Exploring the effects of hyperbaric storage on the optical, structural, mechanical and diffusional properties of food packaging materials

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ABSTRACT

The effect of hyperbaric storage (HS) on food packaging materials was evaluated. PA/PE, PP/EVOH/PE, PET and PLA pouches filled with hydroethanolic simulant (D1) were stored at 0.1 and 200 MPa for up to 35 days and analyzed for optical, structural, mechanical and diffusional properties. HS weakened PLA seals, which easily failed after 7 days releasing the simulant. Both PET and PLA films swelled during HS, reducing PET physical ageing and PLA crystallinity. These structural effects caused PET and PLA mechanical properties to vary during HS, and a slight WVTR increase in PLA. Optical, structural and mechanical properties of multi-material films did not change upon HS. Nevertheless, both PA/PE and PP/EVOH/PE released critical amounts of adhesives after 7 and 35 days under pressure, respectively. Results indicate the critical role of the packaging material of foods intended for HS, and the need for its careful selection in future studies on the topic.

1. Introduction

Hyperbaric storage (HS) is a novel, non-thermal food preservation technology based on the application of moderate hydrostatic pressure (up to 250 MPa) (Santos et al., 2021). From an operational perspective, HS is a simple process comprised by only three steps (Fig. 1) (Basso & Manzocco, 2022).

Food items are first sealed inside flexible plastic pouches with minimal headspace. Then, the packaged items are transferred inside a steel vessel, which is pressurized by pumping a fluid (*e.g.*, water, propylene glycol). When the desired pressure is achieved in the vessel, the pump is switched off and these conditions are maintained for the desired time, which can last up to several months (Lemos, Ribeiro, Delgadillo, & Saraiva, 2020). The key feature of HS is that pressurized storage conditions can be applied at room temperature for long time with very low energetic cost (Bermejo-Prada, Colmant, Otero, & Guignon, 2017). For this reason, during the last decade, HS has been increasingly investigated as a sustainable alternative to refrigeration, showing also potential for non-thermal pasteurization and improvement of food techno-functionality (Basso, Manzocco, Maifreni, & Nicoli, 2021; Basso, Maifreni et al., 2022; Duarte, Pinto, Gomes, Delgadillo, & Saraiva, 2022; Santos et al., 2021).

Despite HS has remarkable demonstrated capabilities, its technological readiness level is still low (around 3–4). This is due to the fact that many technical aspects of the technology still need to be addressed before the latter can be transferred from research laboratories to industry. These aspects primarily include design and construction of feasible HS working units, safety issues related to the handling of pressurized storage vessels, and identification of appropriate food packaging solutions (Basso & Manzocco, 2022; Bermejo-Prada et al., 2017).

The selection of proper packaging materials for food HS is not a trivial issue, since they should: i) guarantee that pressure is uniformly applied without breaking; ii) not undergo pressure-induced defects; iii) prevent mass transfer from the pressurizing fluid to the food and *vice versa.* These requirements could be easily addressed by selecting materials with adequate mechanical and diffusional properties. Nevertheless, literature suggests that pressurizations can induce a complex set of effects in plastic packaging materials, possibly leading to the impairment of barrier properties and even the loss of integrity of the whole packaging (Fraldi et al., 2014; Mensitieri, Scherillo, & Iannace, 2013; Morris, Brody, & Wicker, 2007). Concomitantly to these macroscopic events, pressure affects polymeric materials also at the molecular level. These changes are due to the semicrystalline nature of plastic materials, which are made of an amorphous matrix comprising dispersed, nanoscopic (50 nm) crystalline domains (Lin, Bilotti, Bastiaansen, & Peijs, 2020). When pressure as low as 50 MPa is applied, the free volume of the

amorphous phase of the film is reduced, causing a decrease in the permeation coefficient and an increase in the tendency to crystallize (Galotto et al., 2008; Galotto et al., 2010; Mensitieri et al., 2013). Upon crystallinity enhancement, films might become opaque, more brittle and less permeable, with occasional evidence of localized white staining (Fleckenstein, Sterr, & Langowski, 2014; Juliano, Koutchma, Sui, Barbosa-Cánovas, & Sadler, 2010).

Despite the critical effect of pressure on packaging materials, there is no evidence reporting its extent over wider time scales, typical of HS. In this context, even the less impactful defects occurring at a nanoscopic scale might build up to the point of becoming dramatic during extremely long pressurizations, possibly resulting in visual defects and alterations of packaging mechanical properties. The assessment of the feasibility of packaging materials for HS is thus a gap that must be urgently filled in order to make the technology viable for the industry.

The aim of this work was to explore the effect of hyperbaric storage on selected packaging materials. To this aim, four materials, typically employed in the food industry, were considered as case studies: i) polyamide/low-density polyethylene (PA/PE) was chosen since used in HS studied reported in the literature (Lemos et al., 2020; Santos, Delgadillo, & Saraiva, 2020); ii) polypropylene/ethylene vinyl alcohol/-low-density polyethylene (PP/EVOH/PE) was chosen due to the known capability of EVOH-based multilayer materials to withstand even extreme pressurized conditions (up to 800 MPa) without defects (López-Rubio et al., 2005); iii) polyethylene terephthalate (PET) was chosen based on its widespread use in the food industry (Ashby, 1988); iv) polylactic acid (PLA) was chosen as a bio-based and compostable alternative to PET (Sousa et al., 2021). Sample pouches were then heat-sealed containing D1 food simulant (50 % v/v ethanol). The latter was chosen to emulate perishable, protein-rich foods containing a dispersed lipophilic component (*e.g.*, egg yolk, raw milk, cheese, fat fish) (European Commission, 2011). According to the literature, these matrices would be the most feasible ones for HS, as they would benefit from both the antimicrobial and functionalizing effect of the technology (Basso & Manzocco, 2022). Samples were subjected to HS at 200 MPa at room temperature (20 ± 2 °C). At increasing time for up to 35 days, samples were analyzed for optical, structural, mechanical, and diffusional properties, which were compared to those of control samples stored at atmospheric pressure (0.1 MPa).

