was reported. The physico-chemical and microbiological composition of the glaze without any treatment and glaze with Aceto Balsamico di Modena PGI diluted in ratio of 9:1 and 8:2 with sterile distilled water were described. Then, all the samples were inoculated with type strain of *Gluconoacetobacter johannae* DSM 13595<sup>T</sup>, *Acetobacter pasteurianus* AB0220, *Gluconobacter oxydans* ATCC 621H, *Komagataibacter xylinus* K2G30, and *Komagataeibacter* sp. K1A18. The bacterial growth was tested after incubating the samples at 28°C, 30°C, and 37°C.

In **chapter 3**, the relationship among yeast contamination, viscosity level and glaze with Aceto Balsamico di Modena PGI stability was investigated. *Zygosaccharomyces rouxii* ATCC 56077<sup>T</sup> was used as model microorganisms for glaze contamination. ATCC 56077<sup>T</sup> was inoculated at 10<sup>2</sup> and 10<sup>3</sup> CF/mL in glazes with different viscosity values (1.000, 2.000, 3.000, and 4.000 cps). Glazes with Aceto Balsamico di Modena PGI glazes without any contamination was used as control. Physico-

chemical and microbiological composition were evaluated after 10 and 20 days of incubation, showing low yeast growth in 4.000 cps glazes with Aceto Balsamico di Modena PGI.



## Abstract of the thesis

This PhD thesis was developed in the context of a collaboration between the Department of Life Sciences of the University of Modena and Reggio Emilia and the Acetificio Carandini Emilio S.p.A.

The study concerned the glazes with Aceto Balsamico di Modena PGI, recently developed products, on which there aren't many scientific studies.

The research activity was aimed first at acquiring scientific knowledge on the main factors affecting the microbiological stability of foodstuffs, especially focusing on fruit juices, vinegars and condiments. To this aim, a wide bibliographic research has been done. In fact, different factors, such as type of chemical and physical linkage with water, modifications of the foodstuff consistency, texture and of some microbiological indicators, can contribute to influence the stability of the products.

The glazes with Aceto Balsamico di Modena PGI are a pool of particular foodstuff products made by adding concentrated or cooked grape must and starch (usually modified starch) to Aceto Balsamico di Modena PGI, resulting in a sweet-acid condiment with high density and viscosity. This creamy glaze can be used in a lot of cooking preparation or directly on various dishes. Indeed, the glazes are a "ready to use" products. However, loss of stability has been observed caused by microbial growth; yeasts and bacteria, mainly present in raw material such as grape must, and a low amount of starch which drastically reduces viscosity level contribute to spoilage events. The combination of these two factors lead to undesired fermentations, irreversibly compromising sensorial properties, such as taste and flavour.

To reinforce and confirm some assumptions that have been acquired through literature study, and at the same time to give a starting point about the possible contribution of the viscosity to the microbiological stability, we conducted a first experimental work on glazes.

The importance of the viscosity value (and consequently of the amount of starch addition) has been confirmed by a challenge test that compared the growth of different acetic acid bacteria strains inoculated in the glaze at different dilutions and temperatures. The not diluted samples of the glazes were stable, but when the viscosity decreased (due to the dilution) microbial spoilage arose.

Based on these evidences, the second step of the research aimed at evaluating the role of viscosity in glaze microbial stability. To compare the effect of viscosity, samples with different viscosity levels were inoculated with two different inocula volumes of *Zygosaccharomyces rouxii* ATCC 56077<sup>T</sup>. The final purpose was to find a correlation between the initial contamination, potentially present in

raw materials, and the viscosity level necessary to preserve product stability. To assess the potential role of viscosity in microbiological stability, the analysis of the principal microbial parameters and the most characteristic chemical ones were performed at 0, 10, and 20 days of incubation.

Total mesophilic bacteria and yeasts resulted to be the most present at 20 days of incubation, with molds and acetic acid and lactic acid being significantly less present or even not detected. However, high viscosity glazes deeply reduced microbial spoilage after 10 days of incubation.

Among chemical parameters citric acid, lactic acid and malic acid were the most affected during microbial spoilage in low viscosity samples. Viscosity and tartaric acid were minimally affected.

On the basis of results obtained in this study a number of technological achievements can be transferred in the frame of the industrial production, allowing a more robust knowledge and management of the Glazes with Aceto Balsamico di Modena PGI production.

## Riassunto

Questa tesi di dottorato è stata sviluppata nell'ambito di una collaborazione tra il Dipartimento di Scienze della Vita dell'Università di Modena e Reggio Emilia e l'Acetificio Carandini Emilio S.p.A.

Lo studio ha riguardato le glasse con Aceto Balsamico di Modena IGP, prodotti di recente sviluppo, sui quali non ci sono molti studi scientifici.

L'attività di ricerca è stata finalizzata in primo luogo all'acquisizione di conoscenze scientifiche sui principali fattori che influenzano la stabilità microbiologica degli alimenti, con particolare riferimento a succhi di frutta, aceti e condimenti. A tal fine è stata condotta un'ampia ricerca bibliografica. Infatti, diversi fattori, come il tipo di legame chimico e fisico con l'acqua, le modificazioni della consistenza, della "texture" e di alcuni indicatori microbiologici dell'alimento, possono contribuire ad influenzare la stabilità dei prodotti.

Le glasse con Aceto Balsamico di Modena IGP sono un gruppo di prodotti alimentari particolari realizzati aggiungendo all'Aceto Balsamico di Modena IGP mosto d'uva concentrato o cotto e amido (solitamente amido modificato) per ottenere una sorta di condimento agrodolce ad alta densità e viscosità. Questa glassa cremosa può essere utilizzata in molte preparazioni culinarie o direttamente su vari piatti. Le glasse, infatti, sono un prodotto "pronto all'uso". Tuttavia, è stata osservata una perdita di stabilità causata dalla crescita microbica; lieviti e batteri, presenti principalmente nella materia prima come il mosto d'uva, e una bassa quantità di amido che riduce drasticamente il livello di viscosità contribuiscono ad innescare eventi deterioramento. La combinazione di questi due fattori porta a fermentazioni indesiderate, compromettendo irreversibilmente le proprietà sensoriali, come gusto e sapore.

Per rafforzare e confermare alcune ipotesi acquisite attraverso lo studio della letteratura, e allo stesso tempo per dare uno spunto sul possibile contributo della viscosità alla stabilità microbiologica, abbiamo condotto un primo lavoro sperimentale sulle glasse.

L'importanza del valore della viscosità (e conseguentemente della quantità di amido aggiunta) è stata confermata da un challenge test che ha confrontato la crescita di diversi ceppi di batteri acetici inoculati nella glassa a diverse diluizioni e temperature. I campioni non diluiti delle glasse sono risultati stabili, ma quando la viscosità diminuiva (dovuta alla diluizione) si verificava un deterioramento microbico.

Sulla base di queste evidenze, la seconda fase della ricerca mirava a valutare il ruolo della viscosità nella stabilità microbica della glassa. Per confrontare l'effetto della viscosità, i campioni con diversi livelli di viscosità sono stati inoculati con due diversi volumi di inoculi di *Zygosaccharomyces rouxii*

ATCC 56077<sup>T</sup>. Lo scopo finale era quello di trovare una correlazione tra la contaminazione iniziale, potenzialmente presente nelle materie prime, e il livello di viscosità necessario per preservare la stabilità del prodotto. Per valutare il potenziale ruolo della viscosità nella stabilità microbiologica, è stata eseguita un'analisi dei principali parametri microbici insieme a quelli chimici più caratterizzanti a 0, 10 e 20 giorni di incubazione.

I batteri mesofili totali e i lieviti sono risultati essere i più presenti a 20 giorni di incubazione, con muffe, batteri acetici e lattici significativamente meno presenti o addirittura non rilevati. Tuttavia, le glasse ad alta viscosità hanno registrato una forte riduzione del deterioramento microbico dopo 10 giorni d'incubazione.

Tra i parametri chimici, l'acido citrico, l'acido lattico e l'acido malico sono risultati correlati al deterioramento microbico nelle glasse con Aceto Balsamico di Modena PGI a bassa viscosità.

Sulla base dei risultati ottenuti in questo studio, una serie di conquiste tecnologiche può essere trasferita nell'ambito della produzione industriale, consentendo una più solida conoscenza e gestione della produzione delle Glasse con Aceto Balsamico di Modena IGP.

## **Aims, objectives and strategies**

The aim of this PhD thesis was to investigate the stability of glaze with Aceto Balsamico di Modena PGI and the role of viscosity as a parameter to preserve it against microbial spoilage. Based on previous knowledge, acetic acid bacteria and yeasts were considered as the most common spoilage microorganisms in Balsamic vinegar glaze. To evaluate changes in glaze stability, issues relating physico-chemical features, and the microbiological aspects were set as main parameter to monitor. Indeed, changes in one of these two factors could be related to undesired fermentations which irreversibly compromising product stability.

A challenge test using four genus of acetic acid bacteria was performed on diluted glaze with Aceto Balsamico di Modena PGI incubated at different temperatures This approach was used for evaluating *Zygosaccharomyces rouxii* ATCC 56077<sup>T</sup> spoilage on Balsamic vinegar glazes of four different viscosity. The strategy was to compare the variation of microbiological and physico-chemical parameters on samples of glazes with different viscosity values, in order to find a potential viscosity threshold which could permit to avoid microbial spoilage.

# Chapter 1

## *Introduction*

---

### *1.1 Food Condiments*

Food Condiments are defined as a group of products that cannot be defined as vinegars due to their composition, low acidity or intended use. This category of products may contain a mixture of different types of vinegar (wine, fruit, malt, etc.) along with grape must, fruit juices, sugars, additives, preservatives and food colorings, to achieve the required characteristics. Additionally, the reasons for creating these seasonings are varied: such as, for example, the demand from producers for greater viscosity or a lower acidity or even the need to sell quality products at a lower price, for example, by skipping the long maturation period necessary for aged vinegars. These condiments are considered ready-to-use and marketed in bottles that make their use easy, making them very popular in the catering industry and among consumers. Within the large category of food condiments, the majority of producers of Aceto Balsamico di Modena PGI have developed, over the past two decades, a specific subcategory of vinegar-based condiments, characterized by the use of this famous and distinctive PGI food ingredient.

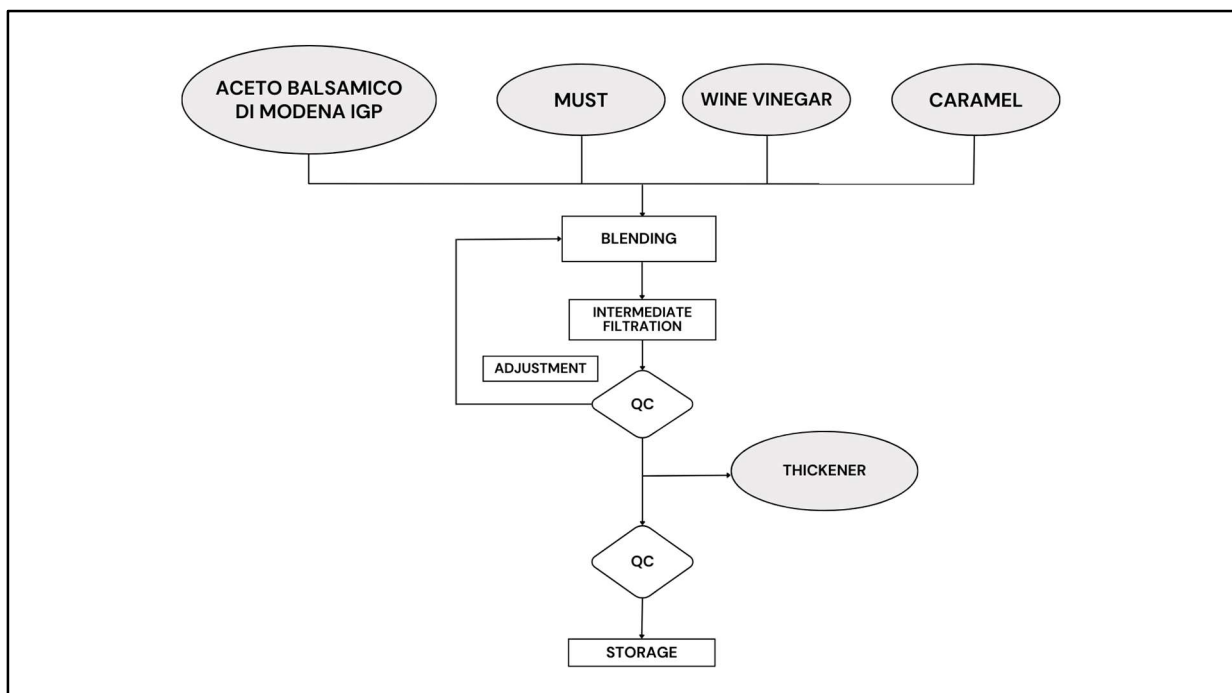
## 1.2 Vinegars towards condiments

The production of vinegar is widespread in many parts of the world and in recent years there has been a growing introduction of new types in the international market, in Europe, North America, Asia, and Latin America. Italy, like Spain and France, bases its high production level on the secondary transformation of wine products. These countries, being major global producers of wine, have managed to develop a well-structured and diversified vinegar industry, also in the commercial proposal that is offered. From this world scenario, a number of products namely condiments are today spread in the market.

## 1.3 The glaze, in particular the Glaze with Aceto Balsamico di Modena PGI

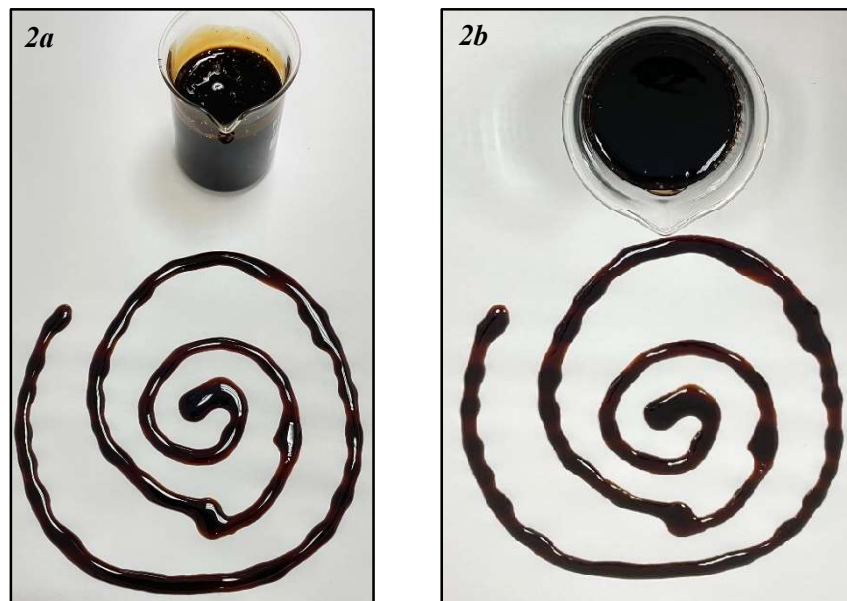
The Glaze with Aceto Balsamico di Modena PGI is a product deriving from the production chain of Aceto Balsamico di Modena PGI, designed to season and flavor any dish.

It's usually composed of a blend of Aceto Balsamico di Modena PGI, with a percentage decided by the company, wine vinegar, concentrated grape must, a thickener which is typically modified starch, xanthan gum or a mixture of both (figure 1). If it's necessary, can be added also caramel and flavourings.



**Figure 1:** Production process flowchart of the glaze with Aceto Balsamico di Modena PGI. The term “QC” stands for quality control

The Glaze with Aceto Balsamico di Modena PGI had, and is still having, great success all over the world, being an innovative product based on raw materials of the Italian tradition, much appreciated by the foreign market. The standard version looks like a black glaze, which recalls the color of its most important component. It is viscous, with a characteristic flavour, with well-balanced and not too accentuated acidity, which goes well with the sweetness brought by the concentrated grape must (figures 2a and 2b).



*Figures 2a and 2b: The typical aspect of the glaze of Aceto Balsamico di Modena PGI*

The organic version of the Glaze with Aceto Balsamico di Modena PGI is also available on the market, produced from certified organic raw materials. The main difference between the organic glaze and the standard one is in the use of xanthan as a thickener instead of modified starch.

Even though, non-organic ingredients are not typically permitted by EU regulation 848/2018 for organic production and related labelling, there are some exception about the use of specific ingredients. Indeed, the regulation does allow the use of specific non-organic ingredients listed in an appendix of approved ingredients, one of which is xanthan gum.

This gives the organic glaze a slightly different consistency, more gelatinous than the standard one, while leaving the taste unchanged.



## ***1.4 Ingredients of Glaze with Aceto Balsamico di Modena PGI***

### *Aceto Balsamico di Modena PGI*

The Aceto Balsamico di Modena IGP, as the consortium mentions, “*is a product obtained from partially fermented and/or cooked and/or concentrated grape musts. The grapes must come exclusively from Lambrusco, Sangiovese, Trebbiano, Albana, Ancellotta, Fontana and Montuni vines*”. On 5 July 2009, Aceto Balsamico di Modena obtained PGI recognition from the European Community, Protected Geographical Indication, and with it a production specification. The latter provides that the must corresponds to at least 20% of the total and that wine vinegar (minimum amount of 10%) and an aliquot of vinegar aged for at least 10 years are added to it.

No other substances can be introduced except a maximum percentage of 2% caramel for colorimetric stabilization according to the regulation. The product obtained must then undergo a "refinement" phase, inside wooden barrels or vats for a minimum period of 60 days before it can be bottled. Vinegar can be subjected to a further period of "aging" which, if it lasts more than three years, gives the Aceto Balsamico di Modena PGI the title of “aged”.

According to the specification, the finished product must have the following characteristics:

- clarity: limpid and brilliant
- colour: intense brown
- smell: characteristic, persistent, intense and delicate, pleasantly acetic, with possible woody notes
- flavor: sweet and sour, balanced, pleasant
- density at 20°C: not less than 1,06 for the refined product
- actual alcoholic strength: not more than 1,5% by volume
- minimum net dry extract: 30 g/L
- minimum total acidity: 6%
- total sulfur dioxide: maximum 100 mg/L
- ashes: minimum 2,5 per mille (g/L)
- reducing sugars: minimum 110 g/L

Packaging for direct consumption can be done in glass, wood and ceramic vessels of various sizes. The specification provides that the assembly of raw materials, the processing, refinement and aging in wooden barrels must take place in the provinces of Modena and Reggio Emilia. Packaging, on the other hand, can also be carried out outside these territories.

Furthermore, the labeling must present the obligatory wording of "Aceto Balsamico di Modena PGI" with also the title "Invecchiato" if it has undergone the aforementioned aging period.

### Wine vinegar

The Article 41 of D.P.R 12.02.1965 of Italian legislation states: "The name "vinegar" or "wine vinegar" is reserved for the product obtained from the acetic fermentation of wines which presents:

- a total acidity expressed in acetic acid not less than 6 grams for hundred milliliters
- a quantity of alcohol not exceeding 1,5 % by volume"

Acetic fermentation is carried out by acetic acid bacteria, i.e. obligate aerobic microorganisms, able to oxidize ethanol and producing acetic acid. These are gram-negative or gram-variable bacteria that belong to the Acetobacteraceae family. The most important genera found in wine are *Acetobacter*, *Gluconobacter*, *Gluconacetobacter* and *Komagataeibacter* (Mamlouk and Gullo 2013).

To produce vinegar normally wines with alcohol grade less than legislative standards or wines with a volatile acidity higher than legal limit are used. The wines must not present alterations of any kind and must have an alcohol content between 5% and 10,5%. If this alcohol content is higher, a blending of the wine with water is allowed in the vinegar factory to bring the product up to production standards. In wines for the production of vinegar it's important to monitor the quantity of sulfur dioxide (SO<sub>2</sub>) normally added in the winemaking processes, as, if present at excessive levels, it causes the inhibition of the acetic acid fermentation. The legislation established a maximum quantity of SO<sub>2</sub> coming from wine of 100mg/L.

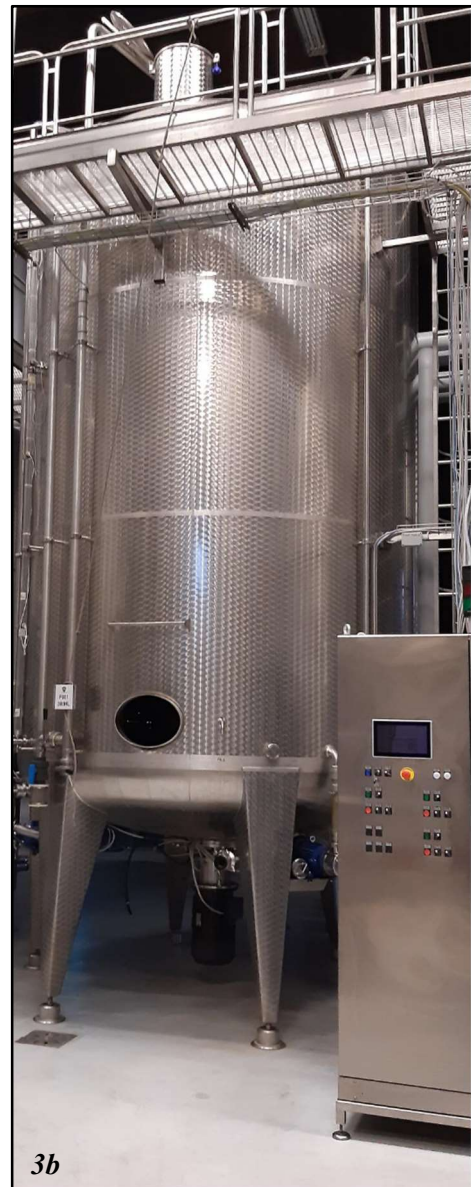
The process of transformation of alcohol into acetic acid can take place through two different methodologies: surface fermentation and submerged fermentation.

In the surface fermentation, the acetic acid bacteria form a biofilm on the surface of the vinegar, of variable thickness and consistency, where the oxidation of the ethanol takes place. However, the extended timing of this process and the absence of controlled conditions could lead to non-conformity of the final product.

The submerged fermentation, on the other hand, allows a quick production of vinegar compliant with legislative standards through efficient control of the aeration, the homogenization of the medium and the temperature. In today's industrial vinegar factories, in fact, stainless bioreactors (Frings) (figures 3a and 3b) are used equipped with turbines for forced aeration, which allows uniform oxygenation of the product, a temperature control system, which keeps it in the optimal production range between

29°C and 32°C (never below 25°C) and loading and unloading pumps. Through these bioreactors the vinegar production times are reduced to 18/30h according to the operating conditions.

The raw vinegar is then clarified and filtered until a clear liquid is obtained, with a light yellow, golden yellow, rosé or red colour, depending on the type of wine being transformed. It has a characteristic and penetrating smell and a sour and spicy taste.



**Figures 3a and 3b:** Bioreactor (Frings) for submerged fermentation (from Acetificio Carandini Emilio S.p.A.)

In addition to water (for a percentage of about 80%) vinegar is composed of a considerable quantity of constituents: the aforementioned acetic acid, tartaric acid, citric acid, malic acid, lactic acid, succinic acid, esters, glycerin, diacetyl, acetone, alcohols, tannins and minerals, such as Fe, Cu, K, Cl<sup>-</sup>, P<sub>2</sub>O<sub>5</sub>.

### Concentrated/Cooked grape must

Grape must is the liquid product obtained after grape treading through mechanical processes such as crushing, pressing or draining. After these physical procedures, the must is dense and very veiled or turbid due to suspended particles deriving from soil, grape skins and stalks. It is then subjected to a filtration process to make it clear. The color of the must can be red, rosé, straw yellow or light yellow depending on the type of grapes used. It is important that the must does not ferment, since an effective alcoholic strength by volume equal to or less than 1% is permitted in the must.

To overcome this problem, the must is stored in special coolers. Furthermore, the use of sulfur dioxide with the aim of inhibiting fermentation is permitted by law.

The must has a complex and very sugary composition (170-230 g/L) which depends on the type of cultivar and the degree of ripeness. The main sugars are glucose and fructose and the other compounds present are: malic acid, tartaric acid, mineral substances, vitamins, aromas, polyphenols and anthocyanins.

Concentrated grape must is obtained by partial dehydration of the grape must using physical methods so that its density is not less than 1,240 g/mL at a temperature of 20°C. The most used method for the concentration of the must is the evaporation by light vacuum treatment at low temperature, between 30°C and 40°C, which allows to avoid the traditional method of concentration at high temperature, which causes changes in the must, including the caramelization of sugars and the formation of furan compounds. To obtain the cooked grape must, the same industrial technique is used, with the only difference that is applied a “thermal flash” at around 100-120°C for 30 seconds in the final part of the process. Alternatively, the refrigeration method can be carried out between -10°C and -13°C; this temperature causes the freezing of the water, which is then removed in the form of ice.

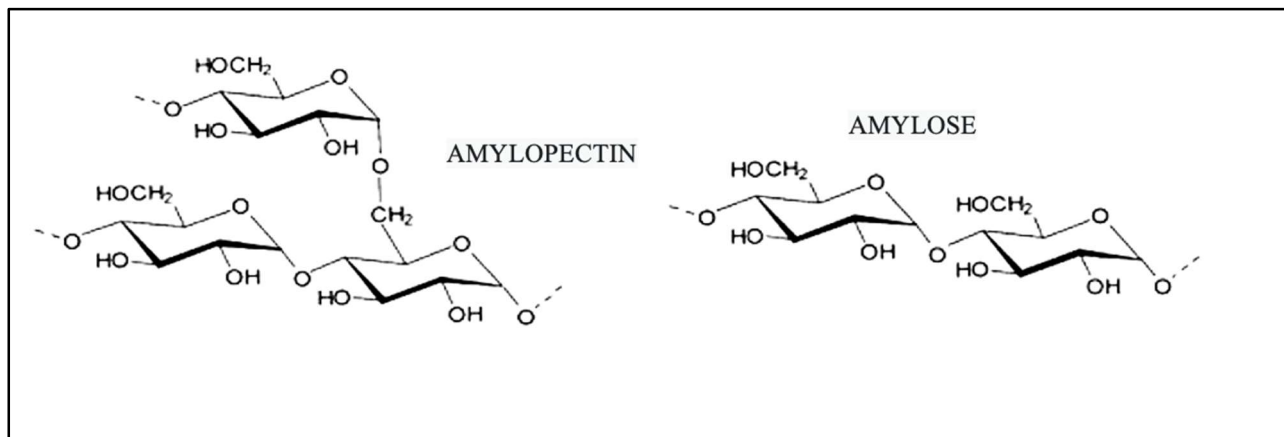
### Modified starch

Modified starch is a very important product for the production of Glaze with Aceto Balsamico di Modena PGI, thanks to its thickening, stabilizing and emulsifying function, which gives the cream the right viscosity and consistency.

Starch is a complex polysaccharide that serves as an energy reserve for plant forms, such as potatoes, corn and maize. After industrial extraction processes, it appears as a white, odorless and tasteless powder. It is a mixture of two different glucose polysaccharides: approximately 80% amylopectin and 20% amylose.

Amylopectin is a branched polysaccharide, consisting of chains of glucose units joined by  $\alpha$  (1→4) glycosidic bonds, which every 24-30 units have branches having  $\alpha$  (1→6) glycosidic bonds (figure 5). Amylose, on the other hand, is made up of unbranched linear chains of glucose linked only by  $\alpha$

(1→4) glycosidic bonds which tend to wind up in a helix, which is made up of six glucose units (figure 4).



*Figure 4: Structure of amylopectin and amylose*

Starch granules are insoluble in cold water due to the hydrogen bonds that join the chains together; however, at temperatures between 55°C and 70°C, these bonds are hydrolysed by water. This temperature is called the "initial gelatinization temperature". This starch gelatinization process is only allowed by heating the aqueous environment with a minimum water concentration of 25%. Under these conditions, the starch granules hydrate and swell and the hydrogen bonds weaken, allowing the release of amylose molecules from the granule, which in solution dissolves in the solvent, thickening and stabilizing the water molecules around it.

This results in increased viscosity and increased structure disorder. Subsequent cooling allows a partial restoration of the ordered structure, which however cannot return to its initial state due to water. Thus, an intermediate structure is established, through a rearrangement of the amylose and amylopectin chains and the exclusion of water.

Modified starches represent a particular class of substances in which the starch molecules are altered in a targeted way, through chemical and physical processes, in order to meet particular needs or requirements of the final production process, such as the thickening, stabilizing and emulsifying function.

Among the main modified starches are:

- Chemically modified starches
- Starches modified with physical treatments
- Starches modified with enzymatic treatments

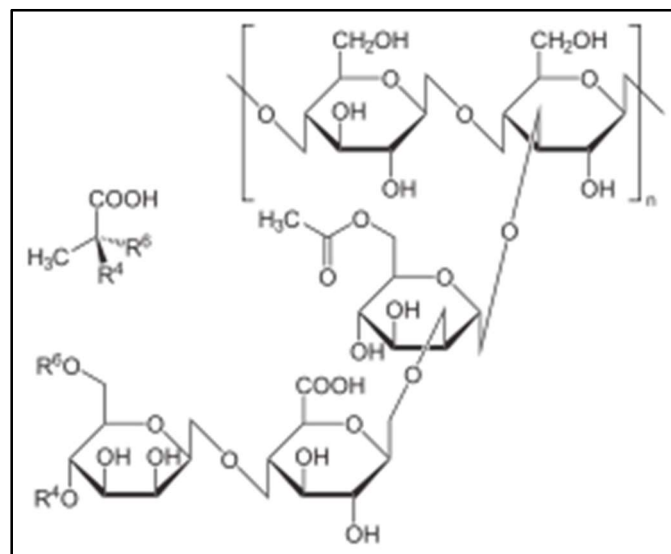
Depending on the structural modifications that are made to the starch, some functional properties may change, such as resistance to overheating or excessive cooling, cold expansion, sterilizability, stability in an acid environment or gelatinization time.

### Xanthan gum

Xanthan gum or "Xanthan" is a polysaccharide normally used as a rheological modifier, i.e. having the ability to modify viscosity. It is mentioned with the wording E415 on the labels of various food products.

As previously mentioned, xanthan is an important ingredient in the organic versions of glazes with Aceto Balsamico di Modena PGI, as it is a thickener considered "Natural" and is permitted by legislation for the production of "Bio" products. It appears as a white/pale yellow powder and is obtained by fermentation process in pure culture of glucose or sucrose by strains of the bacterium *Xanthomonas campestris*, extraction with ethanol or propan-2-ol, dried and ground.

Xanthan gum contains, as the main hexoses, D-glucose and D-mannose, as well as D-glucuronic and pyruvic acids and is separated in the form of sodium, potassium or calcium salts (figure 5).



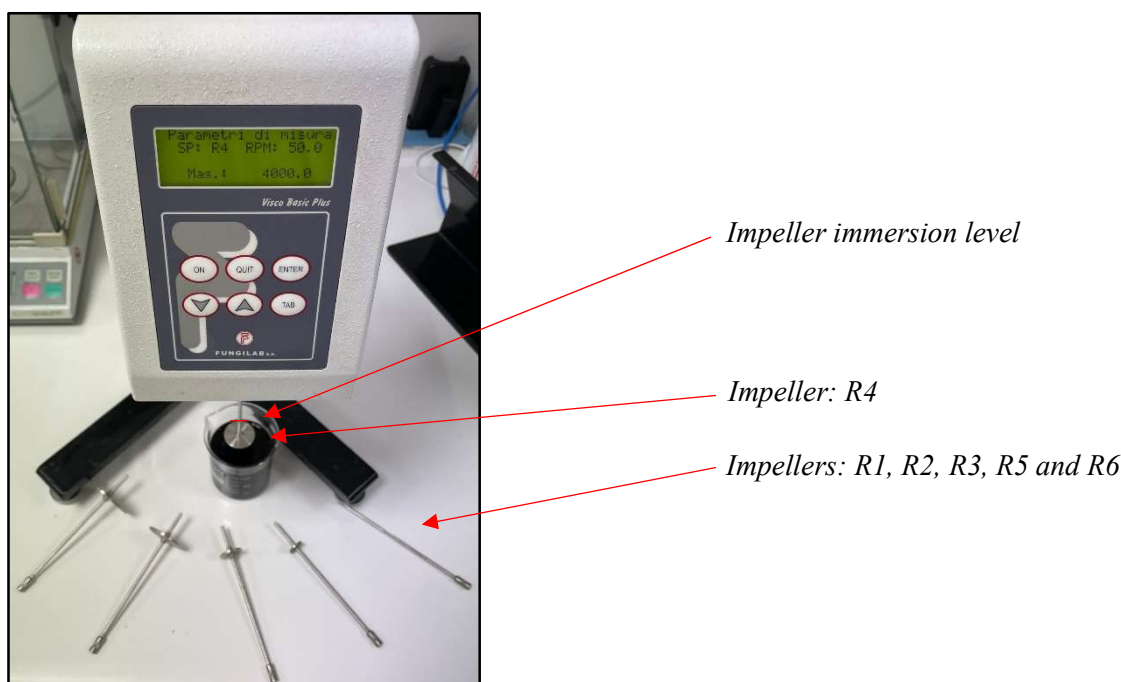
**Figure 5:** Structure of Xanthan Gum

### ***1.5 The stability and safety of the Glaze with Aceto Balsamico di Modena PGI***

In the production of Glazes of Aceto Balsamico di Modena PGI, to ensure a stable, safe and quality product, various physico-chemical parameters are must constantly be monitored, which play an important role in inhibiting the growth of both pathogenic and spoilage microorganisms.

Main stability parameters are total acidity, rotational viscosity, sulfur dioxide, density at 20°C, alcoholic strength, reducing sugars, and pH. The sulfur dioxide is also monitored to ensure the safety for the final consumer of the glazes. Also, the control of the concentration of heavy metals, as Pb, Cu and Zn guarantees the suitability of the consumption of this foodstuff. Some of the parameters just listed will be analyzed in detail below.

By total acidity we mean the sum of the titratable acidities when the pH is brought to 7 by addition of a titrated alkaline solution. Rotational viscosity is a physical quantity that measures the resistance of a fluid to flow. The measurement of this parameter is carried out through the use of a viscometer, i.e. an instrument equipped with a rotor which, turning inside a fluid at a pre-set speed, is able to give a value to the resistance that the latter has to rotation (figure 6).



***Figure 6: Brookfield Rotational Viscosimeter***

With regard to sulfur dioxide, it is a substance with a stabilizing function, which is always monitored, since if it is present in too high concentrations, it can cause health problems as well as making the product unappetizing. Despite this, the legislation does not provide limitations for the product category as can be seen by analyzing Regulation (EU) No. 1129/2011 of the commission of 11 November 2011.

Density at 20°C indicates the ratio between mass and volume of a substance and can be directly measured with a capillary densimeter or calculated from the Brix degree, which is the amount of sugars dissolved in the liquid as determined by a refractometer.

The total dry extract represents the set of non-volatile substances in the product, obtained by removing the volatile substances by heating to 100°C. The net dry extract, on the other hand, is the set of non-volatile substances without reducing sugars.

Heavy metals such as Pb, Zn and Cu are always monitored as they are harmful to health and because they are among the main causes of food safety alerts, as can be seen by analyzing the reports on the "RASFF" portal (Rapid Alert System for Food and Feed), an online platform created by the European Commission in collaboration with "EFSA" (European Food Safety Authority), "ESA" (European Space Agency) and the national health authorities of EU countries, where all the warnings on the safety of food products circulating in the Community are issued.

The alcoholic strength is: "The number of parts by volume of pure alcohol at a temperature of 20°C contained in 100 parts by volume of the product considered at that temperature", and must be declared on the label if higher than 1,2 % Vol as established in Legislative Decree 27 January 1992, n. 109.

Reducing sugars are all those carbohydrates that can be oxidized and have an open chain with an aldehyde or ketone group.

Finally, pH (hydrogen potency) is a measurement that indicates the acidity or basicity of a solution.

A parameter that is not present among those taken into consideration, but which is generally among the pillars of the intrinsic parameters of foods, is the activity of water (aW), i.e. the free water obtained from the ratio between the vapor pressure of the water of the product and the vapor pressure of pure water at the same temperature multiplied by 100. This parameter has a value that varies from 0 to 1, where 1 corresponds to the value of pure water.

The addition of modified starch or xanthan gum to the liquid phase, even though it increases the viscosity value, does not significantly affect free water values. Indeed, no differences are observed in aW value among the liquid phase (before thickener addition) and the final product (ready for packaging). The aW values of Aceto Balsamico di Modena PGI glazes are on average between 0,90 and 0,96.



## ***1.6 Microbiological aspects affecting glazes stability***

Microorganisms, such as bacteria, yeasts and moulds, play a fundamental role in the food industry, as through their specific use together with particular production processes it is possible to obtain foods with a complex physico-chemical profile, with a high nutritional value, such as example vinegar, wine and Parmigiano Reggiano. However, in addition to the useful microorganisms there are, and they are the majority, also those with an altering or pathogenic nature, which can compromise the safety, wholesomeness, stability and therefore the shelf-life of food products. For this reason, in the food industries there is careful monitoring of the latter categories of microorganisms and assiduous work aimed at counteracting their growth and therefore guaranteeing the aspects mentioned above. To do this we rely on intrinsic and extrinsic parameters; the former is characteristic of foods, while the others refer to the environment surrounding them. When the values of these parameters approach the optimal growth values of the microorganisms, the latter tends to have increasingly shorter generation times. Therefore, through a manipulation and a combination of different parameters it is possible to create a hostile environment in the food matrix capable of minimizing the risk of microbial development.

Yeasts and molds are eukaryotic, heterotrophic organisms belonging to the kingdom of *Fungi*. The former are single-celled organisms, widely spread in nature, in the soil and on plants, and reproduce by budding. Generally, they grow in acidic pH conditions (3,5-4) at a temperature between 0 and 45°C with an optimum at 30°C, they grow in aerobic conditions, while they ferment in anaerobic conditions. Some examples of yeasts with a high fermentative metabolism are: *Saccharomyces cerevisiae*, *Saccharomyces ludwigii*, *Schizosaccharomyces pombe*, *Saccharomyces bayanus* or *oviformis*. The potential growth of these yeasts in the glaze with Aceto Balsamico di Modena PGI would compromise the stability of the product, since, following a fermentation process with the consumption of carbohydrates and the consequent formation of alcohol and carbon dioxide, they would cause swelling of the bottles, an excessive alcoholic strength and a reduction of sugars.

Among yeasts, *Zygosaccharomyces rouxii* is often a major spoilage microorganism in high sugared foods, and salad dressing, especially in Glazes with Aceto Balsamico di Modena PGI (Taing et al. 2006; Martorell et al. 2007; Solieri and Giudici, 2008). Main characteristics of *Z. rouxii* are the ability to grow in low a<sub>W</sub> and high osmotic pressure environments (Martorell et al. 2007; Escott et al. 2018). In addition, the species is considered acid-tolerant, being able to grow at pH values of 2,2 (Escott et al. 2018).

*Z. rouxii* has also a high resistance to different chemical compounds used as food preservatives (Frison et al. 2015). Food spoiled from *Z. rouxii* are characterized by rapid reduction in sugar content, and the production of excess gas compromising food-packaging integrity (Escott et al. 2018). Indeed, products contaminated from *Z. rouxii* present in most of the cases “blown” cans or exploding glass bottles.

Molds are multicellular organisms that reproduce through spores. They grow at a pH between 2 and 9,5, with an optimum between 4 and 5, at an optimal temperature between 15°C and 30°C and in aerobic conditions. Some genera of molds such as: *Aspergillum*, *Penicillium*, and *Fusarium* are capable of producing mycotoxins potentially toxic to humans. The potential growth of mold inside the product would cause the formation of a filamentous mass called mycelium, i.e. the typical phenomenon of moulding, with consequent unpleasant alterations to the taste.

Acetic acid bacteria (AAB) are gram negative, catalase positive, strict aerobic bacteria within the family Acetobacteraceae, widespread in sugary, acidic and alcoholic niches. They are known for their ability to partially oxidise carbohydrates and to release the corresponding metabolites (aldehydes, ketones and organic acids) into the surrounding media (Mamlouk and Gullo, 2013). They are beneficial bacteria for vinegar and other fermented beverages (e.g. Kombucha tea). However, in vinegar and condiments they can cause spoilage if the growth in the final products.

Among the current recognized genera, *Acetobacter*, *Gluconobacter*, *Gluconacetobacter* and *Komagataeibacter* include species of interest in the production of fermented products, like vinegars. *Acetobacter* is the first described genus, includes species (i.e. *A. pasteurianus* and *A. aceti*) of interest for the food industry because of the ethanol-oxidizing activity that is beneficial in vinegar production and detrimental in other fermented beverages like wine, beer and cider.

*Gluconacetobacter* and *Komagataeibacter* genera, on the other hand, comprises species with a great variety of physiological abilities including acetic acid and gluconic acid production, nitrogen fixation and bacterial cellulose production. For instance, *Komagataeibacter europaeus* is the main species involved in industrial vinegar production, whereas *K. xylinus* is the model organism for bacterial cellulose production. *Ga. diazotrophicus* instead includes strains of interest for their endophytic activity.

Within *Gluconobacter* genus, *G. oxydans* plays a leading biotechnological role because of its importance for the industrial production of L-ascorbic acid precursors, dihydroxyacetone and gluconic acid (Mamlouk and Gullo, 2013; Sengun and Karabiyikli, 2011).

Lactic acid bacteria are gram positive, catalase negative, and facultative anaerobes; they grow at a pH between 3,2 and 10,5 with an optimum between 5,5 and 6,5 and a temperature between 5 and 53°C (optimal between 30°C and 40°C). The group of lactic acid bacteria includes various genera, such as: *Lactococcus*, *Streptococcus*, *Leuconostoc*, *Weissella*, *Oenococcus* *Pediococcus* and species of lactobacilli. These bacteria are able to ferment glucose and are divided into homofermentative and heterofermentative. The former almost exclusively produces lactic acid, while the latter also produce ethanol, acetic acid and carbon dioxide. Their fermentation process in the glaze with Aceto Balsamico di Modena PGI could cause an increase in acidity and alcoholic strength, a decrease in sugars and an abnormal production of carbon dioxide, which would lead to swelling of the bottles.

Enterococci are gram positive, catalase negative, oxidase negative, non-sporing and facultative anaerobic bacteria. Under the microscope they appear as isolated cocci, in pairs or in chains and some of them are motile. The species belonging to the genus *Enterococcus* grow at a pH between 4,5 and 9,6 and at a temperature between 10°C and 45°C; moreover, they can resist the temperature of 60°C for 30 minutes. These bacteria are also resistant to the animal and human gastrointestinal tract, for this reason they are used as indicators of faecal contamination in food and are an indication of poor sanitation of the production environment.

Coliforms are bacteria belonging to the Enterobacteriaceae family; they are gram negative, non-sporing, facultative anaerobic and catalase positive, generally capable of multiplying at a pH between 4 and 9. Furthermore, they are capable of growing in the presence of bile salts or surfactants and fermenting lactose to produce acids and gases in 48h at temperatures between 30°C and 37°C. Generally, lactose is not present in Glaze with Aceto Balsamico di Modena PGI, except in the case of the variant with Parmigiano Reggiano due to the addition of milk and cheese in the manufacturing process. In this glaze, they could cause an anomalous gas production with consequent swelling of the bottles.

*Escherichia coli* belongs to the above group; it grows in a temperature range between 7 and 46°C, at a pH between 4,3 and 9 with an optimum between 6 and 8 and a minimum required water activity between 0,95 and 0,99. It doesn't have a significant impact on the stability of the products covered by this study, but is monitored for the pathogenic effects it can cause to the body, first of all diarrhea. Staphylococci are pathogenic bacteria, gram positive, anaerobic or facultative anaerobic, lipolytic, resistant up to 7,5% of NaCl and to nitrites; they grow at a temperature between 10°C and 45°C and at a pH between 4 and 9, with an optimum between 7 and 7,5. From a metabolic point of view, they

grow aerobically and ferment glucose anaerobically, forming acetic acid and lactic acid. Their potential growth in the Glaze with Aceto Balsamico di Modena PGI would cause an increase in acidity, but above all a danger to human health because they are the main causes of inflammation and suppuration.

Species of the genus *Salmonella* belong to the family Enterobacteriaceae. These are gram negative, catalase positive, oxidase negative and facultative anaerobic pathogenic bacteria. They grow at a temperature between 6 and 45°C, at a pH range between 4 and 9 with an optimum between 7 and 7,5 and at a minimum water activity between 0,93 and 0,95. Their presence is monitored as they are pathogenic bacteria, causing diarrhea, vomiting, abdominal pain and high fever.

*Listeria* is a gram positive, catalase positive, oxidase positive, facultative anaerobic genus, resistant up to 25% of NaCl, asporigen, pathogenic bacterium belonging to the Corynebacteriaceae family; it develops at temperatures between 1 and 45°C, at a pH between 4,8 and 9, with an optimum at 7, and at a minimum water activity of 0,93. *Listeria monocytogenes* is monitored for its pathogenicity as a cause of meningitis, miscarriages, perinatal septicemia and flu-like symptoms.

Finally, *Bacillus cereus* is a gram positive, facultative anaerobic pathogenic bacterium; it develops at a temperature between 30°C and 55°C and at a pH between 5 and 9,5 with an optimum between 6 and 7. It is also monitored because it causes gastroenteritis.

### ***1.7 Company description***

Acetificio Carandini Emilio S.p.A is located in Via per Formigine 54/A, 41051 Castelnuovo Rangone (Modena).

Thanks to its factories, equipped with cutting-edge systems and one of the best production capacities in the sector that allows it to feed the huge worldwide demand for Aceto Balsamico di Modena PGI and its derivative products as vinegar-based condiments and glazes.



**Figure 7:** *Acetificio Carandini Emilio S.p.A. main site*

In terms of its historical aspect, it is worth remembering that in Modena, since 1641, the Carandini surname held a prestigious role in the production of Aceto Balsamico di Modena PGI, using the traditional methodology.

The vinegar company has two different production sites: the main one, located in Castelnuovo Rangone (figure 7), where most of the production processes such as mixing condiments and glazes, packaging and bottling products for sale together with the most of the physico-chemical analysis.

The other sites is located in the Reggio Emilia countryside, next to Scandiano (figure 8).



*Figure 8: Acetificio Carandini Emilio S.p.A. Scandiano site*

This site is dedicated to the process of turning wine into vinegar (wine acetification) (figure 9), as well as to the storage of most of the raw materials used for the production of vinegar-based condiments and for blending different types of Balsamic Vinegar of Modena PGI (figure 10).



*Figure 9: Bioreactor (Frings) acetification area (Scandiano site)*



*Figure 10: Raw materials external storage steel tanks (Scandiano site)*

The site also houses the production heart of Aceto Balsamico di Modena PGI, with over 200 wooden barrels of varying capacities (from 220L to 120,000L) where this precious vinegar is refined and aged in accordance with PGI production disciplinary (figures 12a, 12b and 12c).



**Figures 11a, 11b, 11c:** Aceto Balsamico di Modena PGI storage area (Scandiano site) in order from left to right: 220L barrique and 120-150hl barrels. (c) Aceto Balsamico di Modena PGI storage area (Scandiano site) - 1.200hl vat

In addition to producing Aceto Balsamico di Modena PGI, the company also offers a wide range of products:

- Wine vinegars and apple cider vinegars
- Glazes with Aceto Balsamico di Modena PGI (mixtures of musts, wine vinegar and Aceto Balsamico di Modena PGI thickened with modified starch in various flavors) (figure 12)
- White glazes (mixtures of musts and wine vinegar thickened with modified starch in various flavors)
- Condiments with Aceto Balsamico di Modena PGI (Aceto Balsamico di Modena PGI mixed with wine vinegar and must in different proportions)
- Condiments and glazes based on apple cider vinegar

All of these types listed above are also available in the certified organic version.



Acetificio Carandini Emilio S.p.A., in its main site in Castelnuovo also has a little production of Aceto Balsamico Tradizionale di Modena D.O.P. Additionally, the company has developed three unique products in recent years:

- Aceto Balsamico di Modena PGI, made with ISO 22005 certified raw materials sourced exclusively from companies in the Modena province.
- Glaze with Aceto Balsamico di Modena PGI and Parmigiano Reggiano DOP (unique in the world).
- 100% organic glaze with Aceto Balsamico di Modena PGI, created using a recently discovered organic modified starch.

*Figure 12: Carandini Glaze with Aceto Balsamico di Modena PGI*

Now, we present some numbers about Acetificio Carandini Emilio S.p.A.:

- Steel tanks storage capacity: more than 9.000.000 liters.
- Wood storage capacity for Aceto Balsamico di Modena PGI: about 3.000.000 liters.
- Aceto Balsamico Tradizionale di Modena DOP batteries capacity: about 9.000 liters.
- Products bottled from all tipologies in 2022: about 9.000.000 liters.

As already mentioned, at the headquarters, there is a laboratory where a series of analyses are carried out for each production phase, aimed at controlling all parameters of interest to ensure the consumer a safe and quality product.

In addition, the modern filtration system guarantees excellent product stability, avoiding contaminations from foreign bodies that can form, for example, during the aging phases of the vinegar inside wooden barrels.

The bottling lines are fully automated and allow a production speed of close to 16.000 bottles/hour. Over the years, the Carandini vinegar factory has become an increasingly synonymous brand of quality and has managed to combine modernity and tradition, bringing typical and ancient-tasting products to the international market, but obtained with increasingly innovative and technological methods. In fact, Acetificio Carandini Emilio S.p.A. has acquired several product and system certifications related to supply chains and quality system, such as:

- PGI certification for the production of Aceto Balsamico di Modena PGI.
- DOP certification for the production of Aceto Balsamico Tradizionale di Modena DOP.
- 848/2018 certification for the production of organic products.
- JAS certification for the production of organic products according to the Japanese standard.
- Naturland standard certification of organic products.
- Halal standard certification.
- ISO 22005:2008 certification for the production of Aceto Balsamico di Modena PGI with Modena grapes.
- ISO 9001:2015 quality system certification.
- BRC quality system certification.
- IFS quality system certification.
- SMETA 4 pillars quality system certification.
- FSMA quality system certification.
- FDA-FSVP quality system certification for USA export



## REFERENCES

“D.Lgs. 27 gennaio 1992, n. 109: Attuazione delle direttive (CEE) n. 395/89 e (CEE) n. 396/89, concernenti l'etichettatura, la presentazione e la pubblicità dei prodotti alimentari.” Gazzetta Ufficiale n. 39 del 17 febbraio 1992 - Supplemento Ordinario.

“Disciplinare di produzione Aceto Balsamico di Modena IGP”.

A.I.S Associazione Italiana Sommelier, “Il Mondo del Sommelier”, Associazione italiana SOM, pubblicazione 11/2004.

Armonini S., “Valutazione della contaminazione da micotossine di prodotti alimentari destinati al consumo umano” Tesi di Laurea AA 2016/17.

Beneventi M., “Determinazione del contenuto di alcuni elementi funzionali in creme di balsamico”, Tesi di Laurea, Modena, AA 2010/11.

Escott, C., Del Fresno, J.M., Loira, I., Morata, A., Suárez-Lepe, J.A. (2018) *Zygosaccharomyces rouxii*: control strategies and applications in food and winemaking. *Fermentation*, 4(3), 69. doi: 10.3390/fermentation4030069

Ferretti M.M., “Determinazione delle caratteristiche fenotipiche e genotipiche di ceppi di *Staphylococcus* in relazione a meticillino- e vancomicina-resistenza”, tesi di laurea, AA 2010/11

Formigaro A., “Studio di sistemi di viscosizzazione di gel idroalcolici con polimeri di origine naturale per la formazione di Gel Igienizzante mani”, Tesi di Laurea, AA 2009/10

Frisón, L.N., Chiericatti, C.A., Aríngoli, E.E., Basílico, J.C., Basílico, M.Z. (2015). Effect of different sanitizers against *Zygosaccharomyces rouxii*. *J Food Sci Technol*, 52(7), 4619-4624. doi: 10.1007/s13197-014-1471-y

Gullo M., Giannone A., Zanichelli G, La produzione industriale dell'aceto: colture indigene e selezionate, Chirotti Editori, 2018.

Gullo M., Giudici P., *Microbiologia Enologica*, “I batteri acetici”, Capitolo/Saggio, 2014

<http://www.agraria.org/viticultura-enologia/analisi-aciditatotale.htm>

<http://www.agraria.org/viticultura-enologia/analisi-estrattosecco.htm>

<http://www.federica.unina.it/agraria/microbiologia-degli-alimenti/batteri-lattici-ecologia-fisiologia-e-tassonomia/>

[https://ec.europa.eu/food/safety/rasff\\_en](https://ec.europa.eu/food/safety/rasff_en)

<https://scoiattolorampante.wordpress.com/2013/12/01/corso-microbiologia-alimentare-enterococchi/>

<https://www.carandini.it/en/company>

<https://www.consorziobalsamico.it/aceto-balsamico-di-modena/come-si-produce/>

Mamlouk, D. & Gullo, M., 2013. Acetic acid bacteria: physiology and carbon sources oxidation. *Indian J. Microbiol.*, Volume 53, p. 377:384.

Martorell, P., Stratford, M., Steels, H., Fernández-Espinar, M.T., Querol, A. (2007). Physiological characterization of spoilage strains of *Zygosaccharomyces bailii* and *Zygosaccharomyces rouxii* isolated from high sugar environments. *Int J Food Microbiol*, 114(2), 234-242. doi: 10.1016/j.ijfoodmicro.2006.09.014

Martorelli P., “Fattori di stabilità delle creme di Aceto Balsamico”, Tesi di Laurea, Reggio Emilia, AA 2017/18

Pompei C., “Operazioni unitarie della tecnologia alimentare”, Casa Editrice Ambrosiana.

Prampolini S., “Parametri chimici e qualità di alimenti: la crema di balsamico”, Tesi di Laurea, Modena, AA 2010/11.

REGULATION (EU) 2018/848 OF THE EUROPEAN PARLIAMENT AND OF THE COUNCIL of 30 May 2018 on organic production and labelling of organic products and repealing Council Regulation (EC) No 834/2007.

REGULATION (EU) No 1129/2011 of 11 November 2011 amending Annex II to Regulation (EC) No 1333/2008 of the European Parliament and of the Council by establishing a Union list of food additives.

Sengun, I.Y., Karabiyikli, S. (2011). Importance of acetic acid bacteria in food industry. *Food Control* 22, 647-656.

Taing, O & Taing, K. (2006). Production of malic and succinic acids by sugar-tolerant yeast *Zygosaccharomyces rouxii*. *Eur Food Res Technol*, 224,343-347

Unioncamere Puglia, Mosto d'uva concentrato, Studio di mercato, Edizione 2009.

Vecchi L., "Determinazione del contenuto di Pb, Cd e Mn nel prodotto Crema di Balsamico, Modena, AA 2010/11.

## Chapter 2

### *First insight into glazes stability*

---

#### **2.1 Introduction**

Worldwide the use of vinegars dates back to very ancient time and today a number of vinegar-based products are available on the market. Next to vinegars which use is consolidated, there are several products, diversified in term of composition and use, grouped as condiments. Condiments deriving from vinegar include the Glaze with Aceto Balsamico di Modena PGI which composition is very diversified and they are very spread on the market.

In this Chapter AAB strains of the four genera mainly occurring in acidic and sugared substrates (*Acetobacter*, *Gluconobacter*, *Gluconacetobacter* e *Komagataeibacter*) were used to test their ability to growth in glazes at different dilutions and storage temperature.

#### **2.2 Materials and method**

##### **2.2.1 Glaze with Aceto Balsamico di Modena IGP - sample**

The work was carried out on a glaze with Aceto Balsamico di Modena PGI, as a representative sample, supplied by Acetificio Carandini Emilio S.p.A. The sample was labeled in the laboratory, as S20BAS.A.

##### **2.2.2 Analytical determinations (pH, titratable acidity and refractometric degree)**

The pH measurement was carried out using the Crison 2002 pH-meter. Titratable acidity was determined by titration with 1 M NaOH until the solution reached a neutral value. The value was expressed as a percentage obtained from the formula shown below.

**Titratable acidity** =  $(V \text{ NaOH} * M \text{ NaOH} / V \text{ sample} + V \text{ H}_2\text{O}) * \text{ACETIC ACID PM}$

The Brix degree measurement was performed using refractometer (ABBE refractometer).

### ***2.2.3 Enzymatic determinations (Ethanol, acetic acid, sucrose, D-fructose and D glucose)***

Ethanol, acetic acid, sucrose, D-fructose and D-glucose were assayed by enzymatic determination using Megazyme (© 2023 Megazyme Ltd) kits. The following kits were used K-ETOH, K-ACET, K-SUFRG. Analyses were performed following the manufacturer's instructions, and measures conducted by the UV-6300PC refractrometer. Data were expressed as grams per liter.

### ***2.2.4 Microbiological Analyses***

#### ***2.2.4.1 Total Mesophilic Bacterial Load***

Total Mesophilic Bacterial Load was assayed in PCA (*Plate Count Agar*) medium, the non-selective medium was sterilized at 121°C for 15 minutes. An aliquote of PCA medium was poured into sterile Petri dishes. Then, 0,1ml of microbial suspension was inoculated in the plate after decimal serial dilutions - as shown in the figure 13 and, through the spreading technique the inoculum was distributed uniformly using a sterile L-shaped spatula. Finally, the plates were incubated at 30°C for 72 hours. The test was conducted in triplicate.



***Figures 13: Serial dilution of the glaze under a laminar flow hood***

#### ***2.2.4.2 Acetic Acid Bacteria growth***

The medium used to search for AAB was GYC agar, table 1 shows the quantities and components used for the preparation of the medium. The medium was sterilized in an autoclave (121°C/15 min.) and then poured into sterile Petri dishes, the microbial suspension was inoculated and distributed uniformly with the spreading technique by spatula. Finally, the plates were incubated at 28°C for 72 h. GYC broth had the same composition, except for agar that was removed.

**Table 1: GYC composition**

<b>Components</b>	<b>Amount (% wt/v)</b>
Glucose	5,0
Yeast Extract	1,0
CaCO <sub>3</sub>	2,0
Agar (for solid medium)	1,5
Distilled Water	1.000 ml

#### **2.2.4.3 Yeasts growth**

The presence of yeasts was checked on YPDA medium (*yeast extract peptone dextrose agar*), which composition is reported in table 2. The YPDA medium was sterilized at 121°C for 15 minutes; it was poured into sterile Petri dishes, and with the spreading technique by spatula, the microbial suspension is inoculated, after serial decimal dilutions. Finally, the plates were incubated at 28°C for 48h.

**Table 2: YPDA composition**

<b>Components</b>	<b>Amount (% wt/v)</b>
Glucose	2,0
Yeast Extract	1,0
Peptone	1,0
Agar	2,0
Distilled Water	1.000 ml

#### **2.2.5 Bacterial strains used in this study**

Table 3 lists the bacterial strains object of this study, obtained from DSMZ-German (Collection of Microorganisms and Cell Cultures) and UMCC (Unimore Microbial Culture Collection) with their main characteristics.

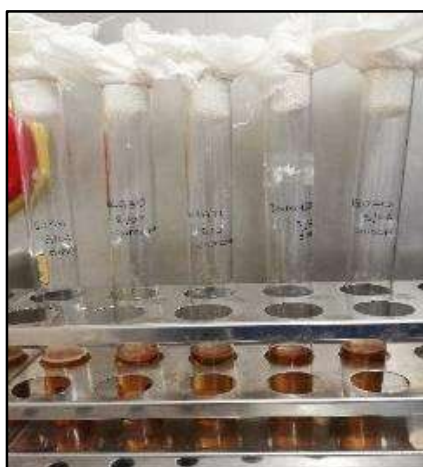
**Table 3: Bacterial strains used in this study**

Strain	Strain designations	Isolation matrix	Culture medium	Cellulose production	References
<i>A. pasteurianus</i>	AB0220 DSM 25273	Wine Vinegar	GYC	-	Gullo et al., 2012
<i>G. oxydans</i>	ATCC 621H UMCC 1754 DSM 2343	Fruit Flower	GYC	-	Gupta et al., 2001
<i>Ga. johannae</i>	DSM13595 <sup>T</sup> NCIB 8036 ATCC 700987	Coffe plants Rizosphere	GYC	+	Fuentes-Ramirez, et al., 2001
<i>K. xylinus</i>	K2G30 UMCC 2756	Kombucha	GYC	+	Gullo et al., 2019
<i>Komagataeibacter sp.</i>	K1A18	Kombucha	GYC	+	La China et al., 2021

### 2.2.5.1 Strain preservation and revitalization

The bacterial strains listed above were stored at a temperature of -80°C in previously prepared vials containing 50% glycerol solution.

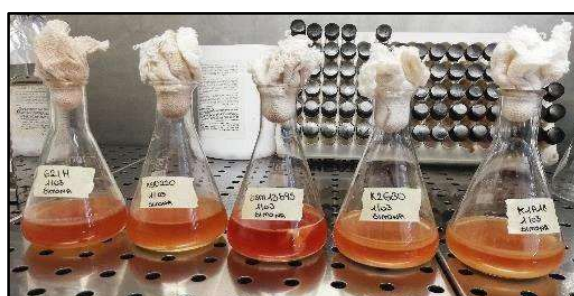
Before use, it is necessary for the bacterial strains to be revitalized. This procedure takes place through the preparation of culture media. In the specific case of this study, 5 bacterial strains were revitalized by initially inoculating 5 ml of medium into tubes and the content of each strain present in the vials, incubated at 28°C for 4 days (figures 14, 15 and 16).



**Figure 14: Revitalization of the strains in a tube**



**Figure 15: Refreshment in 40 ml flasks at 5%**



**Figure 16: Flasks after 4 days of incubation**

After this time, the strains were refreshed in 100 mL flasks with an inoculum equal to 5% of 40 ml (38 ml of GYC medium + 2 ml of strain); they were then incubated at 28°C for 4 days. The culture strains in flasks were refreshed every 4 days until the end of the experimental work.

### **2.2.6 Challenge tests**

Challenge tests are used in microbiology but above all they are essential in the development of new product formulations in food companies.

In this study, the challenge test was set up to verify the stability of the glaze with Aceto Balsamico di Modena IG, by inoculating 5% (v/v) of bacterial culture in the glaze at different dilutions, i.e., in the undiluted product, in a 9:1 dilution, and in an 8:2 dilution; the glaze was diluted with distilled water and filtered. The inocula made in tubes and in triplicates (figure 17) were incubated at three different temperatures: 28°C, 30°C and 37°C for 4 days.



**Figure 17:** Glaze inocula at different dilutions (as is, 9:1, 8:2)

The next step after the tube inoculations was to inoculate 0.1 ml from each tube into the plate and spread the suspension with a sterile L-shaped spatula. This step was done by preparing GYC medium with agar (composition in table 4), used after a sterilization process (121°C for 15 minutes).



*Table 4: GYC agar composition*

Components	Amount (% wt/v)
Glucose	5,0
Yeast Extract	1,0
CaCO <sub>3</sub>	2,0
Agar	1,5
Distilled Water	1.000 ml

For each strain, the work was performed in triplicate, identifying with the letters A, B, C the replicates and with 0, 1 and 2 the corresponding dilution. After plate inoculations, they were placed in different incubators for 5 days at the different temperatures set in the study.

#### **2.2.7. Bacterial cellulose qualitative test**

The qualitative test on bacterial cellulose production was carried out following the procedure used by Navarro (Navarro & Komagata, 1999). The test consists of boiling fragments and films of bacterial cellulose, boiled in water bath tubes for 2 hours, in 4 ml of 5% (v/v) NaOH solution as shown in figure 18. The qualitative test was performed on all plates and tubes that showed growth after inoculation of the product and the bacterial strain.



**Figure 18: Boiling of bacterial cellulose fragments and films**

After boiling, the first step is to remove the liquid fraction from the tubes. After this, the bacterial cellulose, where present, is placed on glass slides and observed. Despite the hostile conditions of the glaze and the thermal stress suffered, the presence of bacterial cellulose was found, especially in some of the strains used in this study.

### ***2.2.8 Quantitative bacterial cellulose test***

The bacterial cellulose films produced in the 8:2 dilution, in tubes, after the inoculation of the *K. xylinus* strain, K2G30, and following incubation at 28°C and 30°C respectively, were dried and weighed following the procedure used by Hwang (Hwang, et al., 1999). The first step was to sample the cellulose in flasks, washing it four times with distilled water. Then, 1M NaOH solution was added and incubated at 80°C for about 20 minutes. At this point, the NaOH was removed and rinsed with distilled water, at least four times at 15 minutes intervals, until neutral pH was reached. Weighing was conducted using an analytical balance (Gibertini E42S, sensitivity 0.1 mg, maximum capacity 240g). All treatments that include the use of NaOH or distilled water must cover the entire bacterial cellulose film. Furthermore, the cellulose film of strain K2G30 of replica C incubated at 30°C, was dried at 20°C with 70% fan speed until constant weight was reached.

### ***2.2.9 Optical microscopy analysis***

Cell morphology and direct observation of samples were carried out using optical microscopy at 100× of magnification, using Nikon Eclipse Ei microscope.

## ***2.3 Results and discussion***

### ***2.3.1 Characterization of the Glaze with Aceto Balsamico di Modena PGI***

Table 5 shows the values of the physico-chemical parameters obtained from analysis carried out on the glaze with Aceto Balsamico di Modena PGI under study. The combination of pH and titratable acidity values are the most important for ensuring the inhibition of microbial growth. A recent study by Lancellotti et al., 2020 reports the chemical characterization of a commercial glaze with Aceto Balsamico di Modena PGI. The sugars, glucose and fructose, represent the main components of the glaze. The presence of these two monosaccharides is known to be due to both the concentrated grape must, which typically contains about 700 g/kg of sugars, and the Aceto Balsamico di Modena, where

the concentration of reducing sugars must not be less than 110 g/kg. L as defined by Regulation (EC) n. 538, 2009.

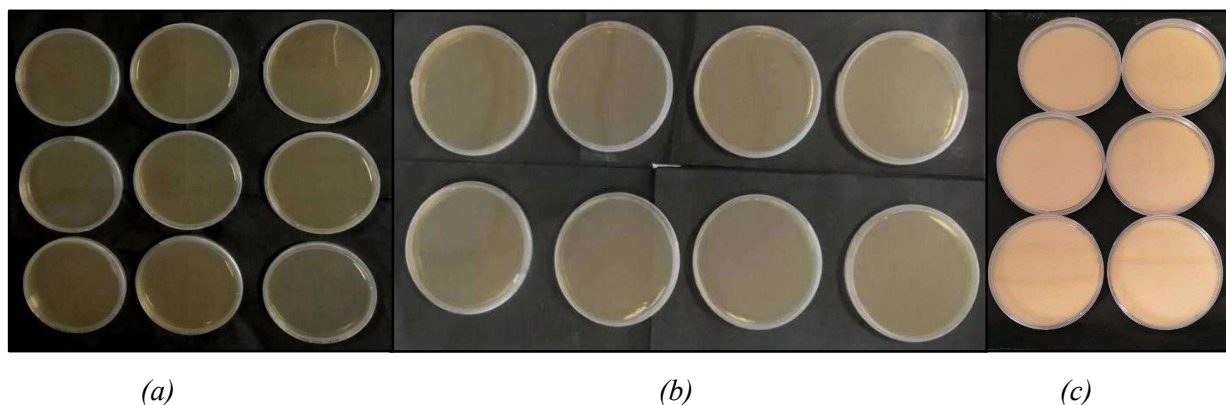
Lancellotti et al., 2020 determined also titratable acidity, which represents the sum of the fixed and volatile acids present in the sample, observing low and similar values among the samples analyzed; also, in our case it resulted to be equal to 2,89%.

**Table 5:** Starting analytical parameters of the glaze (sample S20BAS.A)

<b>Parameter</b>	<b>Mean ± standard deviation</b>
°Brix	44,6 ± 2,04
pH	2,96 ± 0,04
Titratable Acidity (%)	2,89 ± 0,09
Glucose (g/L)	208,29 ± 0,08
Ethanol (g/L)	1,85 ± 0,05
Acetic Acid (g/L)	26,49 ± 0,06

In order to evaluate the microbiological suitability of the glaze with Aceto Balsamico di Modena PGI, samples were first analyzed for the possible presence of bacteria and yeasts. This characterization was aimed at evaluating the microbiological stability of the finished product which could be affected by microbial growth. In fact, the presence of microorganisms in the finished product could potentially harm the preservation of the product. Acetic acid bacteria could be responsible for excessive acidification and production of exopolysaccharides, altering the rheology of the product, whereas, yeast metabolism could also produce unwanted substances, especially CO<sub>2</sub> responsible for bottle swelling or rupture.

As shown in Figure 19 (a, b, and c), research and count of the total mesophilic load, AAB, and yeasts. The total mesophilic load refers to the bacterial load determined for microorganisms that have an optimal growth temperature between 25°C and 40°C. Acetic acid bacteria play an important role in vinegar production, and some are studied as starters in specific fermentations or new formulations of fermented beverages. Given their presence in sugary, acidic and alcoholic environments, their presence in the glaze under study was tested. Finally, the presence yeasts were checked. These microorganisms play an important role in winemaking process and some of them are oxidative because they are harmful to both wines and vinegars as they consume ethanol and oxidize it to CO<sub>2</sub> and H<sub>2</sub>O, negatively impacting product yield. The study of these microorganisms was conducted through microbiological plate tests, and no microbial growth was found in any of the conditions.



**Figure 19:** Petri dishes after incubation for checking total mesophilic load (a), acetic acid bacteria (b) and yeasts (c)

### 2.3.2 Evaluation of AAB growth in glaze at different dilutions and storage temperature by challenge tests

In order to evaluate possible conditions affecting the stability of the glazes, in this study, the behavior of AAB in the glazes with Aceto Balsamico di Modena PGI at different dilutions and incubation temperatures was evaluated. AAB strains belonging to genera *Acetobacter*, *Gluconobacter*, *Gluconacetobacter* and *Komagataeibacter* (table 6) were selected as representative of AAB able to grow in sugared and acetic substrates. Samples at different dilutions were subjected to different thermal stresses.

Acetic acid bacteria include highly versatile organisms capable of producing a wide variety of compounds used in food and beverage, chemical, medical, pharmaceutical and engineering industries. In particular, the genus *Acetobacter* includes species (*A. pasteurianus* and *A. aceti*) of interest to the food industry for their oxidative activity of ethanol, which is positive in the production of vinegar and unfavourable in fermented beverages such as wine, beer, cider (La China, et al., 2018). Within the genus *Gluconobacter*, strains of the species *G. oxydans* play a key biotechnological role as producers of gluconic acid, dihydroxyacetone, precursors of vitamin C (such as L-ascorbic acid) and miglitol (La China et al., 2018). The genera *Gluconacetobacter* and *Komagataeibacter* include highly versatile species with different metabolic abilities, including the production of acetic and gluconic acid, nitrogen fixation, and the production of bacterial cellulose. For example, species belonging to the genus *Komagataeibacter* include *K. europaeus*, which is the main species involved in the industrial production of vinegar (La China et al., 2018); the *K. xylinus* strain, designated K2G30, used in this study is defined as a model organism for bacterial cellulose synthesis. Another strain belonging to this genus, which produced interesting results in the glaze of Aceto Balsamico di Modena PGI, is K1A18, isolated from kombucha but with few references to date. Instead, species

belonging to this genus *Gluconacetobacter*, also a producer of cellulose, includes *Ga. diazotrophicus*, which includes strains of interest for their endophytic activity (Mamlouk & Gullo, 2013).

For the challenge tests, 5% (v/v) inoculum of each culture strain into the glaze was made. Table 6 reports the behavior of the strains after an incubation period of 5 days.

**Table 6:** Microbial growth on plate, X: non-growth; ✓: growth on plate

Strains	10:0 Dilution			9:1 Dilution			8:2 Dilution		
	28°C	30°C	37°C	28°C	30°C	37°C	28°C	30°C	37°C
<b><i>A. pasteurianus</i> AB0220</b>	×	×	×	×	×	✓	✓	✓	✓
<b><i>G. oxydans</i> ATCC 621H</b>	×	×	×	✓	✓	×	✓	✓	×
<b><i>Ga. Johanna</i> DSM 13595<sup>T</sup></b>	×	×	×	×	×	✓	✓	✓	×
<b><i>K. Xylinus</i> K2G30</b>	×	×	×	✓	✓	✓	✓	✓	✓
<b><i>Komagataeibacter</i> sp. K1A18</b>	×	×	✓	✓	✓	✓	✓	✓	✓

The behavior of the different bacterial strains used was highlighted in Table 6 indicating with an (×) the non-growth and with a (✓) the presence in the plate. A first aspect to consider is that in dilution 10:0 no strain showed growth with exception for *K. xylinus*, K1A18, showing adverse conditions for microbial growth. Probably the temperature-dilution combination adopted for the glaze challenge test did not allow the growth of AAB. In this regard, further studies could be aimed at evaluating cellulose as a physical barrier that allows cell growth even in highly selective conditions.

Additionally, the temperature of 37°C, chosen as the limit temperature in the challenge test proved to be critical especially for the strain *G. oxydans*, ATCC 621H, which failed to adapt even in glaze dilution conditions. Another important aspect was the growth of strains belonging to the genus *Komagataeibacter*, respectively K2G30 and K1A18, at different dilutions in the range not only of their ideal temperature but also at 37°C. These results highlight the importance of the correct storage of the product to avoid AAB growth which cause spoilage of the final product. Although AAB are aerobic bacteria, their occurrence and activity is observed in a number of bottled products like wine, vinegar and beer due to the smaller quantities of oxygen in the headspaces of bottles (Bartowsky & Henschke, 2008). Moreover, these data are very important for studying the growth capacity of both strains at this temperature, perhaps on different growth matrices.

### 2.3.2.1 *A. pasteurianus* AB0220

Acetic acid bacteria of the *A. pasteurianus* species are producers of acetic acid, therefore they have the ability to convert ethanol into acetic acid in the presence of oxygen. Strains of *A. pasteurianus* are typically recovered from different kinds of vinegar (Mamlouk & Gullo, 2013). In this study the strain AB0220 (*A. pasteurianus*) was chosen for its ability to grow in vinegars at different composition, also those at high content of grape cooked must (Gullo et al., 2012; Gullo et al., 2009; Gullo et al., 2016). Main phenotypic and technological characteristics of AB0220 strain are reported in tables 7 and 8.

The inoculation of the strain in the 10:0 dilution showed no growth in the plate. However, at a temperature of 28°C (figure 20) it generated small halo visible on the plate, due to its oxidizing activity.

**Table 7:** Phenotypic characteristics of AB0220 (adapted from Gullo et al., 2012)

Feature	AB0220
Shape	Rod
Gram reaction	-
KOH reaction	+
Catalase reaction	+
Production of acetic acid	-
Acetic acid to CO <sub>2</sub> and H <sub>2</sub> O	+
Growth on:	
ethanol	-
methanol	-
Growth on culture media	
GYC	+
AE	-
YPM	+
RAE	-
Frateur medium	+
Modified passmore and Carr medium	+
Carr medium	+
Cellulose production	-
Growth on D-glucose (%)	
20	+
25	+
30	-
Species	<i>Acetobacter pasteurianus</i> Accession number HE650905

**Table 8:** *P* Technological traits of AB0220 (adapted from Gullo et al., 2016)

Medium	Operation mode	Maximum titratable acidity (w/v)
Wine/EtOH 12%	<sup>b</sup> Static after revitalization	6,80 ± 0,34
Wine/EtOH 7.5%	<sup>b</sup> Static fermentation	3,59 ± 0,71
Wine + <sup>a</sup> YE (2%)/EtOH 7.5%	<sup>b</sup> Static fermentation	5,07 ± 0,71
CGM/EtOH 7.5%	<sup>b</sup> Static fermentation	5,35 ± 0,05
SSC-B wine/EtOH 10.5%	<sup>b</sup> Static fermentation	8,90 ± 0,05
SSC-C wine/EtOH 10.5%	<sup>b</sup> Semi-continuous submerged fermentation	9,08 ± 0,09
SSC-E wine-CGM/EtOH 4.60%	<sup>b</sup> Semi-continuous submerged fermentation	6,15 ± 0,06
Wine/EtOH 14.80%	<sup>c</sup> Static fermentation	7,50 ± 0,13
Wine-CGM/EtOH 7.00%	<sup>c</sup> Static fermentation	5,49 ± 0,12

<sup>a</sup>Yeast extract. <sup>b</sup>Experiments conducted at laboratory scale. <sup>c</sup>Experiments conducted at prototype scale.

Particular is its appearance in the C replicate at 30°C (figure 21) where evidently for the cultivation conditions and the inoculation performed in triplicate, *A. pasteurianus* AB0220 was able to visibly grows by creating a halo on the plate.



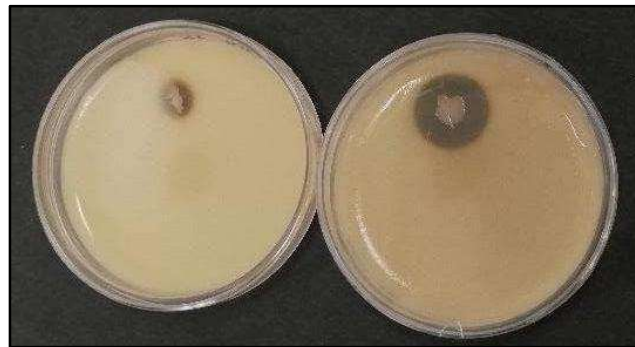
**Figure 20:** *A. pasteurianus*, AB0220, dilution 8:2, replicates A, B, C, 28°C



**Figure 21:** *A. pasteurianus*, AB0220, dilution 8:2, replicates A, B, C, 30°C

### 2.3.2.2 *G. oxydans*, ATCC 621H

Considering *G. oxydans*, ATCC 621H in dilution 9:1 at 30°C (figure 22) no growth was observed due to the harsh conditions of the medium and dilution. On the other hand, the strain showed growth at dilution 8:2, both at 28°C and 30°C where it was able to adapt well to the environment, growing on the plate (figures 23 and 24). This result can be interpreted as the strain's ability to adapt when the viscosity of the glaze decreases with increasing dilution.



**Figure 22:** *G. oxydans*, ATCC 621H, from left 9:1 dilution and 8:2 dilution replica A, 28°C



**Figure 23:** *G. oxydans*, ATCC 621H, 9:1 dilution, replicates A, B, C, 30°C



**Figure 24:** *G. oxydans*, ATCC 621H, 8:2 dilution, replicates A, B, C, 30°C



### 2.3.2.3 *Ga. johannae*, DSM 13595<sup>T</sup>

As can be seen in figure 25, at the dilution of 10:0, no presence of cellulose was found and neither bacterial growth by the *Ga. johannae* DSM 13595T strain even when it was subjected to its optimal growth temperature, perhaps because it was hindered by the high viscosity of the glaze. On the other hand, at the same temperature but at a different dilution (figure 26), it produced bacterial cellulose in the form of fragments, not well distinguishable from the plate but well visible after qualitative test. This is a further confirmation of the difficulty of bacteria to growth on this complex matrix, probably correlated with the viscosity that characterizes it.



**Figure 25:** *Ga. johannae*, DSM 13595T, 10:0 dilution, replicates A, B, C, 30°C



**Figure 26:** *Ga. johannae*, DSM 13595T, 8:2 dilution, replicates A, B, C, 30°C

### 2.3.2.4 *Komagataeibacter* sp., K1A18

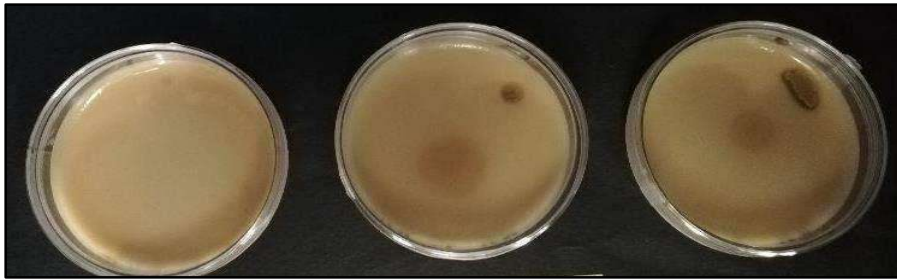
The inoculation of this strain in the starting product with high viscosity, allowed to obtain different results at the three defined temperatures; as for the other strains, in the undiluted product (figure 27) it was not able to develop. At 30°C it produced different films of bacterial cellulose (figure 28 and 29), finally, interesting was its development at 37°C (figure 30), a limit temperature for the growth of AAB.



**Figure 27:** *Komagataeibacter* sp., K1A18, 10:0 dilution, replicates A, B, 28°C



**Figure 28:** *Komagataeibacter* sp., K1A18, 9:1 dilution, replicates A, B, C, 30°C



**Figure 29:** *Komagataeibacter* sp., K1A18, 8:2 dilution, replicates A, B, C, 30°C



**Figure 30:** *Komagataeibacter* sp., K1A18, 9:1 dilution, 37°C

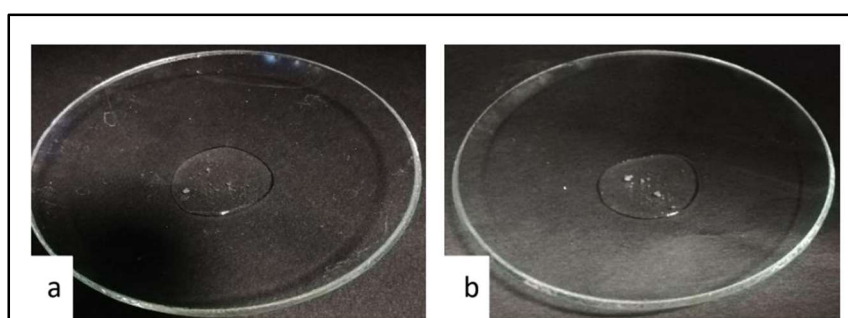
### 2.3.3 Evaluation of bacterial cellulose production

Of the three cellulose-producing strains used in the study, all three were able to synthesize it, although at different yield. Specifically, they are: *Ga. johannae*, with the designation DSM 13595T, *K. xylinus*, with the designation K2G30, isolated from Kombucha tea, a fermented beverage traditionally produced in Asia but native to northern China (Coton et al., 2017). This strain is widely studied for its production of bacterial cellulose, in fact, it is equivalent to  $6,14 \pm 0.02$  g/L when a reduction of pH and gluconic acid formation is observed in glucose-containing / media (Gullo et al., 2019); moreover, this strain will be treated in a separate paragraph because it has been studied under various different physico-chemical aspects in this study; finally, *Komagataeibacter sp.*, with designation K1A18, also isolated from kombucha tea, not yet sequenced and studied in detail but which in liquid state after 6 days is able to produce  $4,0448 \pm 0,0008$  (g/L) of cellulose from film aspect (La China et al., 2021).

**Table 9:** Summarizes a complete overview of cellulose production of the listed strains

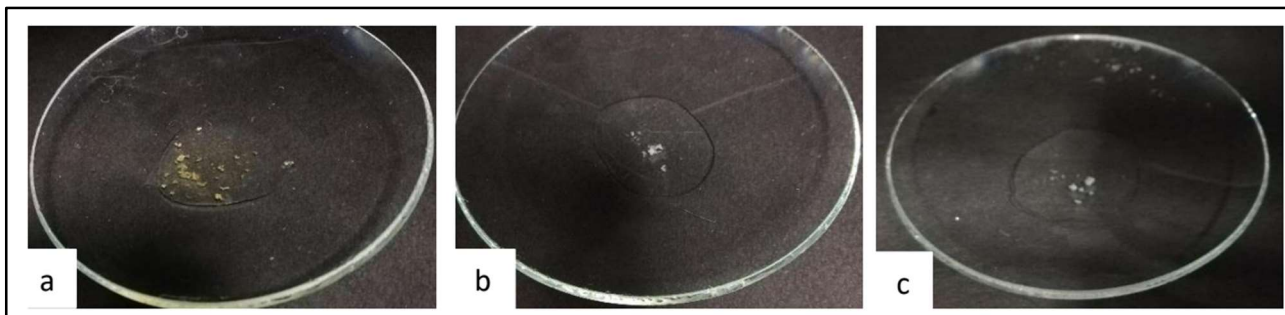
Strains	10:0 Dilution			9:1 Dilution			8:2 Dilution		
	28°C	30°C	37°C	28°C	30°C	37°C	28°C	30°C	37°C
<i>Ga. johannae</i> DSM 13595 <sup>T</sup>	×	×	×	×	×	Fragments	Fragments	Fragments	×
<i>K. xylinus</i> K2G30	×	×	×	Film	Film	×	Film	Film	×
<i>Komagataeibacter</i> <i>sp.</i> K1A18	×	×	×	×	×	Fragments	Fragments	Film	Fragments

As can be seen from table 9, the producer strains used were able to produce different cellulose morphology indicated as film like an uniform and compact layer and fragments. No strains produced cellulose in 10:0 dilution at the three different temperatures probably because the high viscosity which inhibits their growth. While *Komagataeibacter sp.*, K1A18, at 37°C in 9:1 dilution produced cellulose fragments; *Ga. johannae*, DSM 13595T, produced small fragments of bacterial cellulose both in the 9:1 dilution at 37°C and in the 8:2 dilution at 28°C and 30°C (figure 31). The production of bacterial cellulose by *K. xylinus*, K2G30, is analyzed in a separate paragraph.



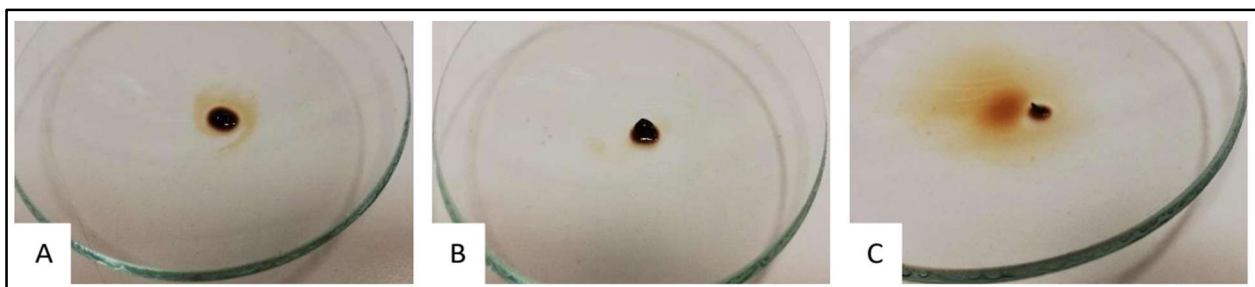
**Figure 31:** DSM 13595T; glaze dilution 9:1 at 37°C (a), glaze dilution 8:2 at 28°C (b)

The cellulose-producing strain, *Komagataeibacter sp.*, K1A18, produced in the plate, in the three replicates (a, b, c) fragments of bacterial cellulose at a temperature of 37°C, as we can see in figure 32, a critical temperature for its growth but important for future studies.



**Figure 32:** 9:1 dilution, at 37°C (a), 8:2 dilution, at 37°C (b), 8:2 dilution, at 28°C (c)

In addition, after 4 days of incubation at 30°C in 8:2 diluted glaze with Aceto Balsamico di Modena PGI, the presence of small exopolysaccharide pellicles was detected in all the biological replicates (figure 33). Bacterial cellulose qualitative test revealed that all the pellicles were bacterial cellulose.



**Figure 33:** A, B, C: 8:2 dilution A, B, C, after incubation at 30°C and after qualitative test

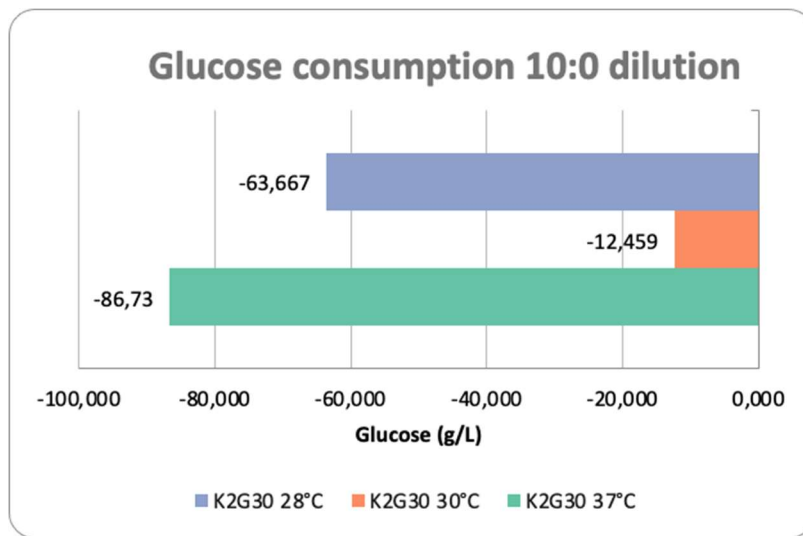
#### **2.3.4 *K. xylinus* K2G30 as a case study in glaze with Aceto Balsamico di Modena PGI**

In order to evaluate the microbial growth in the glaze and observe the changes in its composition due to the potential bacterial activity, the K2G30 strain was used as a case of study. This allowed to obtain information about the critical storage temperature of the product, the variation in the composition of the glaze and its stability. Therefore, in the glaze of Aceto Balsamico di Modena PGI, after the inoculation of the *K. xylinus* strain, K2G30, the parameters: glucose, ethanol and acetic acid were monitored in (g/L). Table 10 shows the respective values of these parameters at our starting conditions, i.e. the glaze at different dilutions (10:0, 8:2, 9:1).

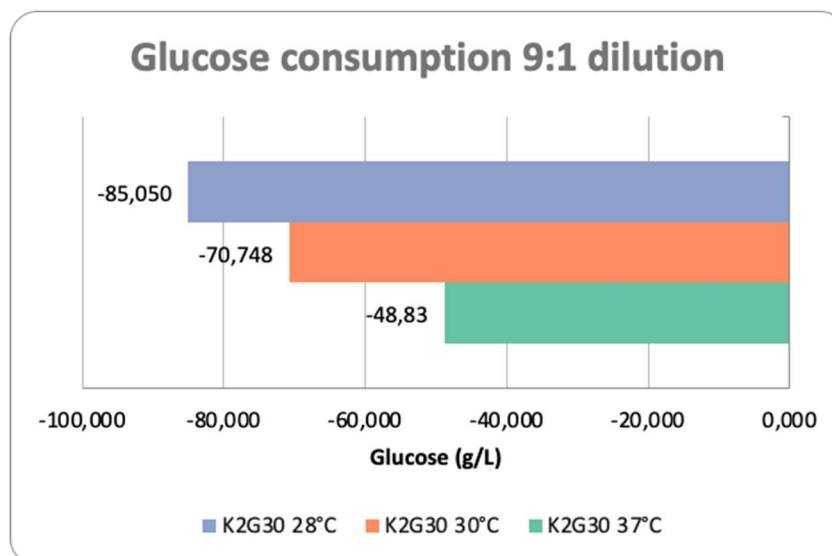
*Table 10: Starting conditions for glucose, ethanol and acetic acid content*

Dilution value	Glucose (g/L)		Ethanol (g/L)		Acetic Acid (g/L)	
	Mean	St. Dev.	Mean	St. Dev.	Mean	St. Dev.
10:00	208,295	1,962	1,854	0,021	26,492	0,612
09:01	187,466	1,766	1,669	0,020	23,843	0,551
08:02	166,636	1,570	1,483	0,017	21,194	0,490

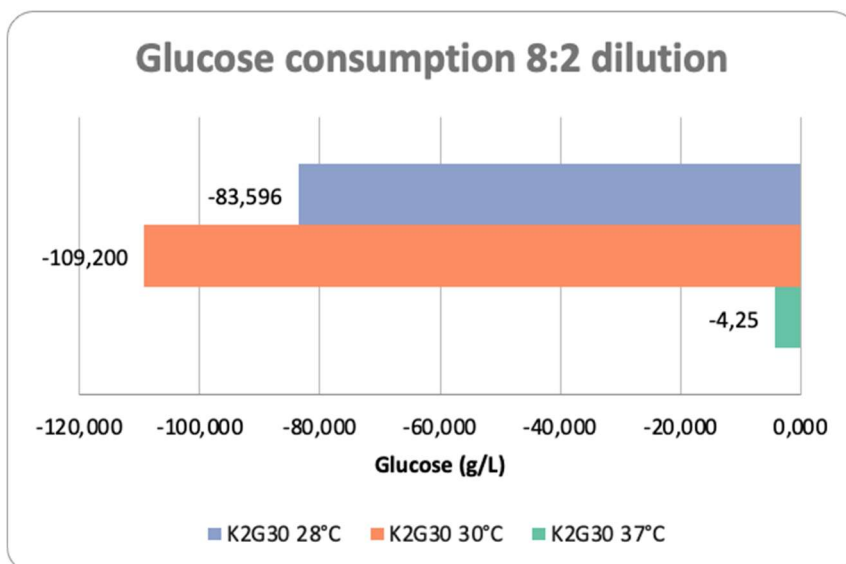
After 5 days, the same parameters reported in table 10 were evaluated to observe any changes and determine if they had altered the matrix.



*Figure 34: Glucose trend at the three temperatures in 10:0 dilution*



*Figure 35: Glucose trend at the three temperatures in 9:1 dilution*



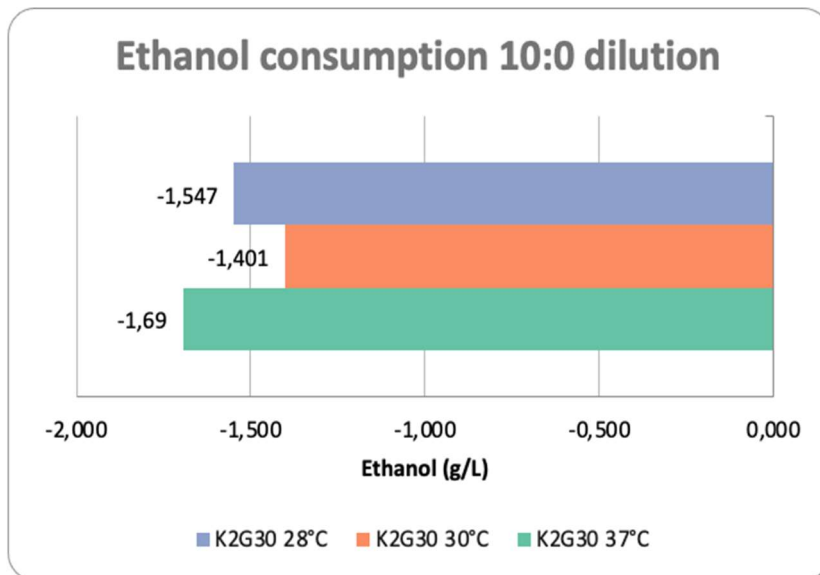
**Figure 36:** Glucose trend at the three temperatures in 8:2 dilution

**Table 11:** Table of percentage change in glucose

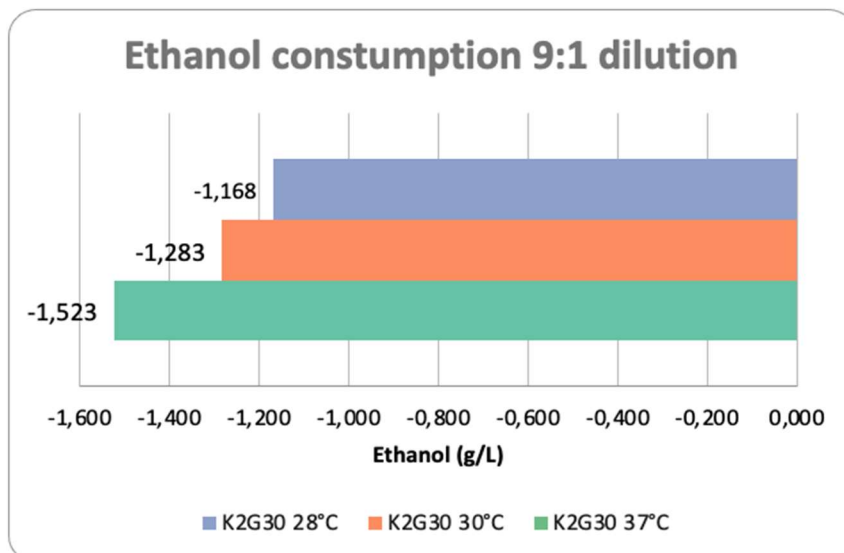
Glucose (%)			
Dilution	K2G30 28°C	K2G30 30°C	K2G30 37°C
10:00	-30,57	-5,98	-41,64
09:01	-45,37	-37,74	-26,05
08:02	-50,17	-65,53	-2,55

Evaluating the glucose consumption from the figures (figures 34, 35 and 36) it is possible to observe that in the 10:0 dilution the glucose consumption by the *K. xylinus* strain, K2G30, at 37°C is higher compared to the consumption at other dilutions. Table 11 highlights that, at the same temperature, the higher is the dilution the higher is the consumption of glucose. Indeed, at 28°C and 30°C K2G30 consumed the highest quantity of glucose in 8:2 glaze with Aceto Balsamico di Modena PGI. On the other hand, at 37°C happen the opposite, with K2G30 consuming more glucose when incubated in the 10:0 diluted glaze with Aceto Balsamico di Modena PGI.

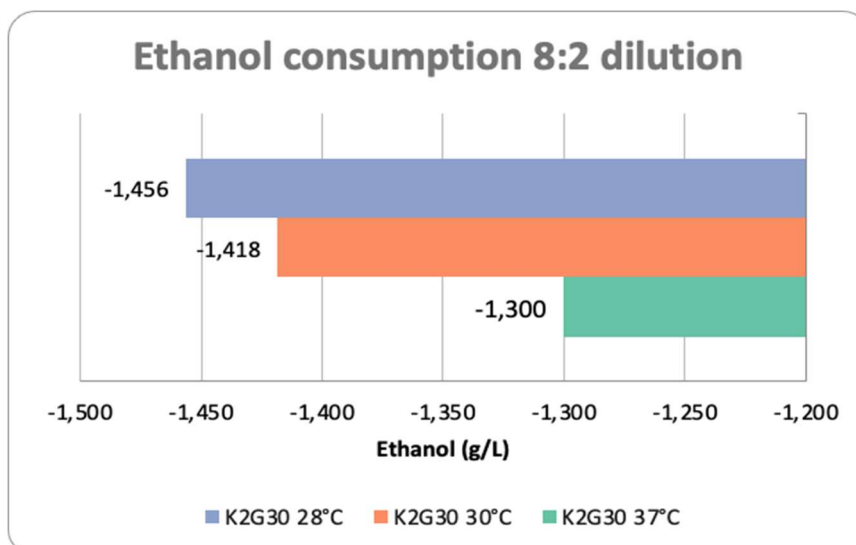
From the figures (figures 37, 38 and 39) on ethanol consumption, a similar trend can be observed at the three different temperatures. A higher consumption is seen at the dilution 10:0 at a temperature of 37°C, as we can see in table 12 where the value is expressed as a percentage.



*Figure 37: Ethanol trend at three temperatures in 10:0 dilution*



*Figure 38: Ethanol trend at three temperatures in 9:1 dilution*



*Figure 39: Ethanol trend at three temperatures in 8:2 dilution*

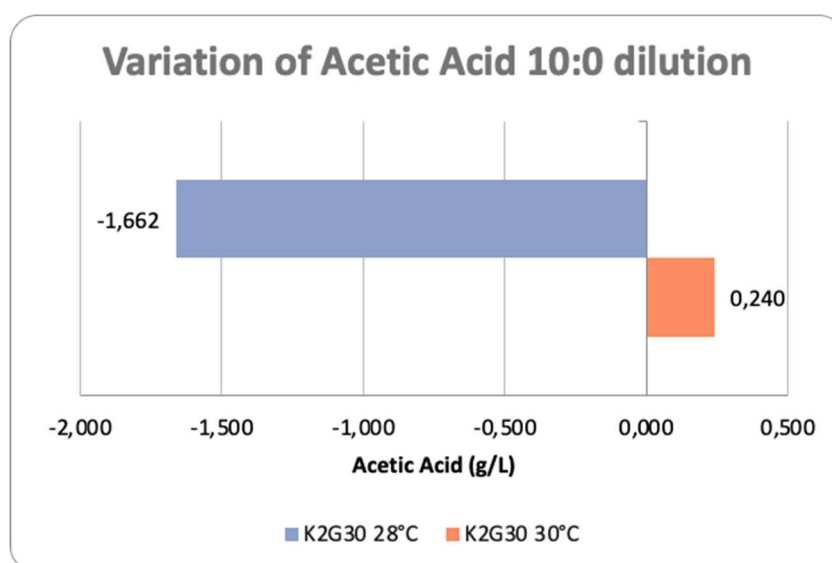
**Table 12:** Table of percentage variation of ethanol

Ethanol (%)			
Dilution	K2G30 28°C	K2G30 30°C	K2G30 37°C
<b>10:00</b>	-83,44	-75,56	-91,39
<b>09:01</b>	-69,98	-76,86	-91,27
<b>08:02</b>	-98,18	-95,62	-87,65

Regarding the trend of acetic acid measured in the product, from table 13 it is possible to observe a higher consumption (expressed in percentage) in dilution 8:2 at 28°C. There is a production of acetic acid in dilution 10:0 when the strain was inoculated and incubated at 30°C; finally, at 37°C, in dilution 10:0 it was not possible to determine the acetic acid content while in the other dilutions an increase in the concentration of acetic acid respectively 2,41% and 6,57% was observed.

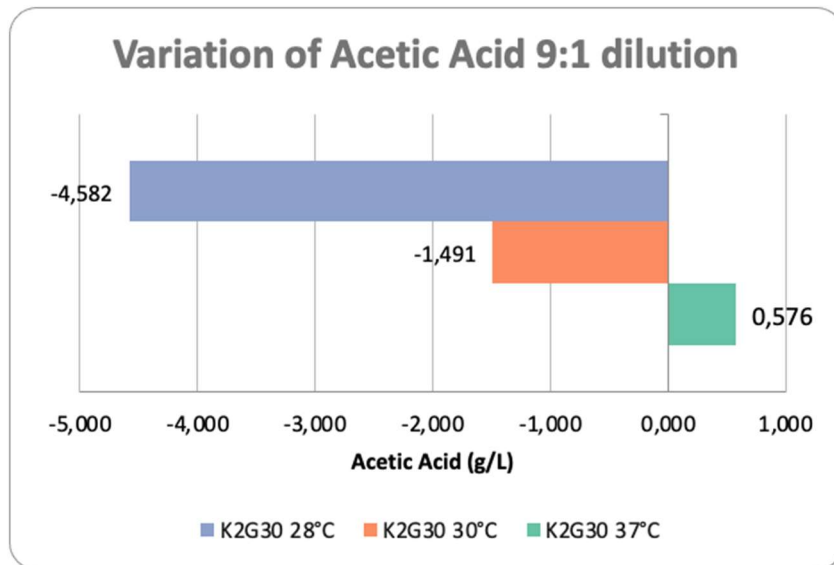
**Table 13:** Table of percentage variation of acetic acid

Acetic acid (%)			
Dilution	K2G30 28°C	K2G30 30°C	K2G30 37°C
<b>10:00</b>	-6,27	+0,9	-
<b>09:01</b>	-19,22	-6,25	+2,41
<b>08:02</b>	-10,29	-12,64	+6,57

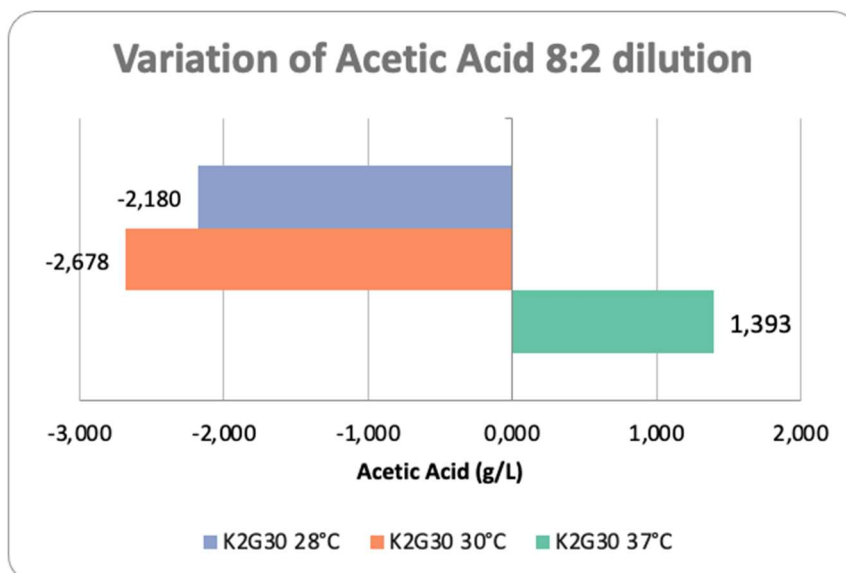


**Figure 40:** Acetic acid trend at three temperatures in 10:0 dilution





**Figure 41:** Acetic acid trend at three temperatures in 9:1 dilution



**Figure 42:** Acetic acid trend at three temperatures in 8:2 dilution

The strain *K. xylinus*, K2G30, that we used has shown cellulose production in dilution 8:2 at 30°C using the different carbon sources present initially in the glaze for growth. Also, from the data obtained during the challenge test, we observed a consumption of ethanol as well as glucose by the strain *K. xylinus*, K2G30, and a reduction in acetic acid concentration (figures 40, 41 and 42).

## 2.4 Conclusion and perspectives

Nowadays, alongside products of consolidated use, there are numerous condiments on the market that are strongly differentiated in terms of composition and use. These, derived from vinegar, are the glazes with Aceto Balsamico di Modena PGI, which are the subject of this study.

This part of the research was aimed at acquiring preliminary information on the microbiological stability of the glaze object of the study. For these reasons, target organism, first considered were AAB and yeasts, which growth is strictly connected to wine, vinegar and cooked must. Since the product and the single ingredients/ intermediary products are all suitable raw materials for AAB growth, strains belonging to the genera *Acetobacter*, *Gluconobacter*, *Gluconacetobacter*, and *Komagataeibacter* were chosen in order to verify their adaptation, growth, and resistance in the culture medium, in the glaze itself, and in the temperature-dilution combination to which it was subjected. From a microbiological point of view, the glaze was found to be stable, but the behavior of the *Komagataeibacter sp.* strain, K1A18, was particularly interesting. When exposed to the critical temperature of 37°C, it was able to produce bacterial cellulose in the form of fragments, as confirmed by qualitative testing. This temperature is a limit for the development of species belonging to the genus *Komagataeibacter*, in fact this and other factors will be the subject of future interesting studies, from a microbiological point of view by testing growth in different matrices. This result highlights the importance of storage condition.

Another result obtained was the study of the *K. xylinus* strain, K2G30, which after inoculation at 5% (v/v) showed production of bacterial cellulose in the form of films at 28°C and 30°C, respectively, using the different carbon sources initially present in the glaze to develop. In addition, the data obtained from the enzymatic analyses performed on it testify that it consumed ethanol and glucose within the matrix.

This study has allowed the evaluation of physico-chemical and biological parameters of interest for the stability of this category of products. Finally, the glaze with Aceto Balsamico di Modena PGI has proven to be an appropriate model for studying AAB under strongly restrictive conditions regarding the compositional parameters of the same.

## REFERENCES

- Bartowsky, E. J., & Henschke, P. A. (2008). Acetic acid bacteria spoilage of bottled red wine -- a review. *International journal of food microbiology*, 125(1), 60–70.
- Coton, M., Pawtowski, A., Taminiau, B., Burgaud, G., Deniel, F., Coulloume-Labarthe, L., ... & Coton, E. (2017). Unraveling microbial ecology of industrial-scale Kombucha fermentations by metabarcoding and culture-based methods. *FEMS microbiology ecology*, 93(5).
- Fuentes-Ramírez, L. E., Bustillos-Cristales, R., Tapia-Hernández, A., Jiménez-Salgado, T., Wang, E. T., Martínez-Romero, E., & Caballero-Mellado, J. (2001). Novel nitrogen-fixing acetic acid bacteria, *Gluconacetobacter johannae* sp. nov. and *Gluconacetobacter azotocaptans* sp. nov., associated with coffee plants. *International Journal of Systematic and Evolutionary Microbiology*, 51(4), 1305-1314.
- Gullo, M., De Vero, L., & Giudici, P. (2009). *Acetobacter pasteurianus* selected strain and acetic acid bacteria species succession in traditional balsamic vinegar. *Applied and Environmental Microbiology*.
- Gullo, M., La China, S., Petroni, G., Di Gregorio, S., & Giudici, P. (2019). Exploring K2G30 genome: a high bacterial cellulose producing strain in glucose and mannitol based media. *Frontiers in microbiology*, 10, 58.
- Gullo, M., Mamlouk, D., De Vero, L., & Giudici, P. (2012). *Acetobacter pasteurianus* strain AB0220: cultivability and phenotypic stability over 9 years of preservation. *Current microbiology*, 64(6), 576-580.
- Gullo, M., Zanichelli, G., Verzelloni, E., Lemmetti, F., & Giudici, P. (2016). Feasible acetic acid fermentations of alcoholic and sugary substrates in combined operation mode. *Process Biochemistry*, 51(9), 1129-1139.
- Gupta, A., Singh, V. K., Qazi, G. N., & Kumar, A. (2001). *Gluconobacter oxydans*: its biotechnological applications. *Journal of molecular microbiology and biotechnology*, 3(3), 445-456.
- Hwang, J. W., Yang, Y. K., Hwang, J. K., Pyun, Y. R., & Kim, Y. S. (1999). Effects of pH and dissolved oxygen on cellulose production by *Acetobacter xylinum* BRC5 in agitated culture. *Journal of Bioscience and Bioengineering*, 88(2), 183-188.

La China, S., De Vero, L., Anguluri, K., Brugnoli, M., Mamlouk, D., & Gullo, M. (2021). Kombucha tea as a reservoir of cellulose producing bacteria: Assessing diversity among Komagataeibacter isolates. *Applied Sciences*, 11(4), 1595.

Lancellotti, L., Ulrici, A., Sighinolfi, S., & Marchetti, A. (2020). Chemical Characterization Of Commercial Glaze with Aceto Balsamico di Modena PGI. *Journal of Food Composition and Analysis*, 94, 103620.

Navarro, R. R. & Komagata, K., 1999. Differentiation of *Gluconacetobacter liquefaciens* and *Gluconacetobacter xylinus* on the basis of DNA base composition, DNA relatedness, and oxidation products from glucose. *J. Gen. Appl. Microbiol.*, Volume 45, p. 7:15.

## Chapter 3

### *Microbiological and chemical study of Glaze with Aceto Balsamico di Modena PGI*

---

#### *3.1 Introduction*

In the world, the use of vinegar dates back to ancient times principally as food preservative. Nowadays vinegar is frequently found on consumer table and in restaurants of different culinary preparations an important ingredient to enhance the taste of dishes. For that reason, alongside the products of consolidated use, there is a great number of diversified condiments on the market that are gaining more and more success. Condiments deriving from vinegar, such as Glazes with Aceto Balsamico di Modena PGI, are the main products on the market. Usually, glazes with Aceto Balsamico di Modena PGI are produced by mixing Aceto Balsamico di Modena PGI, wine vinegar, cooked must with the addition of a thickener (i.e modified starch). Through the addition of modified starch, the Glazes with Aceto Balsamico di Modena PGI increase. The higher is the percentage of modified starch, the thicker is the texture, obtaining a creamy glaze that can be used in several cooking preparation or directly on various dishes. Due to their “ready to eat” purpose, glazes with Aceto Balsamico di Modena PGI are usual stable at high temperature and pressure conditions. However, in combination with high concentration of yeasts (usually present in raw materials) and a low viscosity level due to low modified starch percentage, loss of stability has been observed.

The complexity of the relation between a food product and the spoilage microorganisms is usually determined through a microbial challenge test, which is a study useful to determine the behavior of spoilage microorganisms in a food product. Briefly, in a laboratory scale, a food product is artificially inoculated with specific spoilage microorganisms evaluating if those microorganisms could represent a risk to the quality of the product or the microbial safety.

At industrial level, challenge tests are carried out to ensure compliance with food safety legislation. As is well known, Regulation 2073/2005 (Annex 2) gives producers the possibility to conduct such studies on foods, in particular on ready-to-eat foods, evaluating contamination of *Listeria monocytogenes*, *Salmonella*, *Clostridium botulinum*, and *Bacillus cereus*. In addition, it is possible to carry out tests on other pathogenic and non-pathogenic microorganisms. Besides microbial spoilage, with a challenge test is possible to determine the shelf-life under different storage conditions of the product.

In order to investigate the relationship among yeast contamination, viscosity level and Glaze with Aceto Balsamico di Modena PGI stability, a challenge test was performed. Four glazes with Aceto Balsamico di Modena PGI with different viscosity values (1.000, 2.000, 3.000, and 4.000 cps) were inoculated with  $10^2$  and  $10^3$  CF/ml of *Zygosaccharomyces rouxii* ATCC 56077<sup>T</sup> and incubated at 30°C for 10 and 20 days. Glazes with Aceto Balsamico di Modena PGI without any contamination was used as control.

### 3.2 Materials and method

#### 3.2.1 Samples description and experimental set up

Glazes with Aceto Balsamico di Modena PGI of four different viscosity (1.000, 2.000, 3.000, and 4.000 cps) were prepared (table 14) by *Acetificio Carandini Emilio S.p.A.* In order to standardize glaze with Aceto Balsamico di Modena PGI composition, all the raw materials (grape must, Aceto Balsamico di Modena PGI, wine vinegar, caramel E150D) except for modified starch, were blended and used as the basis for all the four glazes with Aceto Balsamico di Modena PGI. Then the modified starch was added in four concentrations to obtain glazes with Aceto Balsamico di Modena PGI with a viscosity value around of 1.000, 2.000, 3.000, and 4.000 cps. Table 15 reports samples codes and description. All the samples were stored at 4°C until use.

**Table 14:** Glazes with Aceto Balsamico di Modena PGI ingredient and specific formulation. The term “ABM” stands for Aceto Balsamico di Modena

Sample code	Glaze with ABM PGI 1.000 cps	Glaze with ABM PGI 2.000 cps	Glaze with ABM PGI 3.000 cps	Glaze with ABM PGI 4.000 cps
Concentrated grape must	55,03%	54,32%	54,14%	53,79%
Aceto Balsamico di Modena PGI	20,00%	20,00%	20,00%	20,00%
Wine vinegar	19,00%	19,00%	19,00%	19,00%
Caramel E150D	3,00%	3,00%	3,00%	3,00%
Modified starch	2,97%	3,68%	3,86%	4,21%

To evaluate the effect of viscosity level in contrasting microbial growth, three conditions were set for all the glazes with Aceto Balsamico di Modena PGI: zero contamination (ZC) as control,  $10^2$  CF/ml (LC) and  $10^3$  CF/ml (HC) contamination inoculating *Zygosaccharomyces rouxii* ATCC 56077<sup>T</sup>. The latter was chosen based on precious knowledge acquired during company experience. Control and

contaminated samples were incubated at 28°C for 10, and 20 days. All the experiments were performed with three biological replicates.

**Table 15:** Samples coding, description and viscosity value. The term “ABM” stands for Aceto Balsamico di Modena

Sample code	Description	Viscosity
S20Z-1000	Glaze with ABM PGI - zero contamination	1.000 cps
S20L-1000	Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	1.000 cps
S20H-1000	Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	1.000 cps
S20Z-2000	Glaze with ABM PGI - zero contamination	2.000 cps
S20L-2000	Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	2.000 cps
S20H-2000	Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	2.000 cps
S20Z-3000	Glaze with ABM PGI - zero contamination	3.000 cps
S20L-3000	Glaze with ABM PGI- low contamination ( $10^2$ CFU/ml)	3.000 cps
S20H-3000	Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	3.000 cps
S20Z-4000	Glaze with ABM PGI - zero contamination	4.000 cps
S20L-4000	Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	4.000 cps
S20H-4000	Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	4.000 cps

### 3.2.2 Yeast strain and culture conditions

*Z. rouxii* type strain ATCC 56077<sup>T</sup> was obtained from ATCC (American Type Culture Collection) culture collection and cultivated in Yeast Peptone Dextrose (10 g/L yeast extract, 10 g/L peptone, 20 g/L dextrose) (YPD) liquid medium (48h at 28°C). Yeast culture was incubated at 28°C for 48h, repeating inoculation in YPD medium two times. Then, cultures were inoculated in the culture medium to obtain an initial concentration of about  $10^5$  CFU/ml into the first dilution tube. Then, two consecutive serial dilutions were performed prior inoculation in glaze with Aceto Balsamico di Modena PGI. The optical density of the culture in the tube was measured at 600 nm (OD600) with a spectrophotometer UV-6300PC (VWR, United States of America).

### **3.2.3 Microbiological analysis**

Microbiological analyses were performed to assess the microorganisms' growth during incubation period after the glazes opening and inoculation, through total aerobic mesophilic, yeasts, acetic acid bacteria, molds, and lactic acid bacteria counts.

For each sample, serial dilutions were prepared in sterile physiological solution (0,9% w/v). The spread plate method was performed for all the determinations. Total aerobic mesophilic was determined using Plate Count Agar (5 g/L tryptone, 2.5 g/L yeast extract, 1 g/L glucose, 15 g/L agar) (PCA) and the plates were incubated at 30 °C for 48–72 h. Yeasts count was performed in Sabouraud Dextrose Agar (5 g/L pancreatic digest of casein, 5 g/L peptic digest of animal tissue, 40 g/L glucose, 15 g/L agar) (SAB) and the plates were incubated at 28°C for 48–72h.

Molds count was determined using Rose Bengal Agar with chloramphenicol (5 g/L mycological peptone, 1 g/L K<sub>2</sub>HPO<sub>4</sub>, 0,5 g/L MgSO<sub>4</sub>, 10 g/L glucose, 0,05 g/L rose Bengal, 15 g/L agar, 100 mg chloramphenicol) (RBA) and incubating the plates at 25°C for 48h.

AAB and LAB counts were performed in GYC (50 g/L glucose, 10 g/L yeast extract, 15 g/L CaCO<sub>3</sub>, 9 g/L bacteriological agar) and Man Rogosa and Sharpe Agar (10 g/L peptone, 10 g/L beef extract, 5 g/L yeast extract, 20 g/L glucose, 2 g/L K<sub>2</sub>HPO<sub>4</sub>, 5 g/L sodium acetate, 2 g/L ammonium citrate, 0,2 g/L MgSO<sub>4</sub>, 0,05 g/L MnSO<sub>4</sub>, 1 mL Tween ® 80, 15 g/L agar) (MRS) media, respectively. Incubation temperature was set at 28°C for AAB and 30°C for LAB, whereas incubation time was set at 72h for both bacteria.

The media were previously autoclaved at 121°C for 15 min. All experiments were performed in triplicate. The results are expressed as log<sub>10</sub> colony-forming units (CFU) per millilitre of glaze. Media and reagents were purchased from Biolife (Milan, Italy).

### **3.2.4 Physico-chemical analysis**

Physico-chemical analysis were performed by Neutron S.p.A. Water activity (A<sub>w</sub>) at 25°C was determined with 06(S135) rev6 2021) method. Reduction sugars were quantified by 07(S240) 2021 Rev.0 - titrimetric method. Total acidity expressed as g/100 ml of acetic acid was determined through ACIDITA' 2014 Rev.0 - titrimetric method. Relative density at 20°C was determined through a gravimetric method. Alcohol concentration was quantified as %v/v of glaze with Aceto Balsamico di Modena PGI through DM 12/03/1986 SO GU n. 161 14/07/1986 Met V pag 95, OIVMA-AS312-01 Met B R2021 method. Viscosity of glazes with Aceto Balsamico di Modena PGI was determined through VISCOSITA' 2018 Rev.1. Galactose, glucose, sucrose, fructose, lactose, and maltose were



determined with 07(S239) Rev.0 2021 – HPLC-PAD method. Citric acid, lactic acid, malic acid, succinic acid, tartaric acid were determined through AC-ORG Rev.0 2015 - LC-MS/MS method. Ochratoxin A was determined through 03(S130) 2022 Rev.13 – LC-MS/MS method. Finally, pH values were determined with 07(S77) 2012 Rev.1 - potentiometric method.

### **3.3 Results and discussion**

#### **3.3.1 Microbial spoilage**

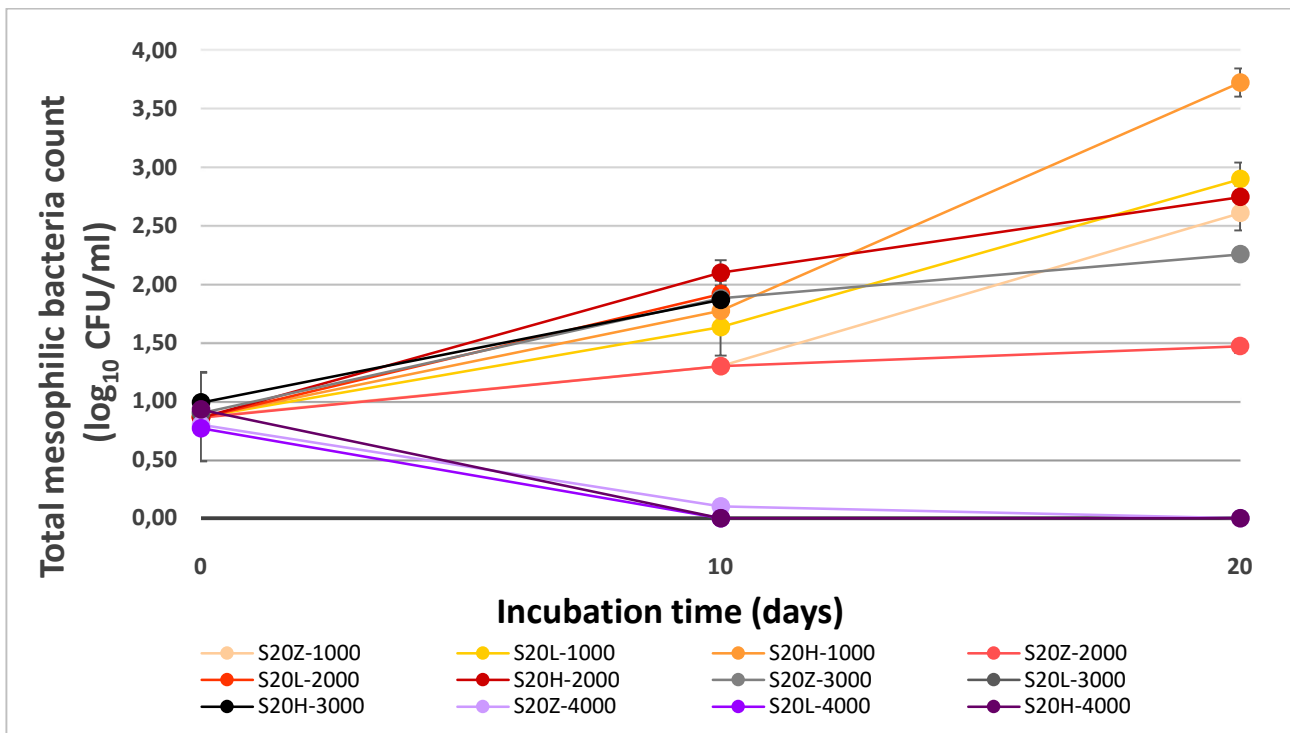
Recent studies show that a large number of yeast species are involved in fermentation including *Zygosaccharomyces bailii*, *Zygosaccharomyces rouxii*, *Zygosaccharomyces pseudorouxii*, *Zygosaccharomyces mellis*, *Zygosaccharomyces bisporus*, *Zygosaccharomyces bisporus*, *Zygosaccharomyces bisporus*, *Zygosaccharomyces bisporus*, *Hanseniaspora valbyensis*, *Hanseniaspora osmophila*, *Candida lactis-condensi*, *Candida stellata*, *Saccharomycodes ludwigii* and *Saccharomyces cerevisiae* (Solieri and Giudici 2008; Garcia et al 2018; Hutchinson et al 2019). Cooked must is a selective medium for yeasts due to its high sugar content and low pH values. In the last two decades, the development of culture-independent techniques, i.e new genomics-based approaches, have clarified the role of yeasts in vinegars and the use of molecular techniques has allowed the characterization of the presence of different strains (Solieri and Giudici 2008).

The spoilage of glazes with Aceto Balsamico di Modena PGI is mainly induced by yeast contamination in must, leading to major physico-chemical changes in the final product and, in the worst cases makes the product as not-edible. Since during glaze production no thermal treatments are performed, the physico-chemical composition of glaze itself play a crucial role in microbiological stability. In this study, we assessed the relevance of glaze final viscosity as factor to control microbial spoilage during storage by inoculating *K. rouxii* ATCC 56077<sup>T</sup> at low ( $10^2$  CFU/ml) and high ( $10^3$  CFU-/ml) contamination levels. *K. rouxii* ATCC 56077<sup>T</sup> was chosen based on literature review, checking yeasts species present as spoiling microorganisms in vinegar, and as micro-flora of vinegars produced with cooked must. Total mesophilic bacteria, yeasts, molds, LAB, and AAB counts were performed.

#### **3.3.2 Total mesophilic count**

The highest total mesophilic count among all glazes with Aceto Balsamico di Modena PGI was observed in S20H-1000, reaching 3.73 log<sub>10</sub> CFU/ml which is lower than the acceptable microbial

growth ( $10^6$  CFU/ml) in food. However, from figure 43 it's evident a straight correlation between viscosity level and the increase in total mesophilic bacteria count. Indeed, we observed a reduction to 0  $\log_{10}$  CFU/ml of TMB in glazes with Aceto Balsamico di Modena PGI at 4.000 cps already after 10 days of incubation. On the other hand, for all the glazes with Aceto Balsamico di Modena PGI at 1.000, 2.000, and 3.000 cps an increase in TMB was observed either after 10 and 20 days of incubation.

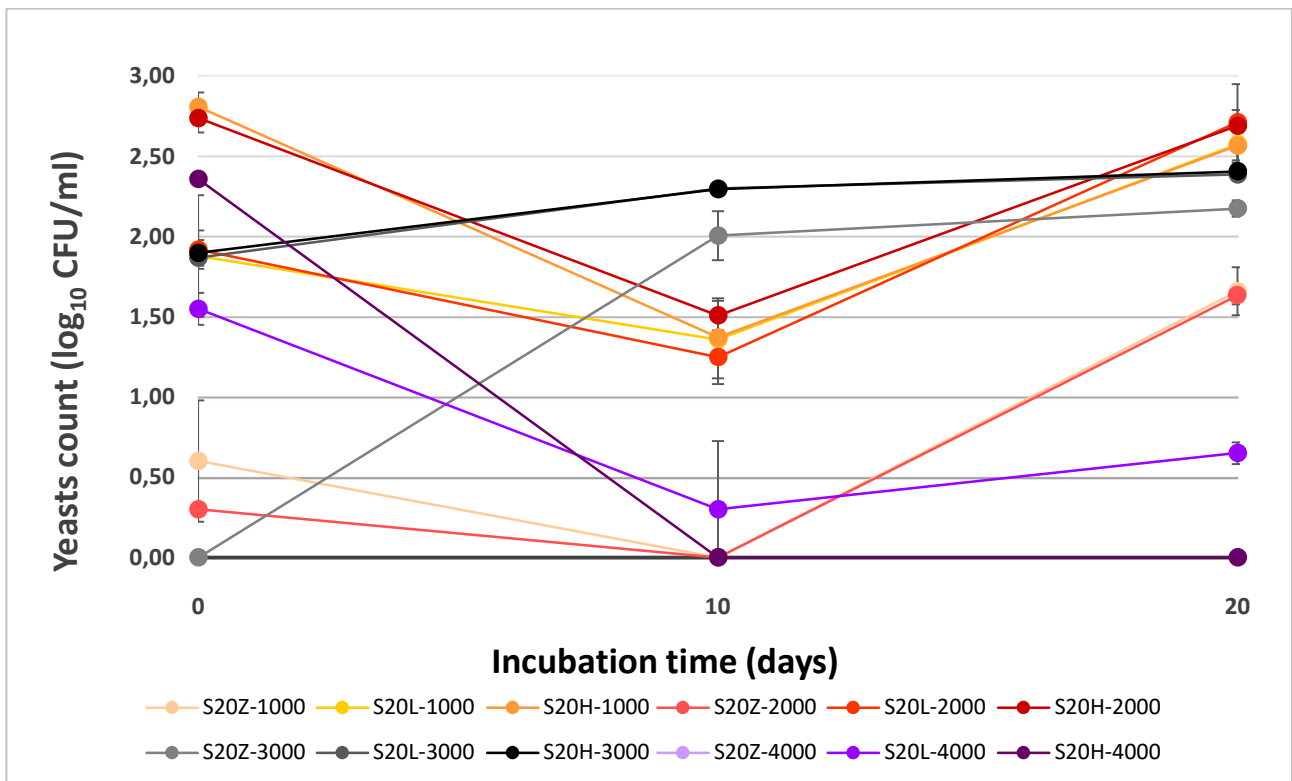


**Figure 43:** Total mesophilic count of four glazes with Aceto Balsamico di Modena PGI at different viscosity value (1.000, 2.000, 3.000, and 4.000 cps) and inoculated with  $10^2$  CFU/ml (S20L) and  $10^3$  CFU/ml (S20H) of *Z. rouxii* ATCC 56077T. Raw glazes with Aceto Balsamico di Modena PGI (S20Z) were used as control. Dots indicate the average total mesophilic count by three replicates. Error bars represent standard deviations

### 3.3.3 Yeasts count

Figure 44 shows changes in yeasts of glazes with Aceto Balsamico di Modena PGI during incubation. Yeasts count decreased sharply at the initial stage but increased slightly from 10 days to 20 days for all the samples of 1.000 and 2.000 cps independently from the contamination level. In 3.000 cps glazes with Aceto Balsamico di Modena PGI with low and high contamination level yeasts count increased moderately over time. On the other hand, we observed a sharp increase in yeasts count for 3.000 cps glaze with Aceto Balsamico di Modena PGI control from the start of incubation until the

first time point. Yeasts count in 4.000 cps glazes with Aceto Balsamico di Modena PGI showed a sharp reduction after 10 days of incubation, reaching 0 log<sub>10</sub> CFU/mL in most of the samples. 4.000 cps glaze with Aceto Balsamico di Modena PGI contaminated with 10<sup>2</sup> of *Z. rouxii* ATCC 56077<sup>T</sup> was the only exception, with yeasts presence being stable from 10 to 20 days of incubation. All the samples presented circular white to cream colonies with regular margins and a convex elevation as showed in figure 45.



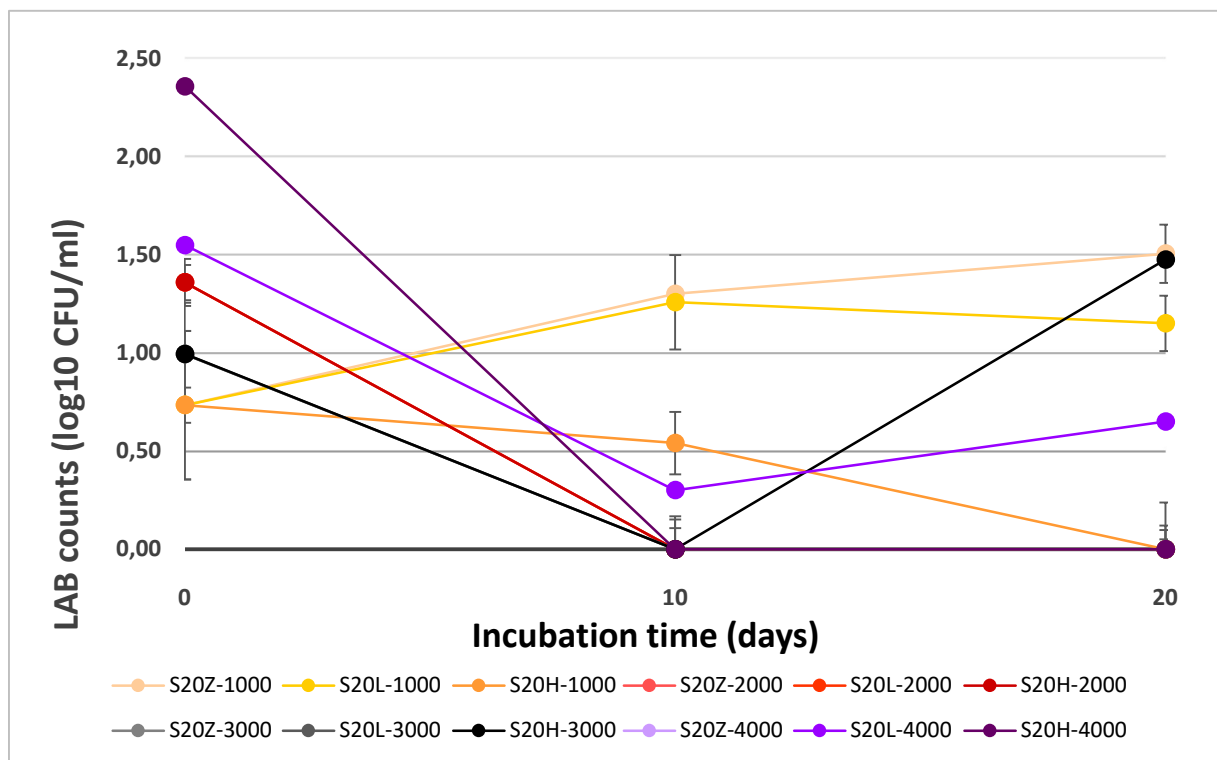
**Figure 44:** Yeasts count of four glazes with Aceto Balsamico di Modena PGI at different viscosity value (1.000, 2.000, 3.000, and 4.000 cps) and inoculated with 10<sup>2</sup> CFU/mL (S20L) and 10<sup>3</sup> CFU/mL (S20H) of *Z. rouxii* ATCC 56077<sup>T</sup>. Raw glazes with Aceto Balsamico di Modena PGI (S20Z) were used as control. Dots indicate the average yeasts count by three replicates. Error bars represent standard deviations



**Figure 45:** Yeast colonies observed in 1000 cps glaze with Aceto Balsamico di Modena PGI inoculated with 10<sup>2</sup> CFU/ml after 48 h of incubation at 30°C

### 3.3.4 Molds, lactic acid bacteria and acetic acid bacteria count

At time 0, very low counts of LAB were observed (figure 46), whereas no presence of molds and AAB was detected in any sample. The latter two microbial groups never showed growth in all the glazes, neither in 1.000 cps glaze. LAB count showed a reduction after 10 days in 2.000, 3.000, 4.000 cps glazes with Aceto Balsamico di Modena PGI, reaching 0 log<sub>10</sub> CFU/ml at 20 days of incubation. On the other hand, 1.000 cps glaze with Aceto Balsamico di Modena PGI revealed an increasing in LAB presence over the 20 days incubation. No AAB and molds growth was observed at 10 days nor at 20 days of incubation.