2. Materials and methods

2.1. Samples preparation

PA/PE (90 μm thickness, PE internal side) were obtained from Savonitti s.a.s. (Goricizza di Codroipo, Italy). PP/EVOH/PE (80 μm thickness, PE internal side) were obtained from Niederwieser Group S.p. A. (Campogalliano, Italy). PET (30 μm thickness) was obtained from DuPont Teijin Films™ (Dumfries, United Kingdom). PLA (thickness 30–35 μm) was obtained from Taghleef Industries (Newark, DE, U.S.A.).

Square (12 × 12 cm) film pieces were cut from each material and assembled to form pouches with 2 dm^2 internal surface. Pouches were welded using a VM-16 (Orved, Musile di Piave, Italy) heat sealer equipped with a 2 × 315 mm NiCr-steel welding bar (maximum sealing temperature = 285 ± 5 °C). Heat-sealing temperature was adjusted using an analogic potentiometer with 10 intensity levels. PLA was welded at an intensity level of 2 for 2.8 s, while PP/EVOH/PE, PA/PE and PET were welded at an intensity level of 4 for 3.8, 4.4 and 5.4 s, respectively. Pouches were then filled with 30 mL aliquots of D1 food simulant, which, in agreement with Annex III of Commission Regulation (EC) No. 10/2011, consisted of a 50 % (v/v) ethanol aqueous solution. To minimize headspace, air bubbles were manually removed from the pouches, which were then heat-sealed applying the same temperature/time combinations used for assembling (Orved VM-16, Musile di Piave, Italy).

2.2. Hyperbaric storage

Pouches were stored at 200 MPa at 20 ± 2 °C in a hyperbaric storage experimental working unit (Comer Srl., Bologna, Italy) comprised by a screw-capped steel vessel (Hystat Slaithwaite, Huddersfield, UK) and a Haskel International (Burbank, CA, USA) pneumatic high pressure multiplier fed with 6.4 bar compressed air. Pressure was increased at a 50 MPa min^{-1} rate, and transmitted inside the vessel with an aqueous solution containing 0.2 % (w/w) potassium sorbate and 0.2 % (w/w) sodium benzoate, which were added to prevent mold growth. At increasing time for up to 35 days, the HS vessel was depressurized (10 MPa s^{-1}) and manually opened. Samples were then withdrawn and carefully dried using paper towels before analysis. Reference samples were stored for analogous times at room pressure and temperature conditions (0.1 MPa, 20 ± 2 °C).

2.3. Lightness

Lightness of films was measured with a tristimulus colorimeter (Chromameter-2 Reflectance, Minolta, Osaka, Japan) equipped with a CR-300 measuring head and a D65 standard illuminant. The instrument was standardized against a reference white tile before analysis. Samples were positioned on top of the reference tile avoiding the formation of wrinkles and air pockets, and analyzed. The L* parameter of the CIELab scale was considered as samples lightness.

2.4. Ultraviolet and visible light barrier and opacity

UV-Vis light transmission was measured on 2 × 2 cm film samples by a spectrophotometer (VWR® Double Beam UV × VIS 6300 PC spectrophotometer, China) in the 200–800 nm wavelength range. Samples opacity value was calculated as:
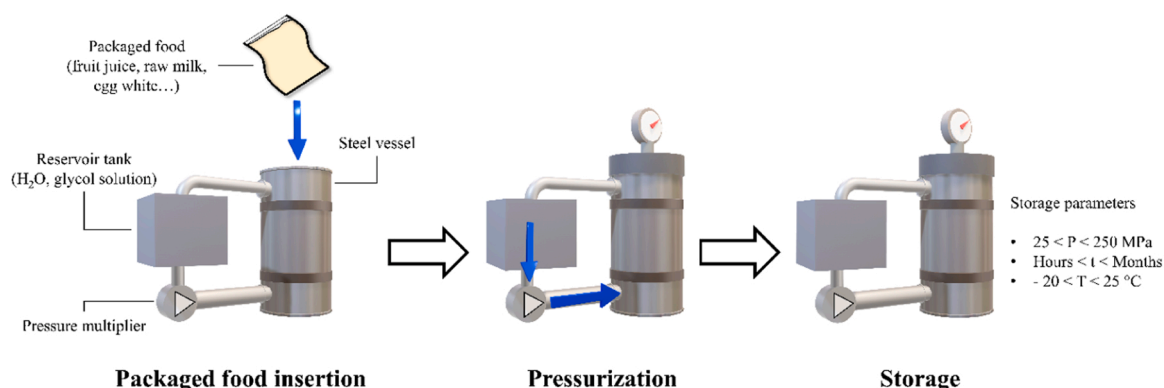

Fig. 1. Schematic representation of a typical hyperbaric storage process.