**Figure 46:** Lactic acid bacteria (LAB) count of four glazes with Aceto Balsamico di Modena PGI at different viscosity value (1.000, 2.000, 3.000, and 4.000 cps) and inoculated with 10<sup>2</sup> CFU/ml (S20L) and 10<sup>3</sup> CFU/ml (S20H) of *Z. rouxii* ATCC 56077T. Raw glazes with Aceto Balsamico di Modena PGI (S20Z) were used as control. Dots indicate the average LAB count by three replicates. Error bars represent standard deviations

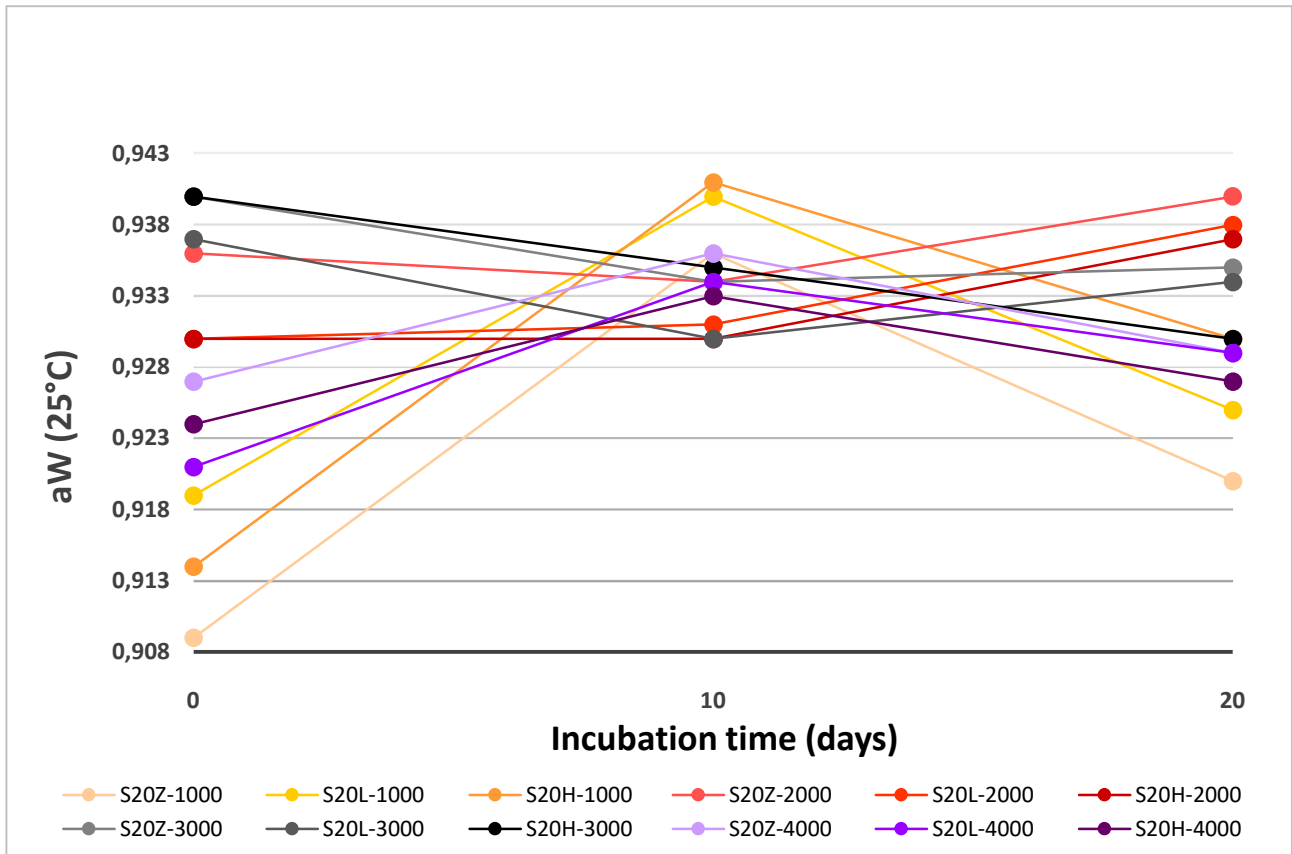
### 3.3.5 Changes in physico-chemical parameter caused by microbial spoilage

#### 3.3.5.1 aW

Balsamic glazes are characterized by high values of water activity (aW), usually above 0,90 (Terpou et al 2019). As Table 16 shows no aW under 0,90 was detected in any samples. A slight increase of aW was observed during incubation time (figure 47).

**Table 16:** aW values of four glazes with Aceto Balsamico di Modena PGI at different viscosity values (1.000, 2.000, 3.000, and 4.000 cps) and inoculated with  $10^2$  CFU/ml (S20B) and  $10^3$  CFU/mL (S20A) of *Z. rouxii* ATCC 56077T. Raw glazes with Aceto Balsamico di Modena PGI (S20Z) were used as control. Values are expressed as average. Measure uncertainty:  $\pm 0,005$ . The term “ABM” stands for Aceto Balsamico di Modena

Sample description	Code	aW (a 25°C)		
		0 days	10 days	20 days
Glaze with ABM PGI - zero contamination	S20Z-1000	0,909	0,936	0,920
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-1000	0,919	0,940	0,925
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-1000	0,914	0,941	0,930
Glaze with ABM PGI - zero contamination	S20Z-2000	0,936	0,934	0,940
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-2000	0,930	0,931	0,938
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-2000	0,930	0,930	0,937
Glaze with ABM PGI - zero contamination	S20Z-3000	0,940	0,934	0,935
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-3000	0,937	0,930	0,934
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-3000	0,940	0,935	0,930
Glaze with ABM PGI - zero contamination	S20Z-4000	0,927	0,936	0,929
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-4000	0,921	0,934	0,929
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-4000	0,924	0,933	0,927



**Figure 47:** *aW* values variation of four glazes with Aceto Balsamico di Modena PGI at different viscosity value (1000, 2000, 3000, and 4000 cps) and inoculated with  $10^2$  CFU/mL (S20B) and  $10^3$  CFU/mL (S20A) of *Z. rouxii* ATCC 56077T. Raw glazes with Aceto Balsamico di Modena PGI (S20Z) were used as control. Dots indicate the mean value on the NEOTRON analysis certificate

Despite having the lowest *aW* values, the lowest viscosity samples (S20-1000) had the highest variability over time. Instead, the S20-4000 samples had a higher initial value of *aW* respect to the S20-1000, but the variation over time is almost null, slightly increasing during incubation. A similar behavior was observed in S20-2000 samples. On the other hand, in S20-3000 was observed a slight reduction after 20 days of incubation, resulting in a final *aW* of 0,93. However, as reported, an *aW* above 0,90 is high enough to permit microbial spoilage during the storage time of a product (Sperber 1983).

Low pH level is fundamental to guarantee microbiological stability, especially for Balsamic glazes. Indeed, a pH level lower than 4 has an adverse effect on the proliferation of pathogenic microorganisms (Sperber 1983; Leistner & Gould 2002).

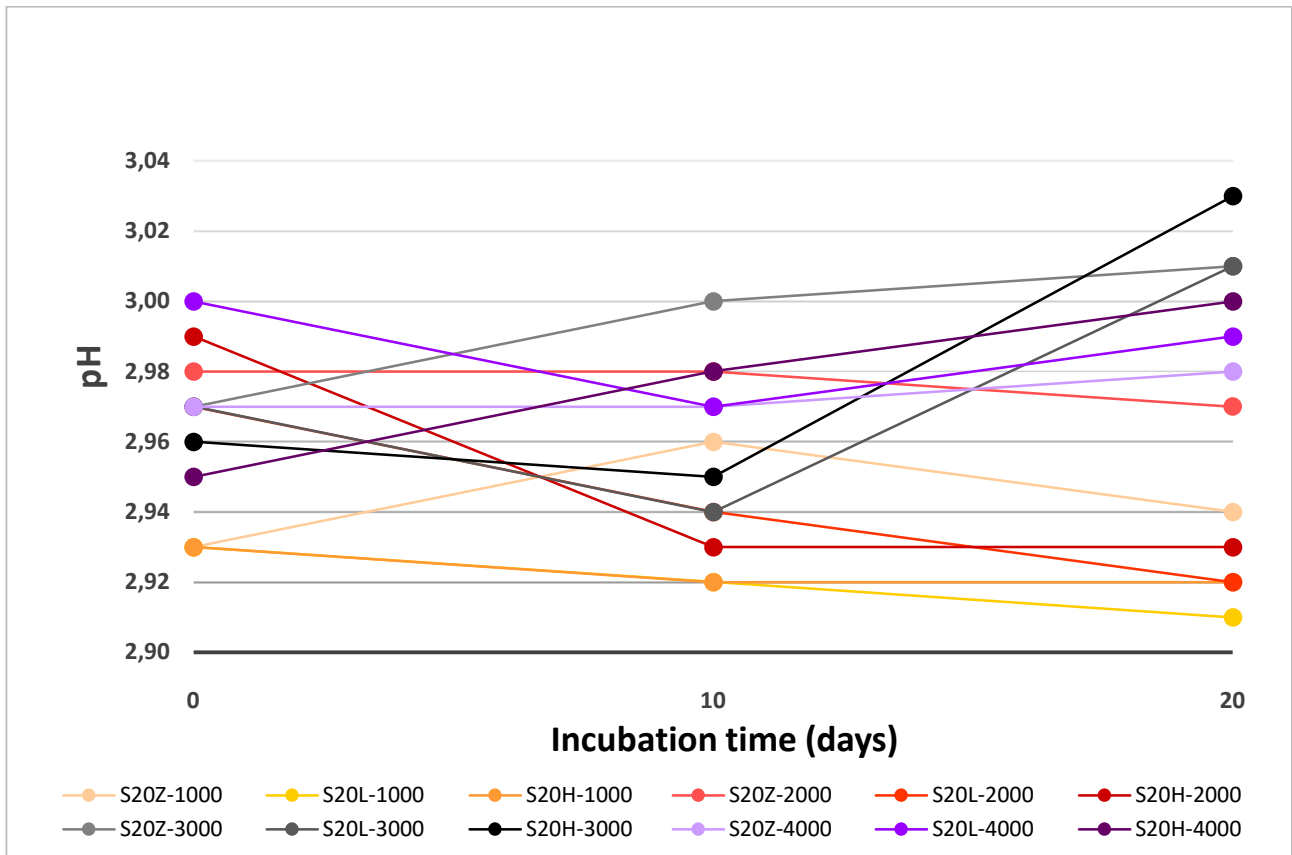
Due to this, it was considered crucial to study the pH trend over time to check for any anomalous variation.

### 3.3.5.2 pH

The analysis confirmed that the pH values of all samples are considerably lower than 4 (table 17 and figure 48) and no increasing trends were observed for any of the samples.

**Table 17:** pH values of four glazes with Aceto Balsamico di Modena PGI at different viscosity values (1.000, 2.000, 3.000, and 4.000 cps) and inoculated with  $10^2$  CFU/mL (S20B) and  $10^3$  CFU/mL (S20A) of *Z. rouxii* ATCC 56077T. Raw glazes with Aceto Balsamico di Modena PGI (S20Z) were used as control. Values are expressed as average. Measure uncertainty:  $\pm 0,04$ . The term “ABM” stands for Aceto Balsamico di Modena

Sample description	Code	pH		
		0 days	10 days	20 days
Glaze with ABM PGI - zero contamination	S20Z-1000	2,93	2,96	2,94
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-1000	2,93	2,92	2,91
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-1000	2,93	2,92	2,92
Glaze with ABM PGI - zero contamination	S20Z-2000	2,98	2,98	2,97
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-2000	2,97	2,94	2,92
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-2000	2,99	2,93	2,93
Glaze with ABM PGI - zero contamination	S20Z-3000	2,97	3,00	3,01
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-3000	2,97	2,94	3,01
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-3000	2,96	2,95	3,03
Glaze with ABM PGI - zero contamination	S20Z-4000	2,97	2,97	2,98
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-4000	3,00	2,97	2,99
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-4000	2,95	2,98	3,00



**Figure 48:** pH values variation of four glazes with Aceto Balsamico di Modena PGI at different viscosity value (1.000, 2.000, 3.000, and 4.000 cps) and inoculated with  $10^2$  CFU/mL (S20B) and  $10^3$  CFU/mL (S20A) of *Z. rouxii* ATCC 56077<sup>T</sup>. Raw glazes with Aceto Balsamico di Modena PGI (S20Z) were used as control. Dots indicate the mean value on the NEOTRON analysis certificate

### 3.3.5.3 Total acidity in acetic acid

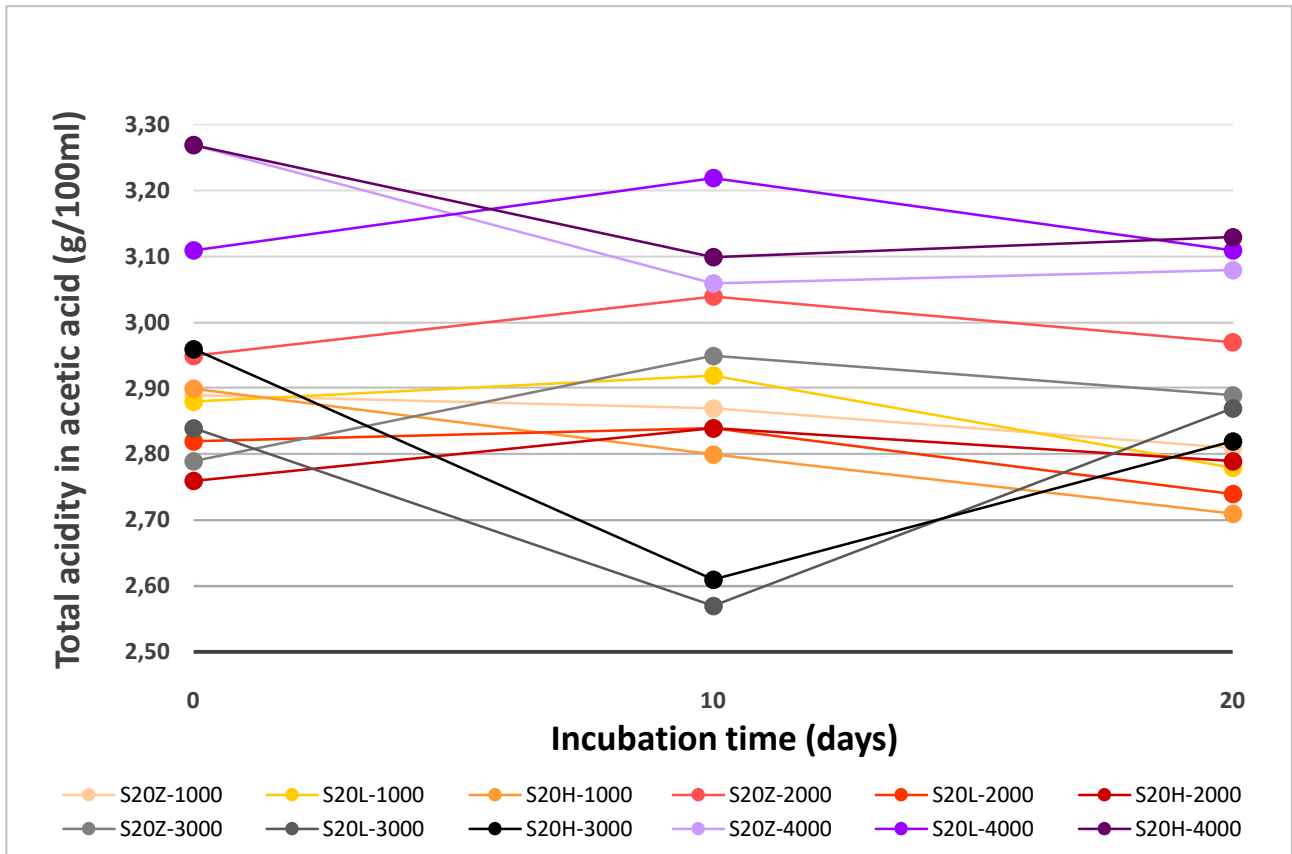
A non-linear trend was observed for total acidity in acetic acid in all the samples. As Figure 49 shows there is no relationship between viscosity values and total acidity. On the other hand, samples with the highest contamination level had the highest reduction in total acidity (table 18), except for S20H-2000.

Considering the uncertainty of the measure ( $\pm 0,06$  g/100ml), the acidity variations can be considered almost null. Due to these considerations seems that viscosity doesn't influence the total acidity variation.



**Table 18:** Total acidity in acetic acid of four glazes with Aceto Balsamico di Modena PGI at different viscosity values (1.000, 2.000, 3.000, and 4.000 cps) and inoculated with  $10^2$  CFU/mL (S20B) and  $10^3$  CFU/mL (S20A) of *Z. rouxii* ATCC 56077<sup>T</sup>. Raw glazes with Aceto Balsamico di Modena PGI (S20Z) were used as control. Values are expressed as average. Measure uncertainty:  $\pm 0,06$ . The term “ABM” stands for Aceto Balsamico di Modena

Sample description	Code	Total acidity in acetic acid (g/100ml)		
		0 days	10 days	20 days
Glaze with ABM PGI - zero contamination	S20Z-1000	2,89	2,87	2,81
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-1000	2,88	2,92	2,78
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-1000	2,90	2,80	2,71
Glaze with ABM PGI - zero contamination	S20Z-2000	2,95	3,04	2,97
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-2000	2,82	2,84	2,74
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-2000	2,76	2,84	2,79
Glaze with ABM PGI - zero contamination	S20Z-3000	2,79	2,95	2,89
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-3000	2,84	2,57	2,87
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-3000	2,96	2,61	2,82
Glaze with ABM PGI - zero contamination	S20Z-4000	3,27	3,06	3,08
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-4000	3,11	3,22	3,11
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-4000	3,27	3,10	3,13



**Figure 49:** Total acidity in acetic acid variation of four glazes with Aceto Balsamico di Modena PGI at different viscosity values (1.000, 2.000, 3.000, and 4.000 cps) and inoculated with  $10^2$  CFU/mL (S20B) and  $10^3$  CFU/mL (S20A) of *Z. rouxii* ATCC 56077<sup>T</sup>. Raw glazes with Aceto Balsamico di Modena PGI (S20Z) were used as control. Dots indicate the mean value on the NEOTRON analysis certificate

### 3.3.5.4 Relative density at 20°C

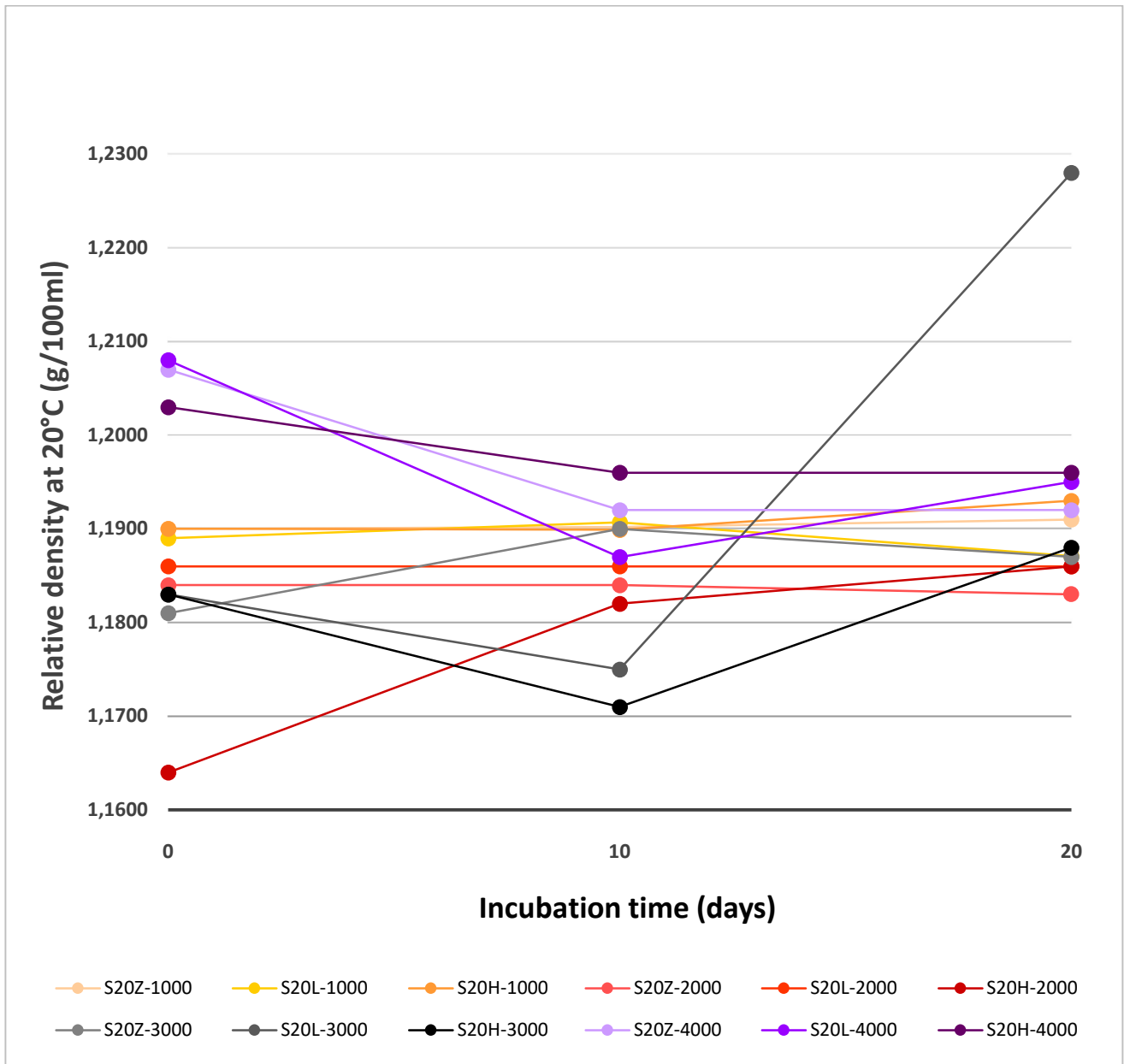
The variations of the density values are not significantly influenced by the levels of contamination and by the level of viscosity of the samples analyzed (table 19). As can be observed (figure 50), the trends are almost linear.

In evaluating the values found, the effect of the dilution caused by the inoculums performed must also be considered.

Only three samples that leave the trend are highlighted and they are S20H-2000, S20L-3000 and less markedly sample S20H-3000 (figure 50).

**Table 19:** Relative density at 20°C of four glazes with Aceto Balsamico di Modena PGI at different viscosity values (1.000, 2.000, 3.000, and 4.000 cps) and inoculated with 10<sup>2</sup> CFU/mL (S20B) and 10<sup>3</sup> CFU/mL (S20A) of *Z. rouxii* ATCC 56077<sup>T</sup>. Raw glazes with Aceto Balsamico di Modena PGI (S20Z) were used as control. Values are expressed as average. Measure uncertainty: from ± 0,00027 to ± 0,001. The term “ABM” stands for Aceto Balsamico di Modena

Sample description	Code	Relative Density at 20°C (g/ml)		
		0 days	10 days	20 days
Glaze with ABM PGI - zero contamination	S20Z-1000	1,1900	1,1902	1,1910
Glaze with ABM PGI - low contamination (10 <sup>2</sup> CFU/ml)	S20L-1000	1,1890	1,1907	1,1871
Glaze with ABM PGI - high contamination (10 <sup>3</sup> CFU/ml)	S20H-1000	1,1900	1,1899	1,1930
Glaze with ABM PGI - zero contamination	S20Z-2000	1,1840	1,1840	1,1830
Glaze with ABM PGI - low contamination (10 <sup>2</sup> CFU/ml)	S20L-2000	1,1860	1,1860	1,1860
Glaze with ABM PGI - high contamination (10 <sup>3</sup> CFU/ml)	S20H-2000	1,1640	1,1820	1,1860
Glaze with ABM PGI - zero contamination	S20Z-3000	1,1810	1,1900	1,1870
Glaze with ABM PGI - low contamination (10 <sup>2</sup> CFU/ml)	S20L-3000	1,1830	1,1750	1,2280
Glaze with ABM PGI - high contamination (10 <sup>3</sup> CFU/ml)	S20H-3000	1,1830	1,1710	1,1880
Glaze with ABM PGI - zero contamination	S20Z-4000	1,2070	1,1920	1,1920
Glaze with ABM PGI - low contamination (10 <sup>2</sup> CFU/ml)	S20L-4000	1,2080	1,1870	1,1950
Glaze with ABM PGI - high contamination (10 <sup>3</sup> CFU/ml)	S20H-4000	1,2030	1,1960	1,1960



**Figure 50:** Relative density at 20°C variation of four glazes with Aceto Balsamico di Modena PGI at different viscosity values (1.000, 2.000, 3.000, and 4.000 cps) and inoculated with  $10^2$  CFU/mL (S20B) and  $10^3$  CFU/mL (S20A) of *Z. rouxii* ATCC 56077<sup>T</sup>. Raw glazes with Aceto Balsamico di Modena PGI (S20Z) were used as control. Dots indicate the mean value on the NEOTRON analysis certificate

### 3.3.5.5 Alcoholic degree

The variations of the alcoholic degree (% Vol) are null. The samples are not influenced by the microbiological spoilage. The initial presence of alcohol is too low to permit at the microorganism to influence this parameter (table 20).

**Table 20:** Alcoholic degree of four glazes with Aceto Balsamico di Modena PGI at different viscosity values (1.000, 2.000, 3.000, and 4.000 cps) and inoculated with  $10^2$  CFU/mL (S20B) and  $10^3$  CFU/mL (S20A) of *Z. rouxii* ATCC 56077<sup>T</sup>. Raw glazes with Aceto Balsamico di Modena PGI (S20Z) were used as control. Values are expressed as average. Measure uncertainty:  $\pm 0,01$ . Quantification limit: 0,07. The term “ABM” stands for Aceto Balsamico di Modena

Sample description	Code	Alcoholic degree (% Vol)		
		0 days	10 days	20 days
Glaze with ABM PGI - zero contamination	S20Z-1000	0,1000	< LQ	0,0900
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-1000	< LQ	< LQ	< LQ
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-1000	0,0900	0,0900	0,1000
Glaze with ABM PGI - zero contamination	S20Z-2000	0,0900	0,1000	< LQ
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-2000	0,1000	0,1100	< LQ
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-2000	0,1000	0,0900	0,0700
Glaze with ABM PGI - zero contamination	S20Z-3000	0,0700	0,0900	0,0900
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-3000	< LQ	0,1100	0,1000
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-3000	0,1000	0,1100	0,0900
Glaze with ABM PGI - zero contamination	S20Z-4000	0,0800	0,0800	0,0900
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-4000	0,0900	0,0800	0,0900
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-4000	0,0800	0,0900	0,0800

### 3.3.5.6 Ochratoxin A

Also Ochratoxin A is not influenced by the microbiological spoilage. Ochratoxin A, being the one most easily present in the grape matrix, was analyzed to verify that the microbiological deterioration

is not able to generate this toxin that at certain concentration is dangerous for the human health (table 21).

**Table 21:** Ochratoxin A of four glazes with Aceto Balsamico di Modena PGI at different viscosity values (1.000, 2.000, 3.000, and 4.000 cps) and inoculated with  $10^2$  CFU/mL (S20B) and  $10^3$  CFU/mL (S20A) of *Z. rouxii* ATCC 56077<sup>T</sup>. Raw glazes with Aceto Balsamico di Modena PGI (S20Z) were used as control. Values are expressed as average. Quantification limit: 0,1. The term “ABM” stands for Aceto Balsamico di Modena

Sample description	Code	Ochratoxin A (µg/kg)		
		0 days	10 days	20 days
Glaze with ABM PGI - zero contamination	S20Z-1000	< LQ	< LQ	< LQ
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-1000	< LQ	< LQ	< LQ
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-1000	< LQ	< LQ	< LQ
Glaze with ABM PGI - zero contamination	S20Z-2000	< LQ	< LQ	< LQ
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-2000	< LQ	< LQ	< LQ
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-2000	< LQ	< LQ	< LQ
Glaze with ABM PGI - zero contamination	S20Z-3000	< LQ	< LQ	< LQ
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-3000	< LQ	< LQ	< LQ
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-3000	< LQ	< LQ	< LQ
Glaze with ABM PGI - zero contamination	S20Z-4000	< LQ	< LQ	< LQ
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-4000	< LQ	< LQ	< LQ
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-4000	< LQ	< LQ	< LQ

### 3.3.5.7 Sugars content and profile

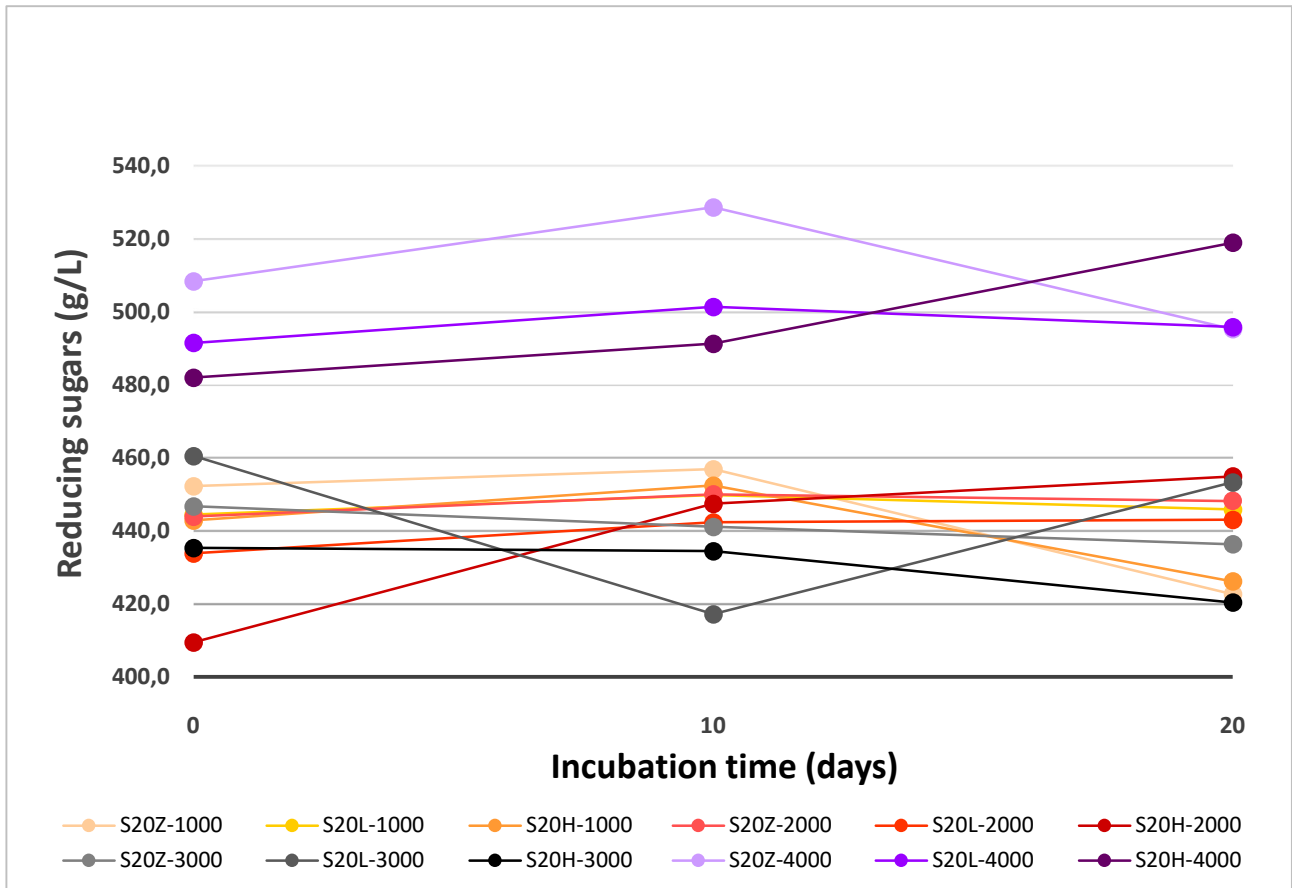
#### 3.3.5.7.1 Reducing sugars

The reducing sugars concentration does not seem to change significantly during the incubation time of the samples (table 22). In fact, also considering the uncertainty of measurement, the glazes that have a more evident oscillation in the reducing sugars value are S20L-2000, S20L-3000 and S20Z-4000 (figure 51).

It is also evident from the samples just highlighted that the variation is not linked to the level of microbial spoilage.

**Table 22:** Reducing sugars of four glazes with Aceto Balsamico di Modena PGI at different viscosity values (1.000, 2.000, 3.000, and 4.000 cps) and inoculated with  $10^2$  CFU/mL (S20B) and  $10^3$  CFU/mL (S20A) of *Z. rouxii* ATCC 56077<sup>T</sup>. Raw glazes with Aceto Balsamico di Modena PGI (S20Z) were used as control. Values are expressed as average. Measure uncertainty: from  $\pm 12$  g/L to  $\pm 15$  g/L. The term “ABM” stands for Aceto Balsamico di Modena

Sample description	Code	Reduction sugars (g/L)		
		0 days	10 days	20 days
Glaze with ABM PGI - zero contamination	S20Z-1000	452,3	457,0	422,7
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-1000	444,5	449,9	445,9
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-1000	442,9	452,5	426,2
Glaze with ABM PGI - zero contamination	S20Z-2000	444,0	450,1	448,2
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-2000	433,9	442,4	443,1
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-2000	409,5	447,5	455,0
Glaze with ABM PGI - zero contamination	S20Z-3000	446,8	441,2	436,4
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-3000	460,6	417,2	453,4
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-3000	435,4	434,5	420,4
Glaze with ABM PGI - zero contamination	S20Z-4000	508,5	528,8	495,4
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-4000	491,6	501,5	495,9
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-4000	482,1	491,4	519,1



**Figure 51:** Reducing sugars variation of four glazes with Aceto Balsamico di Modena PGI at different viscosity values (1.000, 2.000, 3.000, and 4.000 cps) and inoculated with  $10^2$  CFU/mL (S20B) and  $10^3$  CFU/mL (S20A) of *Z. rouxii* ATCC 56077<sup>T</sup>. Raw glazes with Aceto Balsamico di Modena PGI (S20Z) were used as control. Dots indicate the mean value on the NEOTRON analysis certificate

### 3.3.5.7.2 Galactose, Lactose, Maltose and Sucrose content

The glaze of Aceto Balsamico di Modena PGI is a vinegar-based condiment and due its raw materials shouldn't contain sugars other than those which characterize grapes (Glucose and Fructose).

The sugars profile analysis of all the samples confirms the complete absence of Galactose, Sucrose, Lactose and Maltose sugar typology.

The measurements performed also show that these types of sugars are not generated by microbial spoilage during the incubation time (table 23a, 23b, 23c and 23d).



**Table 23a, 23b, 23c and 23d:** Galactose, Sucrose, Lactose and Maltose content of four glazes with Aceto Balsamico di Modena PGI at different viscosity values (1.000, 2.000, 3.000, and 4.000 cps) and inoculated with  $10^2$  CFU/mL (S20B) and  $10^3$  CFU/mL (S20A) of *Z. rouxii* ATCC 56077<sup>T</sup>. Raw glazes with Aceto Balsamico di Modena PGI (S20Z) were used as control. Values are expressed as average. Quantification limit: 0,1. The term “ABM” stands for Aceto Balsamico di Modena

23a	Sample description	Code	Galactose anhydrous (g/100g)		
			0 days	10 days	20 days
	Glaze with ABM PGI - zero contamination	S20Z-1000	< LQ	< LQ	< LQ
	Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-1000	< LQ	< LQ	< LQ
	Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-1000	< LQ	< LQ	< LQ
	Glaze with ABM PGI - zero contamination	S20Z-2000	< LQ	< LQ	< LQ
	Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-2000	< LQ	< LQ	< LQ
	Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-2000	< LQ	< LQ	< LQ
	Glaze with ABM PGI - zero contamination	S20Z-3000	< LQ	< LQ	< LQ
	Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-3000	< LQ	< LQ	< LQ
	Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-3000	< LQ	< LQ	< LQ
	Glaze with ABM PGI - zero contamination	S20Z-4000	< LQ	< LQ	< LQ
	Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-4000	< LQ	< LQ	< LQ
	Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-4000	< LQ	< LQ	< LQ
23b	Sample description	Code	Lactose anhydrous (g/100g)		
			0 days	10 days	20 days
	Glaze with ABM PGI - zero contamination	S20Z-1000	< LQ	< LQ	< LQ
	Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-1000	< LQ	< LQ	< LQ
	Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-1000	< LQ	< LQ	< LQ
	Glaze with ABM PGI - zero contamination	S20Z-2000	< LQ	< LQ	< LQ
	Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-2000	< LQ	< LQ	< LQ
	Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-2000	< LQ	< LQ	< LQ
	Glaze with ABM PGI - zero contamination	S20Z-3000	< LQ	< LQ	< LQ
	Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-3000	< LQ	< LQ	< LQ
	Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-3000	< LQ	< LQ	< LQ
	Glaze with ABM PGI - zero contamination	S20Z-4000	< LQ	< LQ	< LQ
	Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-4000	< LQ	< LQ	< LQ
	Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-4000	< LQ	< LQ	< LQ

23c	Sample description	Code	Maltose anhydrous (g/100g)		
			0 days	10 days	20 days
	Glaze with ABM PGI - zero contamination	S20Z-1000	< LQ	< LQ	< LQ
	Glaze with ABM PGI - low contamination (10 <sup>2</sup> CFU/ml)	S20L-1000	< LQ	< LQ	< LQ
	Glaze with ABM PGI - high contamination (10 <sup>3</sup> CFU/ml)	S20H-1000	< LQ	< LQ	< LQ
	Glaze with ABM PGI - zero contamination	S20Z-2000	< LQ	< LQ	< LQ
	Glaze with ABM PGI - low contamination (10 <sup>2</sup> CFU/ml)	S20L-2000	< LQ	< LQ	< LQ
	Glaze with ABM PGI - high contamination (10 <sup>3</sup> CFU/ml)	S20H-2000	< LQ	< LQ	< LQ
	Glaze with ABM PGI - zero contamination	S20Z-3000	< LQ	< LQ	< LQ
	Glaze with ABM PGI - low contamination (10 <sup>2</sup> CFU/ml)	S20L-3000	< LQ	< LQ	< LQ
	Glaze with ABM PGI - high contamination (10 <sup>3</sup> CFU/ml)	S20H-3000	< LQ	< LQ	< LQ
	Glaze with ABM PGI - zero contamination	S20Z-4000	< LQ	< LQ	< LQ
	Glaze with ABM PGI - low contamination (10 <sup>2</sup> CFU/ml)	S20L-4000	< LQ	< LQ	< LQ
	Glaze with ABM PGI - high contamination (10 <sup>3</sup> CFU/ml)	S20H-4000	< LQ	< LQ	< LQ
23d	Sample description	Code	Sucrose anhydrous (g/100g)		
			0 days	10 days	20 days
	Glaze with ABM PGI - zero contamination	S20Z-1000	< LQ	< LQ	< LQ
	Glaze with ABM PGI - low contamination (10 <sup>2</sup> CFU/ml)	S20L-1000	< LQ	< LQ	< LQ
	Glaze with ABM PGI - high contamination (10 <sup>3</sup> CFU/ml)	S20H-1000	< LQ	< LQ	< LQ
	Glaze with ABM PGI - zero contamination	S20Z-2000	< LQ	< LQ	< LQ
	Glaze with ABM PGI - low contamination (10 <sup>2</sup> CFU/ml)	S20L-2000	< LQ	< LQ	< LQ
	Glaze with ABM PGI - high contamination (10 <sup>3</sup> CFU/ml)	S20H-2000	< LQ	< LQ	< LQ
	Glaze with ABM PGI - zero contamination	S20Z-3000	< LQ	< LQ	< LQ
	Glaze with ABM PGI - low contamination (10 <sup>2</sup> CFU/ml)	S20L-3000	< LQ	< LQ	< LQ
	Glaze with ABM PGI - high contamination (10 <sup>3</sup> CFU/ml)	S20H-3000	< LQ	< LQ	< LQ
	Glaze with ABM PGI - zero contamination	S20Z-4000	< LQ	< LQ	< LQ
	Glaze with ABM PGI - low contamination (10 <sup>2</sup> CFU/ml)	S20L-4000	< LQ	< LQ	< LQ
	Glaze with ABM PGI - high contamination (10 <sup>3</sup> CFU/ml)	S20H-4000	< LQ	< LQ	< LQ

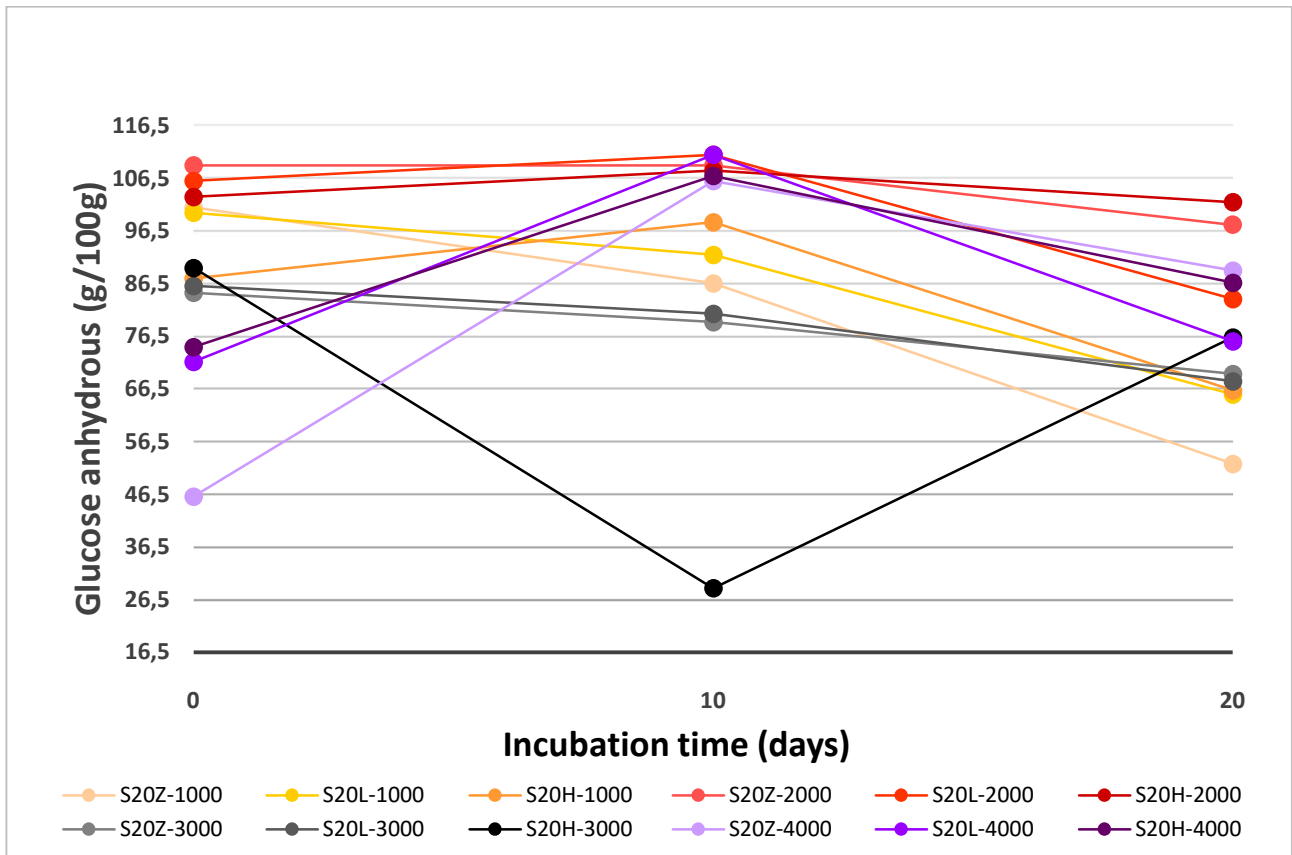
### 3.3.5.7.3 Glucose content

The analysis highlights the glucose presence inside the samples of glaze of Aceto Balsamico di Modena PGI (table 24).

The trend in the figure 52 shows how, regardless of the level of contamination and the viscosity value of the samples, glucose tends to decrease after 10 days of incubation and then tends to return to the initial concentration after 20 days. This highlights how microbial spoilage doesn't affect this parameter.

**Table 24:** Glucose content of four glazes with Aceto Balsamico di Modena PGI at different viscosity values (1.000, 2.000, 3.000, and 4.000 cps) and inoculated with  $10^2$  CFU/mL (S20B) and  $10^3$  CFU/mL (S20A) of *Z. rouxii* ATCC 56077<sup>T</sup>. Raw glazes with Aceto Balsamico di Modena PGI (S20Z) were used as control. Values are expressed as average. Measure uncertainty: from  $\pm 1,2$  g/100g to  $\pm 1,4$  g/100g. The term “ABM” stands for Aceto Balsamico di Modena

Sample description	Code	Glucose anhydrous (g/100g)		
		0 days	10 days	20 days
Glaze with ABM PGI - zero contamination	S20Z-1000	19,2	18,3	18,1
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-1000	19,0	17,5	18,4
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-1000	19,3	17,7	18,1
Glaze with ABM PGI - zero contamination	S20Z-2000	18,4	17,1	18,0
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-2000	17,6	17,4	17,2
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-2000	18,1	17,9	17,1
Glaze with ABM PGI - zero contamination	S20Z-3000	17,2	17,9	16,9
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-3000	17,1	17,3	17,1
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-3000	17,3	17,1	16,9
Glaze with ABM PGI - zero contamination	S20Z-4000	19,3	18,4	18,8
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-4000	18,9	17,8	18,6
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-4000	18,6	18,3	18,4



**Figure 52:** Glucose variation of four glazes with Aceto Balsamico di Modena PGI at different viscosity values (1.000, 2.000, 3.000, and 4.000 cps) and inoculated with  $10^2$  CFU/mL (S20B) and  $10^3$  CFU/mL (S20A) of *Z. rouxii* ATCC 56077<sup>T</sup>. Raw glazes with Aceto Balsamico di Modena PGI (S20Z) were used as control. Dots indicate the mean value on the NEOTRON analysis certificate

### 3.3.5.7.4 Fructose content

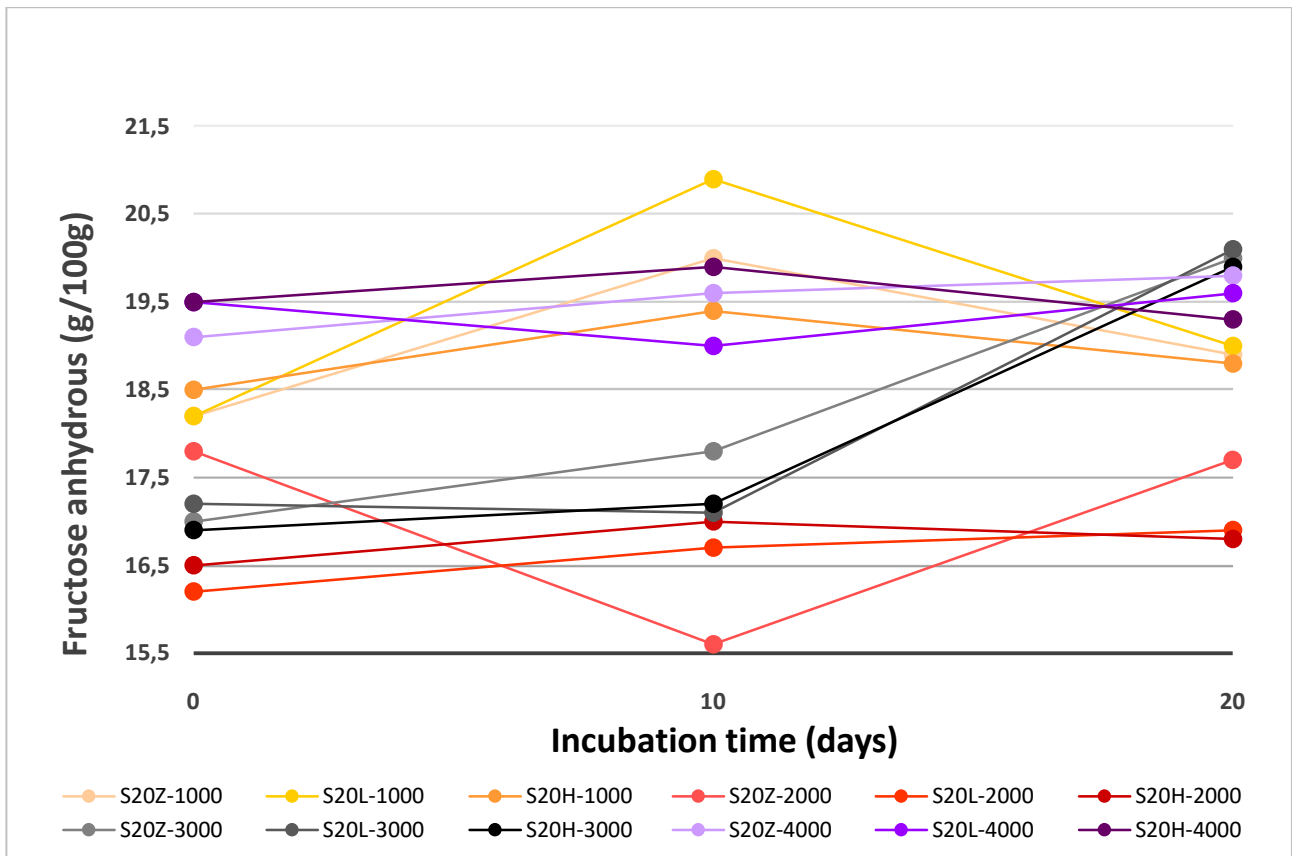
The analysis confirm fructose is present in the sample of glaze of Aceto Balsamico di Modena PGI (table 25).

The trend in the figure 53 shows how, regardless of the level of contamination and the viscosity value of the samples, the concentration of glucose tends to remain more stable during the incubation time. However, there are exceptions to this trend: sample S20Z-2000 that has a sensible decrease after 10 days of incubation to then bring the glucose level back to the initial values; sample S20L-1000 that has a sensible increase after 10 days of incubation to then bring the glucose level back to the initial values and finally the S20-3000 series, all the samples after 10 days of incubation have a surge in fructose value.

**Table 25:** Fructose content of four glazes with Aceto Balsamico di Modena PGI at different viscosity values (1.000, 2.000, 3.000, and 4.000 cps) and inoculated with  $10^2$  CFU/mL (S20B) and  $10^3$  CFU/mL (S20A) of *Z. rouxii* ATCC 56077<sup>T</sup>. Raw glazes with Aceto Balsamico di Modena PGI (S20Z) were used as control. Values are expressed as average. Measure uncertainty: from  $\pm 1,3$  g/100g to  $\pm 1,5$  g/100g. The term “ABM” stands for Aceto Balsamico di Modena

Sample description	Code	Fructose anhydrous (g/100g)		
		0 days	10 days	20 days
Glaze with ABM PGI - zero contamination	S20Z-1000	18,2	20,0	18,9
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-1000	18,2	20,9	19,0
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-1000	18,5	19,4	18,8
Glaze with ABM PGI - zero contamination	S20Z-2000	17,8	15,6	17,7
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-2000	16,2	16,7	16,9
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-2000	16,5	17,0	16,8
Glaze with ABM PGI - zero contamination	S20Z-3000	17,0	17,8	20,0
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-3000	17,2	17,1	20,1
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-3000	16,9	17,2	19,9
Glaze with ABM PGI - zero contamination	S20Z-4000	19,1	19,6	19,8
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-4000	19,5	19,0	19,6
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-4000	19,5	19,9	19,3

The S20-4000 samples are the most stable respect to the variation of the fructose concentration. However, even in this case the microbial spoilage seems not to affect this parameter.



**Figure 53:** Fructose variation of four glazes with Aceto Balsamico di Modena PGI at different viscosity values (1.000, 2.000, 3.000, and 4.000 cps) and inoculated with  $10^2$  CFU/mL (S20B) and  $10^3$  CFU/mL (S20A) of *Z. rouxii* ATCC 56077<sup>T</sup>. Raw glazes with Aceto Balsamico di Modena PGI (S20Z) were used as control. Dots indicate the mean value on the NEOTRON analysis certificate

### 3.3.5.8 Organic acids profile

#### 3.3.5.8.1 Citric acid

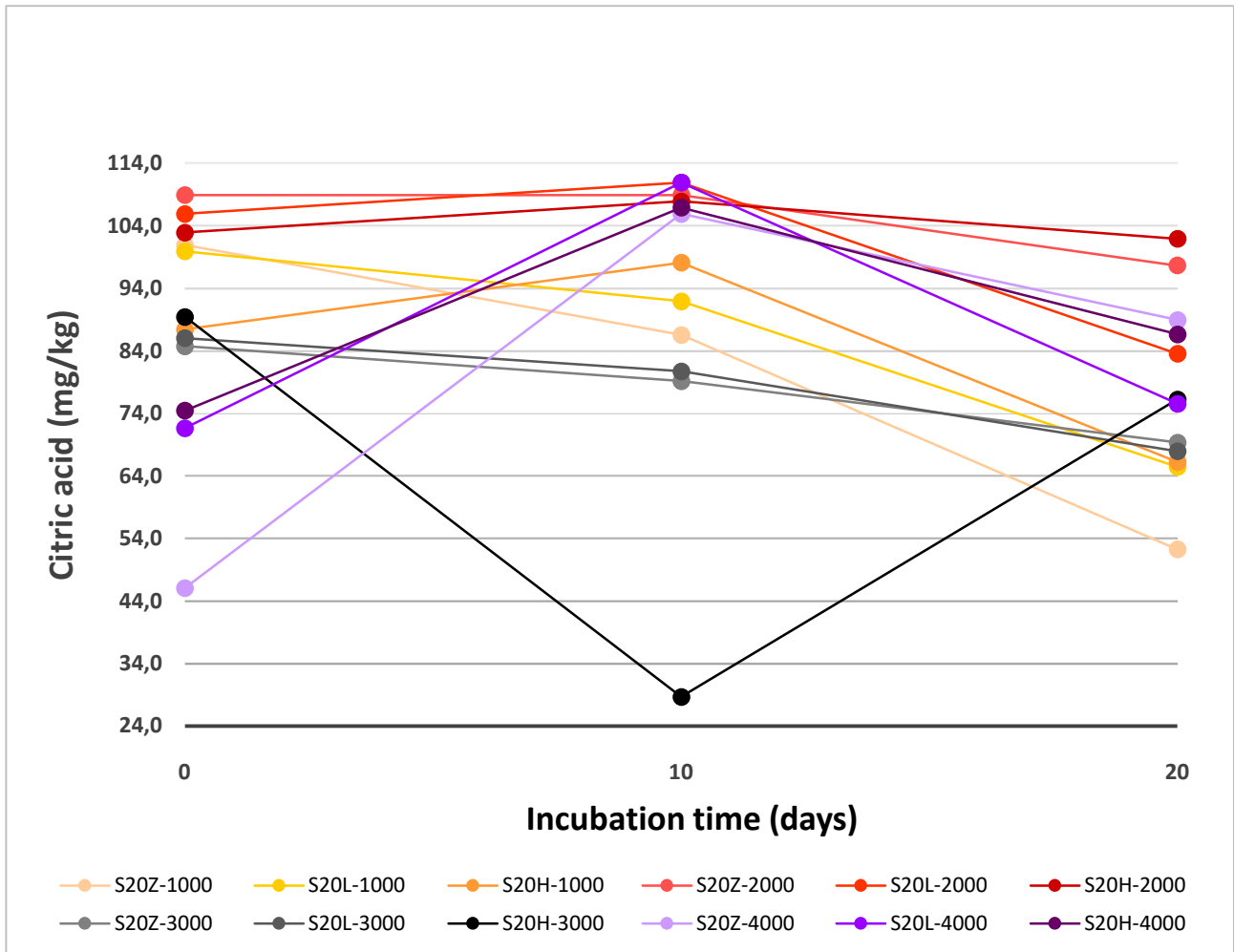
The tendency of the variation of the citric acid content, also considering the measurement uncertainty, doesn't show great changing. However, the trend is that of a slight decrease during the incubation period (table 26).

The most relevant variability is of the samples of the S20-4000 series.

An exception can be noted, it's related to the sample S20H-3000 which is the only one to have a very significant drop at 10 days of incubation time and then return to the initial levels of citric acid at 20 days of incubation time (figure 54).

**Table 26:** Citric acid content of four glazes with Aceto Balsamico di Modena PGI at different viscosity values (1.000, 2.000, 3.000, and 4.000 cps) and inoculated with  $10^2$  CFU/mL (S20B) and  $10^3$  CFU/mL (S20A) of *Z. rouxii* ATCC 56077<sup>T</sup>. Raw glazes with Aceto Balsamico di Modena PGI (S20Z) were used as control. Values are expressed as average. Measure uncertainty: from  $\pm 10$  mg/kg to  $\pm 18$  mg/kg. The term “ABM” stands for Aceto Balsamico di Modena

Sample description	Code	Citric acid (mg/kg)		
		0 days	10 days	20 days
Glaze with ABM PGI - zero contamination	S20Z-1000	101,0	86,6	52,3
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-1000	100,0	92,0	65,5
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-1000	87,5	98,2	66,3
Glaze with ABM PGI - zero contamination	S20Z-2000	109,0	109,0	97,7
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-2000	106,0	111,0	83,6
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-2000	103,0	108,0	102,0
Glaze with ABM PGI - zero contamination	S20Z-3000	84,8	79,2	69,4
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-3000	86,1	80,8	68,0
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-3000	89,5	28,7	76,3
Glaze with ABM PGI - zero contamination	S20Z-4000	46,1	106,0	89,0
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-4000	71,7	111,0	75,6
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-4000	74,5	107,0	86,7



**Figure 54:** Citric acid variation of four glazes with Aceto Balsamico di Modena PGI at different viscosity values (1.000, 2.000, 3.000, and 4.000 cps) and inoculated with  $10^2$  CFU/mL (S20B) and  $10^3$  CFU/mL (S20A) of *Z. rouxii* ATCC 56077<sup>T</sup>. Raw glazes with Aceto Balsamico di Modena PGI (S20Z) were used as control. Dots indicate the mean value on the NEOTRON analysis certificate

### 3.3.5.8.2 Lactic acid

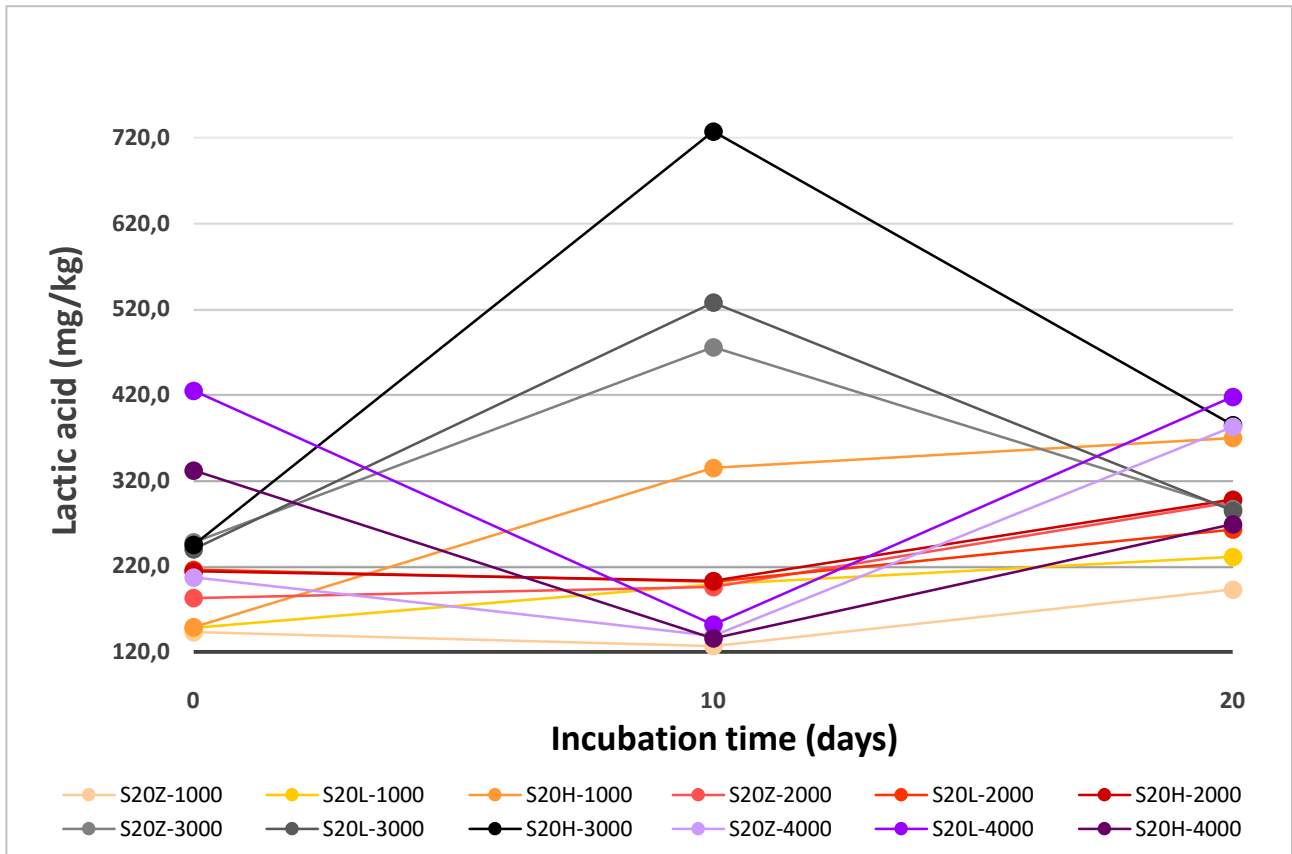
The trend of the lactic acid shows that the level of contamination influences more the glaze samples with high viscosity values (S20-3000 and S20-4000 series). Samples with lower values of viscosity don't have change of lactic acid concentration during the incubation period.



**Table 27:** Lactic acid content of four glazes with Aceto Balsamico di Modena PGI at different viscosity values (1.000, 2.000, 3.000, and 4.000 cps) and inoculated with  $10^2$  CFU/mL (S20B) and  $10^3$  CFU/mL (S20A) of *Z. rouxii* ATCC 56077<sup>T</sup>. Raw glazes with Aceto Balsamico di Modena PGI (S20Z) were used as control. Values are expressed as average. Measure uncertainty: from  $\pm 20$  mg/kg to  $\pm 80$  mg/kg. The term “ABM” stands for Aceto Balsamico di Modena

Sample description	Code	Lactic acid (mg/kg)		
		0 days	10 days	20 days
Glaze with ABM PGI - zero contamination	S20Z-1000	143,0	127,0	193,0
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-1000	148,0	199,0	231,0
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-1000	149,0	335,0	370,0
Glaze with ABM PGI - zero contamination	S20Z-2000	183,0	196,0	295,0
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-2000	216,0	202,0	263,0
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-2000	214,0	203,0	298,0
Glaze with ABM PGI - zero contamination	S20Z-3000	248,0	476,0	287,0
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-3000	240,0	528,0	285,0
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-3000	245,0	728,0	385,0
Glaze with ABM PGI - zero contamination	S20Z-4000	207,0	139,0	383,0
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-4000	425,0	152,0	418,0
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-4000	332,0	136,0	269,0

It is also noted that the S20-3000 and S20-4000 samples have an opposite behavior. The S20-3000 series has an increase at 10 days of incubation and then return to the initial levels of lactic acid, while S20-4000 samples, initially have a decrease of this acid to then return to the initial values at the end of the incubation period (table 27 and figure 55).



**Figure 55:** Lactic acid variation of four glazes with Aceto Balsamico di Modena PGI at different viscosity values (1.000, 2.000, 3.000, and 4.000 cps) and inoculated with  $10^2$  CFU/mL (S20B) and  $10^3$  CFU/mL (S20A) of *Z. rouxii* ATCC 56077<sup>T</sup>. Raw glazes with Aceto Balsamico di Modena PGI (S20Z) were used as control. Dots indicate the mean value on the NEOTRON analysis certificate

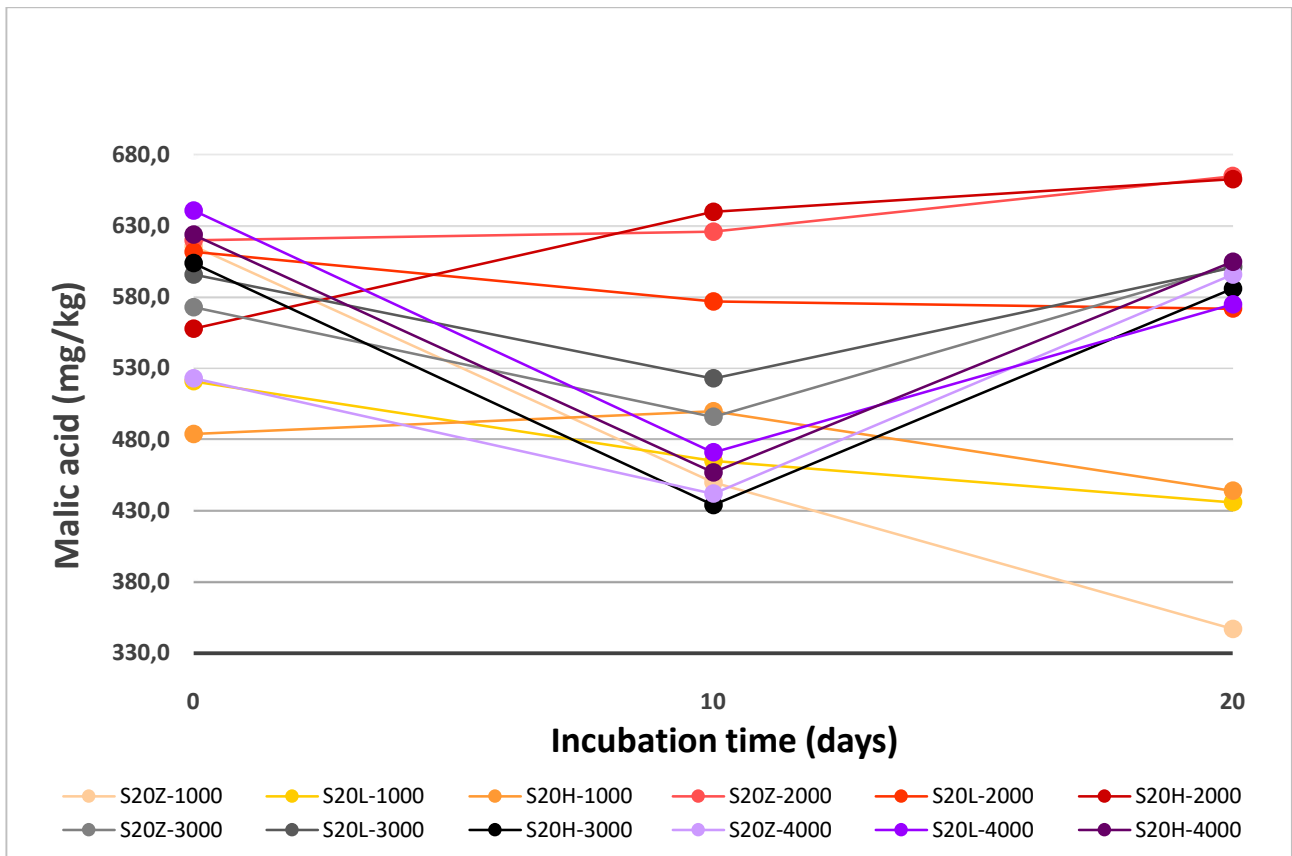
### 3.3.5.8.3 Malic acid

The malic acid trend shows that the contamination doesn't affect the samples of glaze of Aceto Balsamico di Modena PGI.

In fact, the only glaze sample which look like as an outlier, is the S20Z-1000 sample, one of the non-contaminated ones (table 28 and figure 56).

**Table 28:** Malic acid content of four glazes with Aceto Balsamico di Modena PGI at different viscosity values (1.000, 2.000, 3.000, and 4.000 cps) and inoculated with  $10^2$  CFU/mL (S20B) and  $10^3$  CFU/mL (S20A) of *Z. rouxii* ATCC 56077<sup>T</sup>. Raw glazes with Aceto Balsamico di Modena PGI (S20Z) were used as control. Values are expressed as average. Measure uncertainty: from  $\pm 40$  mg/kg to  $\pm 85$  mg/kg. The term “ABM” stands for Aceto Balsamico di Modena

Sample description	Code	Malic acid (mg/kg)		
		0 days	10 days	20 days
Glaze with ABM PGI - zero contamination	S20Z-1000	616,0	450,0	347,0
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-1000	521,0	465,0	436,0
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-1000	484,0	500,0	444,0
Glaze with ABM PGI - zero contamination	S20Z-2000	620,0	626,0	665,0
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-2000	612,0	577,0	572,0
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-2000	558,0	640,0	663,0
Glaze with ABM PGI - zero contamination	S20Z-3000	573,0	496,0	602,0
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-3000	596,0	523,0	601,0
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-3000	604,0	434,0	586,0
Glaze with ABM PGI - zero contamination	S20Z-4000	523,0	442,0	596,0
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-4000	641,0	471,0	575,0
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-4000	624,0	457,0	605,0



**Figure 56:** Malic acid variation of four glazes with Aceto Balsamico di Modena PGI at different viscosity values (1.000, 2.000, 3.000, and 4.000 cps) and inoculated with  $10^2$  CFU/mL (S20B) and  $10^3$  CFU/mL (S20A) of *Z. rouxii* ATCC 56077<sup>T</sup>. Raw glazes with Aceto Balsamico di Modena PGI (S20Z) were used as control. Dots indicate the mean value on the NEOTRON analysis certificate

### 3.3.5.8.4 Succinic acid

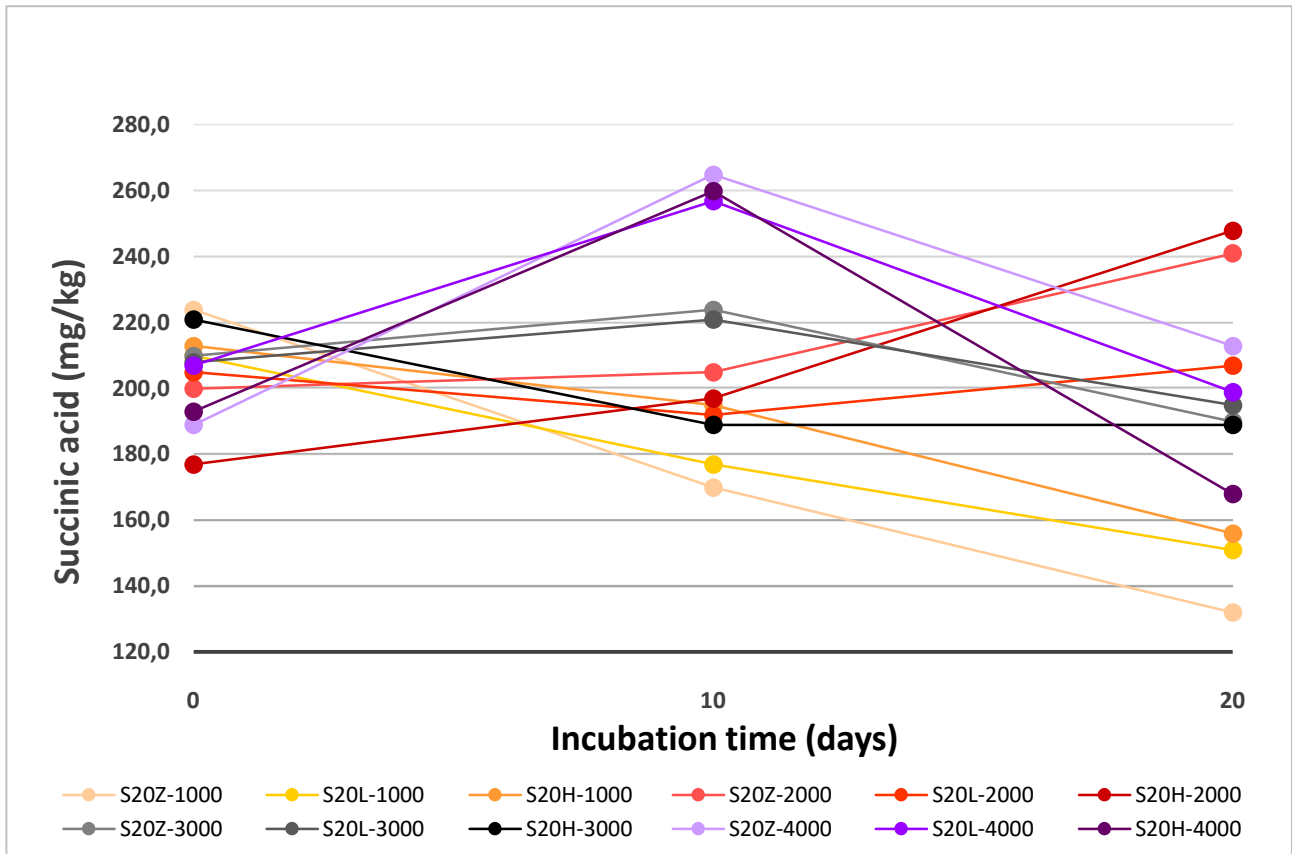
The samples examined regarding the trend of succinic acid during the incubation period have all, with the exception of those of the S20-1000 series, a similar behavior.

The general trend is to maintain the concentration of succinic acid almost constant during the incubation period or in any case bringing the values of the acid back to the initial values.

While the samples of the S20-1000 series show a progressive and irreversible decrease during all the incubation period (table 29 and figure 57).

**Table 29:** Succinic acid content of four glazes with Aceto Balsamico di Modena PGI at different viscosity values (1.000, 2.000, 3.000, and 4.000 cps) and inoculated with  $10^2$  CFU/mL (S20B) and  $10^3$  CFU/mL (S20A) of *Z. rouxii* ATCC 56077<sup>T</sup>. Raw glazes with Aceto Balsamico di Modena PGI (S20Z) were used as control. Values are expressed as average. Measure uncertainty: from  $\pm 20$  mg/kg to  $\pm 40$  mg/kg. The term “ABM” stands for Aceto Balsamico di Modena

Sample description	Code	Succinic acid (mg/kg)		
		0 days	10 days	20 days
Glaze with ABM PGI - zero contamination	S20Z-1000	224,0	170,0	132,0
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-1000	210,0	177,0	151,0
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-1000	213,0	195,0	156,0
Glaze with ABM PGI - zero contamination	S20Z-2000	200,0	205,0	241,0
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-2000	205,0	192,0	207,0
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-2000	177,0	197,0	248,0
Glaze with ABM PGI - zero contamination	S20Z-3000	210,0	224,0	190,0
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-3000	208,0	221,0	195,0
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-3000	221,0	189,0	189,0
Glaze with ABM PGI - zero contamination	S20Z-4000	189,0	265,0	213,0
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-4000	207,0	257,0	199,0
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-4000	193,0	260,0	168,0



**Figure 57:** Succinic acid variation of four glazes with Aceto Balsamico di Modena PGI at different viscosity values (1.000, 2.000, 3.000, and 4.000 cps) and inoculated with  $10^2$  CFU/mL (S20B) and  $10^3$  CFU/mL (S20A) of *Z. rouxii* ATCC 56077<sup>T</sup>. Raw glazes with Aceto Balsamico di Modena PGI (S20Z) were used as control. Dots indicate the mean value on the NEOTRON analysis certificate

### 3.3.5.8.5 Tartaric acid

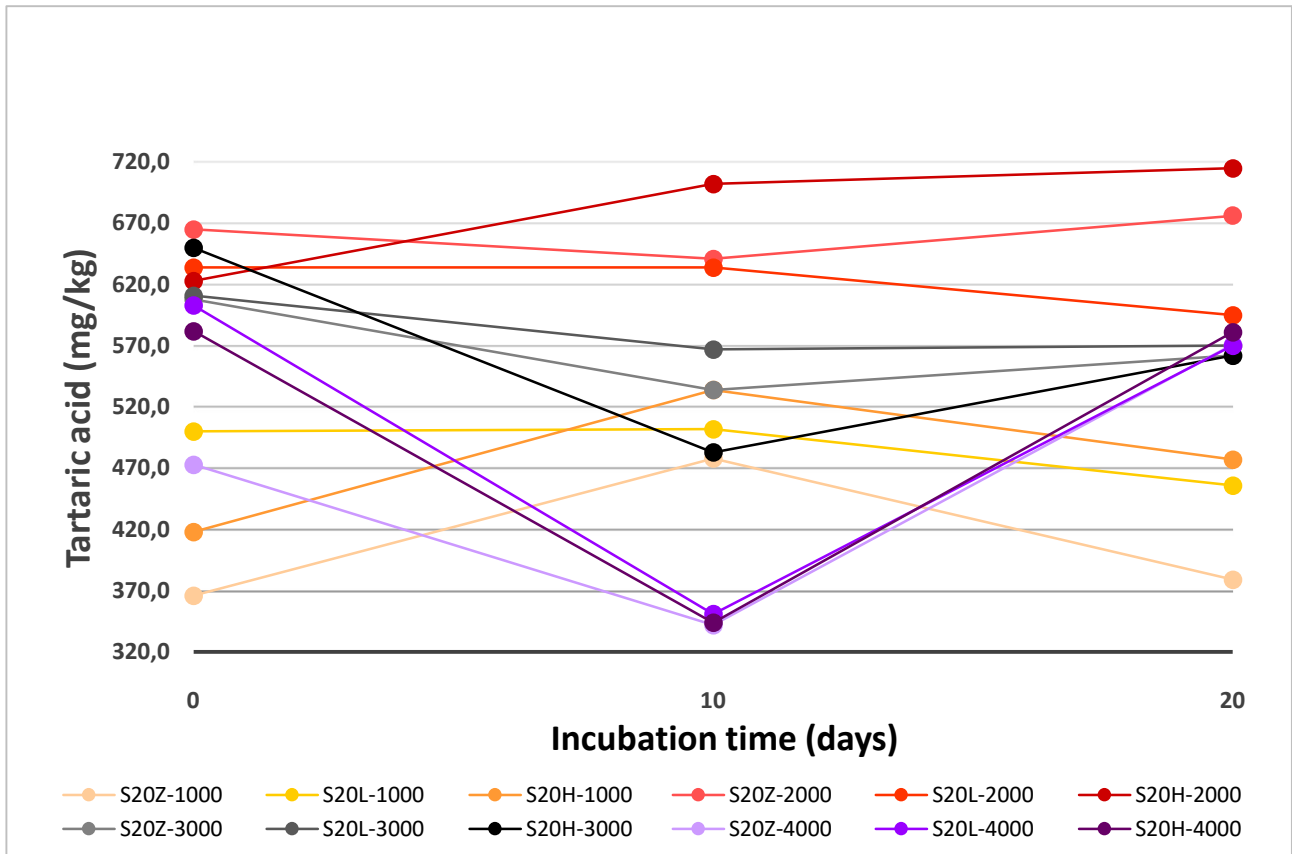
The samples examined regarding the trend of tartaric acid during the incubation period have all, with a similar behavior.

The general trend is to maintain the concentration of tartaric acid almost constant during the incubation period.

The only different trend is registered for the S20-4000 samples. These glazes have a relevant decrease of tartaric acid at 10 days of incubation time, then at 20 days the acid concentration return to the initial values (table 30 and figure 58).

**Table 30:** Tartaric acid content of four glazes with Aceto Balsamico di Modena PGI at different viscosity values (1.000, 2.000, 3.000, and 4.000 cps) and inoculated with  $10^2$  CFU/mL (S20B) and  $10^3$  CFU/mL (S20A) of *Z. rouxii* ATCC 56077<sup>T</sup>. Raw glazes with Aceto Balsamico di Modena PGI (S20Z) were used as control. Values are expressed as average. Measure uncertainty: from  $\pm 50$  mg/kg to  $\pm 90$  mg/kg. The term “ABM” stands for Aceto Balsamico di Modena

Sample description	Code	Tartaric acid (mg/kg)		
		0 days	10 days	20 days
Glaze with ABM PGI - zero contamination	S20Z-1000	366,0	478,0	379,0
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-1000	500,0	502,0	456,0
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-1000	418,0	534,0	477,0
Glaze with ABM PGI - zero contamination	S20Z-2000	665,0	641,0	676,0
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-2000	634,0	634,0	595,0
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-2000	623,0	702,0	715,0
Glaze with ABM PGI - zero contamination	S20Z-3000	608,0	534,0	562,0
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-3000	611,0	567,0	570,0
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-3000	650,0	483,0	562,0
Glaze with ABM PGI - zero contamination	S20Z-4000	473,0	342,0	571,0
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-4000	603,0	351,0	570,0
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-4000	582,0	344,0	581,0



**Figure 58:** Tartaric acid variation of four glazes with Aceto Balsamico di Modena PGI at different viscosity values (1.000, 2.000, 3.000, and 4.000 cps) and inoculated with 10<sup>2</sup> CFU/mL (S20B) and 10<sup>3</sup> CFU/mL (S20A) of *Z. rouxii* ATCC 56077<sup>T</sup>. Raw glazes with Aceto Balsamico di Modena PGI (S20Z) were used as control. Dots indicate the mean value on the NEOTRON analysis certificate

### 3.3.5.9 Brookfield rotational viscosity

The last parameter that is taken into consideration is the Brookfield rotational viscosity. The viscosity is a particular physico-chemical parameter. In fact is derived from the mechanics of fluids. There are two principal categories of fluids: Newtonian fluids and Non-Newtonian fluids. The first ones, that are very rare, are fluids whose viscosity value does not change depending on the measurement conditions and follow law of Stokes of independence of viscosity from position. Instead the Non-Newtonian fluids, that are the majority, are fluids that doesn't follow the law of Stokes and their viscosity value depend on the measurement conditions. The glazes of Aceto Balsamico di Modena PGI are Non-Newtonian fluids, it is for this reason that we attempt, unfortunately in some cases without success, to measure all the samples under the same measurement conditions.



**Table 31:** Brookfield rotational viscosity of four glazes with Aceto Balsamico di Modena PGI at different viscosity values (1.000, 2.000, 3.000, and 4.000 cps) and inoculated with  $10^2$  CFU/mL (S20B) and  $10^3$  CFU/mL (S20A) of *Z. rouxii* ATCC 56077<sup>T</sup>. Raw glazes with Aceto Balsamico di Modena PGI (S20Z) were used as control. Values are expressed as average. The term “ABM” stands for Aceto Balsamico di Modena. The mPa.s and cps unit measure are equivalent.

Measurement condition:

Temperature: 20°C. Impeller: from R2 to R5; specified in the sample code cell (in case of an impeller different from the standard used, it is specified in the cell relating to the measure). RPM: 30 (unless otherwise specified in the cell relating to the measure)

Sample description	Code	Brookfield rotational viscosity (mPa.s)		
		0 days 30 rpm	10 days 30 rpm	20 days 30 rpm
Glaze with ABM PGI - zero contamination	S20Z-1000 (R2)	645	788	935
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-1000 (R2)	700	709	786
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-1000 (R2)	685	665	809
Glaze with ABM PGI - zero contamination	S20Z-2000 (R3)	1.731	1.907	1.764
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-2000 (R3)	1.691	2.012	1.755
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-2000 (R3)	1.721	1.802	1.826
Glaze with ABM PGI - zero contamination	S20Z-3000 (R3)	2.012	2.181	2.159
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-3000 (R3)	2.024	2.244	2.206
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-3000 (R3)	2.058	2.295	2.095
Glaze with ABM PGI - zero contamination	S20Z-4000 (R5)	3.645 <sup>(R4)</sup>	7.132	2.387 <sup>(100 rpm)</sup>
Glaze with ABM PGI - low contamination ( $10^2$ CFU/ml)	S20L-4000 (R5)	4.391 <sup>(60 rpm)</sup>	7.123	6.866
Glaze with ABM PGI - high contamination ( $10^3$ CFU/ml)	S20H-4000 (R5)	4.377 <sup>(60 rpm)</sup>	6.968	6.510

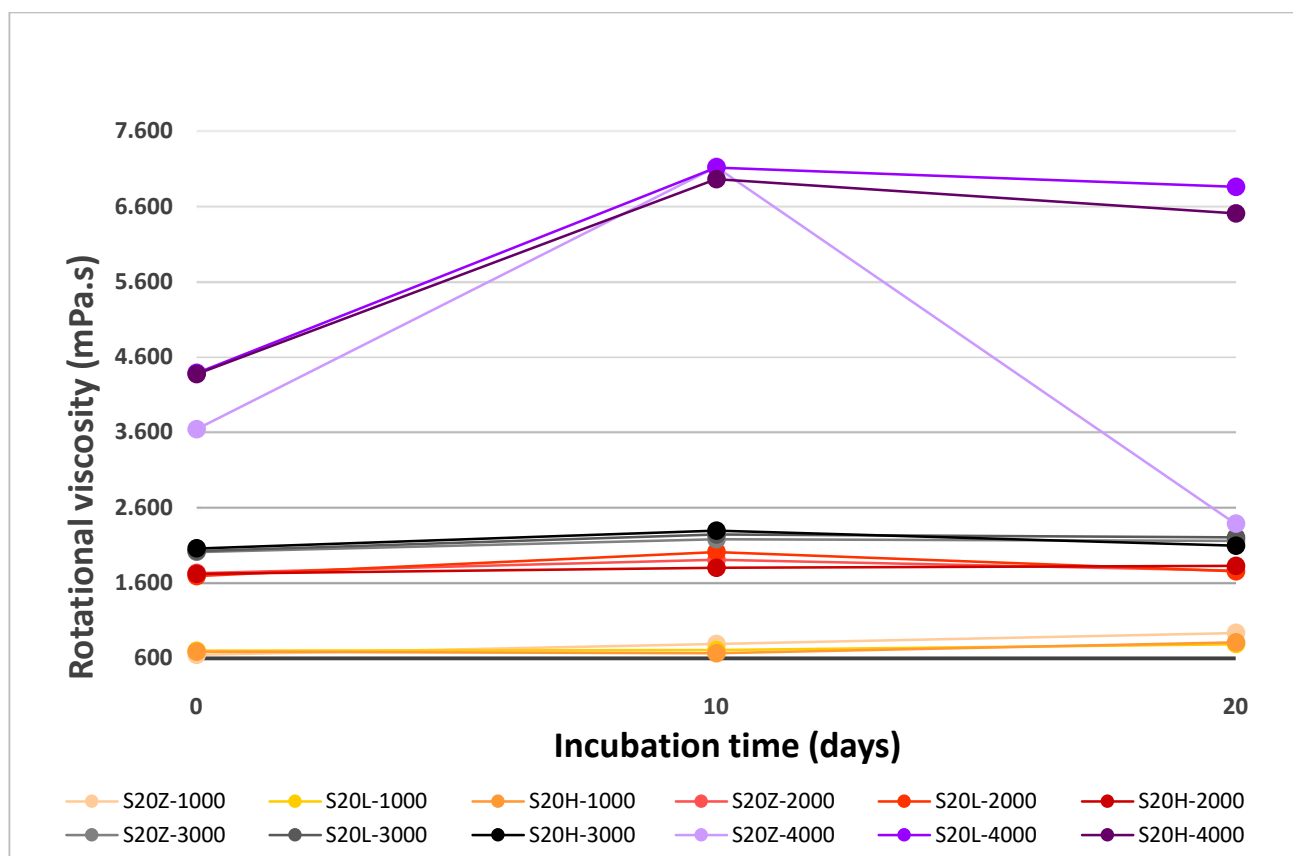
For Brookfield rotational viscosity same measurement conditions means: same sample temperature, same impeller and same rpm. The viscosity of all samples was measured at three different speeds (rpm).

In the table 31 and in the figure 59 it was decided to report only the measurement at one speed (30 rpm) because it turned out to be the speed with which most of the samples were measured. Samples that have not been measured at 30 rpm are properly noted in the table.

Another variant to take into account is the fact that not all samples were measured with the same impeller. This is also indicated in the table.

Unfortunately the choice to change speed and/or impeller depends on the viscosity of the samples. In fact depending on the measuring conditions and the viscosity value of the sample the instrument is unable to return a value.

Although some samples were not measured under the same conditions and therefore some measurements are not comparable to each other, it is still possible by observing the figure 60 to see a fairly precise trend.



**Figure 59:** Brookfield rotational viscosity of four glazes with Aceto Balsamico di Modena PGI at different viscosity values (1.000, 2.000, 3.000, and 4.000 cps) and inoculated with  $10^2$  CFU/mL (S20B) and  $10^3$  CFU/mL (S20A) of *Z. rouxii* ATCC 56077<sup>T</sup>. Raw glazes with Aceto Balsamico di Modena PGI (S20Z) were used as control. Dots indicate the mean value on the NEOTRON analysis certificate

All samples tend not to change viscosity values during the incubation time (figure 60). All samples within the same series were measured with the same rotor and at the same rpm.

The only series which is an exception is the S20-4000 which unfortunately was measured both with different impellers and at different speeds.

In fact it is the only series that does not have a linear trend during the incubation time.

Although some data are not comparable, it is still possible to note that contamination does not influence the viscosity value of the glazes.

Finally, even though in-depth bibliographic research has been done, there are no specific researches regarding a possible stabilizing function of starches in relation to the level of contamination presents in some foods (Taggart 2009, Abbas et al. 2010, Mason 2009 and Anese et al. 1996).

This research work has highlighted how the use of starches, which has the primary purpose of increasing viscosity in foods, also has an important stabilizing function in glazes with Aceto Balsamico di Modena PGI.

In particular, an attempt has been made to evaluate how stabilizing function of viscosity is related or influenced by storage time, product, and contamination level.

This research work confirms that certain types of food can be stored under uncontrolled storage conditions for long time without any heat treatment and still preserve their specific characteristics. Therefore, heat treatments during the production process (for example: pasteurization) are not necessary thanks to viscosity stabilizing and preservative function of starches.

From the results obtained, an in-depth analysis of the viscosity research under various aspects can be taken into consideration:

- evaluate the applicability to other food categories;
- try to define an objective and quantitative correlation between viscosity (and therefore the quantity of starch required), contamination and other instability factors (for example: the storage temperature).

Such insights could lead to the definition of microbiological safety for viscosity values as already defined for the milestone parameters (pH, aW, acidity, etc...) characteristic of self-stabilizing products.

Future research work goal could be to find the minimum viscosity value necessary to ensure food stability over time based on the microbial contamination level.

### ***3.4 Concluding remarks***

Glazes with Aceto Balsamico di Modena PGI are usually a mixture of standard Aceto Balsamico di Modena PGI, wine vinegar, concentrated or cooked grape must and modified starch. During the production process no treatments to reduce microbial load are used meaning the initial stability of raw material is crucial. The results presented in this thesis highlight the crucial role of viscosity in Balsamic glaze stability over storage time. We defined a possible threshold which could be considered as the lowest viscosity value that permit to slow down/control microbial spoilage. Hence, viscosity should be considered as a helper which can maintain the final product stability. As discussed, further studies are necessary to evaluate possible safety thresholds, such as initial microbial load or maximum storage time, in order to improve the applicability at industrial level.

## REFERENCES

- Coton, M., Pawtowski, A., Taminiau, B., Burgaud, G., Deniel, F., Coulloume-Labarthe, L., & Coton, E. (2017). Unraveling microbial ecology of industrial-scale Kombucha fermentations by metabarcoding and culture-based methods. *FEMS microbiology ecology*, 93(5).
- Fuentes-Ramírez, L. E., Bustillos-Cristales, R., Tapia-Hernández, A., Jiménez-Salgado, T., Wang, E. T., Martínez-Romero, E., & Caballero-Mellado, J. (2001). Novel nitrogen-fixing acetic acid bacteria, *Gluconacetobacter johannae* sp. nov. and *Gluconacetobacter azotocaptans* sp. nov., associated with coffee plants. *International Journal of Systematic and Evolutionary Microbiology*, 51(4), 1305-1314.
- García M, Esteve-Zarzoso B, Cabellos JM, Arroyo T (2018) Advances in the study of *Candida stellata*. *Fermentation*, 4(3):74. doi:10.3390/fermentation4030074
- Gullo, M., De Vero, L., & Giudici, P. (2009). *Acetobacter pasteurianus* selected strain and acetic acid bacteria species succession in traditional balsamic vinegar. *Applied and Environmental Microbiology*.
- Gullo, M., La China, S., Petroni, G., Di Gregorio, S., & Giudici, P. (2019). Exploring K2G30 genome: a high bacterial cellulose producing strain in glucose and mannitol based media. *Frontiers in microbiology*, 10, 58.
- Gullo, M., Mamlouk, D., De Vero, L., & Giudici, P. (2012). *Acetobacter pasteurianus* strain AB0220: cultivability and phenotypic stability over 9 years of preservation. *Current microbiology*, 64(6), 576-580.
- Gullo, M., Zanichelli, G., Verzelloni, E., Lemmetti, F., & Giudici, P. (2016). Feasible acetic acid fermentations of alcoholic and sugary substrates in combined operation mode. *Process Biochemistry*, 51(9), 1129-1139.
- Gupta, A., Singh, V. K., Qazi, G. N., & Kumar, A. (2001). *Gluconobacter oxydans*: its biotechnological applications. *Journal of molecular microbiology and biotechnology*, 3(3), 445-456.
- P. Taggart. Starch. IN *Handbook of Hydrocolloids* (Second edition). G.O. Philips and P. A. Williams. Woodhead Publishing Series in Food Science, Technology and Nutrition (2009), pages 108-141

- Hutchinson UF, Ntwampe SKO, Ngongang MM; Chidi BS, Hoff JW, Jolly NP (2019) Product and microbial population kinetics during Balsamic-styled vinegar production. *Journal of Food Science*. doi:10.1111/1750-3841.14429
- Hwang, J. W., Yang, Y. K., Hwang, J. K., Pyun, Y. R., & Kim, Y. S. (1999). Effects of pH and dissolved oxygen on cellulose production by *Acetobacter xylinum* BRC5 in agitated culture. *Journal of Bioscience and Bioengineering*, 88(2), 183-188.
- Abbas, K.A., Khalil, S. K., Hussin, A. S. M. M: (2010). Modified starches and their usages in selected food products: a review study. *Journal of Agricultural Science*, 2(2). doi:10.5539/jas.v2n2p90
- La China, S., De Vero, L., Anguluri, K., Brugnoli, M., Mamlouk, D., & Gullo, M. (2021). Kombucha tea as a reservoir of cellulose producing bacteria: Assessing diversity among *Komagataeibacter* isolates. *Applied Sciences*, 11(4), 1595.
- Lancellotti, L., Ulrici, A., Sighinolfi, S., & Marchetti, A. (2020). Chemical Characterization Of Commercial Glaze with Aceto Balsamico di Modena PGI. *Journal of Food Composition and Analysis*, 94, 103620.
- Leistner, L. & Gould, G.W. “Hurdle technologies: combination treatments for food stability, safety and quality: combination treatments for food stability, safety, and quality”, 2002. Springer New York, NY. doi:10.1007/978-1-4615-0743-7
- Navarro, R. R. & Komagata, K., 1999. Differentiation of *Gluconacetobacter liquefaciens* and *Gluconacetobacter xylinus* on the basis of DNA base composition, DNA relatedness, and oxidation products from glucose. *J. Gen. Appl. Microbiol.*, Volume 45, p. 7:15.
- Solieri L & Giudici P (2008) Yeasts associated to Traditional Balsamic Vinegar: Ecological and technological features. *International Journal of Food Microbiology*, 125(1):36-45. doi:10.1016/j.ijfoodmicro.2007.06.022
- Sperber, E. J. (1983). Influence of water activity on foodborne bacteria — a review. *Journal of Food Protection*, 46(2), 142-150. doi:10.4315/0362-028X-46.2.142
- William R. Mason. Starch use in Foods. IN *Starch (Third Edition)*. James BeMiller and Roy Whistler. Chemistry and Technology. Food Science and Technology (2009), pages 745-795
- Terpou, A.; Mantzourani, I.; Bekatorou, A.; Alexopoulos, A.; Plessas, S. Chapter: Current Trends in Balsamic/Aged Vinegar Production and Research. In “Advances in vinegar production”, 2019.

M. Anese, I. Shtylla, D. Torreggiani, E. Maltini. Water activity and viscosity-relations with glass transition temperatures in model food systems. IN *Thermochimica Acta*. N. Koga and R. Androsch. Volume 275, Issue 1 (1996), pages 131-137

Warren McCabe, Julian Smith, Peter Harriott: *Unit Operations In Chemical Engineering*, 6<sup>a</sup> ed., Tata Mcgraw Hill Publishers, 2005, pp.46-51

Zardetto, S.; Barbanti, D. Shelf life assessment of fresh green pesto using an accelerated test approach. *Food Packag. Shelf Life* 2020,25, 100524

## *Acknowledgements*

First of all, I would like to thank Dr. Maria Gullo who has accompanied and supported me in these 3 years.

It was a point of reference for me to rediscover my passion for research and study.

I thank her in particular for her fundamental contribution in the final phase of drafting the thesis and for her moral support.

Another fundamental person for the finalization of the research work was the PhD student dott. Marcello Brugnoli. Has been fundamental both for the microbiological part but also in the final phase of drafting the thesis of in this research work.

I am also grateful to the student Simona Bruno and the PhD student dott.ssa Elisa Aiello who actively collaborated in the research work and in the drafting of chapter two of this thesis.

I cannot forget the Pradelski family, owners of Acetificio Carandini Emilio S.p.A., together with the CEO, as well as my friend, Stefano Bellei who have decided to support me in this adventure.

Sincere thanks also go to the colleagues of my group who put up with my mental and physical absences because I was involved in this journey.

A special hug to my right arm Dr. Paolo Martorelli who played a key role in the success of the project. Finally, thanks also to Raffaella and Gabriele without whom all this would never have been possible.



## *Ringraziamenti*

Desidero innanzitutto ringraziare la Dott.ssa Maria Gullo che mi ha accompagnato e sopportato in questi 3 anni.

E' stata un punto di riferimento per farmi riscoprire la passione per la ricerca e per lo studio.

La ringrazio in particolare per l'apporto fondamentale nella fase finale di stesura della tesi e per il suo appoggio morale.

Altra persona fondamentale per la finalizzazione del lavoro di ricerca è stato il dottorando Marcello Brugnoli. Fondamentale sia nel lavoro di ricerca vero e proprio, per la parte microbiologica, ma anche nella fase finale di stesura della tesi.

Sono grato anche alla laureanda Simona Bruno e alla dottoranda Elisa Aiello che hanno collaborato attivamente nel lavoro di ricerca e nella stesura del capitolo due di questa tesi.

Non posso dimenticare anche la famiglia Pradelski, proprietari dell'Acetificio Carandini Emilio S.p.A., insieme all'amministratore delegato, oltre che amico Stefano Bellei, che hanno deciso di supportarmi in quest'avventura.

Un sincero ringraziamento va anche ai colleghi del mio gruppo che hanno sopportato le mie assenze sia mentali che fisiche perchè impegnato in questo percorso.

Un abbraccio speciale al mio braccio destro il dott. Paolo Martorelli che ha avuto un ruolo fondamentale per la riuscita del progetto.

Infine un ringraziamento anche a Raffaella e Gabriele senza i quali tutto questo non sarebbe mai stato possibile.

*Appendix A:*

*Neutron SPA analysis certificates*

---

MODENA, li 29/07/2022

Sample arrived on the 19/07/2022  
Registration date 19/07/2022**TEST REPORT nr. 22G12686-In-0**

CUSTOMER

**ACETFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO****SAMPLE 22G12686****MATRIX: Fruit and vegetable concentrates, and  
by-products**Description provided by Customer: GLASSA CON ABM IGP - CONTAMINAZIONE ALTA - VISCOSITA': ~ 1.000 CPS (T=0G) -  
CODICE: S20A-1000Richiesta via Internet n° N0218/22 - 04/07/2022 16:41:22. - Campionamento eseguito da: Committente - Trasporto effettuato da:  
Corriere  
Sample Condition on Receipt: 4°C

ANALYSIS DESCRIPTION	RESULT	U	REC. %	UNIT OF MEASURE	LQ	LD	METHOD	ANALYSES BEGINNING DATE / ENDING DATE
Water activity (Aw) at 25 °C	0,914	± 0,005			0,150		06(S135) rev6 2021	20/07/2022/ 22/07/2022
<b>VINEGAR ANALYSIS</b>								
Reduction sugars (Reducing substances)	442,9	± 13,3		g/l	1,0		07(S240) 2021 Rev.0 - Titrimetrico	21/07/2022/ 26/07/2022
Total Acidity expressed as acetic acid	2,90	± 0,06		g/100 ml	0,10		* ACIDITA 2014 Rev.0 - Titrimetrico	21/07/2022/ 25/07/2022
Relative density at 20°C	1,190	± 0,0003					07(S117) 2015 Rev.1 - Gravimetric	29/07/2022/ 29/07/2022
Alcoholic strength	0,09	± 0,01		% Vol	0,07		DM 12/03/1986 SO GU n 161 14/07/1986 Met V pag 95, OIV- MA-AS312-01 Met B R2021	21/07/2022/ 26/07/2022
pH	2,93	± 0,04		unità	0,50		07(S77) 2012 Rev.1 - Potentiometric	21/07/2022/ 22/07/2022
Brookfield rotational viscosity	Brookfield Rotational Viscosity (T = 20 ° C, impeller R2) 50 rpm: 579 cP (mPa.s) 30 rpm: 685 cP (mPa.s) 20 rpm: 777 cP (mPa.s)						* VISCOSITA 2018 Rev.1	21/07/2022/ 26/07/2022
<b>SUGARS HPAEC-PAD</b>								
Galactose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	21/07/2022/ 25/07/2022
Glucose, anhydrous (HPLC)	19,3	± 1,4		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	21/07/2022/ 25/07/2022
Sucrose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	21/07/2022/ 25/07/2022
Fructose, anhydrous (HPLC)	18,5	± 1,3		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	21/07/2022/ 25/07/2022
Lactose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	21/07/2022/ 25/07/2022
Maltose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	21/07/2022/ 25/07/2022
Citric acid (LCMS) [331]	87,5	± 14,3		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	21/07/2022/ 26/07/2022
Lactic Acid (LCMS) [331]	149	± 22		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	21/07/2022/ 26/07/2022
Malic acid (LCMS) [331]	484	± 61		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	21/07/2022/ 26/07/2022
Succinic acid (LCMS) [331]	213	± 30		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	21/07/2022/ 26/07/2022
Tartaric acid (LCMS) [331]	418	± 54		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	21/07/2022/ 26/07/2022
Ochratoxin A	< LQ			µg/kg	0,10		03(S130) 2022 Rev.12 - LC- MS/MS	21/07/2022/ 26/07/2022

**END TEST REPORT**

The original document is a PDF file with Digital Signature: 22G12686-In-0-S20A-1000 - T0 - UNIMORE-DigitalSignature.pdf

**Next page...**

MODENA, li 29/07/2022

Sample arrived on the 19/07/2022

Registration date 19/07/2022

## TEST REPORT nr. 22G12686-In-0

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**

**SAMPLE 22G12686**

**MATRIX: Fruit and vegetable concentrates, and by-products**

### Notes and method reference:

< LQ: = lower than Quantification Limit.

U: the reported uncertainty is the expanded uncertainty calculated using a coverage factor equal to 2 which gives a reliability of approximately 95%. Please note that results expressed as '<LQ' may not indicate the absence of the searched parameters in the sample.

MICROBIOLOGICAL TESTS: for food and environmental samples, the extended measurement uncertainty was estimated according to ISO 19036:2019 Standard and is based on a standard uncertainty multiplied by a coverage factor of  $K = 2$ , providing a confidence level of approximately 95%. The combined standard uncertainty was assumed to be equal to the standard deviation of intra-laboratory reproducibility. The results of the microbiological tests are calculated according to the ISO 7218: 2007 / Amd 1: 2013 Standard. If the results are reported as <4 (CFU/ml) or <40 (CFU/g), this means that the microorganisms are present in the sample but in amounts less than 4 CFU/ml or 40 CFU/g respectively. For microbiological analyses unless differently reported in the individual test methods, in case of analytical steps foreseen in non-activity days of the laboratory, provisions of the ISO 7218: 2007 / Amd.1 2013 Standard (points 11.2 and 10.2.5) or from specific test methods are applied. In the case of quantitative microbiological tests, these have been set up on a single plate according to ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise expressly requested by current regulations.

In the case of quantitative microbiological tests, these have been set up on a single plate in accordance with ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise explicitly required by current regulations.

For waters, the measurement uncertainty corresponds to the confidence interval calculated according to ISO 8199: 2018 or to the expanded measurement uncertainty estimated according to ISO 29201: 2012. The results are issued in accordance with ISO 8199: 2018. When the number of colonies detected is <3, the result is expressed as "Microorganisms present in the analyzed volume (N ° colonies detected <3 CFU - reference ISO 8199: 2018, paragraph 9.1.8.4.1)".

LQ: Quantification Limit. It is the lowest analyte concentration which can be detected at an acceptable precision (repeatability) and accuracy, under well defined conditions.

LD: Detection Limit. It is the lowest analyte concentration which can be detected but not necessarily quantified, under well defined conditions.

Conformity evaluation: values not complying with laws, decrees, national and EU regulations or specifications supplied by the customer are evaluated case by case, also taking into consideration the uncertainty of measure for each single test and the regulations on rounding-off of values, and pointed out when considered as non conform.

Rec %: Recovery % "+" means that the recovery has been applied to the result. The numeric results between brackets (..) after the expression <LQ are purely indicative of traces that cannot be exactly quantified.

In the case of sampling carried out by Neotron, the laboratory applies the Internal Operating Procedure code: NEOT-DIR/ 006/53.

The laboratory disclaims any responsibility for the information provided by the client reported in this Report which may influence the validity of the results.

Methods marked with an asterisk (\*) are not accredited by ACCREDIA (UNI CEI EN ISO/IEC 17025). The sampling activity is not included within the Scope of Accreditation of Neotron SPA

### NOTES OF PARAMETERS:

[331]: Extended uncertainty calculated following the equation of HORWITZ by using a coverage factor of 2, which guarantee a reliance level of 95%

TEST REPORT VALID FOR ALL LEGAL PURPOSES (Italian R.D. 1-3-1928 n°842 (article 16), - Italian Law 19-7-1957 n°679 articles 16 and 18, Italian Ministerial Decree 25-3-1986).

DATA and SAMPLE STORAGE: Test Reports, Raw data, chromatographic paths and instrumental reports are stored for 5 years. One control sample is stored for 2 months.

Data expressed in this test report refer only to the sample tested in the laboratory. The results reported in this Test Report refer to the sample as received. The description or any other reference concerning the sample are declared by the customer. This Test Report cannot be reproduced except in full. Partial reproductions must be authorized in writing by our laboratory.

THE LABORATORY DIRECTOR: DR. ANDREA RIZZO

THE CHEMIST AUTHORIZED TO SIGN THE TEST REPORTS: DR. MARCO MESCHIARI - N. 514 ORDINE DEI CHIMICI MODENA

Approved by Analysis Manager - laboratory LMAA-Nut

Approved by Analysis Manager - laboratory ICS-HPLC

Approved by Analysis Manager - laboratory LC-MICO

Approved by Analysis Manager - laboratory LC-AN

Approved by Analysis Manager - laboratory FISICHE

Approved by Analysis Manager - laboratory LBM-EL

Approved by Analysis Manager - laboratory LMAA-Bro

NEOTRON SpA - With Sole Shareholder

Stradello Aggazzotti, 104

41126 MODENA - ITALY - Fiscal Code and VAT n° 03807840362

Tel: +39 059461711 - Fax: +39 059461777

www.neotron.it - neotron@neotron.it

Laboratorio Qualificato D.M. 26-2-87 Art. 4 - Legge 46/82 per la Ricerca Applicata e Innovazione Tecnologica.

Regione Emilia Romagna - AUTORIZZAZIONE Autocontrollo N° 008/MO/008

BNN-Monitoring Fruit and Vegetables Approved Laboratory

I-Monitoring EDEKAAG Fruit and Vegetables Registered Laboratory

GMP+ code: GMP051757

MODENA, li 26/10/2022

Sample arrived on the 14/10/2022

Registration date 14/10/2022

**TEST REPORT nr. 22P11008-In-0**

CUSTOMER

**ACETFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**
**SAMPLE 22P11008****MATRIX: Fruit and vegetable concentrates, and by-products**

Description provided by Customer: GLASSA CON ABM IGP - CONTAMINAZIONE ZERO - VISCOSITA': ~ 4.000 CPS (T=20G) - CODICE: S20Z-4000

Richiesta via Internet n° N0386/22 - 28/09/2022 14:04:09. - Campionamento eseguito da: Committente - Trasporto effettuato da: Committente  
Sample Condition on Receipt: 20°C

ANALYSIS DESCRIPTION	RESULT	U	REC. %	UNIT OF MEASURE	LQ	LD	METHOD	ANALYSES BEGINNING DATE / ENDING DATE
Water activity (Aw) at 25 °C	0,929	± 0,005			0,150		06(S135) rev6 2021	17/10/2022 / 18/10/2022
<b>VINEGAR ANALYSIS</b>								
Reduction sugars (Reducing substances)	495,4	± 14,9		g/l	1,0		07(S240) 2021 Rev.0 - Titrimetrico	18/10/2022 / 26/10/2022
Total Acidity expressed as acetic acid	3,08	± 0,06		g/100 ml	0,10		* ACIDITA 2014 Rev.0 - Titrimetrico	18/10/2022 / 19/10/2022
Relative density at 20°C	1,192	± 0,0003					07(S117) 2015 Rev.1 - Gravimetric	25/10/2022 / 25/10/2022
Alcoholic strength	0,09	± 0,01		% Vol	0,07		DM 12/03/1986 SO GU n 161 14/07/1986 Met V pag 95, OIV-MA-AS312-01 Met B R2021	18/10/2022 / 19/10/2022
pH	2,98	± 0,04		unità	0,50		07(S77) 2012 Rev.1 - Potentiometric	18/10/2022 / 19/10/2022
Brookfield rotational viscosity	Brookfield Rotational Viscosity (T = 20 ° C, impeller R5)							
	100 rpm: 2387 cP (mPa.s)							
	60 rpm: 3141 cP (mPa.s)							
	50 rpm: 3494 cP (mPa.s)							
<b>SUGARS HPAEC-PAD</b>								
Galactose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	18/10/2022 / 21/10/2022
Glucose, anhydrous (HPLC)	18,8	± 1,3		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	18/10/2022 / 21/10/2022
Sucrose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	18/10/2022 / 21/10/2022
Fructose, anhydrous (HPLC)	19,8	± 1,4		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	18/10/2022 / 21/10/2022
Lactose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	18/10/2022 / 21/10/2022
Maltose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	18/10/2022 / 21/10/2022
Citric acid (LCMS) [331]	89,0	± 14,5	101	mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	18/10/2022 / 24/10/2022
Lactic Acid (LCMS) [331]	383	± 50	74 +	mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	18/10/2022 / 24/10/2022
Malic acid (LCMS) [331]	596	± 73	90	mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	18/10/2022 / 24/10/2022
Succinic acid (LCMS) [331]	213	± 30	90	mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	18/10/2022 / 24/10/2022
Tartaric acid (LCMS) [331]	571	± 70	80	mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	18/10/2022 / 24/10/2022
Ochratoxin A	< LQ			µg/kg	0,10		03(S130) 2022 Rev.13 - LC-MS/MS	18/10/2022 / 21/10/2022

**END TEST REPORT**

The original document is a PDF file with Digital Signature: 22P11008-In-0-S20Z-4000 - T20 - UNIMORE-DigitalSignature.pdf

**Next page...**

MODENA, li 26/10/2022

Sample arrived on the 14/10/2022

Registration date 14/10/2022

## TEST REPORT nr. 22P11008-In-0

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**

**SAMPLE 22P11008**

**MATRIX: Fruit and vegetable concentrates, and by-products**

### Notes and method reference:

< LQ: = lower than Quantification Limit.

U: the reported uncertainty is the expanded uncertainty calculated using a coverage factor equal to 2 which gives a reliability of approximately 95%. Please note that results expressed as '<LQ' may not indicate the absence of the searched parameters in the sample.

MICROBIOLOGICAL TESTS: for food and environmental samples, the extended measurement uncertainty was estimated according to ISO 19036:2019 Standard and is based on a standard uncertainty multiplied by a coverage factor of K = 2, providing a confidence level of approximately 95%. The combined standard uncertainty was assumed to be equal to the standard deviation of intra-laboratory reproducibility. The results of the microbiological tests are calculated according to the ISO 7218: 2007 / Amd 1: 2013 Standard. If the results are reported as <4 (CFU/ml) or <40 (CFU/g), this means that the microorganisms are present in the sample but in amounts less than 4 CFU/ml or 40 CFU/g respectively. For microbiological analyses unless differently reported in the individual test methods, in case of analytical steps foreseen in non-activity days of the laboratory, provisions of the ISO 7218: 2007 / Amd.1 2013 Standard (points 11.2 and 10.2.5) or from specific test methods are applied. In the case of quantitative microbiological tests, these have been set up on a single plate according to ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise expressly requested by current regulations.

In the case of quantitative microbiological tests, these have been set up on a single plate in accordance with ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise explicitly required by current regulations.

For waters, the measurement uncertainty corresponds to the confidence interval calculated according to ISO 8199: 2018 or to the expanded measurement uncertainty estimated according to ISO 29201: 2012. The results are issued in accordance with ISO 8199: 2018. When the number of colonies detected is <3, the result is expressed as "Microorganisms present in the analyzed volume (N ° colonies detected <3 CFU - reference ISO 8199: 2018, paragraph 9.1.8.4.1)".

LQ: Quantification Limit. It is the lowest analyte concentration which can be detected at an acceptable precision (repeatability) and accuracy, under well defined conditions.

LD: Detection Limit. It is the lowest analyte concentration which can be detected but not necessarily quantified, under well defined conditions. Any fields not filled in the Test Report are to be considered not applicable.

Conformity evaluation: values not complying with laws, decrees, national and EU regulations or specifications supplied by the customer are evaluated case by case, also taking into consideration the uncertainty of measure for each single test and the regulations on rounding-off of values, and pointed out when considered as non conform.

Rec %: Recovery % "+" means that the recovery has been applied to the result. The numeric results between brackets (.) after the expression <LQ are purely indicative of traces that cannot be exactly quantified.

In the case of sampling carried out by Neotron, the laboratory applies the Internal Operating Procedure code: NEOT-DIR/ 006/53.

The laboratory disclaims any responsibility for the information provided by the client reported in this Report which may influence the validity of the results.

Methods marked with an asterisk (\*) are not accredited by ACCREDIA (UNI CEI EN ISO/IEC 17025). The sampling activity is not included within the Scope of Accreditation of Neotron SPA

### NOTES OF PARAMETERS:

[331]: Extended uncertainty calculated following the equation of HORWITZ by using a coverage factor of 2, which guarantee a reliance level of 95%

TEST REPORT VALID FOR ALL LEGAL PURPOSES (Italian R.D. 1-3-1928 n°842 (article 16), - Italian Law 19-7-1957 n°679 articles 16 and 18, Italian Ministerial Decree 25-3-1986).

DATA and SAMPLE STORAGE: Test Reports, Raw data, chromatographic paths and instrumental reports are stored for 5 years. One control sample is stored for 2 months.

Data expressed in this test report refer only to the sample tested in the laboratory. The results reported in this Test Report refer to the sample as received. The description or any other reference concerning the sample are declared by the customer. This Test Report cannot be reproduced except in full. Partial reproductions must be authorized in writing by our laboratory.

THE LABORATORY DIRECTOR: DR. ANDREA RIZZO

THE CHEMIST AUTHORIZED TO SIGN THE TEST REPORTS: DR. MARCO MESCHIARI

NEOTRON SpA - With Sole Shareholder

Stradello Aggazzotti, 104

41126 MODENA - ITALY - Fiscal Code and VAT n° 03807840362

Tel: +39 059461711 - Fax: +39 059461777

www.neotron.it - neotron@neotron.it

Laboratorio Qualificato D.M. 26-2-87 Art. 4 - Legge 46/82 per la Ricerca Applicata e Innovazione Tecnologica.

Regione Emilia Romagna - AUTORIZZAZIONE Autocontrollo N° 008/MO/008

BNN-Monitoring Fruit and Vegetables Approved Laboratory

I-Monitoring EDEKAAG Fruit and Vegetables Registered Laboratory

GMP+ code: GMP051757

MODENA, li 20/10/2022

Sample arrived on the 05/10/2022  
Registration date 05/10/2022**TEST REPORT nr. 22P03162-In-0**

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO****SAMPLE 22P03162****MATRIX: Fruit and vegetable concentrates, and  
by-products**Description provided by Customer: GLASSA CON ABM IGP - CONTAMINAZIONE ZERO - VISCOSITA': ~ 4.000 CPS (T=10G) -  
CODICE: S20Z-4000Richiesta via Internet n° N0385/22 - 28/09/2022 14:04:09. - Campionamento eseguito da: Committente - Trasporto effettuato da:  
Corriere  
Sample Condition on Receipt: 20°C

ANALYSIS DESCRIPTION	RESULT	U	REC. %	UNIT OF MEASURE	LQ	LD	METHOD	ANALYSES BEGINNING DATE / ENDING DATE
Water activity (Aw) at 25 °C	0,936	± 0,005			0,150		06(S135) rev6 2021	05/10/2022/ 11/10/2022
<b>VINEGAR ANALYSIS</b>								
Reduction sugars (Reducing substances)	528,8	± 15,9		g/l	1,0		07(S240) 2021 Rev.0 - Titrimetrico	06/10/2022/ 12/10/2022
Total Acidity expressed as acetic acid	3,06	± 0,06		g/100 ml	0,10		* ACIDITA 2014 Rev.0 - Titrimetrico	06/10/2022/ 06/10/2022
Relative density at 20°C	1,192	± 0,001					07(S117) 2015 Rev.1 - Gravimetric	20/10/2022/ 20/10/2022
Alcoholic strength	0,08	± 0,01		% Vol	0,07		DM 12/03/1986 SO GU n 161 14/07/1986 Met V pag 95, OIV- MA-AS312-01 Met B R2021	06/10/2022/ 12/10/2022
Brookfield rotational viscosity	Brookfield Rotational Viscosity (T = 20 ° C, impeller R5)						* VISCOSITA 2018 Rev.1	06/10/2022/ 12/10/2022
	50 rpm: 5257 cP (mPa.s) 30 rpm: 7132 cP (mPa.s) 20 rpm: 9153 cP (mPa.s)							
pH	2,97	± 0,04		unità	0,50		07(S77) 2012 Rev.1 - Potentiometric	06/10/2022/ 07/10/2022
<b>SUGARS HPAEC-PAD</b>								
Galactose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	06/10/2022/ 12/10/2022
Glucose, anhydrous (HPLC)	18,4	± 1,3		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	06/10/2022/ 12/10/2022
Sucrose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	06/10/2022/ 12/10/2022
Fructose, anhydrous (HPLC)	19,6	± 1,4		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	06/10/2022/ 12/10/2022
Lactose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	06/10/2022/ 12/10/2022
Maltose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	06/10/2022/ 12/10/2022
Citric acid (LCMS) [331]	106	± 17		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	06/10/2022/ 11/10/2022
Lactic Acid (LCMS) [331]	139	± 21		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	06/10/2022/ 11/10/2022
Malic acid (LCMS) [331]	442	± 56		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	06/10/2022/ 11/10/2022
Succinic acid (LCMS) [331]	265	± 37		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	06/10/2022/ 11/10/2022
Tartaric acid (LCMS) [331]	342	± 45		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	06/10/2022/ 11/10/2022
Ochratoxin A	< LQ			µg/kg	0,10		03(S130) 2022 Rev.13 - LC- MS/MS	06/10/2022/ 11/10/2022

**END TEST REPORT**

The original document is a PDF file with Digital Signature: 22P03162-In-0-S20Z-4000 - T10 - UNIMORE-DigitalSignature.pdf

**Next page...**

MODENA, li 20/10/2022

Sample arrived on the 05/10/2022

Registration date 05/10/2022

## TEST REPORT nr. 22P03162-In-0

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**

**SAMPLE 22P03162**

**MATRIX: Fruit and vegetable concentrates, and  
by-products**

### Notes and method reference:

< LQ: = lower than Quantification Limit.

U: the reported uncertainty is the expanded uncertainty calculated using a coverage factor equal to 2 which gives a reliability of approximately 95%. Please note that results expressed as '<LQ' may not indicate the absence of the searched parameters in the sample.

MICROBIOLOGICAL TESTS: for food and environmental samples, the extended measurement uncertainty was estimated according to ISO 19036:2019 Standard and is based on a standard uncertainty multiplied by a coverage factor of K = 2, providing a confidence level of approximately 95%. The combined standard uncertainty was assumed to be equal to the standard deviation of intra-laboratory reproducibility. The results of the microbiological tests are calculated according to the ISO 7218: 2007 / Amd 1: 2013 Standard. If the results are reported as <4 (CFU/ml) or <40 (CFU/g), this means that the microorganisms are present in the sample but in amounts less than 4 CFU/ml or 40 CFU/g respectively. For microbiological analyses unless differently reported in the individual test methods, in case of analytical steps foreseen in non-activity days of the laboratory, provisions of the ISO 7218: 2007 / Amd.1 2013 Standard (points 11.2 and 10.2.5) or from specific test methods are applied. In the case of quantitative microbiological tests, these have been set up on a single plate according to ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise expressly requested by current regulations.

In the case of quantitative microbiological tests, these have been set up on a single plate in accordance with ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise explicitly required by current regulations.

For waters, the measurement uncertainty corresponds to the confidence interval calculated according to ISO 8199: 2018 or to the expanded measurement uncertainty estimated according to ISO 29201: 2012. The results are issued in accordance with ISO 8199: 2018. When the number of colonies detected is <3, the result is expressed as "Microorganisms present in the analyzed volume (N ° colonies detected <3 CFU - reference ISO 8199: 2018, paragraph 9.1.8.4.1)".

LQ: Quantification Limit. It is the lowest analyte concentration which can be detected at an acceptable precision (repeatability) and accuracy, under well defined conditions.

LD: Detection Limit. It is the lowest analyte concentration which can be detected but not necessarily quantified, under well defined conditions. Any fields not filled in the Test Report are to be considered not applicable.

Conformity evaluation: values not complying with laws, decrees, national and EU regulations or specifications supplied by the customer are evaluated case by case, also taking into consideration the uncertainty of measure for each single test and the regulations on rounding-off of values, and pointed out when considered as non conform.

Rec %: Recovery % "+" means that the recovery has been applied to the result. The numeric results between brackets (..) after the expression <LQ are purely indicative of traces that cannot be exactly quantified.

In the case of sampling carried out by Neotron, the laboratory applies the Internal Operating Procedure code: NEOT-DIR/ 006/53.

The laboratory disclaims any responsibility for the information provided by the client reported in this Report which may influence the validity of the results.

Methods marked with an asterisk (\*) are not accredited by ACCREDIA (UNI CEI EN ISO/IEC 17025). The sampling activity is not included within the Scope of Accreditation of Neotron SPA

### NOTES OF PARAMETERS:

[331]: Extended uncertainty calculated following the equation of HORWITZ by using a coverage factor of 2, which guarantee a reliance level of 95%

TEST REPORT VALID FOR ALL LEGAL PURPOSES (Italian R.D. 1-3-1928 n°842 (article 16), - Italian Law 19-7-1957 n°679 articles 16 and 18, Italian Ministerial Decree 25-3-1986).

DATA and SAMPLE STORAGE: Test Reports, Raw data, chromatographic paths and instrumental reports are stored for 5 years. One control sample is stored for 2 months.

Data expressed in this test report refer only to the sample tested in the laboratory. The results reported in this Test Report refer to the sample as received. The description or any other reference concerning the sample are declared by the customer. This Test Report cannot be reproduced except in full. Partial reproductions must be authorized in writing by our laboratory.

THE LABORATORY DIRECTOR: DR. ANDREA RIZZO

THE CHEMIST AUTHORIZED TO SIGN THE TEST REPORTS: DR. MARCO MESCHIARI

NEOTRON SpA - With Sole Shareholder

Stradello Aggazzotti, 104

41126 MODENA - ITALY - Fiscal Code and VAT n° 03807840362

Tel: +39 059461711 - Fax: +39 059461777

www.neotron.it - neotron@neotron.it

Laboratorio Qualificato D.M. 26-2-87 Art. 4 - Legge 46/82 per la Ricerca Applicata e Innovazione Tecnologica.

Regione Emilia Romagna - AUTORIZZAZIONE Autocontrollo N° 008/MO/008

BNN-Monitoring Fruit and Vegetables Approved Laboratory

I-Monitoring EDEKAAG Fruit and Vegetables Registered Laboratory

GMP+ code: GMP051757



MODENA, li 24/10/2022

Sample arrived on the 05/10/2022  
Registration date 05/10/2022**TEST REPORT nr. 22P03163-In-0**

CUSTOMER

**ACETFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO****SAMPLE 22P03163****MATRIX: Fruit and vegetable concentrates, and  
by-products**Description provided by Customer: GLASSA CON ABM IGP - CONTAMINAZIONE ZERO - VISCOSITA': ~ 4.000 CPS (T=0G) -  
CODICE: S20Z-4000Richiesta via Internet n° N0384/22 - 28/09/2022 14:04:08. - Campionamento eseguito da: Committente - Trasporto effettuato da:  
Corriere  
Sample Condition on Receipt: 20°C

ANALYSIS DESCRIPTION	RESULT	U	REC. %	UNIT OF MEASURE	LQ	LD	METHOD	ANALYSES BEGINNING DATE / ENDING DATE
Water activity (Aw) at 25 °C	0,927	± 0,005			0,150		06(S135) rev6 2021	11/10/2022/ 11/10/2022
<b>VINEGAR ANALYSIS</b>								
Reduction sugars (Reducing substances)	508,5	± 15,3		g/l	1,0		07(S240) 2021 Rev.0 - Titrimetrico	10/10/2022/ 12/10/2022
Total Acidity expressed as acetic acid	3,27	± 0,06		g/100 ml	0,10		* ACIDITA 2014 Rev.0 - Titrimetrico	10/10/2022/ 12/10/2022
Relative density at 20°C	1,207	± 0,001					07(S117) 2015 Rev.1 - Gravimetric	20/10/2022/ 20/10/2022
Alcoholic strength	0,08	± 0,01		% Vol	0,07		DM 12/03/1986 SO GU n 161 14/07/1986 Met V pag 95, OIV- MA-AS312-01 Met B R2021	10/10/2022/ 12/10/2022
pH	2,97	± 0,04		unità	0,50		07(S77) 2012 Rev.1 - Potentiometric	10/10/2022/ 21/10/2022
Brookfield rotational viscosity	Brookfield Rotational Viscosity (T = 20 ° C, impeller R4)							
	50 rpm: 2778 cP (mPa.s) 30 rpm: 3645 cP (mPa.s) 20 rpm: 4453 cP (mPa.s)							
<b>SUGARS HPAEC-PAD</b>								
Galactose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	11/10/2022/ 13/10/2022
Glucose, anhydrous (HPLC)	19,3	± 1,4		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	11/10/2022/ 13/10/2022
Sucrose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	11/10/2022/ 13/10/2022
Fructose, anhydrous (HPLC)	19,1	± 1,3		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	11/10/2022/ 13/10/2022
Lactose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	11/10/2022/ 13/10/2022
Maltose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	11/10/2022/ 13/10/2022
Citric acid (LCMS) [331]	46,1	± 8,3	93	mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	11/10/2022/ 18/10/2022
Lactic Acid (LCMS) [331]	207	± 30	64 +	mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	11/10/2022/ 18/10/2022
Malic acid (LCMS) [331]	523	± 65	83	mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	11/10/2022/ 18/10/2022
Succinic acid (LCMS) [331]	189	± 27	83	mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	11/10/2022/ 18/10/2022
Tartaric acid (LCMS) [331]	473	± 60	79	mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	11/10/2022/ 18/10/2022
Ochratoxin A	< LQ			µg/kg	0,10		03(S130) 2022 Rev.13 - LC- MS/MS	11/10/2022/ 13/10/2022

**END TEST REPORT**

The original document is a PDF file with Digital Signature: 22P03163-In-0-S20Z-4000 - T0 - UNIMORE-DigitalSignature.pdf

**Next page...**

MODENA, li 24/10/2022

Sample arrived on the 05/10/2022

Registration date 05/10/2022

## TEST REPORT nr. 22P03163-In-0

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**

**SAMPLE 22P03163**

**MATRIX: Fruit and vegetable concentrates, and by-products**

### Notes and method reference:

< LQ: = lower than Quantification Limit.

U: the reported uncertainty is the expanded uncertainty calculated using a coverage factor equal to 2 which gives a reliability of approximately 95%. Please note that results expressed as '<LQ' may not indicate the absence of the searched parameters in the sample.

MICROBIOLOGICAL TESTS: for food and environmental samples, the extended measurement uncertainty was estimated according to ISO 19036:2019 Standard and is based on a standard uncertainty multiplied by a coverage factor of  $K = 2$ , providing a confidence level of approximately 95%. The combined standard uncertainty was assumed to be equal to the standard deviation of intra-laboratory reproducibility. The results of the microbiological tests are calculated according to the ISO 7218: 2007 / Amd 1: 2013 Standard. If the results are reported as <4 (CFU/ml) or <40 (CFU/g), this means that the microorganisms are present in the sample but in amounts less than 4 CFU/ml or 40 CFU/g respectively. For microbiological analyses unless differently reported in the individual test methods, in case of analytical steps foreseen in non-activity days of the laboratory, provisions of the ISO 7218: 2007 / Amd.1 2013 Standard (points 11.2 and 10.2.5) or from specific test methods are applied. In the case of quantitative microbiological tests, these have been set up on a single plate according to ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise expressly requested by current regulations.

In the case of quantitative microbiological tests, these have been set up on a single plate in accordance with ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise explicitly required by current regulations.

For waters, the measurement uncertainty corresponds to the confidence interval calculated according to ISO 8199: 2018 or to the expanded measurement uncertainty estimated according to ISO 29201: 2012. The results are issued in accordance with ISO 8199: 2018. When the number of colonies detected is <3, the result is expressed as "Microorganisms present in the analyzed volume (N ° colonies detected <3 CFU - reference ISO 8199: 2018, paragraph 9.1.8.4.1)".

LQ: Quantification Limit. It is the lowest analyte concentration which can be detected at an acceptable precision (repeatability) and accuracy, under well defined conditions.

LD: Detection Limit. It is the lowest analyte concentration which can be detected but not necessarily quantified, under well defined conditions. Any fields not filled in the Test Report are to be considered not applicable.

Conformity evaluation: values not complying with laws, decrees, national and EU regulations or specifications supplied by the customer are evaluated case by case, also taking into consideration the uncertainty of measure for each single test and the regulations on rounding-off of values, and pointed out when considered as non conform.

Rec %: Recovery % "+" means that the recovery has been applied to the result. The numeric results between brackets (.) after the expression <LQ are purely indicative of traces that cannot be exactly quantified.

In the case of sampling carried out by Neotron, the laboratory applies the Internal Operating Procedure code: NEOT-DIR/ 006/53.

The laboratory disclaims any responsibility for the information provided by the client reported in this Report which may influence the validity of the results.

Methods marked with an asterisk (\*) are not accredited by ACCREDIA (UNI CEI EN ISO/IEC 17025). The sampling activity is not included within the Scope of Accreditation of Neotron SPA

### NOTES OF PARAMETERS:

[331]: Extended uncertainty calculated following the equation of HORWITZ by using a coverage factor of 2, which guarantee a reliance level of 95%

TEST REPORT VALID FOR ALL LEGAL PURPOSES (Italian R.D. 1-3-1928 n°842 (article 16), - Italian Law 19-7-1957 n°679 articles 16 and 18, Italian Ministerial Decree 25-3-1986).

DATA and SAMPLE STORAGE: Test Reports, Raw data, chromatographic paths and instrumental reports are stored for 5 years. One control sample is stored for 2 months.

Data expressed in this test report refer only to the sample tested in the laboratory. The results reported in this Test Report refer to the sample as received. The description or any other reference concerning the sample are declared by the customer. This Test Report cannot be reproduced except in full. Partial reproductions must be authorized in writing by our laboratory.

THE LABORATORY DIRECTOR: DR. ANDREA RIZZO

THE CHEMIST AUTHORIZED TO SIGN THE TEST REPORTS: DR. MARCO MESCHIARI

NEOTRON SpA - With Sole Shareholder

Stradello Aggazzotti, 104

41126 MODENA - ITALY - Fiscal Code and VAT n° 03807840362

Tel: +39 059461711 - Fax: +39 059461777

www.neotron.it - neotron@neotron.it

Laboratorio Qualificato D.M. 26-2-87 Art. 4 - Legge 46/82 per la Ricerca Applicata e Innovazione Tecnologica.

Regione Emilia Romagna - AUTORIZZAZIONE Autocontrollo N° 008/MO/008

BNN-Monitoring Fruit and Vegetables Approved Laboratory

I-Monitoring EDEKAAG Fruit and Vegetables Registered Laboratory

GMP+ code: GMP051757

MODENA, li 13/10/2022

Sample arrived on the 23/09/2022

Registration date 23/09/2022

**TEST REPORT nr. 22M17704-In-0**

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**
**SAMPLE 22M17704****MATRIX: Fruit and vegetable concentrates, and by-products**

Description provided by Customer: GLASSA CON ABM IGP - CONTAMINAZIONE ZERO - VISCOSITA': ~ 3.000 CPS (T=20G) - CODICE: S20Z-3000

Richiesta via Internet n° N0331/22 - 05/09/2022 13:57:35. - Campionamento eseguito da: Committente - Trasporto effettuato da: Committente  
Sample Condition on Receipt: 20°C

ANALYSIS DESCRIPTION	RESULT	U	REC. %	UNIT OF MEASURE	LQ	LD	METHOD	ANALYSES BEGINNING DATE / ENDING DATE
Water activity (Aw) at 25 °C	0,935	± 0,005			0,150		06(S135) rev6 2021	23/09/2022 / 30/09/2022
<b>VINEGAR ANALYSIS</b>								
Reduction sugars (Reducing substances)	436,4	± 13,1		g/l	1,0		07(S240) 2021 Rev.0 - Titrimetrico	24/09/2022 / 30/09/2022
Total Acidity expressed as acetic acid	2,89	± 0,06		g/100 ml	0,10		* ACIDITA 2014 Rev.0 - Titrimetrico	24/09/2022 / 29/09/2022
Relative density at 20°C	1,187	± 0,001					DM 12/03/1986 SO GU n 161 14/07/1986 Met VI pag 95 p.to 4, OIV-MA-AS2-01A Met B R2021 - densimetry	24/09/2022 / 30/09/2022
Alcoholic strength	0,09	± 0,01		% Vol	0,07		DM 12/03/1986 SO GU n 161 14/07/1986 Met V pag 95, OIV-MA-AS312-01 Met B R2021	24/09/2022 / 30/09/2022
pH	3,01	± 0,04		unità	0,50		07(S77) 2012 Rev.1 - Potentiometric	24/09/2022 / 13/10/2022
Brookfield rotational viscosity	Brookfield Rotational Viscosity (T = 20 ° C, impeller R3)						* VISCOSITA 2018 Rev.1	24/09/2022 / 06/10/2022
	30 rpm: 2159 cP (mPa.s) 20 rpm: 2645 cP (mPa.s) 12 rpm: 3402 cP (mPa.s)							
<b>SUGARS HPAEC-PAD</b>								
Galactose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	24/09/2022 / 12/10/2022
Glucose, anhydrous (HPLC)	16,9	± 1,2		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	24/09/2022 / 12/10/2022
Sucrose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	24/09/2022 / 12/10/2022
Fructose, anhydrous (HPLC)	20,0	± 1,4		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	24/09/2022 / 12/10/2022
Lactose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	24/09/2022 / 12/10/2022
Maltose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	24/09/2022 / 12/10/2022
Citric acid (LCMS) [331]	69,4	± 11,7		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	24/09/2022 / 06/10/2022
Lactic Acid (LCMS) [331]	287	± 39		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	24/09/2022 / 06/10/2022
Malic acid (LCMS) [331]	602	± 73		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	24/09/2022 / 06/10/2022
Succinic acid (LCMS) [331]	190	± 28		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	24/09/2022 / 06/10/2022
Tartaric acid (LCMS) [331]	562	± 69		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	24/09/2022 / 06/10/2022
Ochratoxin A	< LQ			µg/kg	0,10		03(S130) 2022 Rev.13 - LC-MS/MS	24/09/2022 / 05/10/2022

**END TEST REPORT**

The original document is a PDF file with Digital Signature: 22M17704-In-0-S20Z-3000 - T20 - UNIMORE-DigitalSignature.pdf

MODENA, li 13/10/2022

Sample arrived on the 23/09/2022

Registration date 23/09/2022

## TEST REPORT nr. 22M17704-In-0

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**

**SAMPLE 22M17704**

**MATRIX: Fruit and vegetable concentrates, and  
by-products**

### Notes and method reference:

< LQ: = lower than Quantification Limit.