Opacity value $= \dfrac{\log T_{600}}{d}$ (1)

Where T_{600} is the transmittance at 600 nm and d is the film thickness (nm).

2.5. Differential scanning calorimetry (DSC)

Samples were manually cut into approximately 0.3×3 cm stripes, convoluted without wrinkling to fit a 100 μL aluminum crucible (Mettler-Toledo, Greifensee, Switzerland) and weighed to ± 0.0001 g precision. DSC was performed with a DSC 3 Stare System differential scanning calorimeter (Mettler-Toledo, Greifensee, Switzerland). Samples were heated from 0 °C to 100 °C at 10 °C min^{-1} under continuous nitrogen flow (20 L min^{-1}). Glass transition temperature (T_g, °C) and peak enthalpies (ΔH, J g^{-1}) were computed from the thermograms using the program STARe ver. 16.10 (Mettler-Toledo).

2.6. X-ray diffraction (XRD)

Films samples (2×2 cm) were subjected to XRD using an X′Pert PRO diffractometer (Marvel Panalytical, United Kingdom). XRD patterns were recorded using CuKα radiation ($\lambda = 1.54$ Å), at a voltage of 40 kV and a filament emission of 40 mA. Samples were scanned with ramping at 0.5° min^{-1} at an angle (2θ) range of 3–60°. A zero-background holder was used to avoid the detection of peaks not related to samples diffraction. Background noise was quantified by running the diffractometer with empty sample holder and was subtracted to the spectra. Peak elaboration and integration were performed using Origin Pro 2021 (OriginLab, Northampton, MA, USA). Crystallinity index (CI, %) was calculated as:

CI (%) $= \dfrac{S_c}{S_t} * 100$ (2)

Where S_c is the sum of crystalline peaks area and S_t is the sum of total area under the spectra (Mohammadkazemi, Azin, & Ashori, 2015).

2.7. Measurement of mechanical properties

Samples were obtained by cutting films into rectangular strips (10×1.5 cm). Thickness of each sample was estimated by averaging three measurements performed in three random positions with a digital micrometer (IP65, SAMA Tools, Viareggio, Italy). Tensile strength (MPa) and elongation at break (%) were evaluated using a dynamometer (Z1.0, ZwickRoell, Ulm, Germany) equipped with a 1 kN load cell. During the analysis, different test speeds (12.5 mm min^{-1}, 50 mm min^{-1} and 500 mm min^{-1}) were used depending on the percentage of elongation at break of the analyzed material, according to the standard method ASTM D882–12 (ASTM, 2001a). The TestXpert® II 161 software (v 3.31, ZwickRoell GmbH & Co. KG, Ulm, Germany) was used to elaborate the data.

2.8. Determination of water vapor transmission rate (WVTR)

WVTR was determined according to the standard method ASTM E96 (ASTM, 2001b) with some modifications. Glass vials with an internal diameter of 10 mm and a depth of 55 mm were filled with 2 g anhydrous CaCl$_2$ (0 % RH). Samples were then sealed on top of the vials, which were placed in a desiccator containing BaCl$_2$ (90 % RH) at 38 °C. The WVTR was determined by plotting the weight gain of the vials as a function of time for the period of linear ($R^2 > 0.94$) weight increase (up to 16 days), and was calculated as follows:

WVTR $= \dfrac{\Delta W}{\Delta t} \bullet \dfrac{1}{A}$ (3)

Where $\frac{\Delta W}{\Delta t}$ is the slope of the line describing the weight increase of the vials as a function of time (g day^{-1}) and A is the surface area of the exposed film ($7.85 \bullet 10^{-5}$ m^2).

2.9. Assessment of overall migration

Overall migration from the pouches to the food simulant during storage was determined by the thermogravimetric method, adapting the procedure described by Marangoni Júnior et al. (2020). The 30 mL food simulant D1 aliquots contained in each pouch were quantitatively transferred into dry crucibles and evaporated. Dry matter of the simulant ($d.m.$, mg) was then weighed to 0.00001 g precision. Overall migration (OM, mg dm^{-2}) was calculated as:

Overall migration $= \dfrac{d.m}{S_i}$ (4)

Where S_i (2 dm^2) is the internal surface of the pouches.

2.10. Data analysis

Lightness and DSC analyses were performed at least in triplicate. UV-Vis light barrier, opacity, mechanical properties and WVTR were measured at least in quintuple. Quantification of overall migration and XRD analyses were performed in single. All analyses have been performed on three independent experiments. Results are reported as mean $\pm$ standard deviation and were subjected to one-way analysis of variance (ANOVA) and Tukey's Honest Significant Differences test ($p < 0.05$) using R for Windows (v. 4.2.2, The R foundation for statistical computing, Wien, Austria).