U: the reported uncertainty is the expanded uncertainty calculated using a coverage factor equal to 2 which gives a reliability of approximately 95%. Please note that results expressed as '<LQ' may not indicate the absence of the searched parameters in the sample.

MICROBIOLOGICAL TESTS: for food and environmental samples, the extended measurement uncertainty was estimated according to ISO 19036:2019 Standard and is based on a standard uncertainty multiplied by a coverage factor of K = 2, providing a confidence level of approximately 95%. The combined standard uncertainty was assumed to be equal to the standard deviation of intra-laboratory reproducibility. The results of the microbiological tests are calculated according to the ISO 7218: 2007 / Amd 1: 2013 Standard. If the results are reported as <4 (CFU/ml) or <40 (CFU/g), this means that the microorganisms are present in the sample but in amounts less than 4 CFU/ml or 40 CFU/g respectively. For microbiological analyses unless differently reported in the individual test methods, in case of analytical steps foreseen in non-activity days of the laboratory, provisions of the ISO 7218: 2007 / Amd.1 2013 Standard (points 11.2 and 10.2.5) or from specific test methods are applied. In the case of quantitative microbiological tests, these have been set up on a single plate according to ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise expressly requested by current regulations.

In the case of quantitative microbiological tests, these have been set up on a single plate in accordance with ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise explicitly required by current regulations.

For waters, the measurement uncertainty corresponds to the confidence interval calculated according to ISO 8199: 2018 or to the expanded measurement uncertainty estimated according to ISO 29201: 2012. The results are issued in accordance with ISO 8199: 2018. When the number of colonies detected is <3, the result is expressed as "Microorganisms present in the analyzed volume (N ° colonies detected <3 CFU - reference ISO 8199: 2018, paragraph 9.1.8.4.1)".

LQ: Quantification Limit. It is the lowest analyte concentration which can be detected at an acceptable precision (repeatability) and accuracy, under well defined conditions.

LD: Detection Limit. It is the lowest analyte concentration which can be detected but not necessarily quantified, under well defined conditions. Any fields not filled in the Test Report are to be considered not applicable.

Conformity evaluation: values not complying with laws, decrees, national and EU regulations or specifications supplied by the customer are evaluated case by case, also taking into consideration the uncertainty of measure for each single test and the regulations on rounding-off of values, and pointed out when considered as non conform.

Rec %: Recovery % "+" means that the recovery has been applied to the result. The numeric results between brackets (.) after the expression <LQ are purely indicative of traces that cannot be exactly quantified.

In the case of sampling carried out by Neotron, the laboratory applies the Internal Operating Procedure code: NEOT-DIR/ 006/53.

The laboratory disclaims any responsibility for the information provided by the client reported in this Report which may influence the validity of the results.

Methods marked with an asterisk (\*) are not accredited by ACCREDIA (UNI CEI EN ISO/IEC 17025). The sampling activity is not included within the Scope of Accreditation of Neotron SPA

### NOTES OF PARAMETERS:

[331]: Extended uncertainty calculated following the equation of HORWITZ by using a coverage factor of 2, which guarantee a reliance level of 95%

TEST REPORT VALID FOR ALL LEGAL PURPOSES (Italian R.D. 1-3-1928 n°842 (article 16), - Italian Law 19-7-1957 n°679 articles 16 and 18, Italian Ministerial Decree 25-3-1986).

DATA and SAMPLE STORAGE: Test Reports, Raw data, chromatographic paths and instrumental reports are stored for 5 years. One control sample is stored for 2 months.

Data expressed in this test report refer only to the sample tested in the laboratory. The results reported in this Test Report refer to the sample as received. The description or any other reference concerning the sample are declared by the customer. This Test Report cannot be reproduced except in full. Partial reproductions must be authorized in writing by our laboratory.

THE LABORATORY DIRECTOR: DR. ANDREA RIZZO

THE CHEMIST AUTHORIZED TO SIGN THE TEST REPORTS: DR. MARCO MESCHIARI

NEOTRON SpA - With Sole Shareholder

Stradello Aggazzotti, 104

41126 MODENA - ITALY - Fiscal Code and VAT n° 03807840362

Tel: +39 059461711 - Fax: +39 059461777

www.neotron.it - neotron@neotron.it

Laboratorio Qualificato D.M. 26-2-87 Art. 4 - Legge 46/82 per la Ricerca Applicata e Innovazione Tecnologica.

Regione Emilia Romagna - AUTORIZZAZIONE Autocontrollo N° 008/MO/008

BNN-Monitoring Fruit and Vegetables Approved Laboratory

I-Monitoring EDEKAAG Fruit and Vegetables Registered Laboratory

GMP+ code: GMP051757

MODENA, li 26/09/2022

Sample arrived on the 13/09/2022  
Registration date 13/09/2022**TEST REPORT nr. 22M08845-In-0**

CUSTOMER

**ACETFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO****SAMPLE 22M08845****MATRIX: Fruit and vegetable concentrates, and by-products**

Description provided by Customer: GLASSA CON ABM IGP - CONTAMINAZIONE ZERO - VISCOSITA': ~ 3.000 CPS (T=10G) - CODICE: S20Z-3000

Richiesta via Internet n° N0332/22 - 05/09/2022 13:57:35. - Campionamento eseguito da: Committente - Trasporto effettuato da: Corriere  
Sample Condition on Receipt: 23°C

ANALYSIS DESCRIPTION	RESULT	U	REC. %	UNIT OF MEASURE	LQ	LD	METHOD	ANALYSES BEGINNING DATE / ENDING DATE
Water activity (Aw) at 25 °C	0,934	± 0,005			0,150		06(S135) rev6 2021	14/09/2022 / 20/09/2022
<b>VINEGAR ANALYSIS</b>								
Reduction sugars (Reducing substances)	441,2	± 13,2		g/l	1,0		07(S240) 2021 Rev.0 - Titrimetrico	16/09/2022 / 20/09/2022
Total Acidity expressed as acetic acid	2,95	± 0,06		g/100 ml	0,10		* ACIDITA 2014 Rev.0 - Titrimetrico	16/09/2022 / 20/09/2022
Relative density at 20°C	1,190	± 0,001					DM 12/03/1986 SO GU n 161 14/07/1986 Met VI pag 95 p.to 4, OIV-MA-AS2-01A Met B R2021 - densimetry	16/09/2022 / 20/09/2022
Alcoholic strength	0,09	± 0,01		% Vol	0,07		DM 12/03/1986 SO GU n 161 14/07/1986 Met V pag 95, OIV-MA-AS312-01 Met B R2021	16/09/2022 / 20/09/2022
pH	3,00	± 0,04		unità	0,50		07(S77) 2012 Rev.1 - Potentiometric	16/09/2022 / 23/09/2022
Brookfield rotational viscosity	Brookfield Rotational Viscosity (T = 20 ° C, impeller R3)						* VISCOSITA 2018 Rev.1	16/09/2022 / 21/09/2022
	30 rpm: 2181 cP (mPa.s) 20 rpm: 2689 cP (mPa.s) 12 rpm: 3546 cP (mPa.s)							
<b>SUGARS HPAEC-PAD</b>								
Galactose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	16/09/2022 / 20/09/2022
Glucose, anhydrous (HPLC)	17,9	± 1,3		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	16/09/2022 / 20/09/2022
Sucrose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	16/09/2022 / 20/09/2022
Fructose, anhydrous (HPLC)	17,8	± 1,2		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	16/09/2022 / 20/09/2022
Lactose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	16/09/2022 / 20/09/2022
Maltose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	16/09/2022 / 20/09/2022
Citric acid (LCMS) [331]	79,2	± 13,1		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	16/09/2022 / 23/09/2022
Lactic Acid (LCMS) [331]	476	± 60		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	16/09/2022 / 23/09/2022
Malic acid (LCMS) [331]	496	± 62		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	16/09/2022 / 23/09/2022
Succinic acid (LCMS) [331]	224	± 32		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	16/09/2022 / 23/09/2022
Tartaric acid (LCMS) [331]	534	± 66		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	16/09/2022 / 23/09/2022
Ochratoxin A	< LQ			µg/kg	0,10		03(S130) 2022 Rev.13 - LC-MS/MS	16/09/2022 / 20/09/2022

**END TEST REPORT**

The original document is a PDF file with Digital Signature: 22M08845-In-0-S20Z-3000 - T10 - UNIMORE-DigitalSignature.pdf

MODENA, li 26/09/2022

Sample arrived on the 13/09/2022

Registration date 13/09/2022

## TEST REPORT nr. 22M08845-In-0

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**

**SAMPLE 22M08845**

**MATRIX: Fruit and vegetable concentrates, and by-products**

### Notes and method reference:

< LQ: = lower than Quantification Limit.

U: the reported uncertainty is the expanded uncertainty calculated using a coverage factor equal to 2 which gives a reliability of approximately 95%. Please note that results expressed as <LQ> may not indicate the absence of the searched parameters in the sample.

MICROBIOLOGICAL TESTS: for food and environmental samples, the extended measurement uncertainty was estimated according to ISO 19036:2019 Standard and is based on a standard uncertainty multiplied by a coverage factor of K = 2, providing a confidence level of approximately 95%. The combined standard uncertainty was assumed to be equal to the standard deviation of intra-laboratory reproducibility. The results of the microbiological tests are calculated according to the ISO 7218: 2007 / Amd 1: 2013 Standard. If the results are reported as <4 (CFU/ml) or <40 (CFU/g), this means that the microorganisms are present in the sample but in amounts less than 4 CFU/ml or 40 CFU/g respectively. For microbiological analyses unless differently reported in the individual test methods, in case of analytical steps foreseen in non-activity days of the laboratory, provisions of the ISO 7218: 2007 / Amd.1 2013 Standard (points 11.2 and 10.2.5) or from specific test methods are applied. In the case of quantitative microbiological tests, these have been set up on a single plate according to ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise expressly requested by current regulations.

In the case of quantitative microbiological tests, these have been set up on a single plate in accordance with ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise explicitly required by current regulations.

For waters, the measurement uncertainty corresponds to the confidence interval calculated according to ISO 8199: 2018 or to the expanded measurement uncertainty estimated according to ISO 29201: 2012. The results are issued in accordance with ISO 8199: 2018. When the number of colonies detected is <3, the result is expressed as "Microorganisms present in the analyzed volume (N ° colonies detected <3 CFU - reference ISO 8199: 2018, paragraph 9.1.8.4.1)".

LQ: Quantification Limit. It is the lowest analyte concentration which can be detected at an acceptable precision (repeatability) and accuracy, under well defined conditions.

LD: Detection Limit. It is the lowest analyte concentration which can be detected but not necessarily quantified, under well defined conditions. Any fields not filled in in the Test Report are to be considered not applicable.

Conformity evaluation: values not complying with laws, decrees, national and EU regulations or specifications supplied by the customer are evaluated case by case, also taking into consideration the uncertainty of measure for each single test and the regulations on rounding-off of values, and pointed out when considered as non conform.

Rec %: Recovery % "+" means that the recovery has been applied to the result. The numeric results between brackets (.) after the expression <LQ are purely indicative of traces that cannot be exactly quantified.

In the case of sampling carried out by Neotron, the laboratory applies the Internal Operating Procedure code: NEOT-DIR/ 006/53.

The laboratory disclaims any responsibility for the information provided by the client reported in this Report which may influence the validity of the results.

Methods marked with an asterisk (\*) are not accredited by ACCREDIA (UNI CEI EN ISO/IEC 17025). The sampling activity is not included within the Scope of Accreditation of Neotron SPA

### NOTES OF PARAMETERS:

[331]: Extended uncertainty calculated following the equation of HORWITZ by using a coverage factor of 2, which guarantee a reliance level of 95%

TEST REPORT VALID FOR ALL LEGAL PURPOSES (Italian R.D. 1-3-1928 n°842 (article 16), – Italian Law 19-7-1957 n°679 articles 16 and 18, Italian Ministerial Decree 25-3-1986).

DATA and SAMPLE STORAGE: Test Reports, Raw data, chromatographic paths and instrumental reports are stored for 5 years. One control sample is stored for 2 months.

Data expressed in this test report refer only to the sample tested in the laboratory. The results reported in this Test Report refer to the sample as received. The description or any other reference concerning the sample are declared by the customer. This Test Report cannot be reproduced except in full. Partial reproductions must be authorized in writing by our laboratory.

THE LABORATORY DIRECTOR: DR. ANDREA RIZZO

THE CHEMIST AUTHORIZED TO SIGN THE TEST REPORTS: DR. BARBARA MALAGOLI

Approved by Analysis Manager - laboratory LMAA-Nut

Approved by Analysis Manager - laboratory ICS-HPLC

Approved by Analysis Manager - laboratory LC-MICO

Approved by Analysis Manager - laboratory LC-AN

Approved by Analysis Manager - laboratory FISICHE

Approved by Analysis Manager - laboratory LBM-EL

Approved by Analysis Manager - laboratory LMAA-Bro

**NEOTRON SpA** - With Sole Shareholder

Stradello Aggazzotti, 104

41126 MODENA - ITALY - Fiscal Code and VAT n° 03807840362

Tel: +39 059461711 - Fax: +39 059461777

www.neotron.it - neotron@neotron.it

Laboratorio Qualificato D.M. 26-2-87 Art. 4 - Legge 46/82 per la Ricerca Applicata e Innovazione Tecnologica.

Regione Emilia Romagna - AUTORIZZAZIONE Autocontrollo N° 008/MO/008

BNN-Monitoring Fruit and Vegetables Approved Laboratory

I-Monitoring EDEKAAG Fruit and Vegetables Registered Laboratory

GMP+ code: GMP051757

MODENA, li 13/09/2022

Sample arrived on the 05/09/2022

Registration date 05/09/2022

**TEST REPORT nr. 22M02962-In-0**

CUSTOMER

**ACETFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**
**SAMPLE 22M02962****MATRIX: Fruit and vegetable concentrates, and by-products**

Description provided by Customer: GLASSA CON ABM IGP - CONTAMINAZIONE ZERO - VISCOSITA': ~ 3.000 CPS (T=0G) - CODICE: S20Z-3000

Richiesta via Internet n° N0333/22 - 05/09/2022 13:57:35. - Campionamento eseguito da: Committente - Trasporto effettuato da: Committente

Sample Condition on Receipt: refrigerated

ANALYSIS DESCRIPTION	RESULT	U	REC. %	UNIT OF MEASURE	LQ	LD	METHOD	ANALYSES BEGINNING DATE / ENDING DATE
Water activity (Aw) at 25 °C	0,940	± 0,005			0,150		06(S135) rev6 2021	06/09/2022 / 12/09/2022
<b>VINEGAR ANALYSIS</b>								
Reduction sugars (Reducing substances)	446,8	± 13,4		g/l	1,0		07(S240) 2021 Rev.0 - Titrimetric	07/09/2022 / 09/09/2022
Total Acidity expressed as acetic acid	2,79	± 0,05		g/100 ml	0,10		* ACIDITA 2014 Rev.0 - Titrimetric	07/09/2022 / 12/09/2022
Relative density at 20°C	1,181	± 0,001					07(S117) 2015 Rev.1 - Gravimetric	13/09/2022 / 13/09/2022
Alcoholic strength	0,07	± 0,01		% Vol	0,07		DM 12/03/1986 SO GU n 161 14/07/1986 Met V pag 95, OIV-MA-AS312-01 Met B R2021	07/09/2022 / 09/09/2022
pH	2,97	± 0,04		unità	0,50		07(S77) 2012 Rev.1 - Potentiometric	07/09/2022 / 12/09/2022
Brookfield rotational viscosity	Brookfield Rotational Viscosity (T = 20 ° C, impeller R3)						* VISCOSITA 2018 Rev.1	07/09/2022 / 12/09/2022
	30 rpm: 2012 cP (mPa.s)							
	20 rpm: 2446 cP (mPa.s)							
	12 rpm: 3144 cP (mPa.s)							
<b>SUGARS HPAEC-PAD</b>								
Galactose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	07/09/2022 / 13/09/2022
Glucose, anhydrous (HPLC)	17,2	± 1,2		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	07/09/2022 / 13/09/2022
Sucrose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	07/09/2022 / 13/09/2022
Fructose, anhydrous (HPLC)	17,0	± 1,2		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	07/09/2022 / 13/09/2022
Lactose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	07/09/2022 / 13/09/2022
Maltose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	07/09/2022 / 13/09/2022
Citric acid (LCMS) [331]	84,8	± 13,9		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	07/09/2022 / 09/09/2022
Lactic Acid (LCMS) [331]	248	± 35		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	07/09/2022 / 09/09/2022
Malic acid (LCMS) [331]	573	± 70		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	07/09/2022 / 09/09/2022
Succinic acid (LCMS) [331]	210	± 30		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	07/09/2022 / 09/09/2022
Tartaric acid (LCMS) [331]	608	± 74		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	07/09/2022 / 09/09/2022
Ochratoxin A	< LQ			µg/kg	0,10		03(S130) 2022 Rev.13 - LC-MS/MS	07/09/2022 / 09/09/2022

**END TEST REPORT**

The original document is a PDF file with Digital Signature: 22M02962-In-0-S20Z-3000 - T0 - UNIMORE-DigitalSignature.pdf

**Next page...**

MODENA, li 13/09/2022

Sample arrived on the 05/09/2022

Registration date 05/09/2022

## TEST REPORT nr. 22M02962-In-0

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**

**SAMPLE 22M02962**

**MATRIX: Fruit and vegetable concentrates, and by-products**

### Notes and method reference:

< LQ: = lower than Quantification Limit.

U: the reported uncertainty is the expanded uncertainty calculated using a coverage factor equal to 2 which gives a reliability of approximately 95%. Please note that results expressed as '<LQ' may not indicate the absence of the searched parameters in the sample.

MICROBIOLOGICAL TESTS: for food and environmental samples, the extended measurement uncertainty was estimated according to ISO 19036:2019 Standard and is based on a standard uncertainty multiplied by a coverage factor of K = 2, providing a confidence level of approximately 95%. The combined standard uncertainty was assumed to be equal to the standard deviation of intra-laboratory reproducibility. The results of the microbiological tests are calculated according to the ISO 7218: 2007 / Amd 1: 2013 Standard. If the results are reported as <4 (CFU/ml) or <40 (CFU/g), this means that the microorganisms are present in the sample but in amounts less than 4 CFU/ml or 40 CFU/g respectively. For microbiological analyses unless differently reported in the individual test methods, in case of analytical steps foreseen in non-activity days of the laboratory, provisions of the ISO 7218: 2007 / Amd.1 2013 Standard (points 11.2 and 10.2.5) or from specific test methods are applied. In the case of quantitative microbiological tests, these have been set up on a single plate according to ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise expressly requested by current regulations.

In the case of quantitative microbiological tests, these have been set up on a single plate in accordance with ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise explicitly required by current regulations.

For waters, the measurement uncertainty corresponds to the confidence interval calculated according to ISO 8199: 2018 or to the expanded measurement uncertainty estimated according to ISO 29201: 2012. The results are issued in accordance with ISO 8199: 2018. When the number of colonies detected is <3, the result is expressed as "Microorganisms present in the analyzed volume (N ° colonies detected <3 CFU - reference ISO 8199: 2018, paragraph 9.1.8.4.1)".

LQ: Quantification Limit. It is the lowest analyte concentration which can be detected at an acceptable precision (repeatability) and accuracy, under well defined conditions.

LD: Detection Limit. It is the lowest analyte concentration which can be detected but not necessarily quantified, under well defined conditions.

Conformity evaluation: values not complying with laws, decrees, national and EU regulations or specifications supplied by the customer are evaluated case by case, also taking into consideration the uncertainty of measure for each single test and the regulations on rounding-off of values, and pointed out when considered as non conform.

Rec %: Recovery % "+" means that the recovery has been applied to the result. The numeric results between brackets (..) after the expression <LQ are purely indicative of traces that cannot be exactly quantified.

In the case of sampling carried out by Neotron, the laboratory applies the Internal Operating Procedure code: NEOT-DIR/ 006/53.

The laboratory disclaims any responsibility for the information provided by the client reported in this Report which may influence the validity of the results.

Methods marked with an asterisk (\*) are not accredited by ACCREDIA (UNI CEI EN ISO/IEC 17025). The sampling activity is not included within the Scope of Accreditation of Neotron SPA

### NOTES OF PARAMETERS:

[331]: Extended uncertainty calculated following the equation of HORWITZ by using a coverage factor of 2, which guarantee a reliance level of 95%

TEST REPORT VALID FOR ALL LEGAL PURPOSES (Italian R.D. 1-3-1928 n°842 (article 16), - Italian Law 19-7-1957 n°679 articles 16 and 18, Italian Ministerial Decree 25-3-1986).

DATA and SAMPLE STORAGE: Test Reports, Raw data, chromatographic paths and instrumental reports are stored for 5 years. One control sample is stored for 2 months.

Data expressed in this test report refer only to the sample tested in the laboratory. The results reported in this Test Report refer to the sample as received. The description or any other reference concerning the sample are declared by the customer. This Test Report cannot be reproduced except in full. Partial reproductions must be authorized in writing by our laboratory.

THE LABORATORY DIRECTOR: DR. ANDREA RIZZO

THE CHEMIST AUTHORIZED TO SIGN THE TEST REPORTS: DR. MARCO MESCHIARI

Approved by Analysis Manager - laboratory LMAA-Nut  
Approved by Analysis Manager - laboratory ICS-HPLC  
Approved by Analysis Manager - laboratory LC-MICO  
Approved by Analysis Manager - laboratory LC-AN

Approved by Analysis Manager - laboratory FISICHE  
Approved by Analysis Manager - laboratory LBM-EL  
Approved by Analysis Manager - laboratory LMAA-Bro



MODENA, li 13/09/2022

Sample arrived on the 05/09/2022  
Registration date 05/09/2022**TEST REPORT nr. 22M02959-In-0**

CUSTOMER

**ACETFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO****SAMPLE 22M02959****MATRIX: Fruit and vegetable concentrates, and  
by-products**

Description provided by Customer: GLASSA CON ABM IGP - CONTAMINAZIONE ZERO - VISCOSITA': ~ 2.000 CPS (T=20G) - CODICE: S20Z-2000

Richiesta via Internet n° N0315/22 - 25/08/2022 09:11:13. - Campionamento eseguito da: Committente - Trasporto effettuato da: Committente  
Sample Condition on Receipt: Room temperature

ANALYSIS DESCRIPTION	RESULT	U	REC. %	UNIT OF MEASURE	LQ	LD	METHOD	ANALYSES BEGINNING DATE / ENDING DATE
Water activity (Aw) at 25 °C	0,940	± 0,005			0,150		06(S135) rev6 2021	06/09/2022 / 12/09/2022
<b>VINEGAR ANALYSIS</b>								
Reduction sugars (Reducing substances)	448,2	± 13,4		g/l	1,0		07(S240) 2021 Rev.0 - Titrimetrico	07/09/2022 / 09/09/2022
Total Acidity expressed as acetic acid	2,97	± 0,06		g/100 ml	0,10		* ACIDITA 2014 Rev.0 - Titrimetrico	07/09/2022 / 12/09/2022
Relative density at 20°C	1,183	± 0,001					07(S117) 2015 Rev.1 - Gravimetric	13/09/2022 / 13/09/2022
Alcoholic strength	< LQ			% Vol	0,07		DM 12/03/1986 SO GU n 161 14/07/1986 Met V pag 95, OIV-MA-AS312-01 Met B R2021	07/09/2022 / 09/09/2022
Brookfield rotational viscosity	Brookfield Rotational Viscosity (T = 20 ° C, impeller R3)						* VISCOSITA 2018 Rev.1	07/09/2022 / 12/09/2022
	50 rpm: 1390 cP (mPa.s) 30 rpm: 1764 cP (mPa.s) 20 rpm: 2117 cP (mPa.s)							
pH	3,00	± 0,04		unità	0,50		07(S77) 2012 Rev.1 - Potentiometric	07/09/2022 / 12/09/2022
<b>SUGARS HPAEC-PAD</b>								
Galactose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	07/09/2022 / 13/09/2022
Glucose, anhydrous (HPLC)	18,0	± 1,3		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	07/09/2022 / 13/09/2022
Sucrose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	07/09/2022 / 13/09/2022
Fructose, anhydrous (HPLC)	17,7	± 1,2		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	07/09/2022 / 13/09/2022
Lactose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	07/09/2022 / 13/09/2022
Maltose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	07/09/2022 / 13/09/2022
Citric acid (LCMS) [331]	97,7	± 15,7		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	07/09/2022 / 09/09/2022
Lactic Acid (LCMS) [331]	295	± 40		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	07/09/2022 / 09/09/2022
Malic acid (LCMS) [331]	665	± 80		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	07/09/2022 / 09/09/2022
Succinic acid (LCMS) [331]	241	± 34		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	07/09/2022 / 09/09/2022
Tartaric acid (LCMS) [331]	676	± 81		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	07/09/2022 / 09/09/2022
Ochratoxin A	< LQ			µg/kg	0,10		03(S130) 2022 Rev.13 - LC-MS/MS	07/09/2022 / 09/09/2022

**END TEST REPORT**

The original document is a PDF file with Digital Signature: 22M02959-In-0-S20Z-2000 - T20 - UNIMORE-DigitalSignature.pdf

**Next page...**

MODENA, li 13/09/2022

Sample arrived on the 05/09/2022  
Registration date 05/09/2022

**TEST REPORT nr. 22M02959-In-0**

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**

SAMPLE 22M02959

**MATRIX: Fruit and vegetable concentrates, and  
by-products**

## Notes and method reference:

&lt; LQ: = lower than Quantification Limit.

U: the reported uncertainty is the expanded uncertainty calculated using a coverage factor equal to 2 which gives a reliability of approximately 95%. Please note that results expressed as '<LQ' may not indicate the absence of the searched parameters in the sample.

MICROBIOLOGICAL TESTS: for food and environmental samples, the extended measurement uncertainty was estimated according to ISO 19036:2019 Standard and is based on a standard uncertainty multiplied by a coverage factor of K = 2, providing a confidence level of approximately 95%. The combined standard uncertainty was assumed to be equal to the standard deviation of intra-laboratory reproducibility. The results of the microbiological tests are calculated according to the ISO 7218: 2007 / Amd 1: 2013 Standard. If the results are reported as <4 (CFU/ml) or <40 (CFU/g), this means that the microorganisms are present in the sample but in amounts less than 4 CFU/ml or 40 CFU/g respectively. For microbiological analyses unless differently reported in the individual test methods, in case of analytical steps foreseen in non-activity days of the laboratory, provisions of the ISO 7218: 2007 / Amd.1 2013 Standard (points 11.2 and 10.2.5) or from specific test methods are applied. In the case of quantitative microbiological tests, these have been set up on a single plate according to ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise expressly requested by current regulations.

In the case of quantitative microbiological tests, these have been set up on a single plate in accordance with ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise explicitly required by current regulations.

For waters, the measurement uncertainty corresponds to the confidence interval calculated according to ISO 8199: 2018 or to the expanded measurement uncertainty estimated according to ISO 29201: 2012. The results are issued in accordance with ISO 8199: 2018. When the number of colonies detected is <3, the result is expressed as "Microorganisms present in the analyzed volume (N ° colonies detected <3 CFU - reference ISO 8199: 2018, paragraph 9.1.8.4.1)".

LQ: Quantification Limit. It is the lowest analyte concentration which can be detected at an acceptable precision (repeatability) and accuracy, under well defined conditions.

LD: Detection Limit. It is the lowest analyte concentration which can be detected but not necessarily quantified, under well defined conditions.

Conformity evaluation: values not complying with laws, decrees, national and EU regulations or specifications supplied by the customer are evaluated case by case, also taking into consideration the uncertainty of measure for each single test and the regulations on rounding-off of values, and pointed out when considered as non conform.

Rec %: Recovery % "+" means that the recovery has been applied to the result. The numeric results between brackets (..) after the expression <LQ are purely indicative of traces that cannot be exactly quantified.

In the case of sampling carried out by Neotron, the laboratory applies the Internal Operating Procedure code: NEOT-DIR/ 006/53.

The laboratory disclaims any responsibility for the information provided by the client reported in this Report which may influence the validity of the results.

Methods marked with an asterisk (\*) are not accredited by ACCREDIA (UNI CEI EN ISO/IEC 17025). The sampling activity is not included within the Scope of Accreditation of Neotron SPA

## NOTES OF PARAMETERS:

[331]: Extended uncertainty calculated following the equation of HORWITZ by using a coverage factor of 2, which guarantee a reliance level of 95%

TEST REPORT VALID FOR ALL LEGAL PURPOSES (Italian R.D. 1-3-1928 n°842 (article 16), - Italian Law 19-7-1957 n°679 articles 16 and 18, Italian Ministerial Decree 25-3-1986).

DATA and SAMPLE STORAGE: Test Reports, Raw data, chromatographic paths and instrumental reports are stored for 5 years. One control sample is stored for 2 months.

Data expressed in this test report refer only to the sample tested in the laboratory. The results reported in this Test Report refer to the sample as received. The description or any other reference concerning the sample are declared by the customer. This Test Report cannot be reproduced except in full. Partial reproductions must be authorized in writing by our laboratory.

THE LABORATORY DIRECTOR: DR. ANDREA RIZZO

THE CHEMIST AUTHORIZED TO SIGN THE TEST REPORTS: DR. MARCO MESCHIARI

Approved by Analysis Manager - laboratory LMAA-Nut  
Approved by Analysis Manager - laboratory ICS-HPLC  
Approved by Analysis Manager - laboratory LC-MICO  
Approved by Analysis Manager - laboratory LC-AN

Approved by Analysis Manager - laboratory FISICHE  
Approved by Analysis Manager - laboratory LBM-EL  
Approved by Analysis Manager - laboratory LMAA-Bro

NEOTRON SpA - With Sole Shareholder

Stradello Aggazzotti, 104

41126 MODENA - ITALY - Fiscal Code and VAT n° 03807840362

Tel: +39 059461711 - Fax: +39 059461777

www.neotron.it - neotron@neotron.it

Laboratorio Qualificato D.M. 26-2-87 Art. 4 - Legge 46/82 per la Ricerca Applicata e Innovazione Tecnologica.

Regione Emilia Romagna - AUTORIZZAZIONE Autocontrollo N° 008/MO/008

BNN-Monitoring Fruit and Vegetables Approved Laboratory

I-Monitoring EDEKAAG Fruit and Vegetables Registered Laboratory

GMP+ code: GMP051757

MODENA, li 09/09/2022

Sample arrived on the 25/08/2022

Registration date 25/08/2022

**TEST REPORT nr. 22L15875-In-0**

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**

SAMPLE 22L15875

**MATRIX: Fruit and vegetable concentrates, and  
by-products**

Description provided by Customer: GLASSA CON ABM IGP - CONTAMINAZIONE ZERO - VISCOSITA': ~ 2.000 CPS (T=10G) -  
CODICE: S20Z-2000

Richiesta via Internet n° N0305/22 - 23/08/2022 16:48:07. - Campionamento eseguito da: Committente - Trasporto effettuato da:  
Corriere  
Sample Condition on Receipt: 4°C

ANALYSIS DESCRIPTION	RESULT	U	REC. %	UNIT OF MEASURE	LQ	LD	METHOD	ANALYSES BEGINNING DATE / ENDING DATE
Water activity (Aw) at 25 °C	0,934	± 0,005			0,150		06(S135) rev6 2021	26/08/2022 / 26/08/2022
<b>VINEGAR ANALYSIS</b>								
Reduction sugars (Reducing substances)	450,1	± 13,5		g/l	1,0		07(S240) 2021 Rev.0 - Titrimetrico	29/08/2022 / 01/09/2022
Total Acidity expressed as acetic acid	3,04	± 0,06		g/100 ml	0,10		* ACIDITA 2014 Rev.0 - Titrimetrico	29/08/2022 / 01/09/2022
Relative density at 20°C	1,184	± 0,00027					07(S117) 2015 Rev.1 - Gravimetric	02/09/2022 / 02/09/2022
Alcoholic strength	0,10	± 0,01		% Vol	0,07		DM 12/03/1986 SO GU n 161 14/07/1986 Met V pag 95, OIV- MA-AS312-01 Met B R2021	29/08/2022 / 01/09/2022
Brookfield rotational viscosity	Brookfield Rotational Viscosity (T = 20 ° C, impeller R3)							
	50 rpm: 1510 cP (mPa.s) 30 rpm: 1907 cP (mPa.s) 20 rpm: 2336 cP (mPa.s)							
pH	2,98	± 0,04		unità	0,50		07(S77) 2012 Rev.1 - Potentiometric	29/08/2022 / 29/08/2022
<b>SUGARS HPAEC-PAD</b>								
Galactose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	29/08/2022 / 02/09/2022
Glucose, anhydrous (HPLC)	17,1	± 1,2		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	29/08/2022 / 02/09/2022
Sucrose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	29/08/2022 / 02/09/2022
Fructose, anhydrous (HPLC)	15,6	± 1,1		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	29/08/2022 / 02/09/2022
Lactose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	29/08/2022 / 02/09/2022
Maltose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	29/08/2022 / 02/09/2022
Citric acid (LCMS) [331]	109	± 17		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	29/08/2022 / 09/09/2022
Lactic Acid (LCMS) [331]	196	± 28		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	29/08/2022 / 09/09/2022
Malic acid (LCMS) [331]	626	± 76		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	29/08/2022 / 09/09/2022
Succinic acid (LCMS) [331]	205	± 30		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	29/08/2022 / 09/09/2022
Tartaric acid (LCMS) [331]	641	± 78		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	29/08/2022 / 09/09/2022
Ochratoxin A	< LQ			µg/kg	0,10		03(S130) 2022 Rev.13 - LC- MS/MS	29/08/2022 / 31/08/2022

**END TEST REPORT**

The original document is a PDF file with Digital Signature: 22L15875-In-0-S20Z-2000 - T10 - UNIMORE-DigitalSignature.pdf

**Next page...**

MODENA, li 09/09/2022

Sample arrived on the 25/08/2022

Registration date 25/08/2022

## TEST REPORT nr. 22L15875-In-0

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**

**SAMPLE 22L15875**

**MATRIX: Fruit and vegetable concentrates, and by-products**

### Notes and method reference:

< LQ: = lower than Quantification Limit.

U: the reported uncertainty is the expanded uncertainty calculated using a coverage factor equal to 2 which gives a reliability of approximately 95%. Please note that results expressed as '<LQ' may not indicate the absence of the searched parameters in the sample.

MICROBIOLOGICAL TESTS: for food and environmental samples, the extended measurement uncertainty was estimated according to ISO 19036:2019 Standard and is based on a standard uncertainty multiplied by a coverage factor of K = 2, providing a confidence level of approximately 95%. The combined standard uncertainty was assumed to be equal to the standard deviation of intra-laboratory reproducibility. The results of the microbiological tests are calculated according to the ISO 7218: 2007 / Amd 1: 2013 Standard. If the results are reported as <4 (CFU/ml) or <40 (CFU/g), this means that the microorganisms are present in the sample but in amounts less than 4 CFU/ml or 40 CFU/g respectively. For microbiological analyses unless differently reported in the individual test methods, in case of analytical steps foreseen in non-activity days of the laboratory, provisions of the ISO 7218: 2007 / Amd.1 2013 Standard (points 11.2 and 10.2.5) or from specific test methods are applied. In the case of quantitative microbiological tests, these have been set up on a single plate according to ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise expressly requested by current regulations.

In the case of quantitative microbiological tests, these have been set up on a single plate in accordance with ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise explicitly required by current regulations.

For waters, the measurement uncertainty corresponds to the confidence interval calculated according to ISO 8199: 2018 or to the expanded measurement uncertainty estimated according to ISO 29201: 2012. The results are issued in accordance with ISO 8199: 2018. When the number of colonies detected is <3, the result is expressed as "Microorganisms present in the analyzed volume (N ° colonies detected <3 CFU - reference ISO 8199: 2018, paragraph 9.1.8.4.1)".

LQ: Quantification Limit. It is the lowest analyte concentration which can be detected at an acceptable precision (repeatability) and accuracy, under well defined conditions.

LD: Detection Limit. It is the lowest analyte concentration which can be detected but not necessarily quantified, under well defined conditions.

Conformity evaluation: values not complying with laws, decrees, national and EU regulations or specifications supplied by the customer are evaluated case by case, also taking into consideration the uncertainty of measure for each single test and the regulations on rounding-off of values, and pointed out when considered as non conform.

Rec %: Recovery % "+" means that the recovery has been applied to the result. The numeric results between brackets (..) after the expression <LQ are purely indicative of traces that cannot be exactly quantified.

In the case of sampling carried out by Neotron, the laboratory applies the Internal Operating Procedure code: NEOT-DIR/ 006/53.

The laboratory disclaims any responsibility for the information provided by the client reported in this Report which may influence the validity of the results.

Methods marked with an asterisk (\*) are not accredited by ACCREDIA (UNI CEI EN ISO/IEC 17025). The sampling activity is not included within the Scope of Accreditation of Neotron SPA

### NOTES OF PARAMETERS:

[331]: Extended uncertainty calculated following the equation of HORWITZ by using a coverage factor of 2, which guarantee a reliance level of 95%

TEST REPORT VALID FOR ALL LEGAL PURPOSES (Italian R.D. 1-3-1928 n°842 (article 16), - Italian Law 19-7-1957 n°679 articles 16 and 18, Italian Ministerial Decree 25-3-1986).

DATA and SAMPLE STORAGE: Test Reports, Raw data, chromatographic paths and instrumental reports are stored for 5 years. One control sample is stored for 2 months.

Data expressed in this test report refer only to the sample tested in the laboratory. The results reported in this Test Report refer to the sample as received. The description or any other reference concerning the sample are declared by the customer. This Test Report cannot be reproduced except in full. Partial reproductions must be authorized in writing by our laboratory.

THE LABORATORY DIRECTOR: DR. ANDREA RIZZO

THE CHEMIST AUTHORIZED TO SIGN THE TEST REPORTS: DR. BARBARA MALAGOLI

Approved by Analysis Manager - laboratory LMAA-Nut  
Approved by Analysis Manager - laboratory ICS-HPLC  
Approved by Analysis Manager - laboratory LC-MICO  
Approved by Analysis Manager - laboratory LC-AN

Approved by Analysis Manager - laboratory FISICHE  
Approved by Analysis Manager - laboratory LBM-EL  
Approved by Analysis Manager - laboratory LMAA-Bro

MODENA, li 09/09/2022

Sample arrived on the 25/08/2022  
Registration date 25/08/2022**TEST REPORT nr. 22L15873-In-0**

CUSTOMER

**ACETFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO****SAMPLE 22L15873****MATRIX: Fruit and vegetable concentrates, and  
by-products**Description provided by Customer: GLASSA CON ABM IGP - CONTAMINAZIONE ZERO - VISCOSITA': ~ 2.000 CPS (T=0G) -  
CODICE: S20Z-2000Richiesta via Internet n° N0302/22 - 23/08/2022 16:48:06. - Campionamento eseguito da: Committente - Trasporto effettuato da:  
Corriere  
Sample Condition on Receipt: 4°C

ANALYSIS DESCRIPTION	RESULT	U	REC. %	UNIT OF MEASURE	LQ	LD	METHOD	ANALYSES BEGINNING DATE / ENDING DATE
Water activity (Aw) at 25 °C	0,936	± 0,005			0,150		06(S135) rev6 2021	26/08/2022 / 26/08/2022
<b>VINEGAR ANALYSIS</b>								
Reduction sugars (Reducing substances)	444,0	± 13,3		g/l	1,0		07(S240) 2021 Rev.0 - Titrimetrico	29/08/2022 / 01/09/2022
Total Acidity expressed as acetic acid	2,95	± 0,06		g/100 ml	0,10		* ACIDITA 2014 Rev.0 - Titrimetrico	29/08/2022 / 01/09/2022
Relative density at 20°C	1,184	± 0,00027					07(S117) 2015 Rev.1 - Gravimetric	02/09/2022 / 02/09/2022
Alcoholic strength	0,09	± 0,01		% Vol	0,07		DM 12/03/1986 SO GU n 161 14/07/1986 Met V pag 95, OIV- MA-AS312-01 Met B R2021	29/08/2022 / 01/09/2022
pH	2,98	± 0,04		unità	0,50		07(S77) 2012 Rev.1 - Potentiometric	29/08/2022 / 29/08/2022
Brookfield rotational viscosity	Brookfield Rotational Viscosity (T = 20 ° C, impeller R3) 50 rpm: 1379 cP (mPa.s) 30 rpm: 1731 cP (mPa.s) 20 rpm: 2052 cP (mPa.s)						* VISCOSITA 2018 Rev.1	29/08/2022 / 01/09/2022
<b>SUGARS HPAEC-PAD</b>								
Galactose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	29/08/2022 / 02/09/2022
Glucose, anhydrous (HPLC)	18,4	± 1,3		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	29/08/2022 / 02/09/2022
Sucrose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	29/08/2022 / 02/09/2022
Fructose, anhydrous (HPLC)	17,8	± 1,2		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	29/08/2022 / 02/09/2022
Lactose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	29/08/2022 / 02/09/2022
Maltose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	29/08/2022 / 02/09/2022
Citric acid (LCMS) [331]	109	± 17		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	29/08/2022 / 09/09/2022
Lactic Acid (LCMS) [331]	183	± 27		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	29/08/2022 / 09/09/2022
Malic acid (LCMS) [331]	620	± 76		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	29/08/2022 / 09/09/2022
Succinic acid (LCMS) [331]	200	± 29		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	29/08/2022 / 09/09/2022
Tartaric acid (LCMS) [331]	665	± 80		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	29/08/2022 / 09/09/2022
Ochratoxin A	< LQ			µg/kg	0,10		03(S130) 2022 Rev.13 - LC- MS/MS	29/08/2022 / 31/08/2022

**END TEST REPORT**

The original document is a PDF file with Digital Signature: 22L15873-In-0-S20Z-2000 - T0 - UNIMORE-DigitalSignature.pdf

**Next page...**

MODENA, li 09/09/2022

Sample arrived on the 25/08/2022

Registration date 25/08/2022

**TEST REPORT nr. 22L15873-In-0**

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**

SAMPLE 22L15873

**MATRIX: Fruit and vegetable concentrates, and  
by-products**

## Notes and method reference:

&lt; LQ: = lower than Quantification Limit.

U: the reported uncertainty is the expanded uncertainty calculated using a coverage factor equal to 2 which gives a reliability of approximately 95%. Please note that results expressed as '<LQ' may not indicate the absence of the searched parameters in the sample.

MICROBIOLOGICAL TESTS: for food and environmental samples, the extended measurement uncertainty was estimated according to ISO 19036:2019 Standard and is based on a standard uncertainty multiplied by a coverage factor of K = 2, providing a confidence level of approximately 95%. The combined standard uncertainty was assumed to be equal to the standard deviation of intra-laboratory reproducibility. The results of the microbiological tests are calculated according to the ISO 7218: 2007 / Amd 1: 2013 Standard. If the results are reported as <4 (CFU/ml) or <40 (CFU/g), this means that the microorganisms are present in the sample but in amounts less than 4 CFU/ml or 40 CFU/g respectively. For microbiological analyses unless differently reported in the individual test methods, in case of analytical steps foreseen in non-activity days of the laboratory, provisions of the ISO 7218: 2007 / Amd.1 2013 Standard (points 11.2 and 10.2.5) or from specific test methods are applied. In the case of quantitative microbiological tests, these have been set up on a single plate according to ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise expressly requested by current regulations.

In the case of quantitative microbiological tests, these have been set up on a single plate in accordance with ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise explicitly required by current regulations.

For waters, the measurement uncertainty corresponds to the confidence interval calculated according to ISO 8199: 2018 or to the expanded measurement uncertainty estimated according to ISO 29201: 2012. The results are issued in accordance with ISO 8199: 2018. When the number of colonies detected is <3, the result is expressed as "Microorganisms present in the analyzed volume (N ° colonies detected <3 CFU - reference ISO 8199: 2018, paragraph 9.1.8.4.1)".

LQ: Quantification Limit. It is the lowest analyte concentration which can be detected at an acceptable precision (repeatability) and accuracy, under well defined conditions.

LD: Detection Limit. It is the lowest analyte concentration which can be detected but not necessarily quantified, under well defined conditions.

Conformity evaluation: values not complying with laws, decrees, national and EU regulations or specifications supplied by the customer are evaluated case by case, also taking into consideration the uncertainty of measure for each single test and the regulations on rounding-off of values, and pointed out when considered as non conform.

Rec %: Recovery % "+" means that the recovery has been applied to the result. The numeric results between brackets (..) after the expression <LQ are purely indicative of traces that cannot be exactly quantified.

In the case of sampling carried out by Neotron, the laboratory applies the Internal Operating Procedure code: NEOT-DIR/ 006/53.

The laboratory disclaims any responsibility for the information provided by the client reported in this Report which may influence the validity of the results.

Methods marked with an asterisk (\*) are not accredited by ACCREDIA (UNI CEI EN ISO/IEC 17025). The sampling activity is not included within the Scope of Accreditation of Neotron SPA

## NOTES OF PARAMETERS:

[331]: Extended uncertainty calculated following the equation of HORWITZ by using a coverage factor of 2, which guarantee a reliance level of 95%

TEST REPORT VALID FOR ALL LEGAL PURPOSES (Italian R.D. 1-3-1928 n°842 (article 16), - Italian Law 19-7-1957 n°679 articles 16 and 18, Italian Ministerial Decree 25-3-1986).

DATA and SAMPLE STORAGE: Test Reports, Raw data, chromatographic paths and instrumental reports are stored for 5 years. One control sample is stored for 2 months.

Data expressed in this test report refer only to the sample tested in the laboratory. The results reported in this Test Report refer to the sample as received. The description or any other reference concerning the sample are declared by the customer. This Test Report cannot be reproduced except in full. Partial reproductions must be authorized in writing by our laboratory.

THE LABORATORY DIRECTOR: DR. ANDREA RIZZO

THE CHEMIST AUTHORIZED TO SIGN THE TEST REPORTS: DR. BARBARA MALAGOLI

Approved by Analysis Manager - laboratory LMAA-Nut

Approved by Analysis Manager - laboratory ICS-HPLC

Approved by Analysis Manager - laboratory LC-MICO

Approved by Analysis Manager - laboratory LC-AN

Approved by Analysis Manager - laboratory FISICHE

Approved by Analysis Manager - laboratory LBM-EL

Approved by Analysis Manager - laboratory LMAA-Bro

NEOTRON SpA - With Sole Shareholder

Stradello Aggazzotti, 104

41126 MODENA - ITALY - Fiscal Code and VAT n° 03807840362

Tel: +39 059461711 - Fax: +39 059461777

www.neotron.it - neotron@neotron.it

Laboratorio Qualificato D.M. 26-2-87 Art. 4 - Legge 46/82 per la Ricerca Applicata e Innovazione Tecnologica.

Regione Emilia Romagna - AUTORIZZAZIONE Autocontrollo N° 008/MO/008

BNN-Monitoring Fruit and Vegetables Approved Laboratory

I-Monitoring EDEKAAG Fruit and Vegetables Registered Laboratory

GMP+ code: GMP051757

MODENA, li 23/08/2022

Sample arrived on the 05/08/2022

Registration date 08/08/2022

**TEST REPORT nr. 22L05143-In-0**

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**
**SAMPLE 22L05143****MATRIX: Fruit and vegetable concentrates, and by-products**

Description provided by Customer: GLASSA CON ABM IGP - CONTAMINAZIONE ZERO - VISCOSITA': ~ 1.000 CPS (T=20G) - CODICE: S20Z-1000

Richiesta via Internet n° N0255/22 - 20/07/2022 12:55:18. - Campionamento eseguito da: Committente - Trasporto effettuato da: Committente  
Sample Condition on Receipt: 20°C

ANALYSIS DESCRIPTION	RESULT	U	REC. %	UNIT OF MEASURE	LQ	LD	METHOD	ANALYSES BEGINNING DATE / ENDING DATE
Water activity (Aw) at 25 °C	0,920	± 0,005			0,150		06(S135) rev6 2021	08/08/2022 / 08/08/2022
<b>VINEGAR ANALYSIS</b>								
Reduction sugars (Reducing substances)	422,7	± 12,7		g/l	1,0		07(S240) 2021 Rev.0 - Titrimetrico	09/08/2022 / 11/08/2022
Total Acidity expressed as acetic acid	2,81	± 0,05		g/100 ml	0,10		* ACIDITA 2014 Rev.0 - Titrimetrico	09/08/2022 / 11/08/2022
Relative density at 20°C	1,19101	± 0,00027					DM 12/03/1986 SO GU n 161 14/07/1986 Met VI pag 95 p.to 4, OIV-MA-AS2-01A Met B R2021 - densimetry	09/08/2022 / 11/08/2022
Alcoholic strength	0,09	± 0,01		% Vol	0,07		DM 12/03/1986 SO GU n 161 14/07/1986 Met V pag 95, OIV-MA-AS312-01 Met B R2021	09/08/2022 / 11/08/2022
pH	2,94	± 0,04		unità	0,50		07(S77) 2012 Rev.1 - Potentiometric	09/08/2022 / 10/08/2022
Brookfield rotational viscosity	Brookfield Rotational Viscosity (T = 20 ° C, impeller R2)						* VISCOSITA 2018 Rev.1	09/08/2022 / 12/08/2022
	30 rpm: 835 cP (mPa.s)							
	20 rpm: 963 cP (mPa.s)							
	12 rpm: 1161 cP (mPa.s)							
<b>SUGARS HPAEC-PAD</b>								
Galactose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	09/08/2022 / 16/08/2022
Glucose, anhydrous (HPLC)	18,1	± 1,3		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	09/08/2022 / 16/08/2022
Sucrose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	09/08/2022 / 16/08/2022
Fructose, anhydrous (HPLC)	18,9	± 1,3		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	09/08/2022 / 16/08/2022
Lactose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	09/08/2022 / 16/08/2022
Maltose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	09/08/2022 / 16/08/2022
Citric acid (LCMS) [331]	52,3	± 9,2		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	09/08/2022 / 16/08/2022
Lactic Acid (LCMS) [331]	193	± 28		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	09/08/2022 / 16/08/2022
Malic acid (LCMS) [331]	347	± 46		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	09/08/2022 / 16/08/2022
Succinic acid (LCMS) [331]	132	± 20		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	09/08/2022 / 16/08/2022
Tartaric acid (LCMS) [331]	379	± 50		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	09/08/2022 / 16/08/2022
Ochratoxin A	< LQ			µg/kg	0,10		03(S130) 2022 Rev.12 - LC-MS/MS	09/08/2022 / 11/08/2022

**END TEST REPORT**

The original document is a PDF file with Digital Signature: 22L05143-In-0-S20Z-1000 - T20 - UNIMORE-DigitalSignature.pdf

MODENA, li 23/08/2022

Sample arrived on the 05/08/2022

Registration date 08/08/2022

## TEST REPORT nr. 22L05143-In-0

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**

**SAMPLE 22L05143**

**MATRIX: Fruit and vegetable concentrates, and by-products**

### Notes and method reference:

< LQ: = lower than Quantification Limit.

U: the reported uncertainty is the expanded uncertainty calculated using a coverage factor equal to 2 which gives a reliability of approximately 95%. Please note that results expressed as '<LQ' may not indicate the absence of the searched parameters in the sample.

MICROBIOLOGICAL TESTS: for food and environmental samples, the extended measurement uncertainty was estimated according to ISO 19036:2019 Standard and is based on a standard uncertainty multiplied by a coverage factor of K = 2, providing a confidence level of approximately 95%. The combined standard uncertainty was assumed to be equal to the standard deviation of intra-laboratory reproducibility. The results of the microbiological tests are calculated according to the ISO 7218: 2007 / Amd 1: 2013 Standard. If the results are reported as <4 (CFU/ml) or <40 (CFU/g), this means that the microorganisms are present in the sample but in amounts less than 4 CFU/ml or 40 CFU/g respectively. For microbiological analyses unless differently reported in the individual test methods, in case of analytical steps foreseen in non-activity days of the laboratory, provisions of the ISO 7218: 2007 / Amd.1 2013 Standard (points 11.2 and 10.2.5) or from specific test methods are applied. In the case of quantitative microbiological tests, these have been set up on a single plate according to ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise expressly requested by current regulations.

In the case of quantitative microbiological tests, these have been set up on a single plate in accordance with ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise explicitly required by current regulations.

For waters, the measurement uncertainty corresponds to the confidence interval calculated according to ISO 8199: 2018 or to the expanded measurement uncertainty estimated according to ISO 29201: 2012. The results are issued in accordance with ISO 8199: 2018. When the number of colonies detected is <3, the result is expressed as "Microorganisms present in the analyzed volume (N ° colonies detected <3 CFU - reference ISO 8199: 2018, paragraph 9.1.8.4.1)".

LQ: Quantification Limit. It is the lowest analyte concentration which can be detected at an acceptable precision (repeatability) and accuracy, under well defined conditions.

LD: Detection Limit. It is the lowest analyte concentration which can be detected but not necessarily quantified, under well defined conditions.

Conformity evaluation: values not complying with laws, decrees, national and EU regulations or specifications supplied by the customer are evaluated case by case, also taking into consideration the uncertainty of measure for each single test and the regulations on rounding-off of values, and pointed out when considered as non conform.

Rec %: Recovery % "+" means that the recovery has been applied to the result. The numeric results between brackets (..) after the expression <LQ are purely indicative of traces that cannot be exactly quantified.

In the case of sampling carried out by Neotron, the laboratory applies the Internal Operating Procedure code: NEOT-DIR/ 006/53.

The laboratory disclaims any responsibility for the information provided by the client reported in this Report which may influence the validity of the results.

Methods marked with an asterisk (\*) are not accredited by ACCREDIA (UNI CEI EN ISO/IEC 17025). The sampling activity is not included within the Scope of Accreditation of Neotron SPA

### NOTES OF PARAMETERS:

[331]: Extended uncertainty calculated following the equation of HORWITZ by using a coverage factor of 2, which guarantee a reliance level of 95%

TEST REPORT VALID FOR ALL LEGAL PURPOSES (Italian R.D. 1-3-1928 n°842 (article 16), - Italian Law 19-7-1957 n°679 articles 16 and 18, Italian Ministerial Decree 25-3-1986).

DATA and SAMPLE STORAGE: Test Reports, Raw data, chromatographic paths and instrumental reports are stored for 5 years. One control sample is stored for 2 months.

Data expressed in this test report refer only to the sample tested in the laboratory. The results reported in this Test Report refer to the sample as received. The description or any other reference concerning the sample are declared by the customer. This Test Report cannot be reproduced except in full. Partial reproductions must be authorized in writing by our laboratory.

THE LABORATORY DIRECTOR: DR. ANDREA RIZZO

THE CHEMIST AUTHORIZED TO SIGN THE TEST REPORTS: DR. MARCO MESCHIARI - N. 514 ORDINE DEI CHIMICI MODENA

Approved by Analysis Manager - laboratory LMAA-Nut

Approved by Analysis Manager - laboratory ICS-HPLC

Approved by Analysis Manager - laboratory LC-MICO

Approved by Analysis Manager - laboratory LC-AN

Approved by Analysis Manager - laboratory FISICHE

Approved by Analysis Manager - laboratory LBM-EL

Approved by Analysis Manager - laboratory LMAA-Bro

NEOTRON SpA - With Sole Shareholder

Stradello Aggazzotti, 104

41126 MODENA - ITALY - Fiscal Code and VAT n° 03807840362

Tel: +39 059461711 - Fax: +39 059461777

www.neotron.it - neotron@neotron.it

Laboratorio Qualificato D.M. 26-2-87 Art. 4 - Legge 46/82 per la Ricerca Applicata e Innovazione Tecnologica.

Regione Emilia Romagna - AUTORIZZAZIONE Autocontrollo N° 008/MO/008

BNN-Monitoring Fruit and Vegetables Approved Laboratory

I-Monitoring EDEKAAG Fruit and Vegetables Registered Laboratory

GMP+ code: GMP051757



MODENA, li 04/08/2022

Sample arrived on the 26/07/2022  
Registration date 26/07/2022**TEST REPORT nr. 22G17281-In-0**

CUSTOMER

**ACETFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO****SAMPLE 22G17281****MATRIX: Fruit and vegetable concentrates, and by-products**

Description provided by Customer: GLASSA CON ABM IGP - CONTAMINAZIONE ZERO - VISCOSITA': ~ 1.000 CPS (T=10G) - CODICE: S20Z-1000

Richiesta via Internet n° N0254/22 - 20/07/2022 12:55:17. - Campionamento eseguito da: Committente - Trasporto effettuato da: Committente  
Sample Condition on Receipt: 20°C

ANALYSIS DESCRIPTION	RESULT	U	REC. %	UNIT OF MEASURE	LQ	LD	METHOD	ANALYSES BEGINNING DATE / ENDING DATE
Water activity (Aw) at 25 °C	0,936	± 0,005			0,150		06(S135) rev6 2021	27/07/2022 / 29/07/2022
<b>VINEGAR ANALYSIS</b>								
Reduction sugars (Reducing substances)	457,0	± 13,7		g/l	1,0		07(S240) 2021 Rev.0 - Titrimetrico	28/07/2022 / 02/08/2022
Total Acidity expressed as acetic acid	2,87	± 0,06		g/100 ml	0,10		* ACIDITA 2014 Rev.0 - Titrimetrico	28/07/2022 / 01/08/2022
Relative density at 20°C	1,19020	± 0,00027					DM 12/03/1986 SO GU n 161 14/07/1986 Met VI pag 95 p.to 4, OIV-MA-AS2-01A Met B R2021 - densimetry	28/07/2022 / 01/08/2022
Alcoholic strength	< LQ			% Vol	0,07		DM 12/03/1986 SO GU n 161 14/07/1986 Met V pag 95, OIV-MA-AS312-01 Met B R2021	28/07/2022 / 01/08/2022
pH	2,96	± 0,04		unità	0,50		07(S77) 2012 Rev.1 - Potentiometric	28/07/2022 / 01/08/2022
Brookfield rotational viscosity	Brookfield Rotational Viscosity (T = 20 ° C, impeller R2)						* VISCOSITA 2018 Rev.1	28/07/2022 / 04/08/2022
	30 rpm: 788 cP (mPa.s)							
	20 rpm: 903 cP (mPa.s)							
	12 rpm: 1073 cP (mPa.s)							
<b>SUGARS HPAEC-PAD</b>								
Galactose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	28/07/2022 / 02/08/2022
Glucose, anhydrous (HPLC)	18,3	± 1,5		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	28/07/2022 / 02/08/2022
Sucrose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	28/07/2022 / 02/08/2022
Fructose, anhydrous (HPLC)	20,0	± 1,6		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	28/07/2022 / 02/08/2022
Lactose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	28/07/2022 / 02/08/2022
Maltose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	28/07/2022 / 02/08/2022
Citric acid (LCMS) [331]	86,6	± 14,2		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	28/07/2022 / 04/08/2022
Lactic Acid (LCMS) [331]	127	± 20		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	28/07/2022 / 04/08/2022
Malic acid (LCMS) [331]	450	± 58		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	28/07/2022 / 04/08/2022
Succinic acid (LCMS) [331]	170	± 25		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	28/07/2022 / 04/08/2022
Tartaric acid (LCMS) [331]	478	± 60		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	28/07/2022 / 04/08/2022
Ochratoxin A	< LQ			µg/kg	0,10		03(S130) 2022 Rev.12 - LC-MS/MS	28/07/2022 / 02/08/2022

**END TEST REPORT**

The original document is a PDF file with Digital Signature: 22G17281-In-0-S20Z-1000 - T10 - UNIMORE-DigitalSignature.pdf

MODENA, li 04/08/2022

Sample arrived on the 26/07/2022

Registration date 26/07/2022

## TEST REPORT nr. 22G17281-In-0

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**

**SAMPLE 22G17281**

**MATRIX: Fruit and vegetable concentrates, and by-products**

### Notes and method reference:

< LQ: = lower than Quantification Limit.

U: the reported uncertainty is the expanded uncertainty calculated using a coverage factor equal to 2 which gives a reliability of approximately 95%. Please note that results expressed as '<LQ' may not indicate the absence of the searched parameters in the sample.

MICROBIOLOGICAL TESTS: for food and environmental samples, the extended measurement uncertainty was estimated according to ISO 19036:2019 Standard and is based on a standard uncertainty multiplied by a coverage factor of  $K = 2$ , providing a confidence level of approximately 95%. The combined standard uncertainty was assumed to be equal to the standard deviation of intra-laboratory reproducibility. The results of the microbiological tests are calculated according to the ISO 7218: 2007 / Amd 1: 2013 Standard. If the results are reported as <4 (CFU/ml) or <40 (CFU/g), this means that the microorganisms are present in the sample but in amounts less than 4 CFU/ml or 40 CFU/g respectively. For microbiological analyses unless differently reported in the individual test methods, in case of analytical steps foreseen in non-activity days of the laboratory, provisions of the ISO 7218: 2007 / Amd.1 2013 Standard (points 11.2 and 10.2.5) or from specific test methods are applied. In the case of quantitative microbiological tests, these have been set up on a single plate according to ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise expressly requested by current regulations.

In the case of quantitative microbiological tests, these have been set up on a single plate in accordance with ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise explicitly required by current regulations.

For waters, the measurement uncertainty corresponds to the confidence interval calculated according to ISO 8199: 2018 or to the expanded measurement uncertainty estimated according to ISO 29201: 2012. The results are issued in accordance with ISO 8199: 2018. When the number of colonies detected is <3, the result is expressed as "Microorganisms present in the analyzed volume (N ° colonies detected <3 CFU - reference ISO 8199: 2018, paragraph 9.1.8.4.1)".

LQ: Quantification Limit. It is the lowest analyte concentration which can be detected at an acceptable precision (repeatability) and accuracy, under well defined conditions.

LD: Detection Limit. It is the lowest analyte concentration which can be detected but not necessarily quantified, under well defined conditions.

Conformity evaluation: values not complying with laws, decrees, national and EU regulations or specifications supplied by the customer are evaluated case by case, also taking into consideration the uncertainty of measure for each single test and the regulations on rounding-off of values, and pointed out when considered as non conform.

Rec %: Recovery % "+" means that the recovery has been applied to the result. The numeric results between brackets (..) after the expression <LQ are purely indicative of traces that cannot be exactly quantified.

In the case of sampling carried out by Neotron, the laboratory applies the Internal Operating Procedure code: NEOT-DIR/ 006/53.

The laboratory disclaims any responsibility for the information provided by the client reported in this Report which may influence the validity of the results.

Methods marked with an asterisk (\*) are not accredited by ACCREDIA (UNI CEI EN ISO/IEC 17025). The sampling activity is not included within the Scope of Accreditation of Neotron SPA

### NOTES OF PARAMETERS:

[331]: Extended uncertainty calculated following the equation of HORWITZ by using a coverage factor of 2, which guarantee a reliance level of 95%

TEST REPORT VALID FOR ALL LEGAL PURPOSES (Italian R.D. 1-3-1928 n°842 (article 16), – Italian Law 19-7-1957 n°679 articles 16 and 18, Italian Ministerial Decree 25-3-1986).

DATA and SAMPLE STORAGE: Test Reports, Raw data, chromatographic paths and instrumental reports are stored for 5 years. One control sample is stored for 2 months.

Data expressed in this test report refer only to the sample tested in the laboratory. The results reported in this Test Report refer to the sample as received. The description or any other reference concerning the sample are declared by the customer. This Test Report cannot be reproduced except in full. Partial reproductions must be authorized in writing by our laboratory.

THE LABORATORY DIRECTOR: DR. ANDREA RIZZO

THE CHEMIST AUTHORIZED TO SIGN THE TEST REPORTS: DR. MARCO MESCHIARI - N. 514 ORDINE DEI CHIMICI MODENA

Approved by Analysis Manager - laboratory LMAA-Nut

Approved by Analysis Manager - laboratory ICS-HPLC

Approved by Analysis Manager - laboratory LC-MICO

Approved by Analysis Manager - laboratory LC-AN

Approved by Analysis Manager - laboratory FISICHE

Approved by Analysis Manager - laboratory LBM-EL

Approved by Analysis Manager - laboratory LMAA-Bro

NEOTRON SpA - With Sole Shareholder

Stradello Aggazzotti, 104

41126 MODENA - ITALY - Fiscal Code and VAT n° 03807840362

Tel: +39 059461711 - Fax: +39 059461777

www.neotron.it - neotron@neotron.it

Laboratorio Qualificato D.M. 26-2-87 Art. 4 - Legge 46/82 per la Ricerca Applicata e Innovazione Tecnologica.

Regione Emilia Romagna - AUTORIZZAZIONE Autocontrollo N° 008/MO/008

BNN-Monitoring Fruit and Vegetables Approved Laboratory

I-Monitoring EDEKAAG Fruit and Vegetables Registered Laboratory

GMP+ code: GMP051757

MODENA, li 29/07/2022

Sample arrived on the 19/07/2022  
Registration date 19/07/2022**TEST REPORT nr. 22G12684-In-0**

CUSTOMER

**ACETFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO****SAMPLE 22G12684****MATRIX: Fruit and vegetable concentrates, and  
by-products**Description provided by Customer: GLASSA CON ABM IGP - CONTAMINAZIONE ZERO - VISCOSITA': ~ 1.000 CPS (T=0G) -  
CODICE: S20Z-1000Richiesta via Internet n° N0216/22 - 04/07/2022 16:41:56. - Campionamento eseguito da: Committente - Trasporto effettuato da:  
Committente  
Sample Condition on Receipt: 4°C

ANALYSIS DESCRIPTION	RESULT	U	REC. %	UNIT OF MEASURE	LQ	LD	METHOD	ANALYSES BEGINNING DATE / ENDING DATE
Water activity (Aw) at 25 °C	0,909	± 0,005			0,150		06(S135) rev6 2021	20/07/2022/ 22/07/2022
<b>VINEGAR ANALYSIS</b>								
Reduction sugars (Reducing substances)	452,3	± 13,6		g/l	1,0		07(S240) 2021 Rev.0 - Titrimetrico	21/07/2022/ 26/07/2022
Total Acidity expressed as acetic acid	2,89	± 0,06		g/100 ml	0,10		* ACIDITA 2014 Rev.0 - Titrimetrico	21/07/2022/ 25/07/2022
Relative density at 20°C	1,190	± 0,001					07(S117) 2015 Rev.1 - Gravimetric	29/07/2022/ 29/07/2022
Alcoholic strength	0,10	± 0,01		% Vol	0,07		DM 12/03/1986 SO GU n 161 14/07/1986 Met V pag 95, OIV- MA-AS312-01 Met B R2021	21/07/2022/ 26/07/2022
pH	2,93	± 0,04		unità	0,50		07(S77) 2012 Rev.1 - Potentiometric	21/07/2022/ 22/07/2022
Brookfield rotational viscosity	Brookfield Rotational Viscosity (T = 20 ° C, impeller R2) 50 rpm: 548 cP (mPa.s) 30 rpm: 645 cP (mPa.s) 20 rpm: 735 cP (mPa.s)						* VISCOSITA 2018 Rev.1	21/07/2022/ 26/07/2022
<b>SUGARS HPAEC-PAD</b>								
Galactose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	21/07/2022/ 25/07/2022
Glucose, anhydrous (HPLC)	19,2	± 1,3		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	21/07/2022/ 25/07/2022
Sucrose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	21/07/2022/ 25/07/2022
Fructose, anhydrous (HPLC)	18,2	± 1,3		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	21/07/2022/ 25/07/2022
Lactose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	21/07/2022/ 25/07/2022
Maltose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	21/07/2022/ 25/07/2022
Citric acid (LCMS) [331]	101	± 16		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	21/07/2022/ 26/07/2022
Lactic Acid (LCMS) [331]	143	± 22		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	21/07/2022/ 26/07/2022
Malic acid (LCMS) [331]	616	± 75		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	21/07/2022/ 26/07/2022
Succinic acid (LCMS) [331]	224	± 32		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	21/07/2022/ 26/07/2022
Tartaric acid (LCMS) [331]	366	± 48		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	21/07/2022/ 26/07/2022
Ochratoxin A	< LQ			µg/kg	0,10		03(S130) 2022 Rev.12 - LC- MS/MS	21/07/2022/ 26/07/2022

**END TEST REPORT**

The original document is a PDF file with Digital Signature: 22G12684-In-0-S20Z-1000 - T0 - UNIMORE-DigitalSignature.pdf

**Next page...**

MODENA, li 29/07/2022

Sample arrived on the 19/07/2022

Registration date 19/07/2022

## TEST REPORT nr. 22G12684-In-0

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**

**SAMPLE 22G12684**

**MATRIX: Fruit and vegetable concentrates, and by-products**

### Notes and method reference:

< LQ: = lower than Quantification Limit.