3. Results and discussion

3.1. Films integrity and optical properties

The effect of hyperbaric storage on packaging materials was firstly assessed visually, noting that PA/PE, PP/EVOH/PE and PET retained their integrity and transparent aspect without macroscopic defects even after 35 days at 200 MPa. The capability of these materials to withstand pressurized conditions up to 200 MPa has been often reported in the literature, and is primarily due to their inherent elasticity and to the remarkable welding resistance when heat-sealed into pouches (Bamps, Buntinx, & Peeters, 2023; Juliano et al., 2010). By contrast, the seals of PLA pouches were made significantly weaker by HS, as they immediately failed upon samples withdrawal from the hyperbaric chamber. To this regard, some authors have reported pressurization (up to 600 MPa for 20–60 min) to impair packaging seal strength (Dobiáš, Voldřich, Marek, & Chudáčková, 2004; Lambert et al., 2000a; Lambert et al., 2000b; Masuda, Saito, Iwanami, & Hirai, 1992). As Fraldi et al. (2014) have demonstrated, pressure-induced stress concentrates in the welded sides of packaging pouches, where they can even cause macroscopic damages (e.g., delamination, wrinkling). Therefore, PLA seals could have progressively weakened during HS, ultimately failing upon application of even mild shear forces, such as those associated with the manual removal of samples from the hyperbaric vessel.

The optical properties of the packaging films were then instrumentally assessed to detect possible changes related to pressure-induced structural modifications (Table 1).

In agreement with the highly transparent aspect of the samples, the opacity of the unstored films was lower than 5 (Table 1) (Guzman-Puyol, Benítez, & Heredia-Guerrero, 2022). During storage for up to 35 days, samples opacity did not change regardless of the applied pressure. Accordingly, the films UV-C light (≤ 200 nm) transmittance was not affected by storage at all pressures, and remained very low (< 0.5 %) (data not shown). These results indicate that the application of HS did not affect the films see-through aspect, as well as their ability to hurdle

Table 1
Opacity and lightness (L*) of packaging films stored at 0.1 and 200 MPa for up to 35 days at 20 ± 2 °C. Standard deviations in brackets.

Pressure	Time	PA/PE		PP/EVOH/PE		PET		PLA	
(MPa)	(d)	Opacity	L*	Opacity	L*	Opacity	L*	Opacity	L*
0.1	0	1.22[a]	95.25[a]	1.66[a]	96.13[a]	3.56[a]	95.91[a]	2.63[a]	96.57[a]
		(0.02)	(0.18)	(0.05)	(0.05)	(0.25)	(0.14)	(0.24)	(0.01)
	7	1.28[a]	94.94[a]	1.56[a]	95.52[b]	3.48[a]	94.87[cd]	2.92[a]	95.86[c]
		(0.03)	(0.03)	(0.08)	(0.04)	(0.09)	(0.10)	(0.22)	(0.13)
	35	1.27[a]	94.30[b]	1.64[a]	95.54[b]	3.49[a]	94.99[c]	2.49[a]	96.10[b]
		(0.10)	(0.25)	(0.05)	(0.18)	(0.12)	(0.04)	(0.18)	(0.04)
200	7	1.29[a]	93.64[c]	1.84[a]	94.70[c]	3.43[a]	94.63[d]	2.37[a]	95.34[e]
		(0.05)	(0.09)	(0.32)	(0.15)	(0.17)	(0.07)	(0.36)	(0.04)
	35	1.41[a]	94.49[b]	1.68[a]	93.73[d]	3.32[a]	95.38[b]	2.75[a]	95.45[d]
		(0.15)	(0.10)	(0.07)	(0.35)	(0.22)	(0.13)	(0.41)	(0.02)

[a] Different letters in the same column indicate statistically different means (ANOVA; *p < 0.05*).

photo-oxidative reactions upon environmental light exposure (Di Filippo et al., 2021). However, a small but statistically significant (*p* < 0.05, ANOVA) decrease in lightness (L*) was observed in all samples during storage (Table 1). In agreement with the literature, this indicates that the films crystallinity might have decreased. It is in fact known that polymeric matrices with less crystalline domains are less luminous due to their reduced light scattering capability (Yoo, Lee, Holloman, & Pascall, 2009). In PA/PE, PP/EVOH/PE and PLA samples, the decrease in lightness was slightly more pronounced when HS was applied (Table 1), suggesting that pressurization could have favored crystallinity loss in these films. Based on these results, samples were further analyzed by XRD and DSC, in order to clarify the effect of HS on the films structural properties.

3.2. Structural properties

The effect of HS on the crystalline fraction of packaging materials was assessed by XRD. Analyses revealed that storage for up to 35 days did not cause any change in the XRD spectra of PA/PE and PP/EVOH/PE samples (data not shown). On the other hand, storage at atmospheric and hyperbaric conditions significantly affected the crystalline fractions

Fig. 2. X-ray diffractograms of PET (A) and PLA (B) films before (control) and after storage at 0.1 and 200 MPa for 35 days at 20 ± 2 °C.

of both PET and PLA (Fig. 2).

A single, large peak was observed in X-ray diffractograms of control PET and PLA, indicating that both materials were predominantly crystalline (Stetsiv et al., 2021). In agreement with the literature, the XRD peak angles (2θ) of PET and PLA were 26.02 and 16.58°, respectively (Fig. 2) (Ahmed, Mulla, Al-Zuwayed, Joseph, & Auras, 2022; Gaonkar, Murudkar, & Deshpande, 2020). According to the Bragg equation, these angles corresponded to a crystalline interplanar distance (*d*) of 3.42 ± 0.00 and 5.36 ± 0.03 Å in PET and PLA, respectively. As clearly visible from Fig. 2, storage at 0.1 MPa for 35 days did not cause significant changes in PET and PLA XRD peaks positions. Conversely, a peak shift to higher angle was observed for both materials under pressure, indicating that 35 days-HS caused a decrease in the interplanar distance of PET (*d* = 3.36 ± 0.04 Å) and PLA (*d* = 5.18 ± 0.03 Å). This clearly indicates that pressurized storage caused the structure of both materials to become more compact. These results are in agreement with the literature reporting compression of packaging materials even upon brief (15–25 min) hydrostatic pressure processing at 200–450 MPa (Ahmed et al., 2022). As clearly visible from Fig. 2, the intensity and area of PET and PLA XRD peaks decreased during storage at 0.1 MPa. This could be attributed to the swelling of the films with simulant during storage, which would have reduced the abundance of the crystalline fractions (Zhang, Qu, Mosier, Han, & Xiao, 2018). According to Fig. 2, this phenomenon seemed strongly favored by pressure in both materials. This could have been due to the fact that, during storage, pressurized pouches were exposed not only to the simulant on the inside, but were also in contact with the HS pressurizing fluid on the outside. However, regardless of this additional variable, it must be noted that these results are in evident contradiction with the literature, which reports a significant impairment of diffusive phenomena in polymeric films under pressure (Götz & Weisser, 2002; Schmerder, Richter, Langowski, & Ludwig, 2005). Nevertheless, according to Fleckenstein et al. (2014), the enhancement of PET and PLA fluid absorption under HS could have been due to the pressure-induced formation of microscopic irregularities (*e.g.*, valleys, depressions) on the surface of the films. Several Authors have in fact observed this effect even upon brief (5–60 min) pressurizations at pressures as low as 200–300 MPa (Caner, Hernandez, Pascall, & Riemer, 2003; Hoque, McDonagh, Tiwari, Kerry, & Pathania, 2022; Tang, Fan, Fan, Jiang, & Qin, 2020). Based on the changes induced by storage at 0.1 and 200 MPa in PET and PLA XRD peaks (Fig. 2), samples crystallinity index (*CI*) was calculated as reported in Section 2.6. In agreement with the largely crystalline character of the films (Fig. 2), the *CI* of control PET and PLA samples was 77.68 ± 0.25 and 67.30 ± 4.38 %, respectively. Interestingly, no change in *CI* was observed in PET stored for 35 days at 0.1 (77.79 ± 1.10 %) or 200 (75.85 ± 1.95 %) MPa (Fig. 2A). This result indicates that, regardless of storage pressure, the proportionality between the crystalline and the amorphous fractions did not change (Mohammadkazemi et al., 2015). Differently from PET, the application of 35 days-HS to PLA resulted in a pronounced decrease of *CI* (52.65 ± 1.54 %). According to the literature, this can be attributed to
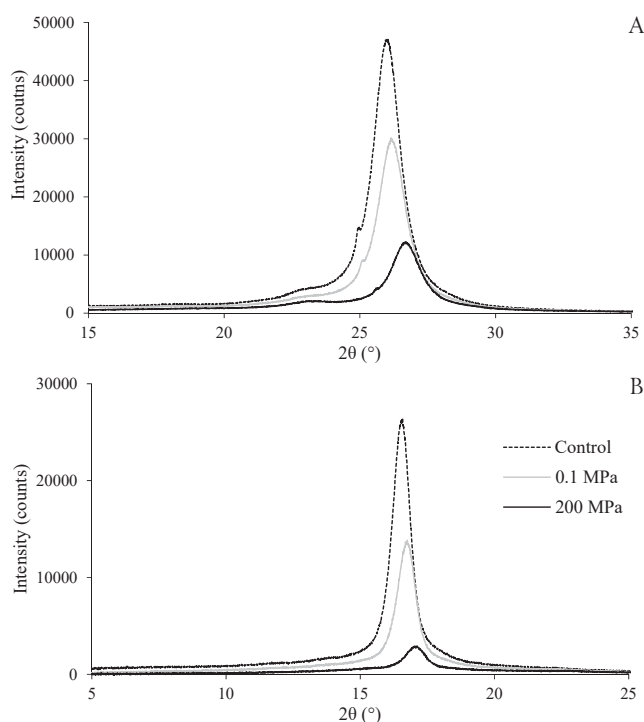
the enhancement of liquid absorption in the material under pressure, which is known to favour structural changes involving the mobilization of the polymeric chains into less ordered structures (Galotto et al., 2008; Galotto, Ulloa, Guarda, Gavara, & Miltz, 2009; Tang et al., 2020). PLA polymers have actually much better affinity for water as compared to PET, determining more intense swelling upon contact with aqueous solutions (*i.e.*, simulant or pressurizing fluid) (Kirchkeszner et al., 2022; Ribitsch et al., 2012).

To evaluate if HS affected the amorphous fraction of the films, DSC analysis was performed. Regardless of the applied pressure, storage for up to 35 days did not modify the thermal behaviour of the amorphous regions of PA/PE, PP/EVOH/PE and PLA (data not shown). Contrarily, interesting changes were observed in the case of PET (Fig. 3).