U: the reported uncertainty is the expanded uncertainty calculated using a coverage factor equal to 2 which gives a reliability of approximately 95%. Please note that results expressed as '<LQ' may not indicate the absence of the searched parameters in the sample.

MICROBIOLOGICAL TESTS: for food and environmental samples, the extended measurement uncertainty was estimated according to ISO 19036:2019 Standard and is based on a standard uncertainty multiplied by a coverage factor of  $K = 2$ , providing a confidence level of approximately 95%. The combined standard uncertainty was assumed to be equal to the standard deviation of intra-laboratory reproducibility. The results of the microbiological tests are calculated according to the ISO 7218: 2007 / Amd 1: 2013 Standard. If the results are reported as <4 (CFU/ml) or <40 (CFU/g), this means that the microorganisms are present in the sample but in amounts less than 4 CFU/ml or 40 CFU/g respectively. For microbiological analyses unless differently reported in the individual test methods, in case of analytical steps foreseen in non-activity days of the laboratory, provisions of the ISO 7218: 2007 / Amd.1 2013 Standard (points 11.2 and 10.2.5) or from specific test methods are applied. In the case of quantitative microbiological tests, these have been set up on a single plate according to ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise expressly requested by current regulations.

In the case of quantitative microbiological tests, these have been set up on a single plate in accordance with ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise explicitly required by current regulations.

For waters, the measurement uncertainty corresponds to the confidence interval calculated according to ISO 8199: 2018 or to the expanded measurement uncertainty estimated according to ISO 29201: 2012. The results are issued in accordance with ISO 8199: 2018. When the number of colonies detected is <3, the result is expressed as "Microorganisms present in the analyzed volume (N ° colonies detected <3 CFU - reference ISO 8199: 2018, paragraph 9.1.8.4.1)".

LQ: Quantification Limit. It is the lowest analyte concentration which can be detected at an acceptable precision (repeatability) and accuracy, under well defined conditions.

LD: Detection Limit. It is the lowest analyte concentration which can be detected but not necessarily quantified, under well defined conditions.

Conformity evaluation: values not complying with laws, decrees, national and EU regulations or specifications supplied by the customer are evaluated case by case, also taking into consideration the uncertainty of measure for each single test and the regulations on rounding-off of values, and pointed out when considered as non conform.

Rec %: Recovery % "+" means that the recovery has been applied to the result. The numeric results between brackets (..) after the expression <LQ are purely indicative of traces that cannot be exactly quantified.

In the case of sampling carried out by Neotron, the laboratory applies the Internal Operating Procedure code: NEOT-DIR/ 006/53.

The laboratory disclaims any responsibility for the information provided by the client reported in this Report which may influence the validity of the results.

Methods marked with an asterisk (\*) are not accredited by ACCREDIA (UNI CEI EN ISO/IEC 17025). The sampling activity is not included within the Scope of Accreditation of Neotron SPA

### NOTES OF PARAMETERS:

[331]: Extended uncertainty calculated following the equation of HORWITZ by using a coverage factor of 2, which guarantee a reliance level of 95%

TEST REPORT VALID FOR ALL LEGAL PURPOSES (Italian R.D. 1-3-1928 n°842 (article 16), – Italian Law 19-7-1957 n°679 articles 16 and 18, Italian Ministerial Decree 25-3-1986).

DATA and SAMPLE STORAGE: Test Reports, Raw data, chromatographic paths and instrumental reports are stored for 5 years. One control sample is stored for 2 months.

Data expressed in this test report refer only to the sample tested in the laboratory. The results reported in this Test Report refer to the sample as received. The description or any other reference concerning the sample are declared by the customer. This Test Report cannot be reproduced except in full. Partial reproductions must be authorized in writing by our laboratory.

THE LABORATORY DIRECTOR: DR. ANDREA RIZZO

THE CHEMIST AUTHORIZED TO SIGN THE TEST REPORTS: DR. MARCO MESCHIARI - N. 514 ORDINE DEI CHIMICI MODENA

Approved by Analysis Manager - laboratory LMAA-Nut

Approved by Analysis Manager - laboratory ICS-HPLC

Approved by Analysis Manager - laboratory LC-MICO

Approved by Analysis Manager - laboratory LC-AN

Approved by Analysis Manager - laboratory FISICHE

Approved by Analysis Manager - laboratory LBM-EL

Approved by Analysis Manager - laboratory LMAA-Bro

NEOTRON SpA - With Sole Shareholder

Stradello Aggazzotti, 104

41126 MODENA - ITALY - Fiscal Code and VAT n° 03807840362

Tel: +39 059461711 - Fax: +39 059461777

www.neotron.it - neotron@neotron.it

Laboratorio Qualificato D.M. 26-2-87 Art. 4 - Legge 46/82 per la Ricerca Applicata e Innovazione Tecnologica.

Regione Emilia Romagna - AUTORIZZAZIONE Autocontrollo N° 008/MO/008

BNN-Monitoring Fruit and Vegetables Approved Laboratory

I-Monitoring EDEKAAG Fruit and Vegetables Registered Laboratory

GMP+ code: GMP051757

MODENA, li 26/10/2022

Sample arrived on the 14/10/2022

Registration date 14/10/2022

**TEST REPORT nr. 22P11007-In-0**

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**

SAMPLE 22P11007

**MATRIX: Fruit and vegetable concentrates, and  
by-products**

Description provided by Customer: GLASSA CON ABM IGP - CONTAMINAZIONE BASSA - VISCOSITA': ~ 4.000 CPS (T=20G) -  
CODICE: S20B-4000

Richiesta via Internet n° N0381/22 - 26/09/2022 09:50:53. - Campionamento eseguito da: Committente - Trasporto effettuato da:  
Committente  
Sample Condition on Receipt: 20°C

ANALYSIS DESCRIPTION	RESULT	U	REC. %	UNIT OF MEASURE	LQ	LD	METHOD	ANALYSES BEGINNING DATE / ENDING DATE
Water activity (Aw) at 25 °C	0,929	± 0,005			0,150		06(S135) rev6 2021	17/10/2022/ 18/10/2022
<b>VINEGAR ANALYSIS</b>								
Reduction sugars (Reducing substances)	495,9	± 14,9		g/l	1,0		07(S240) 2021 Rev.0 - Titrimetrico	18/10/2022/ 26/10/2022
Total Acidity expressed as acetic acid	3,11	± 0,06		g/100 ml	0,10		* ACIDITA 2014 Rev.0 - Titrimetrico	18/10/2022/ 19/10/2022
Relative density at 20°C	1,195	± 0,0003					07(S117) 2015 Rev.1 - Gravimetric	25/10/2022/ 25/10/2022
Alcoholic strength	0,09	± 0,01		% Vol	0,07		DM 12/03/1986 SO GU n 161 14/07/1986 Met V pag 95, OIV- MA-AS312-01 Met B R2021	18/10/2022/ 19/10/2022
Brookfield rotational viscosity	Brookfield Rotational Viscosity (T = 20 °C, impeller R5)						* VISCOSITA 2018 Rev.1	18/10/2022/ 21/10/2022
	50 rpm: 5124 cP (mPa.s) 30 rpm: 6866 cP (mPa.s) 20 rpm: 8731 cP (mPa.s)							
pH	2,99	± 0,04		unità	0,50		07(S77) 2012 Rev.1 - Potentiometric	18/10/2022/ 19/10/2022
<b>SUGARS HPAEC-PAD</b>								
Galactose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	18/10/2022/ 21/10/2022
Glucose, anhydrous (HPLC)	18,6	± 1,3		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	18/10/2022/ 21/10/2022
Sucrose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	18/10/2022/ 21/10/2022
Fructose, anhydrous (HPLC)	19,6	± 1,4		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	18/10/2022/ 21/10/2022
Lactose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	18/10/2022/ 21/10/2022
Maltose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	18/10/2022/ 21/10/2022
Citric acid (LCMS) [331]	75,6	± 12,6	101	mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	18/10/2022/ 24/10/2022
Lactic Acid (LCMS) [331]	418	± 54	74 +	mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	18/10/2022/ 24/10/2022
Malic acid (LCMS) [331]	575	± 71	90	mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	18/10/2022/ 24/10/2022
Succinic acid (LCMS) [331]	199	± 29	90	mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	18/10/2022/ 24/10/2022
Tartaric acid (LCMS) [331]	570	± 70	80	mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	18/10/2022/ 24/10/2022
Ochratoxin A	< LQ			µg/kg	0,10		03(S130) 2022 Rev.13 - LC- MS/MS	18/10/2022/ 21/10/2022

**END TEST REPORT**

The original document is a PDF file with Digital Signature: 22P11007-In-0-S20B-4000 - T20 - UNIMORE-DigitalSignature.pdf

**Next page...**

MODENA, li 26/10/2022

Sample arrived on the 14/10/2022

Registration date 14/10/2022

## TEST REPORT nr. 22P11007-In-0

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**

**SAMPLE 22P11007**

**MATRIX: Fruit and vegetable concentrates, and by-products**

### Notes and method reference:

< LQ: = lower than Quantification Limit.

U: the reported uncertainty is the expanded uncertainty calculated using a coverage factor equal to 2 which gives a reliability of approximately 95%. Please note that results expressed as '<LQ' may not indicate the absence of the searched parameters in the sample.

MICROBIOLOGICAL TESTS: for food and environmental samples, the extended measurement uncertainty was estimated according to ISO 19036:2019 Standard and is based on a standard uncertainty multiplied by a coverage factor of  $K = 2$ , providing a confidence level of approximately 95%. The combined standard uncertainty was assumed to be equal to the standard deviation of intra-laboratory reproducibility. The results of the microbiological tests are calculated according to the ISO 7218: 2007 / Amd 1: 2013 Standard. If the results are reported as <4 (CFU/ml) or <40 (CFU/g), this means that the microorganisms are present in the sample but in amounts less than 4 CFU/ml or 40 CFU/g respectively. For microbiological analyses unless differently reported in the individual test methods, in case of analytical steps foreseen in non-activity days of the laboratory, provisions of the ISO 7218: 2007 / Amd.1 2013 Standard (points 11.2 and 10.2.5) or from specific test methods are applied. In the case of quantitative microbiological tests, these have been set up on a single plate according to ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise expressly requested by current regulations.

In the case of quantitative microbiological tests, these have been set up on a single plate in accordance with ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise explicitly required by current regulations.

For waters, the measurement uncertainty corresponds to the confidence interval calculated according to ISO 8199: 2018 or to the expanded measurement uncertainty estimated according to ISO 29201: 2012. The results are issued in accordance with ISO 8199: 2018. When the number of colonies detected is <3, the result is expressed as "Microorganisms present in the analyzed volume (N ° colonies detected <3 CFU - reference ISO 8199: 2018, paragraph 9.1.8.4.1)".

LQ: Quantification Limit. It is the lowest analyte concentration which can be detected at an acceptable precision (repeatability) and accuracy, under well defined conditions.

LD: Detection Limit. It is the lowest analyte concentration which can be detected but not necessarily quantified, under well defined conditions. Any fields not filled in the Test Report are to be considered not applicable.

Conformity evaluation: values not complying with laws, decrees, national and EU regulations or specifications supplied by the customer are evaluated case by case, also taking into consideration the uncertainty of measure for each single test and the regulations on rounding-off of values, and pointed out when considered as non conform.

Rec %: Recovery % "+" means that the recovery has been applied to the result. The numeric results between brackets (.) after the expression <LQ are purely indicative of traces that cannot be exactly quantified.

In the case of sampling carried out by Neotron, the laboratory applies the Internal Operating Procedure code: NEOT-DIR/ 006/53.

The laboratory disclaims any responsibility for the information provided by the client reported in this Report which may influence the validity of the results.

Methods marked with an asterisk (\*) are not accredited by ACCREDIA (UNI CEI EN ISO/IEC 17025). The sampling activity is not included within the Scope of Accreditation of Neotron SPA

### NOTES OF PARAMETERS:

[331]: Extended uncertainty calculated following the equation of HORWITZ by using a coverage factor of 2, which guarantee a reliance level of 95%

TEST REPORT VALID FOR ALL LEGAL PURPOSES (Italian R.D. 1-3-1928 n°842 (article 16), - Italian Law 19-7-1957 n°679 articles 16 and 18, Italian Ministerial Decree 25-3-1986).

DATA and SAMPLE STORAGE: Test Reports, Raw data, chromatographic paths and instrumental reports are stored for 5 years. One control sample is stored for 2 months.

Data expressed in this test report refer only to the sample tested in the laboratory. The results reported in this Test Report refer to the sample as received. The description or any other reference concerning the sample are declared by the customer. This Test Report cannot be reproduced except in full. Partial reproductions must be authorized in writing by our laboratory.

THE LABORATORY DIRECTOR: DR. ANDREA RIZZO

THE CHEMIST AUTHORIZED TO SIGN THE TEST REPORTS: DR. MARCO MESCHIARI

NEOTRON SpA - With Sole Shareholder

Stradello Aggazzotti, 104

41126 MODENA - ITALY - Fiscal Code and VAT n° 03807840362

Tel: +39 059461711 - Fax: +39 059461777

www.neotron.it - neotron@neotron.it

Laboratorio Qualificato D.M. 26-2-87 Art. 4 - Legge 46/82 per la Ricerca Applicata e Innovazione Tecnologica.

Regione Emilia Romagna - AUTORIZZAZIONE Autocontrollo N° 008/MO/008

BNN-Monitoring Fruit and Vegetables Approved Laboratory

I-Monitoring EDEKAAG Fruit and Vegetables Registered Laboratory

GMP+ code: GMP051757

MODENA, li 20/10/2022

Sample arrived on the 05/10/2022  
Registration date 05/10/2022**TEST REPORT nr. 22P03160-In-0**

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO****SAMPLE 22P03160****MATRIX: Fruit and vegetable concentrates, and  
by-products**

Description provided by Customer: GLASSA CON ABM IGP - CONTAMINAZIONE BASSA - VISCOSITA': ~ 4.000 CPS (T=10G) - CODICE: S20B-4000

Richiesta via Internet n° N0379/22 - 26/09/2022 09:50:53. - Campionamento eseguito da: Committente - Trasporto effettuato da: Corriere  
Sample Condition on Receipt: 20°C

ANALYSIS DESCRIPTION	RESULT	U	REC. %	UNIT OF MEASURE	LQ	LD	METHOD	ANALYSES BEGINNING DATE / ENDING DATE
Water activity (Aw) at 25 °C	0,934	± 0,005			0,150		06(S135) rev6 2021	05/10/2022 / 11/10/2022
<b>VINEGAR ANALYSIS</b>								
Reduction sugars (Reducing substances)	501,5	± 15,0		g/l	1,0		07(S240) 2021 Rev.0 - Titrimetrico	06/10/2022 / 12/10/2022
Total Acidity expressed as acetic acid	3,22	± 0,06		g/100 ml	0,10		* ACIDITA 2014 Rev.0 - Titrimetrico	06/10/2022 / 06/10/2022
Relative density at 20°C	1,187	± 0,001					07(S117) 2015 Rev.1 - Gravimetric	30/12/1899 / 30/12/1899
Alcoholic strength	0,08	± 0,01		% Vol	0,07		DM 12/03/1986 SO GU n 161 14/07/1986 Met V pag 95, OIV-MA-AS312-01 Met B R2021	06/10/2022 / 12/10/2022
Brookfield rotational viscosity	Brookfield Rotational Viscosity (T = 20 ° C, impeller R5)						* VISCOSITA 2018 Rev.1	06/10/2022 / 12/10/2022
	50 rpm: 5278 cP (mPa.s) 30 rpm: 7123 cP (mPa.s) 20 rpm: 9071 cP (mPa.s)							
pH	2,97	± 0,04		unità	0,50		07(S77) 2012 Rev.1 - Potentiometric	06/10/2022 / 07/10/2022
<b>SUGARS HPAEC-PAD</b>								
Galactose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	06/10/2022 / 12/10/2022
Glucose, anhydrous (HPLC)	17,8	± 1,2		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	06/10/2022 / 12/10/2022
Sucrose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	06/10/2022 / 12/10/2022
Fructose, anhydrous (HPLC)	19,0	± 1,3		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	06/10/2022 / 12/10/2022
Lactose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	06/10/2022 / 12/10/2022
Maltose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	06/10/2022 / 12/10/2022
Citric acid (LCMS) [331]	111	± 18		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	06/10/2022 / 11/10/2022
Lactic Acid (LCMS) [331]	152	± 23		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	06/10/2022 / 11/10/2022
Malic acid (LCMS) [331]	471	± 60		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	06/10/2022 / 11/10/2022
Succinic acid (LCMS) [331]	257	± 36		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	06/10/2022 / 11/10/2022
Tartaric acid (LCMS) [331]	351	± 46		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	06/10/2022 / 11/10/2022
Ochratoxin A	< LQ			µg/kg	0,10		03(S130) 2022 Rev.13 - LC-MS/MS	06/10/2022 / 11/10/2022

**END TEST REPORT**

The original document is a PDF file with Digital Signature: 22P03160-In-0-S20B-4000 - T10 - UNIMORE-DigitalSignature.pdf

**Next page...**

MODENA, li 20/10/2022

Sample arrived on the 05/10/2022

Registration date 05/10/2022

**TEST REPORT nr. 22P03160-In-0**

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**

SAMPLE 22P03160

**MATRIX: Fruit and vegetable concentrates, and  
by-products**

## Notes and method reference:

&lt; LQ: = lower than Quantification Limit.

U: the reported uncertainty is the expanded uncertainty calculated using a coverage factor equal to 2 which gives a reliability of approximately 95%. Please note that results expressed as '<LQ' may not indicate the absence of the searched parameters in the sample.

MICROBIOLOGICAL TESTS: for food and environmental samples, the extended measurement uncertainty was estimated according to ISO 19036:2019 Standard and is based on a standard uncertainty multiplied by a coverage factor of K = 2, providing a confidence level of approximately 95%. The combined standard uncertainty was assumed to be equal to the standard deviation of intra-laboratory reproducibility. The results of the microbiological tests are calculated according to the ISO 7218: 2007 / Amd 1: 2013 Standard. If the results are reported as <4 (CFU/ml) or <40 (CFU/g), this means that the microorganisms are present in the sample but in amounts less than 4 CFU/ml or 40 CFU/g respectively. For microbiological analyses unless differently reported in the individual test methods, in case of analytical steps foreseen in non-activity days of the laboratory, provisions of the ISO 7218: 2007 / Amd.1 2013 Standard (points 11.2 and 10.2.5) or from specific test methods are applied. In the case of quantitative microbiological tests, these have been set up on a single plate according to ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise expressly requested by current regulations.

In the case of quantitative microbiological tests, these have been set up on a single plate in accordance with ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise explicitly required by current regulations.

For waters, the measurement uncertainty corresponds to the confidence interval calculated according to ISO 8199: 2018 or to the expanded measurement uncertainty estimated according to ISO 29201: 2012. The results are issued in accordance with ISO 8199: 2018. When the number of colonies detected is <3, the result is expressed as "Microorganisms present in the analyzed volume (N ° colonies detected <3 CFU - reference ISO 8199: 2018, paragraph 9.1.8.4.1)".

LQ: Quantification Limit. It is the lowest analyte concentration which can be detected at an acceptable precision (repeatability) and accuracy, under well defined conditions.

LD: Detection Limit. It is the lowest analyte concentration which can be detected but not necessarily quantified, under well defined conditions. Any fields not filled in the Test Report are to be considered not applicable.

Conformity evaluation: values not complying with laws, decrees, national and EU regulations or specifications supplied by the customer are evaluated case by case, also taking into consideration the uncertainty of measure for each single test and the regulations on rounding-off of values, and pointed out when considered as non conform.

Rec %: Recovery % "+" means that the recovery has been applied to the result. The numeric results between brackets (.) after the expression <LQ are purely indicative of traces that cannot be exactly quantified.

In the case of sampling carried out by Neotron, the laboratory applies the Internal Operating Procedure code: NEOT-DIR/ 006/53.

The laboratory disclaims any responsibility for the information provided by the client reported in this Report which may influence the validity of the results.

Methods marked with an asterisk (\*) are not accredited by ACCREDIA (UNI CEI EN ISO/IEC 17025). The sampling activity is not included within the Scope of Accreditation of Neotron SPA

## NOTES OF PARAMETERS:

[331]: Extended uncertainty calculated following the equation of HORWITZ by using a coverage factor of 2, which guarantee a reliance level of 95%

TEST REPORT VALID FOR ALL LEGAL PURPOSES (Italian R.D. 1-3-1928 n°842 (article 16), - Italian Law 19-7-1957 n°679 articles 16 and 18, Italian Ministerial Decree 25-3-1986).

DATA and SAMPLE STORAGE: Test Reports, Raw data, chromatographic paths and instrumental reports are stored for 5 years. One control sample is stored for 2 months.

Data expressed in this test report refer only to the sample tested in the laboratory. The results reported in this Test Report refer to the sample as received. The description or any other reference concerning the sample are declared by the customer. This Test Report cannot be reproduced except in full. Partial reproductions must be authorized in writing by our laboratory.

THE LABORATORY DIRECTOR: DR. ANDREA RIZZO

THE CHEMIST AUTHORIZED TO SIGN THE TEST REPORTS: DR. MARCO MESCHIARI

NEOTRON SpA - With Sole Shareholder

Stradello Aggazzotti, 104

41126 MODENA - ITALY - Fiscal Code and VAT n° 03807840362

Tel: +39 059461711 - Fax: +39 059461777

www.neotron.it - neotron@neotron.it

Laboratorio Qualificato D.M. 26-2-87 Art. 4 - Legge 46/82 per la Ricerca Applicata e Innovazione Tecnologica.

Regione Emilia Romagna - AUTORIZZAZIONE Autocontrollo N° 008/MO/008

BNN-Monitoring Fruit and Vegetables Approved Laboratory

I-Monitoring EDEKAAG Fruit and Vegetables Registered Laboratory

GMP+ code: GMP051757



MODENA, li 12/10/2022

Sample arrived on the 26/09/2022

Registration date 26/09/2022

**TEST REPORT nr. 22M18693-In-0**

CUSTOMER

**ACETFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**
**SAMPLE 22M18693****MATRIX: Fruit and vegetable concentrates, and by-products**

Description provided by Customer: GLASSA CON ABM IGP - CONTAMINAZIONE BASSA - VISCOSITA': ~ 4.000 CPS (T=0G) - CODICE: S20B-4000

Richiesta via Internet n° N0377/22 - 26/09/2022 09:50:52. - Campionamento eseguito da: Committente - Trasporto effettuato da: Committente  
Sample Condition on Receipt: 5°C

ANALYSIS DESCRIPTION	RESULT	U	REC. %	UNIT OF MEASURE	LQ	LD	METHOD	ANALYSES BEGINNING DATE / ENDING DATE
Water activity (Aw) at 25 °C	0,921	± 0,005			0,150		06(S135) rev6 2021	27/09/2022 / 30/09/2022
<b>VINEGAR ANALYSIS</b>								
Reduction sugars (Reducing substances)	491,6	± 14,7		g/l	1,0		07(S240) 2021 Rev.0 - Titrimetrico	28/09/2022 / 07/10/2022
Total Acidity expressed as acetic acid	3,11	± 0,06		g/100 ml	0,10		* ACIDITA 2014 Rev.0 - Titrimetrico	28/09/2022 / 30/09/2022
Relative density at 20°C	1,208	± 0,001					- Gravimetric	11/10/2022 / 11/10/2022
Alcoholic strength	0,09	± 0,01		% Vol	0,07		DM 12/03/1986 SO GU n 161 14/07/1986 Met V pag 95, OIV-MA-AS312-01 Met B R2021	27/09/2022 / 06/10/2022
Brookfield rotational viscosity	Brookfield Rotational Viscosity (T = 20 ° C, impeller R5)						* VISCOSITA 2018 Rev.1	28/09/2022 / 11/10/2022
	60 rpm: 4391 cP (mPa.s)							
	50 rpm: 4962 cP (mPa.s)							
pH	3,00	± 0,04		unità	0,50		07(S77) 2012 Rev.1 - Potentiometric	28/09/2022 / 03/10/2022
<b>SUGARS HPAEC-PAD</b>								
Galactose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	28/09/2022 / 12/10/2022
Glucose, anhydrous (HPLC)	18,9	± 1,3		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	28/09/2022 / 12/10/2022
Sucrose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	28/09/2022 / 12/10/2022
Fructose, anhydrous (HPLC)	19,5	± 1,4		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	28/09/2022 / 12/10/2022
Lactose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	28/09/2022 / 12/10/2022
Maltose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	28/09/2022 / 12/10/2022
Citric acid (LCMS) [331]	71,7	± 12,0		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	28/09/2022 / 06/10/2022
Lactic Acid (LCMS) [331]	425	± 55		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	28/09/2022 / 06/10/2022
Malic acid (LCMS) [331]	641	± 78		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	28/09/2022 / 06/10/2022
Succinic acid (LCMS) [331]	207	± 30		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	28/09/2022 / 06/10/2022
Tartaric acid (LCMS) [331]	603	± 74		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	28/09/2022 / 06/10/2022
Ochratoxin A	< LQ			µg/kg	0,10		03(S130) 2022 Rev.13 - LC-MS/MS	28/09/2022 / 07/10/2022

**END TEST REPORT**

The original document is a PDF file with Digital Signature: 22M18693-In-0-S20B-4000 - T0 - UNIMORE-DigitalSignature.pdf

**Next page...**

MODENA, li 12/10/2022

Sample arrived on the 26/09/2022

Registration date 26/09/2022

**TEST REPORT nr. 22M18693-In-0**

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**

SAMPLE 22M18693

**MATRIX: Fruit and vegetable concentrates, and  
by-products**

## Notes and method reference:

&lt; LQ: = lower than Quantification Limit.

U: the reported uncertainty is the expanded uncertainty calculated using a coverage factor equal to 2 which gives a reliability of approximately 95%. Please note that results expressed as '<LQ' may not indicate the absence of the searched parameters in the sample.

MICROBIOLOGICAL TESTS: for food and environmental samples, the extended measurement uncertainty was estimated according to ISO 19036:2019 Standard and is based on a standard uncertainty multiplied by a coverage factor of  $K = 2$ , providing a confidence level of approximately 95%. The combined standard uncertainty was assumed to be equal to the standard deviation of intra-laboratory reproducibility. The results of the microbiological tests are calculated according to the ISO 7218: 2007 / Amd 1: 2013 Standard. If the results are reported as <4 (CFU/ml) or <40 (CFU/g), this means that the microorganisms are present in the sample but in amounts less than 4 CFU/ml or 40 CFU/g respectively. For microbiological analyses unless differently reported in the individual test methods, in case of analytical steps foreseen in non-activity days of the laboratory, provisions of the ISO 7218: 2007 / Amd.1 2013 Standard (points 11.2 and 10.2.5) or from specific test methods are applied. In the case of quantitative microbiological tests, these have been set up on a single plate according to ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise expressly requested by current regulations.

In the case of quantitative microbiological tests, these have been set up on a single plate in accordance with ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise explicitly required by current regulations.

For waters, the measurement uncertainty corresponds to the confidence interval calculated according to ISO 8199: 2018 or to the expanded measurement uncertainty estimated according to ISO 29201: 2012. The results are issued in accordance with ISO 8199: 2018. When the number of colonies detected is <3, the result is expressed as "Microorganisms present in the analyzed volume (N ° colonies detected <3 CFU - reference ISO 8199: 2018, paragraph 9.1.8.4.1)".

LQ: Quantification Limit. It is the lowest analyte concentration which can be detected at an acceptable precision (repeatability) and accuracy, under well defined conditions.

LD: Detection Limit. It is the lowest analyte concentration which can be detected but not necessarily quantified, under well defined conditions. Any fields not filled in the Test Report are to be considered not applicable.

Conformity evaluation: values not complying with laws, decrees, national and EU regulations or specifications supplied by the customer are evaluated case by case, also taking into consideration the uncertainty of measure for each single test and the regulations on rounding-off of values, and pointed out when considered as non conform.

Rec %: Recovery % "+" means that the recovery has been applied to the result. The numeric results between brackets (.) after the expression <LQ are purely indicative of traces that cannot be exactly quantified.

In the case of sampling carried out by Neotron, the laboratory applies the Internal Operating Procedure code: NEOT-DIR/ 006/53.

The laboratory disclaims any responsibility for the information provided by the client reported in this Report which may influence the validity of the results.

Methods marked with an asterisk (\*) are not accredited by ACCREDIA (UNI CEI EN ISO/IEC 17025). The sampling activity is not included within the Scope of Accreditation of Neotron SPA

## NOTES OF PARAMETERS:

[331]: Extended uncertainty calculated following the equation of HORWITZ by using a coverage factor of 2, which guarantee a reliance level of 95%

TEST REPORT VALID FOR ALL LEGAL PURPOSES (Italian R.D. 1-3-1928 n°842 (article 16), - Italian Law 19-7-1957 n°679 articles 16 and 18, Italian Ministerial Decree 25-3-1986).

DATA and SAMPLE STORAGE: Test Reports, Raw data, chromatographic paths and instrumental reports are stored for 5 years. One control sample is stored for 2 months.

Data expressed in this test report refer only to the sample tested in the laboratory. The results reported in this Test Report refer to the sample as received. The description or any other reference concerning the sample are declared by the customer. This Test Report cannot be reproduced except in full. Partial reproductions must be authorized in writing by our laboratory.

THE LABORATORY DIRECTOR: DR. ANDREA RIZZO

THE CHEMIST AUTHORIZED TO SIGN THE TEST REPORTS: DR. MARCO MESCHIARI

NEOTRON SpA - With Sole Shareholder

Stradello Aggazzotti, 104

41126 MODENA - ITALY - Fiscal Code and VAT n° 03807840362

Tel: +39 059461711 - Fax: +39 059461777

www.neotron.it - neotron@neotron.it

Laboratorio Qualificato D.M. 26-2-87 Art. 4 - Legge 46/82 per la Ricerca Applicata e Innovazione Tecnologica.

Regione Emilia Romagna - AUTORIZZAZIONE Autocontrollo N° 008/MO/008

BNN-Monitoring Fruit and Vegetables Approved Laboratory

I-Monitoring EDEKAAG Fruit and Vegetables Registered Laboratory

GMP+ code: GMP051757

MODENA, li 13/10/2022

Sample arrived on the 23/09/2022  
Registration date 23/09/2022**TEST REPORT nr. 22M17703-In-0**

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO****SAMPLE 22M17703****MATRIX: Fruit and vegetable concentrates, and by-products**

Description provided by Customer: GLASSA CON ABM IGP - CONTAMINAZIONE BASSA - VISCOSITA': ~ 3.000 CPS (T=20G) - CODICE: S20B-3000

Richiesta via Internet n° N0328/22 - 05/09/2022 13:57:34. - Campionamento eseguito da: Committente - Trasporto effettuato da: Committente  
Sample Condition on Receipt: 20°C

ANALYSIS DESCRIPTION	RESULT	U	REC. %	UNIT OF MEASURE	LQ	LD	METHOD	ANALYSES BEGINNING DATE / ENDING DATE
Water activity (Aw) at 25 °C	0,934	± 0,005			0,150		06(S135) rev6 2021	23/09/2022 / 30/09/2022
<b>VINEGAR ANALYSIS</b>								
Reduction sugars (Reducing substances)	453,4	± 13,6		g/l	1,0		07(S240) 2021 Rev.0 - Titrimetrico	24/09/2022 / 30/09/2022
Total Acidity expressed as acetic acid	2,87	± 0,06		g/100 ml	0,10		* ACIDITA 2014 Rev.0 - Titrimetrico	24/09/2022 / 29/09/2022
Relative density at 20°C	1,228	± 0,001					DM 12/03/1986 SO GU n 161 14/07/1986 Met VI pag 95 p.to 4, OIV-MA-AS2-01A Met B R2021 - densimetry	24/09/2022 / 30/09/2022
Alcoholic strength	0,10	± 0,01		% Vol	0,07		DM 12/03/1986 SO GU n 161 14/07/1986 Met V pag 95, OIV-MA-AS312-01 Met B R2021	24/09/2022 / 30/09/2022
pH	3,01	± 0,04		unità	0,50		07(S77) 2012 Rev.1 - Potentiometric	24/09/2022 / 13/10/2022
Brookfield rotational viscosity	Brookfield Rotational Viscosity (T = 20 ° C, impeller R3)						* VISCOSITA 2018 Rev.1	24/09/2022 / 06/10/2022
	30 rpm: 2206 cP (mPa.s) 20 rpm: 2709 cP (mPa.s) 12 rpm: 3498 cP (mPa.s)							
<b>SUGARS HPAEC-PAD</b>								
Galactose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	24/09/2022 / 12/10/2022
Glucose, anhydrous (HPLC)	17,1	± 1,2		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	24/09/2022 / 12/10/2022
Sucrose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	24/09/2022 / 12/10/2022
Fructose, anhydrous (HPLC)	20,1	± 1,4		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	24/09/2022 / 12/10/2022
Lactose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	24/09/2022 / 12/10/2022
Maltose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	24/09/2022 / 12/10/2022
Citric acid (LCMS) [331]	68,0	± 11,6		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	24/09/2022 / 06/10/2022
Lactic Acid (LCMS) [331]	285	± 39		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	24/09/2022 / 06/10/2022
Malic acid (LCMS) [331]	601	± 73		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	24/09/2022 / 06/10/2022
Succinic acid (LCMS) [331]	195	± 28		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	24/09/2022 / 06/10/2022
Tartaric acid (LCMS) [331]	570	± 70		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	24/09/2022 / 06/10/2022
Ochratoxin A	< LQ			µg/kg	0,10		03(S130) 2022 Rev.13 - LC-MS/MS	24/09/2022 / 05/10/2022

**END TEST REPORT**

The original document is a PDF file with Digital Signature: 22M17703-In-0-S20B-3000 - T20 - UNIMORE-DigitalSignature.pdf

MODENA, li 13/10/2022

Sample arrived on the 23/09/2022

Registration date 23/09/2022

**TEST REPORT nr. 22M17703-In-0**

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**

SAMPLE 22M17703

**MATRIX: Fruit and vegetable concentrates, and  
by-products**

## Notes and method reference:

&lt; LQ: = lower than Quantification Limit.

U: the reported uncertainty is the expanded uncertainty calculated using a coverage factor equal to 2 which gives a reliability of approximately 95%. Please note that results expressed as '<LQ' may not indicate the absence of the searched parameters in the sample.

MICROBIOLOGICAL TESTS: for food and environmental samples, the extended measurement uncertainty was estimated according to ISO 19036:2019 Standard and is based on a standard uncertainty multiplied by a coverage factor of K = 2, providing a confidence level of approximately 95%. The combined standard uncertainty was assumed to be equal to the standard deviation of intra-laboratory reproducibility. The results of the microbiological tests are calculated according to the ISO 7218: 2007 / Amd 1: 2013 Standard. If the results are reported as <4 (CFU/ml) or <40 (CFU/g), this means that the microorganisms are present in the sample but in amounts less than 4 CFU/ml or 40 CFU/g respectively. For microbiological analyses unless differently reported in the individual test methods, in case of analytical steps foreseen in non-activity days of the laboratory, provisions of the ISO 7218: 2007 / Amd.1 2013 Standard (points 11.2 and 10.2.5) or from specific test methods are applied. In the case of quantitative microbiological tests, these have been set up on a single plate according to ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise expressly requested by current regulations.

In the case of quantitative microbiological tests, these have been set up on a single plate in accordance with ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise explicitly required by current regulations.

For waters, the measurement uncertainty corresponds to the confidence interval calculated according to ISO 8199: 2018 or to the expanded measurement uncertainty estimated according to ISO 29201: 2012. The results are issued in accordance with ISO 8199: 2018. When the number of colonies detected is <3, the result is expressed as "Microorganisms present in the analyzed volume (N ° colonies detected <3 CFU - reference ISO 8199: 2018, paragraph 9.1.8.4.1)".

LQ: Quantification Limit. It is the lowest analyte concentration which can be detected at an acceptable precision (repeatability) and accuracy, under well defined conditions.

LD: Detection Limit. It is the lowest analyte concentration which can be detected but not necessarily quantified, under well defined conditions. Any fields not filled in the Test Report are to be considered not applicable.

Conformity evaluation: values not complying with laws, decrees, national and EU regulations or specifications supplied by the customer are evaluated case by case, also taking into consideration the uncertainty of measure for each single test and the regulations on rounding-off of values, and pointed out when considered as non conform.

Rec %: Recovery % "+" means that the recovery has been applied to the result. The numeric results between brackets (.) after the expression <LQ are purely indicative of traces that cannot be exactly quantified.

In the case of sampling carried out by Neotron, the laboratory applies the Internal Operating Procedure code: NEOT-DIR/ 006/53.

The laboratory disclaims any responsibility for the information provided by the client reported in this Report which may influence the validity of the results.

Methods marked with an asterisk (\*) are not accredited by ACCREDIA (UNI CEI EN ISO/IEC 17025). The sampling activity is not included within the Scope of Accreditation of Neotron SPA

## NOTES OF PARAMETERS:

[331]: Extended uncertainty calculated following the equation of HORWITZ by using a coverage factor of 2, which guarantee a reliance level of 95%

TEST REPORT VALID FOR ALL LEGAL PURPOSES (Italian R.D. 1-3-1928 n°842 (article 16), - Italian Law 19-7-1957 n°679 articles 16 and 18, Italian Ministerial Decree 25-3-1986).

DATA and SAMPLE STORAGE: Test Reports, Raw data, chromatographic paths and instrumental reports are stored for 5 years. One control sample is stored for 2 months.

Data expressed in this test report refer only to the sample tested in the laboratory. The results reported in this Test Report refer to the sample as received. The description or any other reference concerning the sample are declared by the customer. This Test Report cannot be reproduced except in full. Partial reproductions must be authorized in writing by our laboratory.

THE LABORATORY DIRECTOR: DR. ANDREA RIZZO

THE CHEMIST AUTHORIZED TO SIGN THE TEST REPORTS: DR. MARCO MESCHIARI

NEOTRON SpA - With Sole Shareholder

Stradello Aggazzotti, 104

41126 MODENA - ITALY - Fiscal Code and VAT n° 03807840362

Tel: +39 059461711 - Fax: +39 059461777

www.neotron.it - neotron@neotron.it

Laboratorio Qualificato D.M. 26-2-87 Art. 4 - Legge 46/82 per la Ricerca Applicata e Innovazione Tecnologica.

Regione Emilia Romagna - AUTORIZZAZIONE Autocontrollo N° 008/MO/008

BNN-Monitoring Fruit and Vegetables Approved Laboratory

I-Monitoring EDEKAAG Fruit and Vegetables Registered Laboratory

GMP+ code: GMP051757

MODENA, li 26/09/2022

Sample arrived on the 13/09/2022

Registration date 13/09/2022

**TEST REPORT nr. 22M08847-In-0**

CUSTOMER

**ACETFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**
**SAMPLE 22M08847****MATRIX: Fruit and vegetable concentrates, and by-products**

Description provided by Customer: GLASSA CON ABM IGP - CONTAMINAZIONE BASSA - VISCOSITA': ~ 3.000 CPS (T=10G) - CODICE: S20B-3000

Richiesta via Internet n° N0329/22 - 05/09/2022 13:57:34. - Campionamento eseguito da: Committente - Trasporto effettuato da: Corriere  
Sample Condition on Receipt: 23°C

ANALYSIS DESCRIPTION	RESULT	U	REC. %	UNIT OF MEASURE	LQ	LD	METHOD	ANALYSES BEGINNING DATE / ENDING DATE
Water activity (Aw) at 25 °C	0,930	± 0,005			0,150		06(S135) rev6 2021	14/09/2022 / 22/09/2022
<b>VINEGAR ANALYSIS</b>								
Reduction sugars (Reducing substances)	417,2	± 12,5		g/l	1,0		07(S240) 2021 Rev.0 - Titrimetrico	16/09/2022 / 20/09/2022
Total Acidity expressed as acetic acid	2,57	± 0,05		g/100 ml	0,10		* ACIDITA 2014 Rev.0 - Titrimetrico	16/09/2022 / 20/09/2022
Relative density at 20°C	1,175	± 0,001					DM 12/03/1986 SO GU n 161 14/07/1986 Met VI pag 95 p.to 4, OIV-MA-AS2-01A Met B R2021 - densimetry	16/09/2022 / 20/09/2022
Alcoholic strength	0,11	± 0,01		% Vol	0,07		DM 12/03/1986 SO GU n 161 14/07/1986 Met V pag 95, OIV-MA-AS312-01 Met B R2021	16/09/2022 / 20/09/2022
Brookfield rotational viscosity	Brookfield Rotational Viscosity (T = 20 ° C, impeller R3)						* VISCOSITA 2018 Rev.1	16/09/2022 / 21/09/2022
	30 rpm: 2244 cP (mPa.s) 20 rpm: 2763 cP (mPa.s) 12 rpm: 3582 cP (mPa.s)							
pH	2,94	± 0,04		unità	0,50		07(S77) 2012 Rev.1 - Potentiometric	16/09/2022 / 23/09/2022
<b>SUGARS HPAEC-PAD</b>								
Galactose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	16/09/2022 / 20/09/2022
Glucose, anhydrous (HPLC)	17,3	± 1,2		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	16/09/2022 / 20/09/2022
Sucrose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	16/09/2022 / 20/09/2022
Fructose, anhydrous (HPLC)	17,1	± 1,2		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	16/09/2022 / 20/09/2022
Lactose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	16/09/2022 / 20/09/2022
Maltose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	16/09/2022 / 20/09/2022
Citric acid (LCMS) [331]	80,8	± 13,3		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	16/09/2022 / 23/09/2022
Lactic Acid (LCMS) [331]	528	± 65		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	16/09/2022 / 23/09/2022
Malic acid (LCMS) [331]	523	± 65		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	16/09/2022 / 23/09/2022
Succinic acid (LCMS) [331]	221	± 31		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	16/09/2022 / 23/09/2022
Tartaric acid (LCMS) [331]	567	± 70		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	16/09/2022 / 23/09/2022
Ochratoxin A	< LQ			µg/kg	0,10		03(S130) 2022 Rev.13 - LC-MS/MS	16/09/2022 / 20/09/2022

**END TEST REPORT**

The original document is a PDF file with Digital Signature: 22M08847-In-0-S20B-3000 - T10 - UNIMORE-DigitalSignature.pdf

MODENA, li 26/09/2022

Sample arrived on the 13/09/2022

Registration date 13/09/2022

## TEST REPORT nr. 22M08847-In-0

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**

**SAMPLE 22M08847**

**MATRIX: Fruit and vegetable concentrates, and by-products**

### Notes and method reference:

< LQ: = lower than Quantification Limit.

U: the reported uncertainty is the expanded uncertainty calculated using a coverage factor equal to 2 which gives a reliability of approximately 95%. Please note that results expressed as '<LQ' may not indicate the absence of the searched parameters in the sample.

MICROBIOLOGICAL TESTS: for food and environmental samples, the extended measurement uncertainty was estimated according to ISO 19036:2019 Standard and is based on a standard uncertainty multiplied by a coverage factor of K = 2, providing a confidence level of approximately 95%. The combined standard uncertainty was assumed to be equal to the standard deviation of intra-laboratory reproducibility. The results of the microbiological tests are calculated according to the ISO 7218: 2007 / Amd 1: 2013 Standard. If the results are reported as <4 (CFU/ml) or <40 (CFU/g), this means that the microorganisms are present in the sample but in amounts less than 4 CFU/ml or 40 CFU/g respectively. For microbiological analyses unless differently reported in the individual test methods, in case of analytical steps foreseen in non-activity days of the laboratory, provisions of the ISO 7218: 2007 / Amd.1 2013 Standard (points 11.2 and 10.2.5) or from specific test methods are applied. In the case of quantitative microbiological tests, these have been set up on a single plate according to ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise expressly requested by current regulations.

In the case of quantitative microbiological tests, these have been set up on a single plate in accordance with ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise explicitly required by current regulations.

For waters, the measurement uncertainty corresponds to the confidence interval calculated according to ISO 8199: 2018 or to the expanded measurement uncertainty estimated according to ISO 29201: 2012. The results are issued in accordance with ISO 8199: 2018. When the number of colonies detected is <3, the result is expressed as "Microorganisms present in the analyzed volume (N ° colonies detected <3 CFU - reference ISO 8199: 2018, paragraph 9.1.8.4.1)".

LQ: Quantification Limit. It is the lowest analyte concentration which can be detected at an acceptable precision (repeatability) and accuracy, under well defined conditions.

LD: Detection Limit. It is the lowest analyte concentration which can be detected but not necessarily quantified, under well defined conditions. Any fields not filled in in the Test Report are to be considered not applicable.

Conformity evaluation: values not complying with laws, decrees, national and EU regulations or specifications supplied by the customer are evaluated case by case, also taking into consideration the uncertainty of measure for each single test and the regulations on rounding-off of values, and pointed out when considered as non conform.

Rec %: Recovery % "+" means that the recovery has been applied to the result. The numeric results between brackets (.) after the expression <LQ are purely indicative of traces that cannot be exactly quantified.

In the case of sampling carried out by Neotron, the laboratory applies the Internal Operating Procedure code: NEOT-DIR/ 006/53.

The laboratory disclaims any responsibility for the information provided by the client reported in this Report which may influence the validity of the results.

Methods marked with an asterisk (\*) are not accredited by ACCREDIA (UNI CEI EN ISO/IEC 17025). The sampling activity is not included within the Scope of Accreditation of Neotron SPA

### NOTES OF PARAMETERS:

[331]: Extended uncertainty calculated following the equation of HORWITZ by using a coverage factor of 2, which guarantee a reliance level of 95%

TEST REPORT VALID FOR ALL LEGAL PURPOSES (Italian R.D. 1-3-1928 n°842 (article 16), - Italian Law 19-7-1957 n°679 articles 16 and 18, Italian Ministerial Decree 25-3-1986).

DATA and SAMPLE STORAGE: Test Reports, Raw data, chromatographic paths and instrumental reports are stored for 5 years. One control sample is stored for 2 months.

Data expressed in this test report refer only to the sample tested in the laboratory. The results reported in this Test Report refer to the sample as received. The description or any other reference concerning the sample are declared by the customer. This Test Report cannot be reproduced except in full. Partial reproductions must be authorized in writing by our laboratory.

THE LABORATORY DIRECTOR: DR. ANDREA RIZZO

THE CHEMIST AUTHORIZED TO SIGN THE TEST REPORTS: DR. BARBARA MALAGOLI

Approved by Analysis Manager - laboratory LMAA-Nut

Approved by Analysis Manager - laboratory ICS-HPLC

Approved by Analysis Manager - laboratory LC-MICO

Approved by Analysis Manager - laboratory LC-AN

Approved by Analysis Manager - laboratory FISICHE

Approved by Analysis Manager - laboratory LBM-EL

Approved by Analysis Manager - laboratory LMAA-Bro

NEOTRON SpA - With Sole Shareholder

Stradello Aggazzotti, 104

41126 MODENA - ITALY - Fiscal Code and VAT n° 03807840362

Tel: +39 059461711 - Fax: +39 059461777

www.neotron.it - neotron@neotron.it

Laboratorio Qualificato D.M. 26-2-87 Art. 4 - Legge 46/82 per la Ricerca Applicata e Innovazione Tecnologica.

Regione Emilia Romagna - AUTORIZZAZIONE Autocontrollo N° 008/MO/008

BNN-Monitoring Fruit and Vegetables Approved Laboratory

I-Monitoring EDEKAAG Fruit and Vegetables Registered Laboratory

GMP+ code: GMP051757

MODENA, li 13/09/2022

Sample arrived on the 05/09/2022  
Registration date 05/09/2022**TEST REPORT nr. 22M02963-In-0**

CUSTOMER

**ACETFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO****SAMPLE 22M02963****MATRIX: Fruit and vegetable concentrates, and  
by-products**Description provided by Customer: GLASSA CON ABM IGP - CONTAMINAZIONE BASSA - VISCOSITA': ~ 3.000 CPS (T=0G) -  
CODICE: S20B-3000Richiesta via Internet n° N0330/22 - 05/09/2022 13:57:34. - Campionamento eseguito da: Committente - Trasporto effettuato da:  
Committente  
Sample Condition on Receipt: refrigerated

ANALYSIS DESCRIPTION	RESULT	U	REC. %	UNIT OF MEASURE	LQ	LD	METHOD	ANALYSES BEGINNING DATE / ENDING DATE
Water activity (Aw) at 25 °C	0,937	± 0,005			0,150		06(S135) rev6 2021	06/09/2022 / 12/09/2022
<b>VINEGAR ANALYSIS</b>								
Reduction sugars (Reducing substances)	460,6	± 13,8		g/l	1,0		07(S240) 2021 Rev.0 - Titrimetrico	07/09/2022 / 09/09/2022
Total Acidity expressed as acetic acid	2,84	± 0,06		g/100 ml	0,10		* ACIDITA 2014 Rev.0 - Titrimetrico	07/09/2022 / 12/09/2022
Relative density at 20°C	1,183	± 0,001					07(S117) 2015 Rev.1 - Gravimetric	13/09/2022 / 13/09/2022
Alcoholic strength	< LQ			% Vol	0,07		DM 12/03/1986 SO GU n 161 14/07/1986 Met V pag 95, OIV- MA-AS312-01 Met B R2021	07/09/2022 / 09/09/2022
pH	2,97	± 0,04		unità	0,50		07(S77) 2012 Rev.1 - Potentiometric	07/09/2022 / 12/09/2022
Brookfield rotational viscosity	Brookfield Rotational Viscosity (T = 20 ° C, impeller R3)  30 rpm: 2024 cP (mPa.s) 20 rpm: 2458 cP (mPa.s) 12 rpm: 3189 cP (mPa.s)						* VISCOSITA 2018 Rev.1	07/09/2022 / 12/09/2022
<b>SUGARS HPAEC-PAD</b>								
Galactose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	07/09/2022 / 13/09/2022
Glucose, anhydrous (HPLC)	17,1	± 1,2		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	07/09/2022 / 13/09/2022
Sucrose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	07/09/2022 / 13/09/2022
Fructose, anhydrous (HPLC)	17,2	± 1,2		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	07/09/2022 / 13/09/2022
Lactose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	07/09/2022 / 13/09/2022
Maltose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	07/09/2022 / 13/09/2022
Citric acid (LCMS) [331]	86,1	± 14,1		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	07/09/2022 / 09/09/2022
Lactic Acid (LCMS) [331]	240	± 34		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	07/09/2022 / 09/09/2022
Malic acid (LCMS) [331]	596	± 73		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	07/09/2022 / 09/09/2022
Succinic acid (LCMS) [331]	208	± 30		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	07/09/2022 / 09/09/2022
Tartaric acid (LCMS) [331]	611	± 75		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	07/09/2022 / 09/09/2022
Ochratoxin A	< LQ			µg/kg	0,10		03(S130) 2022 Rev.13 - LC- MS/MS	07/09/2022 / 09/09/2022

**END TEST REPORT**

The original document is a PDF file with Digital Signature: 22M02963-In-0-S20B-3000 - T0 - UNIMORE-DigitalSignature.pdf

**Next page...**

MODENA, li 13/09/2022

Sample arrived on the 05/09/2022

Registration date 05/09/2022

## TEST REPORT nr. 22M02963-In-0

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**

**SAMPLE 22M02963**

**MATRIX: Fruit and vegetable concentrates, and by-products**

### Notes and method reference:

< LQ: = lower than Quantification Limit.

U: the reported uncertainty is the expanded uncertainty calculated using a coverage factor equal to 2 which gives a reliability of approximately 95%. Please note that results expressed as '<LQ' may not indicate the absence of the searched parameters in the sample.

MICROBIOLOGICAL TESTS: for food and environmental samples, the extended measurement uncertainty was estimated according to ISO 19036:2019 Standard and is based on a standard uncertainty multiplied by a coverage factor of  $K = 2$ , providing a confidence level of approximately 95%. The combined standard uncertainty was assumed to be equal to the standard deviation of intra-laboratory reproducibility. The results of the microbiological tests are calculated according to the ISO 7218: 2007 / Amd 1: 2013 Standard. If the results are reported as <4 (CFU/ml) or <40 (CFU/g), this means that the microorganisms are present in the sample but in amounts less than 4 CFU/ml or 40 CFU/g respectively. For microbiological analyses unless differently reported in the individual test methods, in case of analytical steps foreseen in non-activity days of the laboratory, provisions of the ISO 7218: 2007 / Amd.1 2013 Standard (points 11.2 and 10.2.5) or from specific test methods are applied. In the case of quantitative microbiological tests, these have been set up on a single plate according to ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise expressly requested by current regulations.

In the case of quantitative microbiological tests, these have been set up on a single plate in accordance with ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise explicitly required by current regulations.

For waters, the measurement uncertainty corresponds to the confidence interval calculated according to ISO 8199: 2018 or to the expanded measurement uncertainty estimated according to ISO 29201: 2012. The results are issued in accordance with ISO 8199: 2018. When the number of colonies detected is <3, the result is expressed as "Microorganisms present in the analyzed volume (N ° colonies detected <3 CFU - reference ISO 8199: 2018, paragraph 9.1.8.4.1)".

LQ: Quantification Limit. It is the lowest analyte concentration which can be detected at an acceptable precision (repeatability) and accuracy, under well defined conditions.

LD: Detection Limit. It is the lowest analyte concentration which can be detected but not necessarily quantified, under well defined conditions.

Conformity evaluation: values not complying with laws, decrees, national and EU regulations or specifications supplied by the customer are evaluated case by case, also taking into consideration the uncertainty of measure for each single test and the regulations on rounding-off of values, and pointed out when considered as non conform.

Rec %: Recovery % "+" means that the recovery has been applied to the result. The numeric results between brackets (..) after the expression <LQ are purely indicative of traces that cannot be exactly quantified.

In the case of sampling carried out by Neotron, the laboratory applies the Internal Operating Procedure code: NEOT-DIR/ 006/53.

The laboratory disclaims any responsibility for the information provided by the client reported in this Report which may influence the validity of the results.

Methods marked with an asterisk (\*) are not accredited by ACCREDIA (UNI CEI EN ISO/IEC 17025). The sampling activity is not included within the Scope of Accreditation of Neotron SPA

### NOTES OF PARAMETERS:

[331]: Extended uncertainty calculated following the equation of HORWITZ by using a coverage factor of 2, which guarantee a reliance level of 95%

TEST REPORT VALID FOR ALL LEGAL PURPOSES (Italian R.D. 1-3-1928 n°842 (article 16), - Italian Law 19-7-1957 n°679 articles 16 and 18, Italian Ministerial Decree 25-3-1986).

DATA and SAMPLE STORAGE: Test Reports, Raw data, chromatographic paths and instrumental reports are stored for 5 years. One control sample is stored for 2 months.

Data expressed in this test report refer only to the sample tested in the laboratory. The results reported in this Test Report refer to the sample as received. The description or any other reference concerning the sample are declared by the customer. This Test Report cannot be reproduced except in full. Partial reproductions must be authorized in writing by our laboratory.

THE LABORATORY DIRECTOR: DR. ANDREA RIZZO

THE CHEMIST AUTHORIZED TO SIGN THE TEST REPORTS: DR. MARCO MESCHIARI

Approved by Analysis Manager - laboratory LMAA-Nut  
Approved by Analysis Manager - laboratory ICS-HPLC  
Approved by Analysis Manager - laboratory LC-MICO  
Approved by Analysis Manager - laboratory LC-AN

Approved by Analysis Manager - laboratory FISICHE  
Approved by Analysis Manager - laboratory LBM-EL  
Approved by Analysis Manager - laboratory LMAA-Bro

**NEOTRON SpA** - With Sole Shareholder

Stradello Aggazzotti, 104

41126 MODENA - ITALY - Fiscal Code and VAT n° 03807840362

Tel: +39 059461711 - Fax: +39 059461777

www.neotron.it - neotron@neotron.it

Laboratorio Qualificato D.M. 26-2-87 Art. 4 - Legge 46/82 per la Ricerca Applicata e Innovazione Tecnologica.

Regione Emilia Romagna - AUTORIZZAZIONE Autocontrollo N° 008/MO/008

BNN-Monitoring Fruit and Vegetables Approved Laboratory

I-Monitoring EDEKAAG Fruit and Vegetables Registered Laboratory

GMP+ code: GMP051757



MODENA, li 13/09/2022

Sample arrived on the 05/09/2022  
Registration date 05/09/2022**TEST REPORT nr. 22M02961-In-0**

CUSTOMER

**ACETFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO****SAMPLE 22M02961****MATRIX: Fruit and vegetable concentrates, and  
by-products**

Description provided by Customer: GLASSA CON ABM IGP - CONTAMINAZIONE BASSA - VISCOSITA': ~ 2.000 CPS (T=20G) - CODICE: S20B-2000

Richiesta via Internet n° N0316/22 - 25/08/2022 09:11:13. - Campionamento eseguito da: Committente - Trasporto effettuato da: Committente  
Sample Condition on Receipt: Room temperature

ANALYSIS DESCRIPTION	RESULT	U	REC. %	UNIT OF MEASURE	LQ	LD	METHOD	ANALYSES BEGINNING DATE / ENDING DATE
Water activity (Aw) at 25 °C	0,938	± 0,005			0,150		06(S135) rev6 2021	06/09/2022 / 12/09/2022
<b>VINEGAR ANALYSIS</b>								
Reduction sugars (Reducing substances)	443,1	± 13,3		g/l	1,0		07(S240) 2021 Rev.0 - Titrimetrico	07/09/2022 / 09/09/2022
Total Acidity expressed as acetic acid	2,74	± 0,05		g/100 ml	0,10		* ACIDITA 2014 Rev.0 - Titrimetrico	07/09/2022 / 12/09/2022
Relative density at 20°C	1,186	± 0,001					07(S117) 2015 Rev.1 - Gravimetric	13/09/2022 / 13/09/2022
Alcoholic strength	< LQ			% Vol	0,07		DM 12/03/1986 SO GU n 161 14/07/1986 Met V pag 95, OIV-MA-AS312-01 Met B R2021	07/09/2022 / 09/09/2022
Brookfield rotational viscosity	Brookfield Rotational Viscosity (T = 20 ° C, impeller R3)							
	50 rpm: 1383 cP (mPa.s) 30 rpm: 1755 cP (mPa.s) 20 rpm: 2108 cP (mPa.s)							
pH	2,92	± 0,04		unità	0,50		07(S77) 2012 Rev.1 - Potentiometric	07/09/2022 / 12/09/2022
<b>SUGARS HPAEC-PAD</b>								
Galactose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	07/09/2022 / 13/09/2022
Glucose, anhydrous (HPLC)	17,2	± 1,2		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	07/09/2022 / 13/09/2022
Sucrose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	07/09/2022 / 13/09/2022
Fructose, anhydrous (HPLC)	16,9	± 1,2		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	07/09/2022 / 13/09/2022
Lactose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	07/09/2022 / 13/09/2022
Maltose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	07/09/2022 / 13/09/2022
Citric acid (LCMS) [331]	83,6	± 13,7		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	07/09/2022 / 09/09/2022
Lactic Acid (LCMS) [331]	263	± 36		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	07/09/2022 / 09/09/2022
Malic acid (LCMS) [331]	572	± 70		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	07/09/2022 / 09/09/2022
Succinic acid (LCMS) [331]	207	± 30		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	07/09/2022 / 09/09/2022
Tartaric acid (LCMS) [331]	595	± 73		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	07/09/2022 / 09/09/2022
Ochratoxin A	< LQ			µg/kg	0,10		03(S130) 2022 Rev.13 - LC-MS/MS	07/09/2022 / 09/09/2022

**END TEST REPORT**

The original document is a PDF file with Digital Signature: 22M02961-In-0-S20B-2000 - T20 - UNIMORE-DigitalSignature.pdf

**Next page...**

MODENA, li 13/09/2022

Sample arrived on the 05/09/2022

Registration date 05/09/2022

## TEST REPORT nr. 22M02961-In-0

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**

**SAMPLE 22M02961**

**MATRIX: Fruit and vegetable concentrates, and by-products**

### Notes and method reference:

< LQ: = lower than Quantification Limit.

U: the reported uncertainty is the expanded uncertainty calculated using a coverage factor equal to 2 which gives a reliability of approximately 95%. Please note that results expressed as '<LQ' may not indicate the absence of the searched parameters in the sample.

MICROBIOLOGICAL TESTS: for food and environmental samples, the extended measurement uncertainty was estimated according to ISO 19036:2019 Standard and is based on a standard uncertainty multiplied by a coverage factor of  $K = 2$ , providing a confidence level of approximately 95%. The combined standard uncertainty was assumed to be equal to the standard deviation of intra-laboratory reproducibility. The results of the microbiological tests are calculated according to the ISO 7218: 2007 / Amd 1: 2013 Standard. If the results are reported as <4 (CFU/ml) or <40 (CFU/g), this means that the microorganisms are present in the sample but in amounts less than 4 CFU/ml or 40 CFU/g respectively. For microbiological analyses unless differently reported in the individual test methods, in case of analytical steps foreseen in non-activity days of the laboratory, provisions of the ISO 7218: 2007 / Amd.1 2013 Standard (points 11.2 and 10.2.5) or from specific test methods are applied. In the case of quantitative microbiological tests, these have been set up on a single plate according to ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise expressly requested by current regulations.

In the case of quantitative microbiological tests, these have been set up on a single plate in accordance with ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise explicitly required by current regulations.

For waters, the measurement uncertainty corresponds to the confidence interval calculated according to ISO 8199: 2018 or to the expanded measurement uncertainty estimated according to ISO 29201: 2012. The results are issued in accordance with ISO 8199: 2018. When the number of colonies detected is <3, the result is expressed as "Microorganisms present in the analyzed volume (N ° colonies detected <3 CFU - reference ISO 8199: 2018, paragraph 9.1.8.4.1)".

LQ: Quantification Limit. It is the lowest analyte concentration which can be detected at an acceptable precision (repeatability) and accuracy, under well defined conditions.

LD: Detection Limit. It is the lowest analyte concentration which can be detected but not necessarily quantified, under well defined conditions.

Conformity evaluation: values not complying with laws, decrees, national and EU regulations or specifications supplied by the customer are evaluated case by case, also taking into consideration the uncertainty of measure for each single test and the regulations on rounding-off of values, and pointed out when considered as non conform.

Rec %: Recovery % "+" means that the recovery has been applied to the result. The numeric results between brackets (..) after the expression <LQ are purely indicative of traces that cannot be exactly quantified.

In the case of sampling carried out by Neotron, the laboratory applies the Internal Operating Procedure code: NEOT-DIR/ 006/53.

The laboratory disclaims any responsibility for the information provided by the client reported in this Report which may influence the validity of the results.

Methods marked with an asterisk (\*) are not accredited by ACCREDIA (UNI CEI EN ISO/IEC 17025). The sampling activity is not included within the Scope of Accreditation of Neotron SPA

### NOTES OF PARAMETERS:

[331]: Extended uncertainty calculated following the equation of HORWITZ by using a coverage factor of 2, which guarantee a reliance level of 95%

TEST REPORT VALID FOR ALL LEGAL PURPOSES (Italian R.D. 1-3-1928 n°842 (article 16), - Italian Law 19-7-1957 n°679 articles 16 and 18, Italian Ministerial Decree 25-3-1986).

DATA and SAMPLE STORAGE: Test Reports, Raw data, chromatographic paths and instrumental reports are stored for 5 years. One control sample is stored for 2 months.

Data expressed in this test report refer only to the sample tested in the laboratory. The results reported in this Test Report refer to the sample as received. The description or any other reference concerning the sample are declared by the customer. This Test Report cannot be reproduced except in full. Partial reproductions must be authorized in writing by our laboratory.

THE LABORATORY DIRECTOR: DR. ANDREA RIZZO

THE CHEMIST AUTHORIZED TO SIGN THE TEST REPORTS: DR. MARCO MESCHIARI

Approved by Analysis Manager - laboratory LMAA-Nut  
Approved by Analysis Manager - laboratory ICS-HPLC  
Approved by Analysis Manager - laboratory LC-MICO  
Approved by Analysis Manager - laboratory LC-AN

Approved by Analysis Manager - laboratory FISICHE  
Approved by Analysis Manager - laboratory LBM-EL  
Approved by Analysis Manager - laboratory LMAA-Bro

MODENA, li 09/09/2022

Sample arrived on the 25/08/2022  
Registration date 25/08/2022**TEST REPORT nr. 22L15878-In-0**

CUSTOMER

**ACETFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO****SAMPLE 22L15878****MATRIX: Fruit and vegetable concentrates, and  
by-products**

Description provided by Customer: GLASSA CON ABM IGP - CONTAMINAZIONE BASSA - VISCOSITA': ~ 2.000 CPS (T=10G) - CODICE: S20B-2000

Richiesta via Internet n° N0306/22 - 23/08/2022 16:52:51. - Campionamento eseguito da: Committente - Trasporto effettuato da: Committente  
Sample Condition on Receipt: 4°C

ANALYSIS DESCRIPTION	RESULT	U	REC. %	UNIT OF MEASURE	LQ	LD	METHOD	ANALYSES BEGINNING DATE / ENDING DATE
Water activity (Aw) at 25 °C	0,931	± 0,005			0,150		06(S135) rev6 2021	26/08/2022 / 26/08/2022
<b>VINEGAR ANALYSIS</b>								
Reduction sugars (Reducing substances)	442,4	± 13,3		g/l	1,0		07(S240) 2021 Rev.0 - Titrimetrico	29/08/2022 / 01/09/2022
Total Acidity expressed as acetic acid	2,84	± 0,06		g/100 ml	0,10		* ACIDITA 2014 Rev.0 - Titrimetrico	29/08/2022 / 31/08/2022
Relative density at 20°C	1,186	± 0,00027					07(S117) 2015 Rev.1 - Gravimetric	02/09/2022 / 02/09/2022
Alcoholic strength	0,11	± 0,01		% Vol	0,07		DM 12/03/1986 SO GU n 161 14/07/1986 Met V pag 95, OIV-MA-AS312-01 Met B R2021	29/08/2022 / 01/09/2022
Brookfield rotational viscosity	Brookfield Rotational Viscosity (T = 20 ° C, impeller R3)							
	30 rpm: 2012 cP (mPa.s)							
	20 rpm: 2466 cP (mPa.s)							
	12 rpm: 3191 cP (mPa.s)							
pH	2,94	± 0,04		unità	0,50		07(S77) 2012 Rev.1 - Potentiometric	29/08/2022 / 29/08/2022
<b>SUGARS HPAEC-PAD</b>								
Galactose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	29/08/2022 / 02/09/2022
Glucose, anhydrous (HPLC)	17,4	± 1,2		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	29/08/2022 / 02/09/2022
Sucrose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	29/08/2022 / 02/09/2022
Fructose, anhydrous (HPLC)	16,7	± 1,2		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	29/08/2022 / 02/09/2022
Lactose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	29/08/2022 / 02/09/2022
Maltose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	29/08/2022 / 02/09/2022
Citric acid (LCMS) [331]	111	± 18		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	29/08/2022 / 09/09/2022
Lactic Acid (LCMS) [331]	202	± 29		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	29/08/2022 / 09/09/2022
Malic acid (LCMS) [331]	577	± 71		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	29/08/2022 / 09/09/2022
Succinic acid (LCMS) [331]	192	± 28		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	29/08/2022 / 09/09/2022
Tartaric acid (LCMS) [331]	634	± 77		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	29/08/2022 / 09/09/2022
Ochratoxin A	< LQ			µg/kg	0,10		03(S130) 2022 Rev.13 - LC-MS/MS	29/08/2022 / 31/08/2022

**END TEST REPORT**

The original document is a PDF file with Digital Signature: 22L15878-In-0-S20B-2000 - T10 - UNIMORE-DigitalSignature.pdf

**Next page...**

MODENA, li 09/09/2022

Sample arrived on the 25/08/2022

Registration date 25/08/2022

## TEST REPORT nr. 22L15878-In-0

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**

**SAMPLE 22L15878**

**MATRIX: Fruit and vegetable concentrates, and by-products**

### Notes and method reference:

< LQ: = lower than Quantification Limit.