As visible from Fig. 3, and in agreement with the literature, the glass transition temperature (T_g) of PET control film was around 80 °C (Jog, 1995). In this sample, DSC analysis revealed the presence of a small endothermic peak in correspondence of T_g, indicating that PET films were slightly physically aged before storage trials were performed (Montserrat & Cortés, 1995). Physical ageing is a temperature-dependent phenomenon, affecting polymeric matrices stored for weeks/months below their T_g. It consists in the slow, progressive rearrangement of glassy polymer chains into non-crystalline ordered structures, which require more energy for rubbery transition when heated (Montserrat & Cortés, 1995). Due to physical ageing, polymeric materials usually become denser, with higher tensile strength and Young modulus, but also embrittled (Kong, 1986; Lacatus & Rogers, 1986; Mininni, Moore, Flick, & Petrie, 1973). Data reported in Fig. 3 clearly indicate that storage pressure affected the thermal behavior and physical ageing of PET. To better understand these changes, thermograms were elaborated, focusing the attention on samples T_g and physical ageing enthalpy (ΔH) (Table 2).

As reported in Table 2, the T_g of PET significantly decreased after 7 days of storage at atmospheric pressure (0.1 MPa), without further changes for up to 35 days. When HS was applied, the decrease in T_g after 35 days was more pronounced (Table 2). On the other hand, peak enthalpy (ΔH) progressively increased during storage at atmospheric pressure for up to 35 days (Table 2), indicating a remarkable development of PET physical ageing in these conditions (Montserrat & Cortés, 1995). The application of HS completely hampered the phenomenon, even showing a slight decreasing trend in ΔH under pressure (Table 2). These results suggest that HS was able to promote the plasticization of PET, while concomitantly impairing its physical ageing. Reasonably, both these effects of HS were induced by the enhancement of PET swelling with simulant and pressurizing fluid (Fig. 2). In fact, pressure-induced liquid absorption would have mobilized amorphous PET molecules, decreasing T_g and preventing the formation of non-crystalline supramolecular structures (Feigenbaum, Riquet, & Scholler, 2000; Kirchkeszner et al., 2022). It is interesting to note that
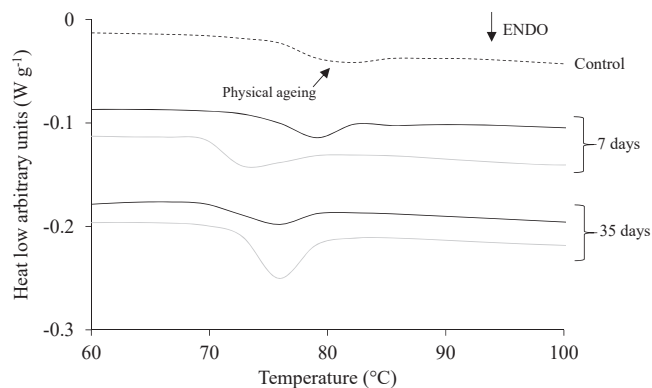
Table 2

T_g and physical ageing peak enthalpy (ΔH) of PET films stored at 0.1 and 200 MPa for up to 35 days at 20 ± 2 °C.

Pressure (MPa)	Time (d)	T_g (°C)	ΔH (J g^{-1})
0.1	0	78.42 ± 0.40^a	0.45 ± 0.02^c
	7	71.16 ± 0.52^{cd}	0.81 ± 0.11^b
	35	71.91 ± 0.06^c	1.04 ± 0.03^a
200	7	74.30 ± 0.06^b	0.31 ± 0.06^c
	35	70.38 ± 0.47^d	0.38 ± 0.05^c

a Different letters in the same column indicate statistically different means (ANOVA; *p < 0.05*).

physical ageing is knowingly associated with a decrease in the specific volume of polymeric matrices (Mininni et al., 1973). Therefore, based on the Le Chatelier principle and Transition State theory, this phenomenon should have been favoured during pressurized storage (Evans & Polanyi, 1935; Le Chatelier, 1891). Coherently with the literature (Fleckenstein et al., 2014), results reported in Table 2 clearly indicate that the outcome of pressurization of packaging materials cannot be predicted based on theoretical kinetic principles solely. In particular, the interaction of the polymeric materials with the packaged foods/simulants appears to be a major source of deviation from ideality, and should thus be carefully considered (Fleckenstein et al., 2014).

3.3. Mechanical properties

In agreement with the absence of structural modifications, PA/PE and PP/EVOH/PE films did not show changes in mechanical properties during storage for up to 35 days, regardless of the applied pressure (data not shown). In particular, PA/PE tensile strength and elongation at break did not vary from ~ 40 MPa and ~ 250 %, respectively. For what concerns PP/EVOH/PE samples, tensile strength and elongation at break values were always very close to ~ 26 MPa and ~ 260 %, respectively. Differently, PET and PLA mechanical properties significantly changed during storage at 0.1 and 200 MPa (Fig. 4).
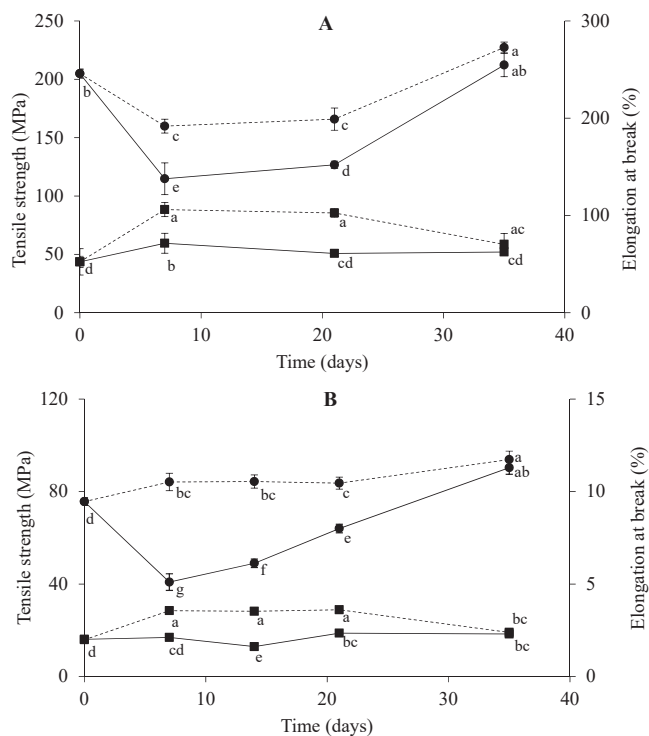[a] Different letters for the same mechanical property for each film

Fig. 3. DSC thermograms of PET stored at 0.1 (–   ) and 200 (– –) MPa for up to 35 days at 20 ± 2 °C.

Fig. 4. Tensile strength (•) and elongation at break (■) of PET (A) and PLA (B) films stored at 0.1 (_) and 200 (_) MPa for up to 35 days at 20 ± 2 °C.

indicate statistically different means (ANOVA; $p < 0.05$).

PET and PLA initially showed values of tensile strength and elongation at break in line with those reported in the literature (Jamshidian et al., 2012; Panowicz et al., 2021). During storage at both 0.1 and 200 MPa, PET mechanical properties showed changes (Fig. 4A) that were likely due to the complex set of modifications in crystalline and amorphous structures (Fig. 2, Table 2). In particular, the tensile strength and the elongation at break of the material decreased and increased, respectively (Fig. 4A). According to (Lim & Hoag, 2013), this was probably due to the plasticization of PET during storage, which was also suggested to occur by data reported in Table 2. However, after 35 days of storage, the mechanical properties of PET kept under hyperbaric conditions were statistically indistinguishable ($p > 0.05$, ANOVA) from those of samples stored at 0.1 MPa, and very close to those of control PET (Fig. 4A). This was probably due to a complex set of several, counteracting structural rearrangements occurring upon prolonged storage of PET samples, which can reportedly occur as a consequence of solvent swelling (Fig. 2) (Li, Wang, Lin, & Hu, 2016).

Concerning PLA, storage at atmospheric pressure for up to 35 days caused a slight increase in tensile strength and elongation at break (Fig. 4B). Differently, in the pressurized films, the variation of mechanical properties during storage resembled the trend exhibited by PET. Nevertheless, PLA showed a slightly more pronounced decrease in tensile strength after 7 days under HS as compared to PET (Fig. 4B). By further prolonging pressurized storage, a trend inversion was observed, which resulted in PLA tensile strength to significantly increase after 35 days. These changes were likely due to the counterbalancing effect of at least two structural events concomitantly occurring in PLA during HS. Based on previous results, it is reasonable that the decrease in PLA crystallinity could have made the material weaker after 7 days, whereas the packing effect of pressure (Fig. 2) might have favoured the enhancement of PLA strength after more prolonged HS.

3.4. Diffusional properties

Based on the commercial relevance of packaging WVTR, and on the regulatory importance of overall migration (European Commission, 2011; Jarvis et al., 2017), analyses were performed not only on PET and PLA, but also on PA/PE and PP/EVOH/PE films (Table 3), regardless of the absence of changes in optical, structural and mechanical properties.

In accordance with the absence of structural modifications in PA/PE and PP/EVOH/PE, the WVTR of both multi-material films did not change during storage for up to 35 days at 0.1 MPa and 200 MPa (Table 3). Differently, when HS was applied to PET, a moderate increase in WVTR was observed after 35 days (Table 3), which was likely related to the swelling of the film with liquid (Fig. 2). In fact, it is widely known that the absorption of water molecules in polymeric materials can significantly impair the barrier properties of polymeric packaging (Juliano et al., 2010). The same mechanism was probably involved in the increase in PLA WVTR after 35 days storage at 0.1 MPa (Table 3). In this case, the fact that PLA barrier properties decayed even without the application of pressure was probably due to the high hydrophilicity of the material, which would have strongly promoted simulant absorption (Scarfato et al., 2017). Unexpectedly, the application of HS limited the degradation of PLA barrier during storage (Table 3). This was likely due to the compaction of the material promoted by hyperbaric conditions (Fig. 2). When pressurized, plastic packaging films are actually known to undergo a reduction of their diffusion coefficient, which is due to the shrinkage of the intermolecular voids embedded in their matrix (Götz & Weisser, 2002). This effect has been frequently observed even upon brief (5–30 min) pressurizations at 50–200 MPa in a wide range of food packaging films (including PLA) (Fleckenstein et al., 2014; Mauricio-Iglesias, Peyron, Chalier, & Gontard, 2011; Schmerder et al., 2005; Yoo et al., 2009). It is thus likely that, in the specific case of hyperbarically-stored PLA (Table 3), the compression induced by HS counteracted the barrier loss promoted by the material swelling with

Table 3

Water vapour transmission rate (g day^{-1} m^{-2}) and overall migration (mg dm^{-2}) of PA/PE, PP/EVOH/PE, PLA and PET stored at 0.1 and 200 MPa for up to 35 days at 20 ± 2 °C.