U: the reported uncertainty is the expanded uncertainty calculated using a coverage factor equal to 2 which gives a reliability of approximately 95%. Please note that results expressed as '<LQ' may not indicate the absence of the searched parameters in the sample.

MICROBIOLOGICAL TESTS: for food and environmental samples, the extended measurement uncertainty was estimated according to ISO 19036:2019 Standard and is based on a standard uncertainty multiplied by a coverage factor of K = 2, providing a confidence level of approximately 95%. The combined standard uncertainty was assumed to be equal to the standard deviation of intra-laboratory reproducibility. The results of the microbiological tests are calculated according to the ISO 7218: 2007 / Amd 1: 2013 Standard. If the results are reported as <4 (CFU/ml) or <40 (CFU/g), this means that the microorganisms are present in the sample but in amounts less than 4 CFU/ml or 40 CFU/g respectively. For microbiological analyses unless differently reported in the individual test methods, in case of analytical steps foreseen in non-activity days of the laboratory, provisions of the ISO 7218: 2007 / Amd.1 2013 Standard (points 11.2 and 10.2.5) or from specific test methods are applied. In the case of quantitative microbiological tests, these have been set up on a single plate according to ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise expressly requested by current regulations.

In the case of quantitative microbiological tests, these have been set up on a single plate in accordance with ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise explicitly required by current regulations.

For waters, the measurement uncertainty corresponds to the confidence interval calculated according to ISO 8199: 2018 or to the expanded measurement uncertainty estimated according to ISO 29201: 2012. The results are issued in accordance with ISO 8199: 2018. When the number of colonies detected is <3, the result is expressed as "Microorganisms present in the analyzed volume (N ° colonies detected <3 CFU - reference ISO 8199: 2018, paragraph 9.1.8.4.1)".

LQ: Quantification Limit. It is the lowest analyte concentration which can be detected at an acceptable precision (repeatability) and accuracy, under well defined conditions.

LD: Detection Limit. It is the lowest analyte concentration which can be detected but not necessarily quantified, under well defined conditions.

Conformity evaluation: values not complying with laws, decrees, national and EU regulations or specifications supplied by the customer are evaluated case by case, also taking into consideration the uncertainty of measure for each single test and the regulations on rounding-off of values, and pointed out when considered as non conform.

Rec %: Recovery % "+" means that the recovery has been applied to the result. The numeric results between brackets (..) after the expression <LQ are purely indicative of traces that cannot be exactly quantified.

In the case of sampling carried out by Neotron, the laboratory applies the Internal Operating Procedure code: NEOT-DIR/ 006/53.

The laboratory disclaims any responsibility for the information provided by the client reported in this Report which may influence the validity of the results.

Methods marked with an asterisk (\*) are not accredited by ACCREDIA (UNI CEI EN ISO/IEC 17025). The sampling activity is not included within the Scope of Accreditation of Neotron SPA

### NOTES OF PARAMETERS:

[331]: Extended uncertainty calculated following the equation of HORWITZ by using a coverage factor of 2, which guarantee a reliance level of 95%

TEST REPORT VALID FOR ALL LEGAL PURPOSES (Italian R.D. 1-3-1928 n°842 (article 16), - Italian Law 19-7-1957 n°679 articles 16 and 18, Italian Ministerial Decree 25-3-1986).

DATA and SAMPLE STORAGE: Test Reports, Raw data, chromatographic paths and instrumental reports are stored for 5 years. One control sample is stored for 2 months.

Data expressed in this test report refer only to the sample tested in the laboratory. The results reported in this Test Report refer to the sample as received. The description or any other reference concerning the sample are declared by the customer. This Test Report cannot be reproduced except in full. Partial reproductions must be authorized in writing by our laboratory.

THE LABORATORY DIRECTOR: DR. ANDREA RIZZO

THE CHEMIST AUTHORIZED TO SIGN THE TEST REPORTS: DR. BARBARA MALAGOLI

Approved by Analysis Manager - laboratory LMAA-Nut  
Approved by Analysis Manager - laboratory ICS-HPLC  
Approved by Analysis Manager - laboratory LC-MICO  
Approved by Analysis Manager - laboratory LC-AN

Approved by Analysis Manager - laboratory FISICHE  
Approved by Analysis Manager - laboratory LBM-EL  
Approved by Analysis Manager - laboratory LMAA-Bro

NEOTRON SpA - With Sole Shareholder

Stradello Aggazzotti, 104

41126 MODENA - ITALY - Fiscal Code and VAT n° 03807840362

Tel: +39 059461711 - Fax: +39 059461777

www.neotron.it - neotron@neotron.it

Laboratorio Qualificato D.M. 26-2-87 Art. 4 - Legge 46/82 per la Ricerca Applicata e Innovazione Tecnologica.

Regione Emilia Romagna - AUTORIZZAZIONE Autocontrollo N° 008/MO/008

BNN-Monitoring Fruit and Vegetables Approved Laboratory

I-Monitoring EDEKAAG Fruit and Vegetables Registered Laboratory

GMP+ code: GMP051757

MODENA, li 09/09/2022

Sample arrived on the 25/08/2022  
Registration date 25/08/2022**TEST REPORT nr. 22L15877-In-0**

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO****SAMPLE 22L15877****MATRIX: Fruit and vegetable concentrates, and  
by-products**Description provided by Customer: GLASSA CON ABM IGP - CONTAMINAZIONE BASSA - VISCOSITA': ~ 2.000 CPS (T=0G) -  
CODICE: S20B-2000Richiesta via Internet n° N0303/22 - 23/08/2022 16:52:11. - Campionamento eseguito da: Committente - Trasporto effettuato da:  
Committente  
Sample Condition on Receipt: 4°C

ANALYSIS DESCRIPTION	RESULT	U	REC. %	UNIT OF MEASURE	LQ	LD	METHOD	ANALYSES BEGINNING DATE / ENDING DATE
Water activity (Aw) at 25 °C	0,930	± 0,005			0,150		06(S135) rev6 2021	26/08/2022 / 26/08/2022
<b>VINEGAR ANALYSIS</b>								
Reduction sugars (Reducing substances)	433,9	± 13,0		g/l	1,0		07(S240) 2021 Rev.0 - Titrimetrico	29/08/2022 / 01/09/2022
Total Acidity expressed as acetic acid	2,82	± 0,05		g/100 ml	0,10		* ACIDITA 2014 Rev.0 - Titrimetrico	29/08/2022 / 31/08/2022
Relative density at 20°C	1,186	± 0,0003					07(S117) 2015 Rev.1 - Gravimetric	02/09/2022 / 02/09/2022
Alcoholic strength	0,10	± 0,01		% Vol	0,07		DM 12/03/1986 SO GU n 161 14/07/1986 Met V pag 95, OIV- MA-AS312-01 Met B R2021	29/08/2022 / 01/09/2022
Brookfield rotational viscosity	Brookfield Rotational Viscosity (T = 20 ° C, impeller R3)							
	50 rpm: 1351 cP (mPa.s) 30 rpm: 1691 cP (mPa.s) 20 rpm: 2041 cP (mPa.s)							
pH	2,97	± 0,04		unità	0,50		07(S77) 2012 Rev.1 - Potentiometric	29/08/2022 / 29/08/2022
<b>SUGARS HPAEC-PAD</b>								
Galactose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	29/08/2022 / 02/09/2022
Glucose, anhydrous (HPLC)	17,6	± 1,2		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	29/08/2022 / 02/09/2022
Sucrose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	29/08/2022 / 02/09/2022
Fructose, anhydrous (HPLC)	16,2	± 1,1		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	29/08/2022 / 02/09/2022
Lactose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	29/08/2022 / 02/09/2022
Maltose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	29/08/2022 / 02/09/2022
Citric acid (LCMS) [331]	106	± 17		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	29/08/2022 / 09/09/2022
Lactic Acid (LCMS) [331]	216	± 31		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	29/08/2022 / 09/09/2022
Malic acid (LCMS) [331]	612	± 75		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	29/08/2022 / 09/09/2022
Succinic acid (LCMS) [331]	205	± 30		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	29/08/2022 / 09/09/2022
Tartaric acid (LCMS) [331]	634	± 77		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	29/08/2022 / 09/09/2022
Ochratoxin A	< LQ			µg/kg	0,10		03(S130) 2022 Rev.13 - LC- MS/MS	29/08/2022 / 31/08/2022

**END TEST REPORT**

The original document is a PDF file with Digital Signature: 22L15877-In-0-S20B-2000 - T0 - UNIMORE-DigitalSignature.pdf

**Next page...**

MODENA, li 09/09/2022

Sample arrived on the 25/08/2022

Registration date 25/08/2022

## TEST REPORT nr. 22L15877-In-0

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**

**SAMPLE 22L15877**

**MATRIX: Fruit and vegetable concentrates, and by-products**

### Notes and method reference:

< LQ: = lower than Quantification Limit.

U: the reported uncertainty is the expanded uncertainty calculated using a coverage factor equal to 2 which gives a reliability of approximately 95%. Please note that results expressed as '<LQ' may not indicate the absence of the searched parameters in the sample.

MICROBIOLOGICAL TESTS: for food and environmental samples, the extended measurement uncertainty was estimated according to ISO 19036:2019 Standard and is based on a standard uncertainty multiplied by a coverage factor of K = 2, providing a confidence level of approximately 95%. The combined standard uncertainty was assumed to be equal to the standard deviation of intra-laboratory reproducibility. The results of the microbiological tests are calculated according to the ISO 7218: 2007 / Amd 1: 2013 Standard. If the results are reported as <4 (CFU/ml) or <40 (CFU/g), this means that the microorganisms are present in the sample but in amounts less than 4 CFU/ml or 40 CFU/g respectively. For microbiological analyses unless differently reported in the individual test methods, in case of analytical steps foreseen in non-activity days of the laboratory, provisions of the ISO 7218: 2007 / Amd.1 2013 Standard (points 11.2 and 10.2.5) or from specific test methods are applied. In the case of quantitative microbiological tests, these have been set up on a single plate according to ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise expressly requested by current regulations.

In the case of quantitative microbiological tests, these have been set up on a single plate in accordance with ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise explicitly required by current regulations.

For waters, the measurement uncertainty corresponds to the confidence interval calculated according to ISO 8199: 2018 or to the expanded measurement uncertainty estimated according to ISO 29201: 2012. The results are issued in accordance with ISO 8199: 2018. When the number of colonies detected is <3, the result is expressed as "Microorganisms present in the analyzed volume (N ° colonies detected <3 CFU - reference ISO 8199: 2018, paragraph 9.1.8.4.1)".

LQ: Quantification Limit. It is the lowest analyte concentration which can be detected at an acceptable precision (repeatability) and accuracy, under well defined conditions.

LD: Detection Limit. It is the lowest analyte concentration which can be detected but not necessarily quantified, under well defined conditions.

Conformity evaluation: values not complying with laws, decrees, national and EU regulations or specifications supplied by the customer are evaluated case by case, also taking into consideration the uncertainty of measure for each single test and the regulations on rounding-off of values, and pointed out when considered as non conform.

Rec %: Recovery % "+" means that the recovery has been applied to the result. The numeric results between brackets (..) after the expression <LQ are purely indicative of traces that cannot be exactly quantified.

In the case of sampling carried out by Neotron, the laboratory applies the Internal Operating Procedure code: NEOT-DIR/ 006/53.

The laboratory disclaims any responsibility for the information provided by the client reported in this Report which may influence the validity of the results.

Methods marked with an asterisk (\*) are not accredited by ACCREDIA (UNI CEI EN ISO/IEC 17025). The sampling activity is not included within the Scope of Accreditation of Neotron SPA

### NOTES OF PARAMETERS:

[331]: Extended uncertainty calculated following the equation of HORWITZ by using a coverage factor of 2, which guarantee a reliance level of 95%

TEST REPORT VALID FOR ALL LEGAL PURPOSES (Italian R.D. 1-3-1928 n°842 (article 16), – Italian Law 19-7-1957 n°679 articles 16 and 18, Italian Ministerial Decree 25-3-1986).

DATA and SAMPLE STORAGE: Test Reports, Raw data, chromatographic paths and instrumental reports are stored for 5 years. One control sample is stored for 2 months.

Data expressed in this test report refer only to the sample tested in the laboratory. The results reported in this Test Report refer to the sample as received. The description or any other reference concerning the sample are declared by the customer. This Test Report cannot be reproduced except in full. Partial reproductions must be authorized in writing by our laboratory.

THE LABORATORY DIRECTOR: DR. ANDREA RIZZO

THE CHEMIST AUTHORIZED TO SIGN THE TEST REPORTS: DR. BARBARA MALAGOLI

Approved by Analysis Manager - laboratory LMAA-Nut  
Approved by Analysis Manager - laboratory ICS-HPLC  
Approved by Analysis Manager - laboratory LC-MICO  
Approved by Analysis Manager - laboratory LC-AN

Approved by Analysis Manager - laboratory FISICHE  
Approved by Analysis Manager - laboratory LBM-EL  
Approved by Analysis Manager - laboratory LMAA-Bro

MODENA, li 23/08/2022

Sample arrived on the 05/08/2022

Registration date 08/08/2022

**TEST REPORT nr. 22L05144-In-0**

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**

SAMPLE 22L05144

**MATRIX: Fruit and vegetable concentrates, and  
by-products**

Description provided by Customer: GLASSA CON ABM IGP - CONTAMINAZIONE BASSA - VISCOSITA': ~ 1.000 CPS (T=20G) - CODICE: S20B-1000

Richiesta via Internet n° N0257/22 - 20/07/2022 12:56:04. - Campionamento eseguito da: Committente - Trasporto effettuato da: Corriere  
Sample Condition on Receipt: 20°C

ANALYSIS DESCRIPTION	RESULT	U	REC. %	UNIT OF MEASURE	LQ	LD	METHOD	ANALYSES BEGINNING DATE / ENDING DATE
Water activity (Aw) at 25 °C	0,925	± 0,005			0,150		06(S135) rev6 2021	08/08/2022 / 08/08/2022
<b>VINEGAR ANALYSIS</b>								
Reduction sugars (Reducing substances)	445,9	± 13,4		g/l	1,0		07(S240) 2021 Rev.0 - Titrimetrico	09/08/2022 / 11/08/2022
Total Acidity expressed as acetic acid	2,78	± 0,05		g/100 ml	0,10		* ACIDITA 2014 Rev.0 - Titrimetrico	09/08/2022 / 11/08/2022
Relative density at 20°C	1,18711	± 0,00027					DM 12/03/1986 SO GU n 161 14/07/1986 Met VI pag 95 p.to 4, OIV-MA-AS2-01A Met B R2021 - densimetry	09/08/2022 / 11/08/2022
Alcoholic strength	< LQ			% Vol	0,07		DM 12/03/1986 SO GU n 161 14/07/1986 Met V pag 95, OIV-MA-AS312-01 Met B R2021	09/08/2022 / 11/08/2022
pH	2,91	± 0,04		unità	0,50		07(S77) 2012 Rev.1 - Potentiometric	09/08/2022 / 10/08/2022
Brookfield rotational viscosity	Brookfield Rotational Viscosity (T = 20 ° C, impeller R2)						* VISCOSITA 2018 Rev.1	09/08/2022 / 12/08/2022
	30 rpm: 786 cP (mPa.s) 20 rpm: 908 cP (mPa.s) 12 rpm: 1091 cP (mPa.s)							
<b>SUGARS HPAEC-PAD</b>								
Galactose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	09/08/2022 / 16/08/2022
Glucose, anhydrous (HPLC)	18,4	± 1,3		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	09/08/2022 / 16/08/2022
Sucrose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	09/08/2022 / 16/08/2022
Fructose, anhydrous (HPLC)	19,0	± 1,3		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	09/08/2022 / 16/08/2022
Lactose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	09/08/2022 / 16/08/2022
Maltose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	09/08/2022 / 16/08/2022
Citric acid (LCMS) [331]	65,5	± 11,2		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	09/08/2022 / 16/08/2022
Lactic Acid (LCMS) [331]	231	± 33		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	09/08/2022 / 16/08/2022
Malic acid (LCMS) [331]	436	± 56		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	09/08/2022 / 16/08/2022
Succinic acid (LCMS) [331]	151	± 23		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	09/08/2022 / 16/08/2022
Tartaric acid (LCMS) [331]	456	± 58		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	09/08/2022 / 16/08/2022
Ochratoxin A	< LQ			µg/kg	0,10		03(S130) 2022 Rev.12 - LC-MS/MS	09/08/2022 / 11/08/2022

**END TEST REPORT**

The original document is a PDF file with Digital Signature: 22L05144-In-0-S20B-1000 - T20 - UNIMORE-DigitalSignature.pdf

NEOTRON SpA - With Sole Shareholder

Stradello Aggazzotti, 104

41126 MODENA - ITALY - Fiscal Code and VAT n° 03807840362

Tel: +39 059461711 - Fax: +39 059461777

www.neotron.it - neotron@neotron.it

Laboratorio Qualificato D.M. 26-2-87 Art. 4 - Legge 46/82 per la Ricerca Applicata e Innovazione Tecnologica.

Regione Emilia Romagna - AUTORIZZAZIONE Autocontrollo N° 008/MO/008

BNN-Monitoring Fruit and Vegetables Approved Laboratory

I-Monitoring EDEKAAG Fruit and Vegetables Registered Laboratory

GMP+ code: GMP051757

Next page...

MODENA, li 23/08/2022

Sample arrived on the 05/08/2022

Registration date 08/08/2022

**TEST REPORT nr. 22L05144-In-0**

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**

SAMPLE 22L05144

**MATRIX: Fruit and vegetable concentrates, and  
by-products**

## Notes and method reference:

&lt; LQ: = lower than Quantification Limit.

U: the reported uncertainty is the expanded uncertainty calculated using a coverage factor equal to 2 which gives a reliability of approximately 95%. Please note that results expressed as '<LQ' may not indicate the absence of the searched parameters in the sample.

MICROBIOLOGICAL TESTS: for food and environmental samples, the extended measurement uncertainty was estimated according to ISO 19036:2019 Standard and is based on a standard uncertainty multiplied by a coverage factor of K = 2, providing a confidence level of approximately 95%. The combined standard uncertainty was assumed to be equal to the standard deviation of intra-laboratory reproducibility. The results of the microbiological tests are calculated according to the ISO 7218: 2007 / Amd 1: 2013 Standard. If the results are reported as <4 (CFU/ml) or <40 (CFU/g), this means that the microorganisms are present in the sample but in amounts less than 4 CFU/ml or 40 CFU/g respectively. For microbiological analyses unless differently reported in the individual test methods, in case of analytical steps foreseen in non-activity days of the laboratory, provisions of the ISO 7218: 2007 / Amd.1 2013 Standard (points 11.2 and 10.2.5) or from specific test methods are applied. In the case of quantitative microbiological tests, these have been set up on a single plate according to ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise expressly requested by current regulations.

In the case of quantitative microbiological tests, these have been set up on a single plate in accordance with ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise explicitly required by current regulations.

For waters, the measurement uncertainty corresponds to the confidence interval calculated according to ISO 8199: 2018 or to the expanded measurement uncertainty estimated according to ISO 29201: 2012. The results are issued in accordance with ISO 8199: 2018. When the number of colonies detected is <3, the result is expressed as "Microorganisms present in the analyzed volume (N ° colonies detected <3 CFU - reference ISO 8199: 2018, paragraph 9.1.8.4.1)".

LQ: Quantification Limit. It is the lowest analyte concentration which can be detected at an acceptable precision (repeatability) and accuracy, under well defined conditions.

LD: Detection Limit. It is the lowest analyte concentration which can be detected but not necessarily quantified, under well defined conditions.

Conformity evaluation: values not complying with laws, decrees, national and EU regulations or specifications supplied by the customer are evaluated case by case, also taking into consideration the uncertainty of measure for each single test and the regulations on rounding-off of values, and pointed out when considered as non conform.

Rec %: Recovery % "+" means that the recovery has been applied to the result. The numeric results between brackets (..) after the expression <LQ are purely indicative of traces that cannot be exactly quantified.

In the case of sampling carried out by Neotron, the laboratory applies the Internal Operating Procedure code: NEOT-DIR/ 006/53.

The laboratory disclaims any responsibility for the information provided by the client reported in this Report which may influence the validity of the results.

Methods marked with an asterisk (\*) are not accredited by ACCREDIA (UNI CEI EN ISO/IEC 17025). The sampling activity is not included within the Scope of Accreditation of Neotron SPA

## NOTES OF PARAMETERS:

[331]: Extended uncertainty calculated following the equation of HORWITZ by using a coverage factor of 2, which guarantee a reliance level of 95%

TEST REPORT VALID FOR ALL LEGAL PURPOSES (Italian R.D. 1-3-1928 n°842 (article 16), - Italian Law 19-7-1957 n°679 articles 16 and 18, Italian Ministerial Decree 25-3-1986).

DATA and SAMPLE STORAGE: Test Reports, Raw data, chromatographic paths and instrumental reports are stored for 5 years. One control sample is stored for 2 months.

Data expressed in this test report refer only to the sample tested in the laboratory. The results reported in this Test Report refer to the sample as received. The description or any other reference concerning the sample are declared by the customer. This Test Report cannot be reproduced except in full. Partial reproductions must be authorized in writing by our laboratory.

THE LABORATORY DIRECTOR: DR. ANDREA RIZZO

THE CHEMIST AUTHORIZED TO SIGN THE TEST REPORTS: DR. MARCO MESCHIARI - N. 514 ORDINE DEI CHIMICI MODENA

Approved by Analysis Manager - laboratory LMAA-Nut

Approved by Analysis Manager - laboratory ICS-HPLC

Approved by Analysis Manager - laboratory LC-MICO

Approved by Analysis Manager - laboratory LC-AN

Approved by Analysis Manager - laboratory FISICHE

Approved by Analysis Manager - laboratory LBM-EL

Approved by Analysis Manager - laboratory LMAA-Bro

NEOTRON SpA - With Sole Shareholder

Stradello Aggazzotti, 104

41126 MODENA - ITALY - Fiscal Code and VAT n° 03807840362

Tel: +39 059461711 - Fax: +39 059461777

www.neotron.it - neotron@neotron.it

Laboratorio Qualificato D.M. 26-2-87 Art. 4 - Legge 46/82 per la Ricerca Applicata e Innovazione Tecnologica.

Regione Emilia Romagna - AUTORIZZAZIONE Autocontrollo N° 008/MO/008

BNN-Monitoring Fruit and Vegetables Approved Laboratory

I-Monitoring EDEKAAG Fruit and Vegetables Registered Laboratory

GMP+ code: GMP051757



MODENA, li 04/08/2022

Sample arrived on the 26/07/2022  
Registration date 26/07/2022**TEST REPORT nr. 22G17282-In-0**

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO****SAMPLE 22G17282****MATRIX: Fruit and vegetable concentrates, and by-products**

Description provided by Customer: GLASSA CON ABM IGP - CONTAMINAZIONE BASSA - VISCOSITA': ~ 1.000 CPS (T=10G) - CODICE: S20B-1000

Richiesta via Internet n° N0256/22 - 20/07/2022 12:56:03. - Campionamento eseguito da: Committente - Trasporto effettuato da: Committente  
Sample Condition on Receipt: 20°C

ANALYSIS DESCRIPTION	RESULT	U	REC. %	UNIT OF MEASURE	LQ	LD	METHOD	ANALYSES BEGINNING DATE / ENDING DATE
Water activity (Aw) at 25 °C	0,940	± 0,005			0,150		06(S135) rev6 2021	27/07/2022 / 29/07/2022
<b>VINEGAR ANALYSIS</b>								
Reduction sugars (Reducing substances)	449,9	± 13,5		g/l	1,0		07(S240) 2021 Rev.0 - Titrimetrico	28/07/2022 / 02/08/2022
Total Acidity expressed as acetic acid	2,92	± 0,06		g/100 ml	0,10		* ACIDITA 2014 Rev.0 - Titrimetrico	28/07/2022 / 01/08/2022
Relative density at 20°C	1,19067	± 0,00027					DM 12/03/1986 SO GU n 161 14/07/1986 Met VI pag 95 p.to 4, OIV-MA-AS2-01A Met B R2021 - densimetry	28/07/2022 / 01/08/2022
Alcoholic strength	< LQ			% Vol	0,07		DM 12/03/1986 SO GU n 161 14/07/1986 Met V pag 95, OIV-MA-AS312-01 Met B R2021	28/07/2022 / 01/08/2022
pH	2,92	± 0,04		unità	0,50		07(S77) 2012 Rev.1 - Potentiometric	28/07/2022 / 01/08/2022
Brookfield rotational viscosity	Brookfield Rotational Viscosity (T = 20 ° C, impeller R2) 30 rpm: 709 cP (mPa.s) 20 rpm: 806 cP (mPa.s) 12 rpm: 1027 cP (mPa.s)						* VISCOSITA 2018 Rev.1	28/07/2022 / 04/08/2022
<b>SUGARS HPAEC-PAD</b>								
Galactose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	28/07/2022 / 02/08/2022
Glucose, anhydrous (HPLC)	17,5	± 1,4		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	28/07/2022 / 02/08/2022
Sucrose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	28/07/2022 / 02/08/2022
Fructose, anhydrous (HPLC)	20,9	± 1,7		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	28/07/2022 / 02/08/2022
Lactose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	28/07/2022 / 02/08/2022
Maltose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	28/07/2022 / 02/08/2022
Citric acid (LCMS) [331]	92,0	± 14,9		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	28/07/2022 / 04/08/2022
Lactic Acid (LCMS) [331]	199	± 29		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	28/07/2022 / 04/08/2022
Malic acid (LCMS) [331]	465	± 59		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	28/07/2022 / 04/08/2022
Succinic acid (LCMS) [331]	177	± 26		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	28/07/2022 / 04/08/2022
Tartaric acid (LCMS) [331]	502	± 63		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	28/07/2022 / 04/08/2022
Ochratoxin A	< LQ			µg/kg	0,10		03(S130) 2022 Rev.12 - LC-MS/MS	28/07/2022 / 02/08/2022

**END TEST REPORT**

The original document is a PDF file with Digital Signature: 22G17282-In-0-S20B-1000 - T10 - UNIMORE-DigitalSignature.pdf

MODENA, li 04/08/2022

Sample arrived on the 26/07/2022

Registration date 26/07/2022

**TEST REPORT nr. 22G17282-In-0**

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**

SAMPLE 22G17282

**MATRIX: Fruit and vegetable concentrates, and  
by-products**

## Notes and method reference:

&lt; LQ: = lower than Quantification Limit.

U: the reported uncertainty is the expanded uncertainty calculated using a coverage factor equal to 2 which gives a reliability of approximately 95%. Please note that results expressed as '<LQ' may not indicate the absence of the searched parameters in the sample.

MICROBIOLOGICAL TESTS: for food and environmental samples, the extended measurement uncertainty was estimated according to ISO 19036:2019 Standard and is based on a standard uncertainty multiplied by a coverage factor of K = 2, providing a confidence level of approximately 95%. The combined standard uncertainty was assumed to be equal to the standard deviation of intra-laboratory reproducibility. The results of the microbiological tests are calculated according to the ISO 7218: 2007 / Amd 1: 2013 Standard. If the results are reported as <4 (CFU/ml) or <40 (CFU/g), this means that the microorganisms are present in the sample but in amounts less than 4 CFU/ml or 40 CFU/g respectively. For microbiological analyses unless differently reported in the individual test methods, in case of analytical steps foreseen in non-activity days of the laboratory, provisions of the ISO 7218: 2007 / Amd.1 2013 Standard (points 11.2 and 10.2.5) or from specific test methods are applied. In the case of quantitative microbiological tests, these have been set up on a single plate according to ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise expressly requested by current regulations.

In the case of quantitative microbiological tests, these have been set up on a single plate in accordance with ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise explicitly required by current regulations.

For waters, the measurement uncertainty corresponds to the confidence interval calculated according to ISO 8199: 2018 or to the expanded measurement uncertainty estimated according to ISO 29201: 2012. The results are issued in accordance with ISO 8199: 2018. When the number of colonies detected is <3, the result is expressed as "Microorganisms present in the analyzed volume (N ° colonies detected <3 CFU - reference ISO 8199: 2018, paragraph 9.1.8.4.1)".

LQ: Quantification Limit. It is the lowest analyte concentration which can be detected at an acceptable precision (repeatability) and accuracy, under well defined conditions.

LD: Detection Limit. It is the lowest analyte concentration which can be detected but not necessarily quantified, under well defined conditions.

Conformity evaluation: values not complying with laws, decrees, national and EU regulations or specifications supplied by the customer are evaluated case by case, also taking into consideration the uncertainty of measure for each single test and the regulations on rounding-off of values, and pointed out when considered as non conform.

Rec %: Recovery % "+" means that the recovery has been applied to the result. The numeric results between brackets (..) after the expression <LQ are purely indicative of traces that cannot be exactly quantified.

In the case of sampling carried out by Neotron, the laboratory applies the Internal Operating Procedure code: NEOT-DIR/ 006/53.

The laboratory disclaims any responsibility for the information provided by the client reported in this Report which may influence the validity of the results.

Methods marked with an asterisk (\*) are not accredited by ACCREDIA (UNI CEI EN ISO/IEC 17025). The sampling activity is not included within the Scope of Accreditation of Neotron SPA

## NOTES OF PARAMETERS:

[331]: Extended uncertainty calculated following the equation of HORWITZ by using a coverage factor of 2, which guarantee a reliance level of 95%

TEST REPORT VALID FOR ALL LEGAL PURPOSES (Italian R.D. 1-3-1928 n°842 (article 16), - Italian Law 19-7-1957 n°679 articles 16 and 18, Italian Ministerial Decree 25-3-1986).

DATA and SAMPLE STORAGE: Test Reports, Raw data, chromatographic paths and instrumental reports are stored for 5 years. One control sample is stored for 2 months.

Data expressed in this test report refer only to the sample tested in the laboratory. The results reported in this Test Report refer to the sample as received. The description or any other reference concerning the sample are declared by the customer. This Test Report cannot be reproduced except in full. Partial reproductions must be authorized in writing by our laboratory.

THE LABORATORY DIRECTOR: DR. ANDREA RIZZO

THE CHEMIST AUTHORIZED TO SIGN THE TEST REPORTS: DR. MARCO MESCHIARI - N. 514 ORDINE DEI CHIMICI MODENA

Approved by Analysis Manager - laboratory LMAA-Nut

Approved by Analysis Manager - laboratory ICS-HPLC

Approved by Analysis Manager - laboratory LC-MICO

Approved by Analysis Manager - laboratory LC-AN

Approved by Analysis Manager - laboratory FISICHE

Approved by Analysis Manager - laboratory LBM-EL

Approved by Analysis Manager - laboratory LMAA-Bro

NEOTRON SpA - With Sole Shareholder

Stradello Aggazzotti, 104

41126 MODENA - ITALY - Fiscal Code and VAT n° 03807840362

Tel: +39 059461711 - Fax: +39 059461777

www.neotron.it - neotron@neotron.it

Laboratorio Qualificato D.M. 26-2-87 Art. 4 - Legge 46/82 per la Ricerca Applicata e Innovazione Tecnologica.

Regione Emilia Romagna - AUTORIZZAZIONE Autocontrollo N° 008/MO/008

BNN-Monitoring Fruit and Vegetables Approved Laboratory

I-Monitoring EDEKAAG Fruit and Vegetables Registered Laboratory

GMP+ code: GMP051757

MODENA, li 29/07/2022

Sample arrived on the 19/07/2022  
Registration date 19/07/2022**TEST REPORT nr. 22G12685-In-0**

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO****SAMPLE 22G12685****MATRIX: Fruit and vegetable concentrates, and  
by-products**

Description provided by Customer: GLASSA CON ABM IGP - CONTAMINAZIONE BASSA - VISCOSITA': ~ 1.000 CPS (T=0G) - CODICE: S20B-1000

Richiesta via Internet n° N0217/22 - 04/07/2022 16:41:22. - Campionamento eseguito da: Committente - Trasporto effettuato da: Committente  
Sample Condition on Receipt: 4°C

ANALYSIS DESCRIPTION	RESULT	U	REC. %	UNIT OF MEASURE	LQ	LD	METHOD	ANALYSES BEGINNING DATE / ENDING DATE
Water activity (Aw) at 25 °C	0,919	± 0,005			0,150		06(S135) rev6 2021	20/07/2022 / 22/07/2022
<b>VINEGAR ANALYSIS</b>								
Reduction sugars (Reducing substances)	444,5	± 13,3		g/l	1,0		07(S240) 2021 Rev.0 - Titrimetrico	21/07/2022 / 26/07/2022
Total Acidity expressed as acetic acid	2,88	± 0,06		g/100 ml	0,10		* ACIDITA 2014 Rev.0 - Titrimetrico	21/07/2022 / 25/07/2022
Relative density at 20°C	1,189	± 0,0003					07(S117) 2015 Rev.1 - Gravimetric	29/07/2022 / 29/07/2022
Alcoholic strength	< LQ			% Vol	0,07		DM 12/03/1986 SO GU n 161 14/07/1986 Met V pag 95, OIV-MA-AS312-01 Met B R2021	21/07/2022 / 26/07/2022
Brookfield rotational viscosity	Brookfield Rotational Viscosity (T = 20 ° C, impeller R2)							
	50 rpm: 568 cP (mPa.s) 30 rpm: 700 cP (mPa.s) 20 rpm: 759 cP (mPa.s)							
pH	2,93	± 0,04		unità	0,50		07(S77) 2012 Rev.1 - Potentiometric	21/07/2022 / 22/07/2022
<b>SUGARS HPAEC-PAD</b>								
Galactose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	21/07/2022 / 25/07/2022
Glucose, anhydrous (HPLC)	19,0	± 1,3		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	21/07/2022 / 25/07/2022
Sucrose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	21/07/2022 / 25/07/2022
Fructose, anhydrous (HPLC)	18,2	± 1,3		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	21/07/2022 / 25/07/2022
Lactose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	21/07/2022 / 25/07/2022
Maltose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	21/07/2022 / 25/07/2022
Citric acid (LCMS) [331]	100	± 16		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	21/07/2022 / 26/07/2022
Lactic Acid (LCMS) [331]	148	± 22		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	21/07/2022 / 26/07/2022
Malic acid (LCMS) [331]	521	± 65		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	21/07/2022 / 26/07/2022
Succinic acid (LCMS) [331]	210	± 30		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	21/07/2022 / 26/07/2022
Tartaric acid (LCMS) [331]	500	± 63		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	21/07/2022 / 26/07/2022
Ochratoxin A	< LQ			µg/kg	0,10		03(S130) 2022 Rev.12 - LC-MS/MS	21/07/2022 / 26/07/2022

**END TEST REPORT**

The original document is a PDF file with Digital Signature: 22G12685-In-0-S20B-1000 - T0 - UNIMORE-DigitalSignature.pdf

**Next page...**

MODENA, li 29/07/2022

Sample arrived on the 19/07/2022

Registration date 19/07/2022

## TEST REPORT nr. 22G12685-In-0

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**

**SAMPLE 22G12685**

**MATRIX: Fruit and vegetable concentrates, and by-products**

### Notes and method reference:

< LQ: = lower than Quantification Limit.

U: the reported uncertainty is the expanded uncertainty calculated using a coverage factor equal to 2 which gives a reliability of approximately 95%. Please note that results expressed as '<LQ' may not indicate the absence of the searched parameters in the sample.

MICROBIOLOGICAL TESTS: for food and environmental samples, the extended measurement uncertainty was estimated according to ISO 19036:2019 Standard and is based on a standard uncertainty multiplied by a coverage factor of K = 2, providing a confidence level of approximately 95%. The combined standard uncertainty was assumed to be equal to the standard deviation of intra-laboratory reproducibility. The results of the microbiological tests are calculated according to the ISO 7218: 2007 / Amd 1: 2013 Standard. If the results are reported as <4 (CFU/ml) or <40 (CFU/g), this means that the microorganisms are present in the sample but in amounts less than 4 CFU/ml or 40 CFU/g respectively. For microbiological analyses unless differently reported in the individual test methods, in case of analytical steps foreseen in non-activity days of the laboratory, provisions of the ISO 7218: 2007 / Amd.1 2013 Standard (points 11.2 and 10.2.5) or from specific test methods are applied. In the case of quantitative microbiological tests, these have been set up on a single plate according to ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise expressly requested by current regulations.

In the case of quantitative microbiological tests, these have been set up on a single plate in accordance with ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise explicitly required by current regulations.

For waters, the measurement uncertainty corresponds to the confidence interval calculated according to ISO 8199: 2018 or to the expanded measurement uncertainty estimated according to ISO 29201: 2012. The results are issued in accordance with ISO 8199: 2018. When the number of colonies detected is <3, the result is expressed as "Microorganisms present in the analyzed volume (N ° colonies detected <3 CFU - reference ISO 8199: 2018, paragraph 9.1.8.4.1)".

LQ: Quantification Limit. It is the lowest analyte concentration which can be detected at an acceptable precision (repeatability) and accuracy, under well defined conditions.

LD: Detection Limit. It is the lowest analyte concentration which can be detected but not necessarily quantified, under well defined conditions.

Conformity evaluation: values not complying with laws, decrees, national and EU regulations or specifications supplied by the customer are evaluated case by case, also taking into consideration the uncertainty of measure for each single test and the regulations on rounding-off of values, and pointed out when considered as non conform.

Rec %: Recovery % "+" means that the recovery has been applied to the result. The numeric results between brackets (..) after the expression <LQ are purely indicative of traces that cannot be exactly quantified.

In the case of sampling carried out by Neotron, the laboratory applies the Internal Operating Procedure code: NEOT-DIR/ 006/53.

The laboratory disclaims any responsibility for the information provided by the client reported in this Report which may influence the validity of the results.

Methods marked with an asterisk (\*) are not accredited by ACCREDIA (UNI CEI EN ISO/IEC 17025). The sampling activity is not included within the Scope of Accreditation of Neotron SPA

### NOTES OF PARAMETERS:

[331]: Extended uncertainty calculated following the equation of HORWITZ by using a coverage factor of 2, which guarantee a reliance level of 95%

TEST REPORT VALID FOR ALL LEGAL PURPOSES (Italian R.D. 1-3-1928 n°842 (article 16), - Italian Law 19-7-1957 n°679 articles 16 and 18, Italian Ministerial Decree 25-3-1986).

DATA and SAMPLE STORAGE: Test Reports, Raw data, chromatographic paths and instrumental reports are stored for 5 years. One control sample is stored for 2 months.

Data expressed in this test report refer only to the sample tested in the laboratory. The results reported in this Test Report refer to the sample as received. The description or any other reference concerning the sample are declared by the customer. This Test Report cannot be reproduced except in full. Partial reproductions must be authorized in writing by our laboratory.

THE LABORATORY DIRECTOR: DR. ANDREA RIZZO

THE CHEMIST AUTHORIZED TO SIGN THE TEST REPORTS: DR. MARCO MESCHIARI - N. 514 ORDINE DEI CHIMICI MODENA

Approved by Analysis Manager - laboratory LMAA-Nut

Approved by Analysis Manager - laboratory ICS-HPLC

Approved by Analysis Manager - laboratory LC-MICO

Approved by Analysis Manager - laboratory LC-AN

Approved by Analysis Manager - laboratory FISICHE

Approved by Analysis Manager - laboratory LBM-EL

Approved by Analysis Manager - laboratory LMAA-Bro

NEOTRON SpA - With Sole Shareholder

Stradello Aggazzotti, 104

41126 MODENA - ITALY - Fiscal Code and VAT n° 03807840362

Tel: +39 059461711 - Fax: +39 059461777

www.neotron.it - neotron@neotron.it

Laboratorio Qualificato D.M. 26-2-87 Art. 4 - Legge 46/82 per la Ricerca Applicata e Innovazione Tecnologica.

Regione Emilia Romagna - AUTORIZZAZIONE Autocontrollo N° 008/MO/008

BNN-Monitoring Fruit and Vegetables Approved Laboratory

I-Monitoring EDEKAAG Fruit and Vegetables Registered Laboratory

GMP+ code: GMP051757

MODENA, li 26/10/2022

Sample arrived on the 14/10/2022

Registration date 14/10/2022

**TEST REPORT nr. 22P11006-In-0**

CUSTOMER

**ACETFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**
**SAMPLE 22P11006****MATRIX: Fruit and vegetable concentrates, and by-products**

Description provided by Customer: GLASSA CON ABM IGP - CONTAMINAZIONE ALTA - VISCOSITA': ~ 4.000 CPS (T=10G) - CODICE: S20A-4000

Richiesta via Internet n° N0382/22 - 26/09/2022 09:50:54. - Campionamento eseguito da: Committente - Trasporto effettuato da: Committente  
Sample Condition on Receipt: 20°C

ANALYSIS DESCRIPTION	RESULT	U	REC. %	UNIT OF MEASURE	LQ	LD	METHOD	ANALYSES BEGINNING DATE / ENDING DATE
Water activity (Aw) at 25 °C	0,927	± 0,005			0,150		06(S135) rev6 2021	17/10/2022 / 18/10/2022
<b>VINEGAR ANALYSIS</b>								
Reduction sugars (Reducing substances)	519,1	± 15,6		g/l	1,0		07(S240) 2021 Rev.0 - Titrimetrico	18/10/2022 / 26/10/2022
Total Acidity expressed as acetic acid	3,13	± 0,06		g/100 ml	0,10		* ACIDITA 2014 Rev.0 - Titrimetrico	18/10/2022 / 19/10/2022
Relative density at 20°C	1,196	± 0,0003					07(S117) 2015 Rev.1 - Gravimetric	25/10/2022 / 25/10/2022
Alcoholic strength	0,08	± 0,01		% Vol	0,07		DM 12/03/1986 SO GU n 161 14/07/1986 Met V pag 95, OIV-MA-AS312-01 Met B R2021	18/10/2022 / 19/10/2022
pH	3,00	± 0,04		unità	0,50		07(S77) 2012 Rev.1 - Potentiometric	18/10/2022 / 19/10/2022
Brookfield rotational viscosity	Brookfield Rotational Viscosity (T = 20 ° C, impeller R5)							
	50 rpm: 4823 cP (mPa.s)							
	30 rpm: 6510 cP (mPa.s)							
	20 rpm: 8281 cP (mPa.s)							
<b>SUGARS HPAEC-PAD</b>								
Galactose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	18/10/2022 / 21/10/2022
Glucose, anhydrous (HPLC)	18,4	± 1,3		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	18/10/2022 / 21/10/2022
Sucrose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	18/10/2022 / 21/10/2022
Fructose, anhydrous (HPLC)	19,3	± 1,4		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	18/10/2022 / 21/10/2022
Lactose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	18/10/2022 / 21/10/2022
Maltose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	18/10/2022 / 21/10/2022
Citric acid (LCMS) [331]	86,7	± 14,1	101	mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	18/10/2022 / 24/10/2022
Lactic Acid (LCMS) [331]	269	± 37	74 +	mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	18/10/2022 / 24/10/2022
Malic acid (LCMS) [331]	605	± 74	90	mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	18/10/2022 / 24/10/2022
Succinic acid (LCMS) [331]	168	± 25	90	mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	18/10/2022 / 24/10/2022
Tartaric acid (LCMS) [331]	581	± 71	80	mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	18/10/2022 / 24/10/2022
Ochratoxin A	< LQ			µg/kg	0,10		03(S130) 2022 Rev.13 - LC-MS/MS	18/10/2022 / 21/10/2022

**END TEST REPORT**

The original document is a PDF file with Digital Signature: 22P11006-In-0-S20A-4000 - T20 - UNIMORE-DigitalSignature.pdf

**Next page...**

MODENA, li 26/10/2022

Sample arrived on the 14/10/2022

Registration date 14/10/2022

## TEST REPORT nr. 22P11006-In-0

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**

**SAMPLE 22P11006**

**MATRIX: Fruit and vegetable concentrates, and by-products**

### Notes and method reference:

< LQ: = lower than Quantification Limit.

U: the reported uncertainty is the expanded uncertainty calculated using a coverage factor equal to 2 which gives a reliability of approximately 95%. Please note that results expressed as '<LQ' may not indicate the absence of the searched parameters in the sample.

MICROBIOLOGICAL TESTS: for food and environmental samples, the extended measurement uncertainty was estimated according to ISO 19036:2019 Standard and is based on a standard uncertainty multiplied by a coverage factor of  $K = 2$ , providing a confidence level of approximately 95%. The combined standard uncertainty was assumed to be equal to the standard deviation of intra-laboratory reproducibility. The results of the microbiological tests are calculated according to the ISO 7218: 2007 / Amd 1: 2013 Standard. If the results are reported as <4 (CFU/ml) or <40 (CFU/g), this means that the microorganisms are present in the sample but in amounts less than 4 CFU/ml or 40 CFU/g respectively. For microbiological analyses unless differently reported in the individual test methods, in case of analytical steps foreseen in non-activity days of the laboratory, provisions of the ISO 7218: 2007 / Amd.1 2013 Standard (points 11.2 and 10.2.5) or from specific test methods are applied. In the case of quantitative microbiological tests, these have been set up on a single plate according to ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise expressly requested by current regulations.

In the case of quantitative microbiological tests, these have been set up on a single plate in accordance with ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise explicitly required by current regulations.

For waters, the measurement uncertainty corresponds to the confidence interval calculated according to ISO 8199: 2018 or to the expanded measurement uncertainty estimated according to ISO 29201: 2012. The results are issued in accordance with ISO 8199: 2018. When the number of colonies detected is <3, the result is expressed as "Microorganisms present in the analyzed volume (N ° colonies detected <3 CFU - reference ISO 8199: 2018, paragraph 9.1.8.4.1)".

LQ: Quantification Limit. It is the lowest analyte concentration which can be detected at an acceptable precision (repeatability) and accuracy, under well defined conditions.

LD: Detection Limit. It is the lowest analyte concentration which can be detected but not necessarily quantified, under well defined conditions. Any fields not filled in the Test Report are to be considered not applicable.

Conformity evaluation: values not complying with laws, decrees, national and EU regulations or specifications supplied by the customer are evaluated case by case, also taking into consideration the uncertainty of measure for each single test and the regulations on rounding-off of values, and pointed out when considered as non conform.

Rec %: Recovery % "+" means that the recovery has been applied to the result. The numeric results between brackets (.) after the expression <LQ are purely indicative of traces that cannot be exactly quantified.

In the case of sampling carried out by Neotron, the laboratory applies the Internal Operating Procedure code: NEOT-DIR/ 006/53.

The laboratory disclaims any responsibility for the information provided by the client reported in this Report which may influence the validity of the results.

Methods marked with an asterisk (\*) are not accredited by ACCREDIA (UNI CEI EN ISO/IEC 17025). The sampling activity is not included within the Scope of Accreditation of Neotron SPA

### NOTES OF PARAMETERS:

[331]: Extended uncertainty calculated following the equation of HORWITZ by using a coverage factor of 2, which guarantee a reliance level of 95%

TEST REPORT VALID FOR ALL LEGAL PURPOSES (Italian R.D. 1-3-1928 n°842 (article 16), - Italian Law 19-7-1957 n°679 articles 16 and 18, Italian Ministerial Decree 25-3-1986).

DATA and SAMPLE STORAGE: Test Reports, Raw data, chromatographic paths and instrumental reports are stored for 5 years. One control sample is stored for 2 months.

Data expressed in this test report refer only to the sample tested in the laboratory. The results reported in this Test Report refer to the sample as received. The description or any other reference concerning the sample are declared by the customer. This Test Report cannot be reproduced except in full. Partial reproductions must be authorized in writing by our laboratory.

THE LABORATORY DIRECTOR: DR. ANDREA RIZZO

THE CHEMIST AUTHORIZED TO SIGN THE TEST REPORTS: DR. MARCO MESCHIARI

NEOTRON SpA - With Sole Shareholder

Stradello Aggazzotti, 104

41126 MODENA - ITALY - Fiscal Code and VAT n° 03807840362

Tel: +39 059461711 - Fax: +39 059461777

www.neotron.it - neotron@neotron.it

Laboratorio Qualificato D.M. 26-2-87 Art. 4 - Legge 46/82 per la Ricerca Applicata e Innovazione Tecnologica.

Regione Emilia Romagna - AUTORIZZAZIONE Autocontrollo N° 008/MO/008

BNN-Monitoring Fruit and Vegetables Approved Laboratory

I-Monitoring EDEKAAG Fruit and Vegetables Registered Laboratory

GMP+ code: GMP051757

MODENA, li 20/10/2022

Sample arrived on the 05/10/2022  
Registration date 05/10/2022**TEST REPORT nr. 22P03161-In-0**

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO****SAMPLE 22P03161****MATRIX: Fruit and vegetable concentrates, and  
by-products**

Description provided by Customer: GLASSA CON ABM IGP - CONTAMINAZIONE ALTA - VISCOSITA': ~ 4.000 CPS (T=10G) - CODICE: S20A-4000

Richiesta via Internet n° N0380/22 - 26/09/2022 09:50:53. - Campionamento eseguito da: Committente - Trasporto effettuato da: Corriere  
Sample Condition on Receipt: 20°C

ANALYSIS DESCRIPTION	RESULT	U	REC. %	UNIT OF MEASURE	LQ	LD	METHOD	ANALYSES BEGINNING DATE / ENDING DATE
Water activity (Aw) at 25 °C	0,933	± 0,005			0,150		06(S135) rev6 2021	05/10/2022 / 11/10/2022
<b>VINEGAR ANALYSIS</b>								
Reduction sugars (Reducing substances)	491,4	± 14,7		g/l	1,0		07(S240) 2021 Rev.0 - Titrimetrico	06/10/2022 / 12/10/2022
Total Acidity expressed as acetic acid	3,10	± 0,06		g/100 ml	0,10		* ACIDITA 2014 Rev.0 - Titrimetrico	06/10/2022 / 06/10/2022
Relative density at 20°C	1,196	± 0,001					07(S117) 2015 Rev.1 - Gravimetric	20/10/2022 / 20/10/2022
Alcoholic strength	0,09	± 0,01		% Vol	0,07		DM 12/03/1986 SO GU n 161 14/07/1986 Met V pag 95, OIV-MA-AS312-01 Met B R2021	06/10/2022 / 12/10/2022
Brookfield rotational viscosity	Brookfield Rotational Viscosity (T = 20 ° C, impeller R5)							
	50 rpm: 5140 cP (mPa.s)							
	30 rpm: 6968 cP (mPa.s)							
	20 rpm: 8940 cP (mPa.s)							
pH	2,98	± 0,04		unità	0,50		07(S77) 2012 Rev.1 - Potentiometric	06/10/2022 / 07/10/2022
<b>SUGARS HPAEC-PAD</b>								
Galactose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	06/10/2022 / 12/10/2022
Glucose, anhydrous (HPLC)	18,3	± 1,3		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	06/10/2022 / 12/10/2022
Sucrose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	06/10/2022 / 12/10/2022
Fructose, anhydrous (HPLC)	19,9	± 1,4		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	06/10/2022 / 12/10/2022
Lactose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	06/10/2022 / 12/10/2022
Maltose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	06/10/2022 / 12/10/2022
Citric acid (LCMS) [331]	107	± 17		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	06/10/2022 / 11/10/2022
Lactic Acid (LCMS) [331]	136	± 21		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	06/10/2022 / 11/10/2022
Malic acid (LCMS) [331]	457	± 58		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	06/10/2022 / 11/10/2022
Succinic acid (LCMS) [331]	260	± 36		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	06/10/2022 / 11/10/2022
Tartaric acid (LCMS) [331]	344	± 46		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	06/10/2022 / 11/10/2022
Ochratoxin A	< LQ			µg/kg	0,10		03(S130) 2022 Rev.13 - LC-MS/MS	06/10/2022 / 11/10/2022

**END TEST REPORT**

The original document is a PDF file with Digital Signature: 22P03161-In-0-S20A-4000 - T10 - UNIMORE-DigitalSignature.pdf

**Next page...**

MODENA, li 20/10/2022

Sample arrived on the 05/10/2022

Registration date 05/10/2022

## TEST REPORT nr. 22P03161-In-0

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**

**SAMPLE 22P03161**

**MATRIX: Fruit and vegetable concentrates, and by-products**

### Notes and method reference:

< LQ: = lower than Quantification Limit.

U: the reported uncertainty is the expanded uncertainty calculated using a coverage factor equal to 2 which gives a reliability of approximately 95%. Please note that results expressed as '<LQ' may not indicate the absence of the searched parameters in the sample.

MICROBIOLOGICAL TESTS: for food and environmental samples, the extended measurement uncertainty was estimated according to ISO 19036:2019 Standard and is based on a standard uncertainty multiplied by a coverage factor of  $K = 2$ , providing a confidence level of approximately 95%. The combined standard uncertainty was assumed to be equal to the standard deviation of intra-laboratory reproducibility. The results of the microbiological tests are calculated according to the ISO 7218: 2007 / Amd 1: 2013 Standard. If the results are reported as <4 (CFU/ml) or <40 (CFU/g), this means that the microorganisms are present in the sample but in amounts less than 4 CFU/ml or 40 CFU/g respectively. For microbiological analyses unless differently reported in the individual test methods, in case of analytical steps foreseen in non-activity days of the laboratory, provisions of the ISO 7218: 2007 / Amd.1 2013 Standard (points 11.2 and 10.2.5) or from specific test methods are applied. In the case of quantitative microbiological tests, these have been set up on a single plate according to ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise expressly requested by current regulations.

In the case of quantitative microbiological tests, these have been set up on a single plate in accordance with ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise explicitly required by current regulations.

For waters, the measurement uncertainty corresponds to the confidence interval calculated according to ISO 8199: 2018 or to the expanded measurement uncertainty estimated according to ISO 29201: 2012. The results are issued in accordance with ISO 8199: 2018. When the number of colonies detected is <3, the result is expressed as "Microorganisms present in the analyzed volume (N ° colonies detected <3 CFU - reference ISO 8199: 2018, paragraph 9.1.8.4.1)".

LQ: Quantification Limit. It is the lowest analyte concentration which can be detected at an acceptable precision (repeatability) and accuracy, under well defined conditions.

LD: Detection Limit. It is the lowest analyte concentration which can be detected but not necessarily quantified, under well defined conditions. Any fields not filled in the Test Report are to be considered not applicable.

Conformity evaluation: values not complying with laws, decrees, national and EU regulations or specifications supplied by the customer are evaluated case by case, also taking into consideration the uncertainty of measure for each single test and the regulations on rounding-off of values, and pointed out when considered as non conform.

Rec %: Recovery % "+" means that the recovery has been applied to the result. The numeric results between brackets (.) after the expression <LQ are purely indicative of traces that cannot be exactly quantified.

In the case of sampling carried out by Neotron, the laboratory applies the Internal Operating Procedure code: NEOT-DIR/ 006/53.

The laboratory disclaims any responsibility for the information provided by the client reported in this Report which may influence the validity of the results.

Methods marked with an asterisk (\*) are not accredited by ACCREDIA (UNI CEI EN ISO/IEC 17025). The sampling activity is not included within the Scope of Accreditation of Neotron SPA

### NOTES OF PARAMETERS:

[331]: Extended uncertainty calculated following the equation of HORWITZ by using a coverage factor of 2, which guarantee a reliance level of 95%

TEST REPORT VALID FOR ALL LEGAL PURPOSES (Italian R.D. 1-3-1928 n°842 (article 16), - Italian Law 19-7-1957 n°679 articles 16 and 18, Italian Ministerial Decree 25-3-1986).

DATA and SAMPLE STORAGE: Test Reports, Raw data, chromatographic paths and instrumental reports are stored for 5 years. One control sample is stored for 2 months.

Data expressed in this test report refer only to the sample tested in the laboratory. The results reported in this Test Report refer to the sample as received. The description or any other reference concerning the sample are declared by the customer. This Test Report cannot be reproduced except in full. Partial reproductions must be authorized in writing by our laboratory.

THE LABORATORY DIRECTOR: DR. ANDREA RIZZO

THE CHEMIST AUTHORIZED TO SIGN THE TEST REPORTS: DR. MARCO MESCHIARI

NEOTRON SpA - With Sole Shareholder

Stradello Aggazzotti, 104

41126 MODENA - ITALY - Fiscal Code and VAT n° 03807840362

Tel: +39 059461711 - Fax: +39 059461777

www.neotron.it - neotron@neotron.it

Laboratorio Qualificato D.M. 26-2-87 Art. 4 - Legge 46/82 per la Ricerca Applicata e Innovazione Tecnologica.

Regione Emilia Romagna - AUTORIZZAZIONE Autocontrollo N° 008/MO/008

BNN-Monitoring Fruit and Vegetables Approved Laboratory

I-Monitoring EDEKAAG Fruit and Vegetables Registered Laboratory

GMP+ code: GMP051757



MODENA, li 12/10/2022

Sample arrived on the 26/09/2022  
Registration date 26/09/2022**TEST REPORT nr. 22M18694-In-0**

CUSTOMER

**ACETFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO****SAMPLE 22M18694****MATRIX: Fruit and vegetable concentrates, and  
by-products**Description provided by Customer: GLASSA CON ABM IGP - CONTAMINAZIONE ALTA - VISCOSITA': ~ 4.000 CPS (T=0G) -  
CODICE: S20A-4000Richiesta via Internet n° N0378/22 - 26/09/2022 09:50:53. - Campionamento eseguito da: Committente - Trasporto effettuato da:  
Committente  
Sample Condition on Receipt: 5°C

ANALYSIS DESCRIPTION	RESULT	U	REC. %	UNIT OF MEASURE	LQ	LD	METHOD	ANALYSES BEGINNING DATE / ENDING DATE
Water activity (Aw) at 25 °C	0,924	± 0,005			0,150		06(S135) rev6 2021	27/09/2022 / 30/09/2022
<b>VINEGAR ANALYSIS</b>								
Reduction sugars (Reducing substances)	482,1	± 14,5		g/l	1,0		07(S240) 2021 Rev.0 - Titrimetrico	28/09/2022 / 07/10/2022
Total Acidity expressed as acetic acid	3,27	± 0,06		g/100 ml	0,10		* ACIDITA 2014 Rev.0 - Titrimetrico	28/09/2022 / 30/09/2022
Relative density at 20°C	1,203	± 0,0003					- Gravimetric	11/10/2022 / 11/10/2022
Alcoholic strength	0,08	± 0,01		% Vol	0,07		DM 12/03/1986 SO GU n 161 14/07/1986 Met V pag 95, OIV- MA-AS312-01 Met B R2021	27/09/2022 / 06/10/2022
pH	2,95	± 0,04		unità	0,50		07(S77) 2012 Rev.1 - Potentiometric	28/09/2022 / 03/10/2022
Brookfield rotational viscosity	Brookfield Rotational Viscosity (T = 20 ° C, impeller R5) 60 rpm: 4377 cP (mPa.s) 50 rpm: 4898 cP (mPa.s)							
<b>SUGARS HPAEC-PAD</b>								
Galactose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	28/09/2022 / 12/10/2022
Glucose, anhydrous (HPLC)	18,6	± 1,3		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	28/09/2022 / 12/10/2022
Sucrose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	28/09/2022 / 12/10/2022
Fructose, anhydrous (HPLC)	19,5	± 1,4		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	28/09/2022 / 12/10/2022
Lactose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	28/09/2022 / 12/10/2022
Maltose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	28/09/2022 / 12/10/2022
Citric acid (LCMS) [331]	74,5	± 12,4		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	28/09/2022 / 06/10/2022
Lactic Acid (LCMS) [331]	332	± 44		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	28/09/2022 / 06/10/2022
Malic acid (LCMS) [331]	624	± 76		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	28/09/2022 / 06/10/2022
Succinic acid (LCMS) [331]	193	± 28		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	28/09/2022 / 06/10/2022
Tartaric acid (LCMS) [331]	582	± 72		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	28/09/2022 / 06/10/2022
Ochratoxin A	< LQ			µg/kg	0,10		03(S130) 2022 Rev.13 - LC- MS/MS	28/09/2022 / 07/10/2022

**END TEST REPORT**

The original document is a PDF file with Digital Signature: 22M18694-In-0-S20A-4000 - T0 - UNIMORE-DigitalSignature.pdf

**Next page...**

MODENA, li 12/10/2022

Sample arrived on the 26/09/2022

Registration date 26/09/2022

## TEST REPORT nr. 22M18694-In-0

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**

**SAMPLE 22M18694**

**MATRIX: Fruit and vegetable concentrates, and by-products**

### Notes and method reference:

< LQ: = lower than Quantification Limit.

U: the reported uncertainty is the expanded uncertainty calculated using a coverage factor equal to 2 which gives a reliability of approximately 95%. Please note that results expressed as '<LQ' may not indicate the absence of the searched parameters in the sample.

MICROBIOLOGICAL TESTS: for food and environmental samples, the extended measurement uncertainty was estimated according to ISO 19036:2019 Standard and is based on a standard uncertainty multiplied by a coverage factor of K = 2, providing a confidence level of approximately 95%. The combined standard uncertainty was assumed to be equal to the standard deviation of intra-laboratory reproducibility. The results of the microbiological tests are calculated according to the ISO 7218: 2007 / Amd 1: 2013 Standard. If the results are reported as <4 (CFU/ml) or <40 (CFU/g), this means that the microorganisms are present in the sample but in amounts less than 4 CFU/ml or 40 CFU/g respectively. For microbiological analyses unless differently reported in the individual test methods, in case of analytical steps foreseen in non-activity days of the laboratory, provisions of the ISO 7218: 2007 / Amd.1 2013 Standard (points 11.2 and 10.2.5) or from specific test methods are applied. In the case of quantitative microbiological tests, these have been set up on a single plate according to ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise expressly requested by current regulations.

In the case of quantitative microbiological tests, these have been set up on a single plate in accordance with ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise explicitly required by current regulations.

For waters, the measurement uncertainty corresponds to the confidence interval calculated according to ISO 8199: 2018 or to the expanded measurement uncertainty estimated according to ISO 29201: 2012. The results are issued in accordance with ISO 8199: 2018. When the number of colonies detected is <3, the result is expressed as "Microorganisms present in the analyzed volume (N ° colonies detected <3 CFU - reference ISO 8199: 2018, paragraph 9.1.8.4.1)".

LQ: Quantification Limit. It is the lowest analyte concentration which can be detected at an acceptable precision (repeatability) and accuracy, under well defined conditions.

LD: Detection Limit. It is the lowest analyte concentration which can be detected but not necessarily quantified, under well defined conditions. Any fields not filled in the Test Report are to be considered not applicable.

Conformity evaluation: values not complying with laws, decrees, national and EU regulations or specifications supplied by the customer are evaluated case by case, also taking into consideration the uncertainty of measure for each single test and the regulations on rounding-off of values, and pointed out when considered as non conform.

Rec %: Recovery % "+" means that the recovery has been applied to the result. The numeric results between brackets (.) after the expression <LQ are purely indicative of traces that cannot be exactly quantified.

In the case of sampling carried out by Neotron, the laboratory applies the Internal Operating Procedure code: NEOT-DIR/ 006/53.

The laboratory disclaims any responsibility for the information provided by the client reported in this Report which may influence the validity of the results.

Methods marked with an asterisk (\*) are not accredited by ACCREDIA (UNI CEI EN ISO/IEC 17025). The sampling activity is not included within the Scope of Accreditation of Neotron SPA

### NOTES OF PARAMETERS:

[331]: Extended uncertainty calculated following the equation of HORWITZ by using a coverage factor of 2, which guarantee a reliance level of 95%

TEST REPORT VALID FOR ALL LEGAL PURPOSES (Italian R.D. 1-3-1928 n°842 (article 16), - Italian Law 19-7-1957 n°679 articles 16 and 18, Italian Ministerial Decree 25-3-1986).

DATA and SAMPLE STORAGE: Test Reports, Raw data, chromatographic paths and instrumental reports are stored for 5 years. One control sample is stored for 2 months.

Data expressed in this test report refer only to the sample tested in the laboratory. The results reported in this Test Report refer to the sample as received. The description or any other reference concerning the sample are declared by the customer. This Test Report cannot be reproduced except in full. Partial reproductions must be authorized in writing by our laboratory.

THE LABORATORY DIRECTOR: DR. ANDREA RIZZO

THE CHEMIST AUTHORIZED TO SIGN THE TEST REPORTS: DR. MARCO MESCHIARI

NEOTRON SpA - With Sole Shareholder

Stradello Aggazzotti, 104

41126 MODENA - ITALY - Fiscal Code and VAT n° 03807840362

Tel: +39 059461711 - Fax: +39 059461777

www.neotron.it - neotron@neotron.it

Laboratorio Qualificato D.M. 26-2-87 Art. 4 - Legge 46/82 per la Ricerca Applicata e Innovazione Tecnologica.

Regione Emilia Romagna - AUTORIZZAZIONE Autocontrollo N° 008/MO/008

BNN-Monitoring Fruit and Vegetables Approved Laboratory

I-Monitoring EDEKAAG Fruit and Vegetables Registered Laboratory

GMP+ code: GMP051757

MODENA, li 13/10/2022

Sample arrived on the 23/09/2022  
Registration date 23/09/2022**TEST REPORT nr. 22M17702-In-0**

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO****SAMPLE 22M17702****MATRIX: Fruit and vegetable concentrates, and by-products**

Description provided by Customer: GLASSA CON ABM IGP - CONTAMINAZIONE ALTA - VISCOSITA': ~ 3.000 CPS (T=20G) - CODICE: S20A-3000

Richiesta via Internet n° N0325/22 - 05/09/2022 13:57:33. - Campionamento eseguito da: Committente - Trasporto effettuato da: Committente  
Sample Condition on Receipt: 20°C

ANALYSIS DESCRIPTION	RESULT	U	REC. %	UNIT OF MEASURE	LQ	LD	METHOD	ANALYSES BEGINNING DATE / ENDING DATE
Water activity (Aw) at 25 °C	0,930	± 0,005			0,150		06(S135) rev6 2021	23/09/2022 / 30/09/2022
<b>VINEGAR ANALYSIS</b>								
Reduction sugars (Reducing substances)	420,4	± 12,6		g/l	1,0		07(S240) 2021 Rev.0 - Titrimetrico	24/09/2022 / 30/09/2022
Total Acidity expressed as acetic acid	2,82	± 0,05		g/100 ml	0,10		* ACIDITA 2014 Rev.0 - Titrimetrico	24/09/2022 / 29/09/2022
Relative density at 20°C	1,188	± 0,001					DM 12/03/1986 SO GU n 161 14/07/1986 Met VI pag 95 p.to 4, OIV-MA-AS2-01A Met B R2021 - densimetry	24/09/2022 / 30/09/2022
Alcoholic strength	0,09	± 0,01		% Vol	0,07		DM 12/03/1986 SO GU n 161 14/07/1986 Met V pag 95, OIV-MA-AS312-01 Met B R2021	24/09/2022 / 30/09/2022
Brookfield rotational viscosity	Brookfield Rotational Viscosity (T = 20 ° C, impeller R3)						* VISCOSITA 2018 Rev.1	24/09/2022 / 06/10/2022
	30 rpm: 2095 cP (mPa.s) 20 rpm: 2560 cP (mPa.s) 12 rpm: 3317 cP (mPa.s)							
pH	3,03	± 0,04		unità	0,50		07(S77) 2012 Rev.1 - Potentiometric	24/09/2022 / 13/10/2022
<b>SUGARS HPAEC-PAD</b>								
Galactose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	24/09/2022 / 12/10/2022
Glucose, anhydrous (HPLC)	16,9	± 1,2		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	24/09/2022 / 12/10/2022
Sucrose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	24/09/2022 / 12/10/2022
Fructose, anhydrous (HPLC)	19,9	± 1,4		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	24/09/2022 / 12/10/2022
Lactose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	24/09/2022 / 12/10/2022
Maltose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	24/09/2022 / 12/10/2022
Citric acid (LCMS) [331]	76,3	± 12,7		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	24/09/2022 / 06/10/2022
Lactic Acid (LCMS) [331]	385	± 50		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	24/09/2022 / 06/10/2022
Malic acid (LCMS) [331]	586	± 72		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	24/09/2022 / 06/10/2022
Succinic acid (LCMS) [331]	189	± 27		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	24/09/2022 / 06/10/2022
Tartaric acid (LCMS) [331]	562	± 69		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	24/09/2022 / 06/10/2022
Ochratoxin A	< LQ			µg/kg	0,10		03(S130) 2022 Rev.13 - LC-MS/MS	24/09/2022 / 06/10/2022

**END TEST REPORT**

The original document is a PDF file with Digital Signature: 22M17702-In-0-S20A-3000 - T20 - UNIMORE-DigitalSignature.pdf

MODENA, li 13/10/2022

Sample arrived on the 23/09/2022

Registration date 23/09/2022

**TEST REPORT nr. 22M17702-In-0**

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**

SAMPLE 22M17702

**MATRIX: Fruit and vegetable concentrates, and  
by-products**

## Notes and method reference:

&lt; LQ: = lower than Quantification Limit.

U: the reported uncertainty is the expanded uncertainty calculated using a coverage factor equal to 2 which gives a reliability of approximately 95%. Please note that results expressed as '&lt;LQ' may not indicate the absence of the searched parameters in the sample.

MICROBIOLOGICAL TESTS: for food and environmental samples, the extended measurement uncertainty was estimated according to ISO 19036:2019 Standard and is based on a standard uncertainty multiplied by a coverage factor of K = 2, providing a confidence level of approximately 95%. The combined standard uncertainty was assumed to be equal to the standard deviation of intra-laboratory reproducibility. The results of the microbiological tests are calculated according to the ISO 7218: 2007 / Amd 1: 2013 Standard. If the results are reported as <4 (CFU/ml) or <40 (CFU/g), this means that the microorganisms are present in the sample but in amounts less than 4 CFU/ml or 40 CFU/g respectively. For microbiological analyses unless differently reported in the individual test methods, in case of analytical steps foreseen in non-activity days of the laboratory, provisions of the ISO 7218: 2007 / Amd.1 2013 Standard (points 11.2 and 10.2.5) or from specific test methods are applied. In the case of quantitative microbiological tests, these have been set up on a single plate according to ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise expressly requested by current regulations.

In the case of quantitative microbiological tests, these have been set up on a single plate in accordance with ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise explicitly required by current regulations.

For waters, the measurement uncertainty corresponds to the confidence interval calculated according to ISO 8199: 2018 or to the expanded measurement uncertainty estimated according to ISO 29201: 2012. The results are issued in accordance with ISO 8199: 2018. When the number of colonies detected is <3, the result is expressed as "Microorganisms present in the analyzed volume (N ° colonies detected <3 CFU - reference ISO 8199: 2018, paragraph 9.1.8.4.1)".

LQ: Quantification Limit. It is the lowest analyte concentration which can be detected at an acceptable precision (repeatability) and accuracy, under well defined conditions.

LD: Detection Limit. It is the lowest analyte concentration which can be detected but not necessarily quantified, under well defined conditions. Any fields not filled in the Test Report are to be considered not applicable.

Conformity evaluation: values not complying with laws, decrees, national and EU regulations or specifications supplied by the customer are evaluated case by case, also taking into consideration the uncertainty of measure for each single test and the regulations on rounding-off of values, and pointed out when considered as non conform.

Rec %: Recovery % "+" means that the recovery has been applied to the result. The numeric results between brackets (.) after the expression <LQ are purely indicative of traces that cannot be exactly quantified.

In the case of sampling carried out by Neotron, the laboratory applies the Internal Operating Procedure code: NEOT-DIR/ 006/53.

The laboratory disclaims any responsibility for the information provided by the client reported in this Report which may influence the validity of the results.

Methods marked with an asterisk (\*) are not accredited by ACCREDIA (UNI CEI EN ISO/IEC 17025). The sampling activity is not included within the Scope of Accreditation of Neotron SPA

## NOTES OF PARAMETERS:

[331]: Extended uncertainty calculated following the equation of HORWITZ by using a coverage factor of 2, which guarantee a reliance level of 95%

TEST REPORT VALID FOR ALL LEGAL PURPOSES (Italian R.D. 1-3-1928 n°842 (article 16), - Italian Law 19-7-1957 n°679 articles 16 and 18, Italian Ministerial Decree 25-3-1986).

DATA and SAMPLE STORAGE: Test Reports, Raw data, chromatographic paths and instrumental reports are stored for 5 years. One control sample is stored for 2 months.

Data expressed in this test report refer only to the sample tested in the laboratory. The results reported in this Test Report refer to the sample as received. The description or any other reference concerning the sample are declared by the customer. This Test Report cannot be reproduced except in full. Partial reproductions must be authorized in writing by our laboratory.

THE LABORATORY DIRECTOR: DR. ANDREA RIZZO

THE CHEMIST AUTHORIZED TO SIGN THE TEST REPORTS: DR. MARCO MESCHIARI

NEOTRON SpA - With Sole Shareholder

Stradello Aggazzotti, 104

41126 MODENA - ITALY - Fiscal Code and VAT n° 03807840362

Tel: +39 059461711 - Fax: +39 059461777

www.neotron.it - neotron@neotron.it

Laboratorio Qualificato D.M. 26-2-87 Art. 4 - Legge 46/82 per la Ricerca Applicata e Innovazione Tecnologica.

Regione Emilia Romagna - AUTORIZZAZIONE Autocontrollo N° 008/MO/008

BNN-Monitoring Fruit and Vegetables Approved Laboratory

I-Monitoring EDEKAAG Fruit and Vegetables Registered Laboratory

GMP+ code: GMP051757

MODENA, li 04/10/2022

Sample arrived on the 13/09/2022  
Registration date 13/09/2022**TEST REPORT nr. 22M08846-In-0**

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO****SAMPLE 22M08846****MATRIX: Fruit and vegetable concentrates, and by-products**

Description provided by Customer: GLASSA CON ABM IGP - CONTAMINAZIONE ALTA - VISCOSITA': ~ 3.000 CPS (T=10G) - CODICE: S20A-3000

Richiesta via Internet n° N0326/22 - 05/09/2022 13:59:07. - Campionamento eseguito da: Committente - Trasporto effettuato da: Committente  
Sample Condition on Receipt: 23°C

ANALYSIS DESCRIPTION	RESULT	U	REC. %	UNIT OF MEASURE	LQ	LD	METHOD	ANALYSES BEGINNING DATE / ENDING DATE
Water activity (Aw) at 25 °C	0,935	± 0,005			0,150		06(S135) rev6 2021	14/09/2022 / 20/09/2022
<b>VINEGAR ANALYSIS</b>								
Reduction sugars (Reducing substances)	434,5	± 13,0		g/l	1,0		07(S240) 2021 Rev.0 - Titrimetrico	16/09/2022 / 20/09/2022
Total Acidity expressed as acetic acid	2,61	± 0,05		g/100 ml	0,10		* ACIDITA 2014 Rev.0 - Titrimetrico	16/09/2022 / 20/09/2022
Relative density at 20°C	1,171	± 0,001					DM 12/03/1986 SO GU n 161 14/07/1986 Met VI pag 95 p.to 4, OIV-MA-AS2-01A Met B R2021 - densimetry	16/09/2022 / 20/09/2022
Alcoholic strength	0,11	± 0,01		% Vol	0,07		DM 12/03/1986 SO GU n 161 14/07/1986 Met V pag 95, OIV-MA-AS312-01 Met B R2021	16/09/2022 / 20/09/2022
pH	2,95	± 0,04		unità	0,50		07(S77) 2012 Rev.1 - Potentiometric	16/09/2022 / 23/09/2022
Brookfield rotational viscosity	Brookfield Rotational Viscosity (T = 20 ° C, impeller R3) 30 rpm: 2295 cP (mPa.s) 20 rpm: 2877 cP (mPa.s) 12 rpm: 3801 cP (mPa.s)						* VISCOSITA 2018 Rev.1	16/09/2022 / 21/09/2022
<b>SUGARS HPAEC-PAD</b>								
Galactose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	16/09/2022 / 20/09/2022
Glucose, anhydrous (HPLC)	17,1	± 1,2		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	16/09/2022 / 20/09/2022
Sucrose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	16/09/2022 / 20/09/2022
Fructose, anhydrous (HPLC)	17,2	± 1,2		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	16/09/2022 / 20/09/2022
Lactose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	16/09/2022 / 20/09/2022
Maltose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	16/09/2022 / 20/09/2022
Citric acid (LCMS) [331]	28,7	± 5,5		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	16/09/2022 / 04/10/2022
Lactic Acid (LCMS) [331]	728	± 87		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	16/09/2022 / 04/10/2022
Malic acid (LCMS) [331]	434	± 56		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	16/09/2022 / 04/10/2022
Succinic acid (LCMS) [331]	189	± 27		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	16/09/2022 / 04/10/2022
Tartaric acid (LCMS) [331]	483	± 61		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	16/09/2022 / 04/10/2022
Ochratoxin A	< LQ			µg/kg	0,10		03(S130) 2022 Rev.13 - LC-MS/MS	16/09/2022 / 20/09/2022

**END TEST REPORT**

The original document is a PDF file with Digital Signature: 22M08846-In-0-S20A-3000 - T10 - UNIMORE-DigitalSignature.pdf

MODENA, li 04/10/2022

Sample arrived on the 13/09/2022

Registration date 13/09/2022

## TEST REPORT nr. 22M08846-In-0

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**

**SAMPLE 22M08846**

**MATRIX: Fruit and vegetable concentrates, and by-products**

### Notes and method reference:

< LQ: = lower than Quantification Limit.

U: the reported uncertainty is the expanded uncertainty calculated using a coverage factor equal to 2 which gives a reliability of approximately 95%. Please note that results expressed as '<LQ' may not indicate the absence of the searched parameters in the sample.

MICROBIOLOGICAL TESTS: for food and environmental samples, the extended measurement uncertainty was estimated according to ISO 19036:2019 Standard and is based on a standard uncertainty multiplied by a coverage factor of K = 2, providing a confidence level of approximately 95%. The combined standard uncertainty was assumed to be equal to the standard deviation of intra-laboratory reproducibility. The results of the microbiological tests are calculated according to the ISO 7218: 2007 / Amd 1: 2013 Standard. If the results are reported as <4 (CFU/ml) or <40 (CFU/g), this means that the microorganisms are present in the sample but in amounts less than 4 CFU/ml or 40 CFU/g respectively. For microbiological analyses unless differently reported in the individual test methods, in case of analytical steps foreseen in non-activity days of the laboratory, provisions of the ISO 7218: 2007 / Amd.1 2013 Standard (points 11.2 and 10.2.5) or from specific test methods are applied. In the case of quantitative microbiological tests, these have been set up on a single plate according to ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise expressly requested by current regulations.

In the case of quantitative microbiological tests, these have been set up on a single plate in accordance with ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise explicitly required by current regulations.

For waters, the measurement uncertainty corresponds to the confidence interval calculated according to ISO 8199: 2018 or to the expanded measurement uncertainty estimated according to ISO 29201: 2012. The results are issued in accordance with ISO 8199: 2018. When the number of colonies detected is <3, the result is expressed as "Microorganisms present in the analyzed volume (N ° colonies detected <3 CFU - reference ISO 8199: 2018, paragraph 9.1.8.4.1)".

LQ: Quantification Limit. It is the lowest analyte concentration which can be detected at an acceptable precision (repeatability) and accuracy, under well defined conditions.

LD: Detection Limit. It is the lowest analyte concentration which can be detected but not necessarily quantified, under well defined conditions. Any fields not filled in the Test Report are to be considered not applicable.

Conformity evaluation: values not complying with laws, decrees, national and EU regulations or specifications supplied by the customer are evaluated case by case, also taking into consideration the uncertainty of measure for each single test and the regulations on rounding-off of values, and pointed out when considered as non conform.

Rec %: Recovery % "+" means that the recovery has been applied to the result. The numeric results between brackets (..) after the expression <LQ are purely indicative of traces that cannot be exactly quantified.

In the case of sampling carried out by Neotron, the laboratory applies the Internal Operating Procedure code: NEOT-DIR/ 006/53.

The laboratory disclaims any responsibility for the information provided by the client reported in this Report which may influence the validity of the results.

Methods marked with an asterisk (\*) are not accredited by ACCREDIA (UNI CEI EN ISO/IEC 17025). The sampling activity is not included within the Scope of Accreditation of Neotron SPA

### NOTES OF PARAMETERS:

[331]: Extended uncertainty calculated following the equation of HORWITZ by using a coverage factor of 2, which guarantee a reliance level of 95%

TEST REPORT VALID FOR ALL LEGAL PURPOSES (Italian R.D. 1-3-1928 n°842 (article 16), - Italian Law 19-7-1957 n°679 articles 16 and 18, Italian Ministerial Decree 25-3-1986).

DATA and SAMPLE STORAGE: Test Reports, Raw data, chromatographic paths and instrumental reports are stored for 5 years. One control sample is stored for 2 months.

Data expressed in this test report refer only to the sample tested in the laboratory. The results reported in this Test Report refer to the sample as received. The description or any other reference concerning the sample are declared by the customer. This Test Report cannot be reproduced except in full. Partial reproductions must be authorized in writing by our laboratory.

THE LABORATORY DIRECTOR: DR. ANDREA RIZZO

THE CHEMIST AUTHORIZED TO SIGN THE TEST REPORTS: DR. MARCO MESCHIARI

NEOTRON SpA - With Sole Shareholder

Stradello Aggazzotti, 104

41126 MODENA - ITALY - Fiscal Code and VAT n° 03807840362

Tel: +39 059461711 - Fax: +39 059461777

www.neotron.it - neotron@neotron.it

Laboratorio Qualificato D.M. 26-2-87 Art. 4 - Legge 46/82 per la Ricerca Applicata e Innovazione Tecnologica.

Regione Emilia Romagna - AUTORIZZAZIONE Autocontrollo N° 008/MO/008

BNN-Monitoring Fruit and Vegetables Approved Laboratory

I-Monitoring EDEKAAG Fruit and Vegetables Registered Laboratory

GMP+ code: GMP051757

MODENA, li 13/09/2022

Sample arrived on the 05/09/2022

Registration date 05/09/2022

**TEST REPORT nr. 22M02964-In-0**

CUSTOMER

**ACETFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**

SAMPLE 22M02964

**MATRIX: Fruit and vegetable concentrates, and  
by-products**

Description provided by Customer: GLASSA CON ABM IGP - CONTAMINAZIONE ALTA - VISCOSITA': ~ 3.000 CPS (T=0G) -  
CODICE: S20A-3000

Richiesta via Internet n° N0327/22 - 05/09/2022 13:57:34. - Campionamento eseguito da: Committente - Trasporto effettuato da:  
Committente  
Sample Condition on Receipt: refrigerated

ANALYSIS DESCRIPTION	RESULT	U	REC. %	UNIT OF MEASURE	LQ	LD	METHOD	ANALYSES BEGINNING DATE / ENDING DATE
Water activity (Aw) at 25 °C	0,940	± 0,005			0,150		06(S135) rev6 2021	06/09/2022 / 12/09/2022
<b>VINEGAR ANALYSIS</b>								
Reduction sugars (Reducing substances)	435,4	± 13,1		g/l	1,0		07(S240) 2021 Rev.0 - Titrimetric	07/09/2022 / 09/09/2022
Total Acidity expressed as acetic acid	2,96	± 0,06		g/100 ml	0,10		* ACIDITA 2014 Rev.0 - Titrimetric	07/09/2022 / 12/09/2022
Relative density at 20°C	1,183	± 0,001					07(S117) 2015 Rev.1 - Gravimetric	13/09/2022 / 13/09/2022
Alcoholic strength	0,10	± 0,01		% Vol	0,07		DM 12/03/1986 SO GU n 161 14/07/1986 Met V pag 95, OIV- MA-AS312-01 Met B R2021	07/09/2022 / 09/09/2022
Brookfield rotational viscosity	Brookfield Rotational Viscosity (T = 20 ° C, impeller R3)							
	30 rpm: 2058 cP (mPa.s) 20 rpm: 2488 cP (mPa.s) 12 rpm: 3178 cP (mPa.s)							
pH	2,96	± 0,04		unità	0,50		07(S77) 2012 Rev.1 - Potentiometric	07/09/2022 / 12/09/2022
<b>SUGARS HPAEC-PAD</b>								
Galactose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	07/09/2022 / 13/09/2022
Glucose, anhydrous (HPLC)	17,3	± 1,2		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	07/09/2022 / 13/09/2022
Sucrose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	07/09/2022 / 13/09/2022
Fructose, anhydrous (HPLC)	16,9	± 1,2		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	07/09/2022 / 13/09/2022
Lactose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	07/09/2022 / 13/09/2022
Maltose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	07/09/2022 / 13/09/2022
Citric acid (LCMS) [331]	89,5	± 14,6		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	07/09/2022 / 09/09/2022
Lactic Acid (LCMS) [331]	245	± 34		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	07/09/2022 / 09/09/2022
Malic acid (LCMS) [331]	604	± 74		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	07/09/2022 / 09/09/2022
Succinic acid (LCMS) [331]	221	± 31		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	07/09/2022 / 09/09/2022
Tartaric acid (LCMS) [331]	650	± 79		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	07/09/2022 / 09/09/2022
Ochratoxin A	< LQ			µg/kg	0,10		03(S130) 2022 Rev.13 - LC- MS/MS	07/09/2022 / 09/09/2022

**END TEST REPORT**

The original document is a PDF file with Digital Signature: 22M02964-In-0-S20A-3000 - T0 - UNIMORE-DigitalSignature.pdf

**Next page...**

MODENA, li 13/09/2022

Sample arrived on the 05/09/2022

Registration date 05/09/2022

## TEST REPORT nr. 22M02964-In-0

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**

**SAMPLE 22M02964**

**MATRIX: Fruit and vegetable concentrates, and by-products**

### Notes and method reference:

< LQ: = lower than Quantification Limit.

U: the reported uncertainty is the expanded uncertainty calculated using a coverage factor equal to 2 which gives a reliability of approximately 95%. Please note that results expressed as '<LQ' may not indicate the absence of the searched parameters in the sample.

MICROBIOLOGICAL TESTS: for food and environmental samples, the extended measurement uncertainty was estimated according to ISO 19036:2019 Standard and is based on a standard uncertainty multiplied by a coverage factor of K = 2, providing a confidence level of approximately 95%. The combined standard uncertainty was assumed to be equal to the standard deviation of intra-laboratory reproducibility. The results of the microbiological tests are calculated according to the ISO 7218: 2007 / Amd 1: 2013 Standard. If the results are reported as <4 (CFU/ml) or <40 (CFU/g), this means that the microorganisms are present in the sample but in amounts less than 4 CFU/ml or 40 CFU/g respectively. For microbiological analyses unless differently reported in the individual test methods, in case of analytical steps foreseen in non-activity days of the laboratory, provisions of the ISO 7218: 2007 / Amd.1 2013 Standard (points 11.2 and 10.2.5) or from specific test methods are applied. In the case of quantitative microbiological tests, these have been set up on a single plate according to ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise expressly requested by current regulations.

In the case of quantitative microbiological tests, these have been set up on a single plate in accordance with ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise explicitly required by current regulations.

For waters, the measurement uncertainty corresponds to the confidence interval calculated according to ISO 8199: 2018 or to the expanded measurement uncertainty estimated according to ISO 29201: 2012. The results are issued in accordance with ISO 8199: 2018. When the number of colonies detected is <3, the result is expressed as "Microorganisms present in the analyzed volume (N ° colonies detected <3 CFU - reference ISO 8199: 2018, paragraph 9.1.8.4.1)".

LQ: Quantification Limit. It is the lowest analyte concentration which can be detected at an acceptable precision (repeatability) and accuracy, under well defined conditions.

LD: Detection Limit. It is the lowest analyte concentration which can be detected but not necessarily quantified, under well defined conditions.

Conformity evaluation: values not complying with laws, decrees, national and EU regulations or specifications supplied by the customer are evaluated case by case, also taking into consideration the uncertainty of measure for each single test and the regulations on rounding-off of values, and pointed out when considered as non conform.

Rec %: Recovery % "+" means that the recovery has been applied to the result. The numeric results between brackets (..) after the expression <LQ are purely indicative of traces that cannot be exactly quantified.

In the case of sampling carried out by Neotron, the laboratory applies the Internal Operating Procedure code: NEOT-DIR/ 006/53.

The laboratory disclaims any responsibility for the information provided by the client reported in this Report which may influence the validity of the results.

Methods marked with an asterisk (\*) are not accredited by ACCREDIA (UNI CEI EN ISO/IEC 17025). The sampling activity is not included within the Scope of Accreditation of Neotron SPA

### NOTES OF PARAMETERS:

[331]: Extended uncertainty calculated following the equation of HORWITZ by using a coverage factor of 2, which guarantee a reliance level of 95%

TEST REPORT VALID FOR ALL LEGAL PURPOSES (Italian R.D. 1-3-1928 n°842 (article 16), – Italian Law 19-7-1957 n°679 articles 16 and 18, Italian Ministerial Decree 25-3-1986).

DATA and SAMPLE STORAGE: Test Reports, Raw data, chromatographic paths and instrumental reports are stored for 5 years. One control sample is stored for 2 months.

Data expressed in this test report refer only to the sample tested in the laboratory. The results reported in this Test Report refer to the sample as received. The description or any other reference concerning the sample are declared by the customer. This Test Report cannot be reproduced except in full. Partial reproductions must be authorized in writing by our laboratory.

THE LABORATORY DIRECTOR: DR. ANDREA RIZZO

THE CHEMIST AUTHORIZED TO SIGN THE TEST REPORTS: DR. MARCO MESCHIARI

Approved by Analysis Manager - laboratory LMAA-Nut  
Approved by Analysis Manager - laboratory ICS-HPLC  
Approved by Analysis Manager - laboratory LC-MICO  
Approved by Analysis Manager - laboratory LC-AN

Approved by Analysis Manager - laboratory FISICHE  
Approved by Analysis Manager - laboratory LBM-EL  
Approved by Analysis Manager - laboratory LMAA-Bro



MODENA, li 13/09/2022

Sample arrived on the 05/09/2022

Registration date 05/09/2022

**TEST REPORT nr. 22M02960-In-0**

CUSTOMER

**ACETFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**
**SAMPLE 22M02960****MATRIX: Fruit and vegetable concentrates, and by-products**

Description provided by Customer: GLASSA CON ABM IGP - CONTAMINAZIONE ALTA - VISCOSITA': ~ 2.000 CPS (T=20G) - CODICE: S20A-2000

Richiesta via Internet n° N0317/22 - 25/08/2022 09:11:14. - Campionamento eseguito da: Committente - Trasporto effettuato da: Committente

Sample Condition on Receipt: Room temperature

ANALYSIS DESCRIPTION	RESULT	U	REC. %	UNIT OF MEASURE	LQ	LD	METHOD	ANALYSES BEGINNING DATE / ENDING DATE
Water activity (Aw) at 25 °C	0,937	± 0,005			0,150		06(S135) rev6 2021	06/09/2022 / 12/09/2022
<b>VINEGAR ANALYSIS</b>								
Reduction sugars (Reducing substances)	455,0	± 13,7		g/l	1,0		07(S240) 2021 Rev.0 - Titrimetrico	07/09/2022 / 09/09/2022
Total Acidity expressed as acetic acid	2,79	± 0,05		g/100 ml	0,10		* ACIDITA 2014 Rev.0 - Titrimetrico	07/09/2022 / 12/09/2022
Relative density at 20°C	1,186	± 0,001					07(S117) 2015 Rev.1 - Gravimetric	13/09/2022 / 13/09/2022
Alcoholic strength	0,07	± 0,01		% Vol	0,07		DM 12/03/1986 SO GU n 161 14/07/1986 Met V pag 95, OIV-MA-AS312-01 Met B R2021	07/09/2022 / 09/09/2022
pH	2,93	± 0,04		unità	0,50		07(S77) 2012 Rev.1 - Potentiometric	07/09/2022 / 12/09/2022
Brookfield rotational viscosity	Brookfield Rotational Viscosity (T = 20 ° C, impeller R3)						* VISCOSITA 2018 Rev.1	07/09/2022 / 12/09/2022
	50 rpm: 1434 cP (mPa.s)							
	30 rpm: 1826 cP (mPa.s)							
	20 rpm: 2213 cP (mPa.s)							
<b>SUGARS HPAEC-PAD</b>								
Galactose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	07/09/2022 / 13/09/2022
Glucose, anhydrous (HPLC)	17,1	± 1,2		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	07/09/2022 / 13/09/2022
Sucrose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	07/09/2022 / 13/09/2022
Fructose, anhydrous (HPLC)	16,8	± 1,2		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	07/09/2022 / 13/09/2022
Lactose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	07/09/2022 / 13/09/2022
Maltose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	07/09/2022 / 13/09/2022
Citric acid (LCMS) [331]	102	± 16		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	07/09/2022 / 09/09/2022
Lactic Acid (LCMS) [331]	298	± 41		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	07/09/2022 / 09/09/2022
Malic acid (LCMS) [331]	663	± 80		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	07/09/2022 / 09/09/2022
Succinic acid (LCMS) [331]	248	± 35		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	07/09/2022 / 09/09/2022
Tartaric acid (LCMS) [331]	715	± 85		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	07/09/2022 / 09/09/2022
Ochratoxin A	< LQ			µg/kg	0,10		03(S130) 2022 Rev.13 - LC-MS/MS	07/09/2022 / 09/09/2022

**END TEST REPORT**

The original document is a PDF file with Digital Signature: 22M02960-In-0-S20A-2000 - T20 - UNIMORE-DigitalSignature.pdf

**Next page...**

MODENA, li 13/09/2022

Sample arrived on the 05/09/2022

Registration date 05/09/2022

## TEST REPORT nr. 22M02960-In-0

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**

**SAMPLE 22M02960**

**MATRIX: Fruit and vegetable concentrates, and by-products**

### Notes and method reference:

< LQ: = lower than Quantification Limit.

U: the reported uncertainty is the expanded uncertainty calculated using a coverage factor equal to 2 which gives a reliability of approximately 95%. Please note that results expressed as '<LQ' may not indicate the absence of the searched parameters in the sample.

MICROBIOLOGICAL TESTS: for food and environmental samples, the extended measurement uncertainty was estimated according to ISO 19036:2019 Standard and is based on a standard uncertainty multiplied by a coverage factor of  $K = 2$ , providing a confidence level of approximately 95%. The combined standard uncertainty was assumed to be equal to the standard deviation of intra-laboratory reproducibility. The results of the microbiological tests are calculated according to the ISO 7218: 2007 / Amd 1: 2013 Standard. If the results are reported as <4 (CFU/ml) or <40 (CFU/g), this means that the microorganisms are present in the sample but in amounts less than 4 CFU/ml or 40 CFU/g respectively. For microbiological analyses unless differently reported in the individual test methods, in case of analytical steps foreseen in non-activity days of the laboratory, provisions of the ISO 7218: 2007 / Amd.1 2013 Standard (points 11.2 and 10.2.5) or from specific test methods are applied. In the case of quantitative microbiological tests, these have been set up on a single plate according to ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise expressly requested by current regulations.

In the case of quantitative microbiological tests, these have been set up on a single plate in accordance with ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise explicitly required by current regulations.

For waters, the measurement uncertainty corresponds to the confidence interval calculated according to ISO 8199: 2018 or to the expanded measurement uncertainty estimated according to ISO 29201: 2012. The results are issued in accordance with ISO 8199: 2018. When the number of colonies detected is <3, the result is expressed as "Microorganisms present in the analyzed volume (N ° colonies detected <3 CFU - reference ISO 8199: 2018, paragraph 9.1.8.4.1)".

LQ: Quantification Limit. It is the lowest analyte concentration which can be detected at an acceptable precision (repeatability) and accuracy, under well defined conditions.

LD: Detection Limit. It is the lowest analyte concentration which can be detected but not necessarily quantified, under well defined conditions.

Conformity evaluation: values not complying with laws, decrees, national and EU regulations or specifications supplied by the customer are evaluated case by case, also taking into consideration the uncertainty of measure for each single test and the regulations on rounding-off of values, and pointed out when considered as non conform.

Rec %: Recovery % "+" means that the recovery has been applied to the result. The numeric results between brackets (..) after the expression <LQ are purely indicative of traces that cannot be exactly quantified.

In the case of sampling carried out by Neotron, the laboratory applies the Internal Operating Procedure code: NEOT-DIR/ 006/53.

The laboratory disclaims any responsibility for the information provided by the client reported in this Report which may influence the validity of the results.

Methods marked with an asterisk (\*) are not accredited by ACCREDIA (UNI CEI EN ISO/IEC 17025). The sampling activity is not included within the Scope of Accreditation of Neotron SPA

### NOTES OF PARAMETERS:

[331]: Extended uncertainty calculated following the equation of HORWITZ by using a coverage factor of 2, which guarantee a reliance level of 95%

TEST REPORT VALID FOR ALL LEGAL PURPOSES (Italian R.D. 1-3-1928 n°842 (article 16), - Italian Law 19-7-1957 n°679 articles 16 and 18, Italian Ministerial Decree 25-3-1986).

DATA and SAMPLE STORAGE: Test Reports, Raw data, chromatographic paths and instrumental reports are stored for 5 years. One control sample is stored for 2 months.

Data expressed in this test report refer only to the sample tested in the laboratory. The results reported in this Test Report refer to the sample as received. The description or any other reference concerning the sample are declared by the customer. This Test Report cannot be reproduced except in full. Partial reproductions must be authorized in writing by our laboratory.

THE LABORATORY DIRECTOR: DR. ANDREA RIZZO

THE CHEMIST AUTHORIZED TO SIGN THE TEST REPORTS: DR. MARCO MESCHIARI

Approved by Analysis Manager - laboratory LMAA-Nut  
Approved by Analysis Manager - laboratory ICS-HPLC  
Approved by Analysis Manager - laboratory LC-MICO  
Approved by Analysis Manager - laboratory LC-AN

Approved by Analysis Manager - laboratory FISICHE  
Approved by Analysis Manager - laboratory LBM-EL  
Approved by Analysis Manager - laboratory LMAA-Bro

**NEOTRON SpA** - With Sole Shareholder

Stradello Aggazzotti, 104

41126 MODENA - ITALY - Fiscal Code and VAT n° 03807840362

Tel: +39 059461711 - Fax: +39 059461777

www.neotron.it - neotron@neotron.it

Laboratorio Qualificato D.M. 26-2-87 Art. 4 - Legge 46/82 per la Ricerca Applicata e Innovazione Tecnologica.

Regione Emilia Romagna - AUTORIZZAZIONE Autocontrollo N° 008/MO/008

BNN-Monitoring Fruit and Vegetables Approved Laboratory

I-Monitoring EDEKAAG Fruit and Vegetables Registered Laboratory

GMP+ code: GMP051757

MODENA, li 09/09/2022

Sample arrived on the 25/08/2022  
Registration date 25/08/2022**TEST REPORT nr. 22L15876-In-0**

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO****SAMPLE 22L15876****MATRIX: Fruit and vegetable concentrates, and  
by-products**Description provided by Customer: GLASSA CON ABM IGP - CONTAMINAZIONE ALTA - VISCOSITA': ~ 2.000 CPS (T=10G) -  
CODICE: S20A-2000Richiesta via Internet n° N0307/22 - 23/08/2022 16:48:08. - Campionamento eseguito da: Committente - Trasporto effettuato da:  
Corriere  
Sample Condition on Receipt: 4°C

ANALYSIS DESCRIPTION	RESULT	U	REC. %	UNIT OF MEASURE	LQ	LD	METHOD	ANALYSES BEGINNING DATE / ENDING DATE
Water activity (Aw) at 25 °C	0,930	± 0,005			0,150		06(S135) rev6 2021	26/08/2022 / 26/08/2022
<b>VINEGAR ANALYSIS</b>								
Reduction sugars (Reducing substances)	447,5	± 13,4		g/l	1,0		07(S240) 2021 Rev.0 - Titrimetrico	29/08/2022 / 01/09/2022
Total Acidity expressed as acetic acid	2,84	± 0,06		g/100 ml	0,10		* ACIDITA 2014 Rev.0 - Titrimetrico	29/08/2022 / 01/09/2022
Relative density at 20°C	1,182	± 0,0003					07(S117) 2015 Rev.1 - Gravimetric	02/09/2022 / 02/09/2022
Alcoholic strength	0,09	± 0,01		% Vol	0,07		DM 12/03/1986 SO GU n 161 14/07/1986 Met V pag 95, OIV- MA-AS312-01 Met B R2021	29/08/2022 / 01/09/2022
Brookfield rotational viscosity	Brookfield Rotational Viscosity (T = 20 ° C, impeller R3) 50 rpm: 1440 cP (mPa.s) 30 rpm: 1802 cP (mPa.s) 20 rpm: 2154 cP (mPa.s)							
pH	2,93	± 0,04		unità	0,50		07(S77) 2012 Rev.1 - Potentiometric	29/08/2022 / 29/08/2022
<b>SUGARS HPAEC-PAD</b>								
Galactose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	29/08/2022 / 02/09/2022
Glucose, anhydrous (HPLC)	17,9	± 1,3		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	29/08/2022 / 02/09/2022
Sucrose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	29/08/2022 / 02/09/2022
Fructose, anhydrous (HPLC)	17,0	± 1,2		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	29/08/2022 / 02/09/2022
Lactose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	29/08/2022 / 02/09/2022
Maltose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	29/08/2022 / 02/09/2022
Citric acid (LCMS) [331]	108	± 17		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	29/08/2022 / 09/09/2022
Lactic Acid (LCMS) [331]	203	± 29		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	29/08/2022 / 09/09/2022
Malic acid (LCMS) [331]	640	± 77		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	29/08/2022 / 09/09/2022
Succinic acid (LCMS) [331]	197	± 28		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	29/08/2022 / 09/09/2022
Tartaric acid (LCMS) [331]	702	± 84		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	29/08/2022 / 09/09/2022
Ochratoxin A	< LQ			µg/kg	0,10		03(S130) 2022 Rev.13 - LC- MS/MS	29/08/2022 / 31/08/2022

**END TEST REPORT**

The original document is a PDF file with Digital Signature: 22L15876-In-0-S20A-2000 - T10 - UNIMORE-DigitalSignature.pdf

**Next page...**

MODENA, li 09/09/2022

Sample arrived on the 25/08/2022

Registration date 25/08/2022

## TEST REPORT nr. 22L15876-In-0

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**

**SAMPLE 22L15876**

**MATRIX: Fruit and vegetable concentrates, and by-products**

### Notes and method reference:

< LQ: = lower than Quantification Limit.

U: the reported uncertainty is the expanded uncertainty calculated using a coverage factor equal to 2 which gives a reliability of approximately 95%. Please note that results expressed as '<LQ' may not indicate the absence of the searched parameters in the sample.

MICROBIOLOGICAL TESTS: for food and environmental samples, the extended measurement uncertainty was estimated according to ISO 19036:2019 Standard and is based on a standard uncertainty multiplied by a coverage factor of K = 2, providing a confidence level of approximately 95%. The combined standard uncertainty was assumed to be equal to the standard deviation of intra-laboratory reproducibility. The results of the microbiological tests are calculated according to the ISO 7218: 2007 / Amd 1: 2013 Standard. If the results are reported as <4 (CFU/ml) or <40 (CFU/g), this means that the microorganisms are present in the sample but in amounts less than 4 CFU/ml or 40 CFU/g respectively. For microbiological analyses unless differently reported in the individual test methods, in case of analytical steps foreseen in non-activity days of the laboratory, provisions of the ISO 7218: 2007 / Amd.1 2013 Standard (points 11.2 and 10.2.5) or from specific test methods are applied. In the case of quantitative microbiological tests, these have been set up on a single plate according to ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise expressly requested by current regulations.

In the case of quantitative microbiological tests, these have been set up on a single plate in accordance with ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise explicitly required by current regulations.

For waters, the measurement uncertainty corresponds to the confidence interval calculated according to ISO 8199: 2018 or to the expanded measurement uncertainty estimated according to ISO 29201: 2012. The results are issued in accordance with ISO 8199: 2018. When the number of colonies detected is <3, the result is expressed as "Microorganisms present in the analyzed volume (N ° colonies detected <3 CFU - reference ISO 8199: 2018, paragraph 9.1.8.4.1)".

LQ: Quantification Limit. It is the lowest analyte concentration which can be detected at an acceptable precision (repeatability) and accuracy, under well defined conditions.

LD: Detection Limit. It is the lowest analyte concentration which can be detected but not necessarily quantified, under well defined conditions.

Conformity evaluation: values not complying with laws, decrees, national and EU regulations or specifications supplied by the customer are evaluated case by case, also taking into consideration the uncertainty of measure for each single test and the regulations on rounding-off of values, and pointed out when considered as non conform.

Rec %: Recovery % "+" means that the recovery has been applied to the result. The numeric results between brackets (..) after the expression <LQ are purely indicative of traces that cannot be exactly quantified.

In the case of sampling carried out by Neotron, the laboratory applies the Internal Operating Procedure code: NEOT-DIR/ 006/53.

The laboratory disclaims any responsibility for the information provided by the client reported in this Report which may influence the validity of the results.

Methods marked with an asterisk (\*) are not accredited by ACCREDIA (UNI CEI EN ISO/IEC 17025). The sampling activity is not included within the Scope of Accreditation of Neotron SPA

### NOTES OF PARAMETERS:

[331]: Extended uncertainty calculated following the equation of HORWITZ by using a coverage factor of 2, which guarantee a reliance level of 95%

TEST REPORT VALID FOR ALL LEGAL PURPOSES (Italian R.D. 1-3-1928 n°842 (article 16), - Italian Law 19-7-1957 n°679 articles 16 and 18, Italian Ministerial Decree 25-3-1986).

DATA and SAMPLE STORAGE: Test Reports, Raw data, chromatographic paths and instrumental reports are stored for 5 years. One control sample is stored for 2 months.

Data expressed in this test report refer only to the sample tested in the laboratory. The results reported in this Test Report refer to the sample as received. The description or any other reference concerning the sample are declared by the customer. This Test Report cannot be reproduced except in full. Partial reproductions must be authorized in writing by our laboratory.

THE LABORATORY DIRECTOR: DR. ANDREA RIZZO

THE CHEMIST AUTHORIZED TO SIGN THE TEST REPORTS: DR. BARBARA MALAGOLI

Approved by Analysis Manager - laboratory LMAA-Nut

Approved by Analysis Manager - laboratory ICS-HPLC

Approved by Analysis Manager - laboratory LC-MICO

Approved by Analysis Manager - laboratory LC-AN

Approved by Analysis Manager - laboratory FISICHE

Approved by Analysis Manager - laboratory LBM-EL

Approved by Analysis Manager - laboratory LMAA-Bro

NEOTRON SpA - With Sole Shareholder

Stradello Aggazzotti, 104

41126 MODENA - ITALY - Fiscal Code and VAT n° 03807840362

Tel: +39 059461711 - Fax: +39 059461777

www.neotron.it - neotron@neotron.it

Laboratorio Qualificato D.M. 26-2-87 Art. 4 - Legge 46/82 per la Ricerca Applicata e Innovazione Tecnologica.

Regione Emilia Romagna - AUTORIZZAZIONE Autocontrollo N° 008/MO/008

BNN-Monitoring Fruit and Vegetables Approved Laboratory

I-Monitoring EDEKAAG Fruit and Vegetables Registered Laboratory

GMP+ code: GMP051757

MODENA, li 09/09/2022

Sample arrived on the 25/08/2022

Registration date 25/08/2022

**TEST REPORT nr. 22L15874-In-0**

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**

SAMPLE 22L15874

**MATRIX: Fruit and vegetable concentrates, and  
by-products**

Description provided by Customer: GLASSA CON ABM IGP - CONTAMINAZIONE ALTA - VISCOSITA': ~ 2.000 CPS (T=0G) -  
CODICE: S20A-2000

Richiesta via Internet n° N0304/22 - 23/08/2022 16:48:07. - Campionamento eseguito da: Committente - Trasporto effettuato da:  
Corriere  
Sample Condition on Receipt: 4°C

ANALYSIS DESCRIPTION	RESULT	U	REC. %	UNIT OF MEASURE	LQ	LD	METHOD	ANALYSES BEGINNING DATE / ENDING DATE
Water activity (Aw) at 25 °C	0,930	± 0,005			0,150		06(S135) rev6 2021	26/08/2022 / 26/08/2022
<b>VINEGAR ANALYSIS</b>								
Reduction sugars (Reducing substances)	409,5	± 12,3		g/l	1,0		07(S240) 2021 Rev.0 - Titrimetrico	29/08/2022 / 01/09/2022
Total Acidity expressed as acetic acid	2,76	± 0,05		g/100 ml	0,10		* ACIDITA 2014 Rev.0 - Titrimetrico	29/08/2022 / 01/09/2022
Relative density at 20°C	1,164	± 0,00027					07(S117) 2015 Rev.1 - Gravimetric	02/09/2022 / 02/09/2022
Alcoholic strength	0,10	± 0,01		% Vol	0,07		DM 12/03/1986 SO GU n 161 14/07/1986 Met V pag 95, OIV- MA-AS312-01 Met B R2021	29/08/2022 / 01/09/2022
Brookfield rotational viscosity	Brookfield Rotational Viscosity (T = 20 ° C, impeller R3)							
	50 rpm: 1563 cP (mPa.s) 30 rpm: 1721 cP (mPa.s) 20 rpm: 2040 cP (mPa.s)							
pH	2,99	± 0,04		unità	0,50		07(S77) 2012 Rev.1 - Potentiometric	29/08/2022 / 29/08/2022
<b>SUGARS HPAEC-PAD</b>								
Galactose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	29/08/2022 / 02/09/2022
Glucose, anhydrous (HPLC)	18,1	± 1,3		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	29/08/2022 / 02/09/2022
Sucrose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	29/08/2022 / 02/09/2022
Fructose, anhydrous (HPLC)	16,5	± 1,2		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	29/08/2022 / 02/09/2022
Lactose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	29/08/2022 / 02/09/2022
Maltose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	29/08/2022 / 02/09/2022
Citric acid (LCMS) [331]	103	± 16		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	29/08/2022 / 09/09/2022
Lactic Acid (LCMS) [331]	214	± 31		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	29/08/2022 / 09/09/2022
Malic acid (LCMS) [331]	558	± 69		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	29/08/2022 / 09/09/2022
Succinic acid (LCMS) [331]	177	± 26		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	29/08/2022 / 09/09/2022
Tartaric acid (LCMS) [331]	623	± 75		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	29/08/2022 / 09/09/2022
Ochratoxin A	< LQ			µg/kg	0,10		03(S130) 2022 Rev.13 - LC- MS/MS	29/08/2022 / 31/08/2022

**END TEST REPORT**

The original document is a PDF file with Digital Signature: 22L15874-In-0-S20A-2000 - T0 - UNIMORE-DigitalSignature.pdf

**Next page...**

MODENA, li 09/09/2022

Sample arrived on the 25/08/2022  
Registration date 25/08/2022

## TEST REPORT nr. 22L15874-In-0

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**

**SAMPLE 22L15874**

**MATRIX: Fruit and vegetable concentrates, and by-products**

### Notes and method reference:

< LQ: = lower than Quantification Limit.

U: the reported uncertainty is the expanded uncertainty calculated using a coverage factor equal to 2 which gives a reliability of approximately 95%. Please note that results expressed as '<LQ' may not indicate the absence of the searched parameters in the sample.

MICROBIOLOGICAL TESTS: for food and environmental samples, the extended measurement uncertainty was estimated according to ISO 19036:2019 Standard and is based on a standard uncertainty multiplied by a coverage factor of K = 2, providing a confidence level of approximately 95%. The combined standard uncertainty was assumed to be equal to the standard deviation of intra-laboratory reproducibility. The results of the microbiological tests are calculated according to the ISO 7218: 2007 / Amd 1: 2013 Standard. If the results are reported as <4 (CFU/ml) or <40 (CFU/g), this means that the microorganisms are present in the sample but in amounts less than 4 CFU/ml or 40 CFU/g respectively. For microbiological analyses unless differently reported in the individual test methods, in case of analytical steps foreseen in non-activity days of the laboratory, provisions of the ISO 7218: 2007 / Amd.1 2013 Standard (points 11.2 and 10.2.5) or from specific test methods are applied. In the case of quantitative microbiological tests, these have been set up on a single plate according to ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise expressly requested by current regulations.

In the case of quantitative microbiological tests, these have been set up on a single plate in accordance with ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise explicitly required by current regulations.

For waters, the measurement uncertainty corresponds to the confidence interval calculated according to ISO 8199: 2018 or to the expanded measurement uncertainty estimated according to ISO 29201: 2012. The results are issued in accordance with ISO 8199: 2018. When the number of colonies detected is <3, the result is expressed as "Microorganisms present in the analyzed volume (N ° colonies detected <3 CFU - reference ISO 8199: 2018, paragraph 9.1.8.4.1)".

LQ: Quantification Limit. It is the lowest analyte concentration which can be detected at an acceptable precision (repeatability) and accuracy, under well defined conditions.

LD: Detection Limit. It is the lowest analyte concentration which can be detected but not necessarily quantified, under well defined conditions.

Conformity evaluation: values not complying with laws, decrees, national and EU regulations or specifications supplied by the customer are evaluated case by case, also taking into consideration the uncertainty of measure for each single test and the regulations on rounding-off of values, and pointed out when considered as non conform.

Rec %: Recovery % "+" means that the recovery has been applied to the result. The numeric results between brackets (..) after the expression <LQ are purely indicative of traces that cannot be exactly quantified.

In the case of sampling carried out by Neotron, the laboratory applies the Internal Operating Procedure code: NEOT-DIR/ 006/53.

The laboratory disclaims any responsibility for the information provided by the client reported in this Report which may influence the validity of the results.

Methods marked with an asterisk (\*) are not accredited by ACCREDIA (UNI CEI EN ISO/IEC 17025). The sampling activity is not included within the Scope of Accreditation of Neotron SPA

### NOTES OF PARAMETERS:

[331]: Extended uncertainty calculated following the equation of HORWITZ by using a coverage factor of 2, which guarantee a reliance level of 95%

TEST REPORT VALID FOR ALL LEGAL PURPOSES (Italian R.D. 1-3-1928 n°842 (article 16), - Italian Law 19-7-1957 n°679 articles 16 and 18, Italian Ministerial Decree 25-3-1986).

DATA and SAMPLE STORAGE: Test Reports, Raw data, chromatographic paths and instrumental reports are stored for 5 years. One control sample is stored for 2 months.

Data expressed in this test report refer only to the sample tested in the laboratory. The results reported in this Test Report refer to the sample as received. The description or any other reference concerning the sample are declared by the customer. This Test Report cannot be reproduced except in full. Partial reproductions must be authorized in writing by our laboratory.

THE LABORATORY DIRECTOR: DR. ANDREA RIZZO

THE CHEMIST AUTHORIZED TO SIGN THE TEST REPORTS: DR. BARBARA MALAGOLI

Approved by Analysis Manager - laboratory LMAA-Nut  
Approved by Analysis Manager - laboratory ICS-HPLC  
Approved by Analysis Manager - laboratory LC-MICO  
Approved by Analysis Manager - laboratory LC-AN

Approved by Analysis Manager - laboratory FISICHE  
Approved by Analysis Manager - laboratory LBM-EL  
Approved by Analysis Manager - laboratory LMAA-Bro

**NEOTRON SpA** - With Sole Shareholder

Stradello Aggazzotti, 104

41126 MODENA - ITALY - Fiscal Code and VAT n° 03807840362

Tel: +39 059461711 - Fax: +39 059461777

www.neotron.it - neotron@neotron.it

Laboratorio Qualificato D.M. 26-2-87 Art. 4 - Legge 46/82 per la Ricerca Applicata e Innovazione Tecnologica.

Regione Emilia Romagna - AUTORIZZAZIONE Autocontrollo N° 008/MO/008

BNN-Monitoring Fruit and Vegetables Approved Laboratory

I-Monitoring EDEKAAG Fruit and Vegetables Registered Laboratory

GMP+ code: GMP051757

MODENA, li 23/08/2022

Sample arrived on the 05/08/2022  
Registration date 08/08/2022

**TEST REPORT nr. 22L05145-In-0**

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**

**SAMPLE 22L05145**

**MATRIX: Fruit and vegetable concentrates, and by-products**

Description provided by Customer: GLASSA CON ABM IGP - CONTAMINAZIONE ALTA - VISCOSITA': ~ 1.000 CPS (T=20G) - CODICE: S20A-1000

Richiesta via Internet n° N0259/22 - 20/07/2022 12:56:56. - Campionamento eseguito da: Committente - Trasporto effettuato da: Corriere  
Sample Condition on Receipt: 20°C

ANALYSIS DESCRIPTION	RESULT	U	REC. %	UNIT OF MEASURE	LQ	LD	METHOD	ANALYSES BEGINNING DATE / ENDING DATE
Water activity (Aw) at 25 °C	0,930	± 0,005			0,150		06(S135) rev6 2021	08/08/2022 / 08/08/2022
<b>VINEGAR ANALYSIS</b>								
Reduction sugars (Reducing substances)	426,2	± 12,8		g/l	1,0		07(S240) 2021 Rev.0 - Titrimetrico	09/08/2022 / 11/08/2022
Total Acidity expressed as acetic acid	2,71	± 0,05		g/100 ml	0,10		* ACIDITA 2014 Rev.0 - Titrimetrico	09/08/2022 / 11/08/2022
Relative density at 20°C	1,19302	± 0,00027					DM 12/03/1986 SO GU n 161 14/07/1986 Met VI pag 95 p.to 4, OIV-MA-AS2-01A Met B R2021 - densimetry	09/08/2022 / 11/08/2022
Alcoholic strength	0,10	± 0,01		% Vol	0,07		DM 12/03/1986 SO GU n 161 14/07/1986 Met V pag 95, OIV-MA-AS312-01 Met B R2021	09/08/2022 / 11/08/2022
Brookfield rotational viscosity	Brookfield Rotational Viscosity (T = 20 ° C, impeller R2)						* VISCOSITA 2018 Rev.1	09/08/2022 / 12/08/2022
	30 rpm: 809 cP (mPa.s) 20 rpm: 933 cP (mPa.s) 12 rpm: 1110 cP (mPa.s)							
pH	2,92	± 0,04		unità	0,50		07(S77) 2012 Rev.1 - Potentiometric	09/08/2022 / 10/08/2022
<b>SUGARS HPAEC-PAD</b>								
Galactose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	09/08/2022 / 16/08/2022
Glucose, anhydrous (HPLC)	18,1	± 1,3		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	09/08/2022 / 16/08/2022
Sucrose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	09/08/2022 / 16/08/2022
Fructose, anhydrous (HPLC)	18,8	± 1,3		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	09/08/2022 / 16/08/2022
Lactose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	09/08/2022 / 16/08/2022
Maltose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC-PAD	09/08/2022 / 16/08/2022
Citric acid (LCMS) [331]	66,3	± 11,3		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	09/08/2022 / 16/08/2022
Lactic Acid (LCMS) [331]	370	± 48		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	09/08/2022 / 16/08/2022
Malic acid (LCMS) [331]	444	± 57		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	09/08/2022 / 16/08/2022
Succinic acid (LCMS) [331]	156	± 23		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	09/08/2022 / 16/08/2022
Tartaric acid (LCMS) [331]	477	± 60		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	09/08/2022 / 16/08/2022
Ochratoxin A	< LQ			µg/kg	0,10		03(S130) 2022 Rev.12 - LC-MS/MS	09/08/2022 / 11/08/2022

END TEST REPORT

The original document is a PDF file with Digital Signature: 22L05145-In-0-S20A-1000 - T20 - UNIMORE-DigitalSignature.pdf

MODENA, li 23/08/2022

Sample arrived on the 05/08/2022

Registration date 08/08/2022

**TEST REPORT nr. 22L05145-In-0**

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**

SAMPLE 22L05145

**MATRIX: Fruit and vegetable concentrates, and  
by-products**

## Notes and method reference:

&lt; LQ: = lower than Quantification Limit.

U: the reported uncertainty is the expanded uncertainty calculated using a coverage factor equal to 2 which gives a reliability of approximately 95%. Please note that results expressed as '<LQ' may not indicate the absence of the searched parameters in the sample.

MICROBIOLOGICAL TESTS: for food and environmental samples, the extended measurement uncertainty was estimated according to ISO 19036:2019 Standard and is based on a standard uncertainty multiplied by a coverage factor of K = 2, providing a confidence level of approximately 95%. The combined standard uncertainty was assumed to be equal to the standard deviation of intra-laboratory reproducibility. The results of the microbiological tests are calculated according to the ISO 7218: 2007 / Amd 1: 2013 Standard. If the results are reported as <4 (CFU/ml) or <40 (CFU/g), this means that the microorganisms are present in the sample but in amounts less than 4 CFU/ml or 40 CFU/g respectively. For microbiological analyses unless differently reported in the individual test methods, in case of analytical steps foreseen in non-activity days of the laboratory, provisions of the ISO 7218: 2007 / Amd.1 2013 Standard (points 11.2 and 10.2.5) or from specific test methods are applied. In the case of quantitative microbiological tests, these have been set up on a single plate according to ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise expressly requested by current regulations.

In the case of quantitative microbiological tests, these have been set up on a single plate in accordance with ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise explicitly required by current regulations.

For waters, the measurement uncertainty corresponds to the confidence interval calculated according to ISO 8199: 2018 or to the expanded measurement uncertainty estimated according to ISO 29201: 2012. The results are issued in accordance with ISO 8199: 2018. When the number of colonies detected is <3, the result is expressed as "Microorganisms present in the analyzed volume (N ° colonies detected <3 CFU - reference ISO 8199: 2018, paragraph 9.1.8.4.1)".

LQ: Quantification Limit. It is the lowest analyte concentration which can be detected at an acceptable precision (repeatability) and accuracy, under well defined conditions.

LD: Detection Limit. It is the lowest analyte concentration which can be detected but not necessarily quantified, under well defined conditions.

Conformity evaluation: values not complying with laws, decrees, national and EU regulations or specifications supplied by the customer are evaluated case by case, also taking into consideration the uncertainty of measure for each single test and the regulations on rounding-off of values, and pointed out when considered as non conform.

Rec %: Recovery % "+" means that the recovery has been applied to the result. The numeric results between brackets (..) after the expression <LQ are purely indicative of traces that cannot be exactly quantified.

In the case of sampling carried out by Neotron, the laboratory applies the Internal Operating Procedure code: NEOT-DIR/ 006/53.

The laboratory disclaims any responsibility for the information provided by the client reported in this Report which may influence the validity of the results.

Methods marked with an asterisk (\*) are not accredited by ACCREDIA (UNI CEI EN ISO/IEC 17025). The sampling activity is not included within the Scope of Accreditation of Neotron SPA

## NOTES OF PARAMETERS:

[331]: Extended uncertainty calculated following the equation of HORWITZ by using a coverage factor of 2, which guarantee a reliance level of 95%

TEST REPORT VALID FOR ALL LEGAL PURPOSES (Italian R.D. 1-3-1928 n°842 (article 16), - Italian Law 19-7-1957 n°679 articles 16 and 18, Italian Ministerial Decree 25-3-1986).

DATA and SAMPLE STORAGE: Test Reports, Raw data, chromatographic paths and instrumental reports are stored for 5 years. One control sample is stored for 2 months.

Data expressed in this test report refer only to the sample tested in the laboratory. The results reported in this Test Report refer to the sample as received. The description or any other reference concerning the sample are declared by the customer. This Test Report cannot be reproduced except in full. Partial reproductions must be authorized in writing by our laboratory.

THE LABORATORY DIRECTOR: DR. ANDREA RIZZO

THE CHEMIST AUTHORIZED TO SIGN THE TEST REPORTS: DR. MARCO MESCHIARI - N. 514 ORDINE DEI CHIMICI MODENA

Approved by Analysis Manager - laboratory LMAA-Nut

Approved by Analysis Manager - laboratory ICS-HPLC

Approved by Analysis Manager - laboratory LC-MICO

Approved by Analysis Manager - laboratory LC-AN

Approved by Analysis Manager - laboratory FISICHE

Approved by Analysis Manager - laboratory LBM-EL

Approved by Analysis Manager - laboratory LMAA-Bro

NEOTRON SpA - With Sole Shareholder

Stradello Aggazzotti, 104

41126 MODENA - ITALY - Fiscal Code and VAT n° 03807840362

Tel: +39 059461711 - Fax: +39 059461777

www.neotron.it - neotron@neotron.it

Laboratorio Qualificato D.M. 26-2-87 Art. 4 - Legge 46/82 per la Ricerca Applicata e Innovazione Tecnologica.

Regione Emilia Romagna - AUTORIZZAZIONE Autocontrollo N° 008/MO/008

BNN-Monitoring Fruit and Vegetables Approved Laboratory

I-Monitoring EDEKAAG Fruit and Vegetables Registered Laboratory

GMP+ code: GMP051757



MODENA, li 04/08/2022

Sample arrived on the 26/07/2022

Registration date 26/07/2022

**TEST REPORT nr. 22G17283-In-0**

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**

SAMPLE 22G17283

**MATRIX: Fruit and vegetable concentrates, and  
by-products**

Description provided by Customer: GLASSA CON ABM IGP - CONTAMINAZIONE ALTA - VISCOSITA': ~ 1.000 CPS (T=10G) -  
CODICE: S20A-1000

Richiesta via Internet n° N0258/22 - 20/07/2022 12:56:55. - Campionamento eseguito da: Committente - Trasporto effettuato da:  
Committente  
Sample Condition on Receipt: 20°C

ANALYSIS DESCRIPTION	RESULT	U	REC. %	UNIT OF MEASURE	LQ	LD	METHOD	ANALYSES BEGINNING DATE / ENDING DATE
Water activity (Aw) at 25 °C	0,941	± 0,005			0,150		06(S135) rev6 2021	27/07/2022 / 29/07/2022
<b>VINEGAR ANALYSIS</b>								
Reduction sugars (Reducing substances)	452,5	± 13,6		g/l	1,0		07(S240) 2021 Rev.0 - Titrimetrico	28/07/2022 / 02/08/2022
Total Acidity expressed as acetic acid	2,80	± 0,05		g/100 ml	0,10		* ACIDITA 2014 Rev.0 - Titrimetrico	28/07/2022 / 01/08/2022
Relative density at 20°C	1,18987	± 0,00027					DM 12/03/1986 SO GU n 161 14/07/1986 Met VI pag 95 p.to 4, OIV-MA-AS2-01A Met B R2021 - densimetry	28/07/2022 / 01/08/2022
Alcoholic strength	0,09	± 0,011		% Vol	0,07		DM 12/03/1986 SO GU n 161 14/07/1986 Met V pag 95, OIV- MA-AS312-01 Met B R2021	28/07/2022 / 01/08/2022
Brookfield rotational viscosity	Brookfield Rotational Viscosity (T = 20 ° C, impeller R2)						* VISCOSITA 2018 Rev.1	28/07/2022 / 04/08/2022
	50 rpm: 564 cP (mPa.s)							
	30 rpm: 665 cP (mPa.s)							
	20 rpm: 755 cP (mPa.s)							
pH	2,92	± 0,04		unità	0,50		07(S77) 2012 Rev.1 - Potentiometric	28/07/2022 / 01/08/2022
<b>SUGARS HPAEC-PAD</b>								
Galactose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	28/07/2022 / 02/08/2022
Glucose, anhydrous (HPLC)	17,7	± 1,2		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	28/07/2022 / 02/08/2022
Sucrose, anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	28/07/2022 / 02/08/2022
Fructose, anhydrous (HPLC)	19,4	± 1,4		g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	28/07/2022 / 02/08/2022
Lactose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	28/07/2022 / 02/08/2022
Maltose anhydrous (HPLC)	< LQ			g/100 g	0,10		07(S239) Rev.0 2021 - HPLC- PAD	28/07/2022 / 02/08/2022
Citric acid (LCMS) [331]	98,2	± 15,7		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	28/07/2022 / 04/08/2022
Lactic Acid (LCMS) [331]	335	± 45		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	28/07/2022 / 04/08/2022
Malic acid (LCMS) [331]	500	± 63		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	28/07/2022 / 04/08/2022
Succinic acid (LCMS) [331]	195	± 28		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	28/07/2022 / 04/08/2022
Tartaric acid (LCMS) [331]	534	± 66		mg/kg	5,0		* AC-ORG Rev.0 2015 - LC-MS/MS	28/07/2022 / 04/08/2022
Ochratoxin A	< LQ			µg/kg	0,10		03(S130) 2022 Rev.12 - LC- MS/MS	28/07/2022 / 02/08/2022

**END TEST REPORT**

The original document is a PDF file with Digital Signature: 22G17283-In-0-S20A-1000 - T10 - UNIMORE-DigitalSignature.pdf

MODENA, li 04/08/2022

Sample arrived on the 26/07/2022

Registration date 26/07/2022

**TEST REPORT nr. 22G17283-In-0**

CUSTOMER

**ACETIFICIO CARANDINI EMILIO S.p.A.**  
**Via per Formigine, 54/A**  
**41051 CASTELNUOVO RANGONE MO**

SAMPLE 22G17283

**MATRIX: Fruit and vegetable concentrates, and  
by-products**

## Notes and method reference:

&lt; LQ: = lower than Quantification Limit.

U: the reported uncertainty is the expanded uncertainty calculated using a coverage factor equal to 2 which gives a reliability of approximately 95%. Please note that results expressed as '<LQ' may not indicate the absence of the searched parameters in the sample.

MICROBIOLOGICAL TESTS: for food and environmental samples, the extended measurement uncertainty was estimated according to ISO 19036:2019 Standard and is based on a standard uncertainty multiplied by a coverage factor of K = 2, providing a confidence level of approximately 95%. The combined standard uncertainty was assumed to be equal to the standard deviation of intra-laboratory reproducibility. The results of the microbiological tests are calculated according to the ISO 7218: 2007 / Amd 1: 2013 Standard. If the results are reported as <4 (CFU/ml) or <40 (CFU/g), this means that the microorganisms are present in the sample but in amounts less than 4 CFU/ml or 40 CFU/g respectively. For microbiological analyses unless differently reported in the individual test methods, in case of analytical steps foreseen in non-activity days of the laboratory, provisions of the ISO 7218: 2007 / Amd.1 2013 Standard (points 11.2 and 10.2.5) or from specific test methods are applied. In the case of quantitative microbiological tests, these have been set up on a single plate according to ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise expressly requested by current regulations.

In the case of quantitative microbiological tests, these have been set up on a single plate in accordance with ISO 7218:2007/Amd.1 2013 par. 10.2.2 unless otherwise explicitly required by current regulations.

For waters, the measurement uncertainty corresponds to the confidence interval calculated according to ISO 8199: 2018 or to the expanded measurement uncertainty estimated according to ISO 29201: 2012. The results are issued in accordance with ISO 8199: 2018. When the number of colonies detected is <3, the result is expressed as "Microorganisms present in the analyzed volume (N ° colonies detected <3 CFU - reference ISO 8199: 2018, paragraph 9.1.8.4.1)".

LQ: Quantification Limit. It is the lowest analyte concentration which can be detected at an acceptable precision (repeatability) and accuracy, under well defined conditions.

LD: Detection Limit. It is the lowest analyte concentration which can be detected but not necessarily quantified, under well defined conditions.

Conformity evaluation: values not complying with laws, decrees, national and EU regulations or specifications supplied by the customer are evaluated case by case, also taking into consideration the uncertainty of measure for each single test and the regulations on rounding-off of values, and pointed out when considered as non conform.

Rec %: Recovery % "+" means that the recovery has been applied to the result. The numeric results between brackets (..) after the expression <LQ are purely indicative of traces that cannot be exactly quantified.

In the case of sampling carried out by Neotron, the laboratory applies the Internal Operating Procedure code: NEOT-DIR/ 006/53.

The laboratory disclaims any responsibility for the information provided by the client reported in this Report which may influence the validity of the results.

Methods marked with an asterisk (\*) are not accredited by ACCREDIA (UNI CEI EN ISO/IEC 17025). The sampling activity is not included within the Scope of Accreditation of Neotron SPA

## NOTES OF PARAMETERS:

[331]: Extended uncertainty calculated following the equation of HORWITZ by using a coverage factor of 2, which guarantee a reliance level of 95%

TEST REPORT VALID FOR ALL LEGAL PURPOSES (Italian R.D. 1-3-1928 n°842 (article 16), - Italian Law 19-7-1957 n°679 articles 16 and 18, Italian Ministerial Decree 25-3-1986).

DATA and SAMPLE STORAGE: Test Reports, Raw data, chromatographic paths and instrumental reports are stored for 5 years. One control sample is stored for 2 months.

Data expressed in this test report refer only to the sample tested in the laboratory. The results reported in this Test Report refer to the sample as received. The description or any other reference concerning the sample are declared by the customer. This Test Report cannot be reproduced except in full. Partial reproductions must be authorized in writing by our laboratory.

THE LABORATORY DIRECTOR: DR. ANDREA RIZZO

THE CHEMIST AUTHORIZED TO SIGN THE TEST REPORTS: DR. MARCO MESCHIARI - N. 514 ORDINE DEI CHIMICI MODENA

Approved by Analysis Manager - laboratory LMAA-Nut

Approved by Analysis Manager - laboratory ICS-HPLC

Approved by Analysis Manager - laboratory LC-MICO

Approved by Analysis Manager - laboratory LC-AN

Approved by Analysis Manager - laboratory FISICHE

Approved by Analysis Manager - laboratory LBM-EL

Approved by Analysis Manager - laboratory LMAA-Bro

NEOTRON SpA - With Sole Shareholder

Stradello Aggazzotti, 104

41126 MODENA - ITALY - Fiscal Code and VAT n° 03807840362

Tel: +39 059461711 - Fax: +39 059461777

www.neotron.it - neotron@neotron.it

Laboratorio Qualificato D.M. 26-2-87 Art. 4 - Legge 46/82 per la Ricerca Applicata e Innovazione Tecnologica.

Regione Emilia Romagna - AUTORIZZAZIONE Autocontrollo N° 008/MO/008

BNN-Monitoring Fruit and Vegetables Approved Laboratory

I-Monitoring EDEKAAG Fruit and Vegetables Registered Laboratory

GMP+ code: GMP051757