Pressure (MPa)	Time (d)	PA/PE	PP/EVOH/PE	PET	PLA
			WVTR (g day^{-1} m^{-2})		
0.1	0	11.21 ± 1.06[a]	7.13 ± 0.70[a]	42.67 ± 1.64[ab]	300.00 ± 16.30[c]
	7	12.10 ± 0.74[a]	8.41 ± 1.14[a]	41.40 ± 2.65[b]	339.81 ± 7.60[ab]
	21	12.31 ± 0.74[a]	8.41 ± 1.14[a]	42.88 ± 4.80[ab]	346.82 ± 9.44[ab]
	35	12.74 ± 2.38[a]	9.17 ± 0.57[a]	40.45 ± 1.60[b]	350.64 ± 8.41[a]
200	14	10.70 ± 1.71[a]	8.66 ± 1.07[a]	42.29 ± 3.97[ab]	321.87 ± 6.41[bc]
	35	10.45 ± 1.40[a]	7.64 ± 1.56[a]	48.73 ± 3.18[a]	320.59 ± 17.39[bc]
			Overall migration (mg dm^{-2})		
0.1	7	2.06 ± 0.39[b]	1.92 ± 0.29[a]	0.90 ± 0.05[bc]	2.17 ± 0.23[ab]
	14	1.93 ± 0.16[b]	1.15 ± 0.00[a]	1.94 ± 0.29[ab]	2.20 ± 0.13[a]
	21	3.19 ± 0.48[b]	2.21 ± 0.17[a]	1.21 ± 0.11[ac]	0.92 ± 0.08[bc]
	35	1.71 ± 0.18[b]	2.28 ± 0.80[a]	1.75 ± 0.45[ab]	1.71 ± 0.77[ac]
200	7	485.25 ± 52.14[a]	2.95 ± 1.10[a]	0.43 ± 0.04[c]	0.79 ± 0.35[c]
	14	432.30 ± 32.15[a]	1.19 ± 0.16[a]	1.26 ± 0.24[bc]	O.d.s.
	21	428.07 ± 19.53[a]	2.82 ± 1.71[a]	1.99 ± 0.21[a]	O.d.s.
	35	397.82 ± 33.10[a]	251.61 ± 310.17*	1.89 ± 1.81*	O.d.s.

[a] Different letters in the same column indicate statistically different means (ANOVA; $p < 0.05$)

* Sample not included in statistical analysis

O.d.s. Sample opened during storage

simulant and pressurizing fluid, ultimately limiting its increase in WVTR.

Concerning overall migration, no significant changes ($p > 0.05$, ANOVA) were observed in samples subjected to storage at atmospheric pressure for up to 35 days (Table 3). The same results were obtained even in PET and PLA subjected to HS for up to 35 and 7 days, respectively. This is in agreement with several studies reporting that pressurization up to 600 MPa does not affect the capability of packaging materials to retain migrating molecules like monomers and additives (Juliano et al., 2010). Nevertheless, it must be noted that, since PLA seals failed after 7 days under pressure solely, it remains unclear whether this material could beget critical migration upon longer HS. Different from single-material films, the overall migration of multi-material packaging significantly increased during HS (Table 3). In particular, the application of pressurized storage to PA/PE caused a release of substances about 50 times higher than the legal limit of 10 mg dm^{-2} (European Commission, 2011) after only 7 days. Migrants were likely represented by adhesives located between the film layers, and their diffusion in the simulant was probably promoted by the swelling of PA/PE upon pressurization (Galotto et al., 2010). To the best of our knowledge, migration phenomena of such extent have never been observed upon short-term pressurizations (30–60 min), even at pressure levels as high as 800 MPa (Juliano et al., 2010). This suggests that pressure-induced migration of adhesives or additives from the packaging materials is highly time-dependent, thus being potentially critical in the HS context. Compared to PA/PE, PP/EVOH/PE was much less prone to releasing substances under HS, since no changes in overall migration were observed for up to 21 days (Table 3). Nevertheless, it must be noted that, after 35 days under pressure, a critical amount of migrants (470.9 mg dm^{-2}) was found in the simulant contained in one

of the pressurized PP/EVOH/PE pouches (Table 3). As for PA/PE, these compounds were likely adhesives sited between film layers, which migrated into the simulant upon diffusion through the polymeric matrix (Galotto et al., 2010). Although limited to only a single sample, this result indicates the possibility of critical migration of substances if PP/EVOH/PE is kept for prolonged times under pressure.

4. Conclusions

This work reported for the first time the effects of hyperbaric storage (200 MPa, up to 35 days) on selected food packaging materials. Multi-material films do not seem to suffer significant changes in optical, structural and mechanical properties under pressurized conditions, but could release critical amounts of migrants (*e.g.*, adhesive compounds). On the other hand, mono-material, adhesive-free PET and PLA films could undergo intense structural modifications upon simulant swelling during hyperbaric storage. These effects may result in complex changes in mechanical properties, and in barrier capacity loss, but are not expected to beget migration. Nevertheless, the mechanical strain occurring during HS can cause PLA packaging seals to significantly weaken and easily fail.

Based on these results, future studies on hyperbaric storage should take into account the selection of appropriate packaging solutions as a crucial aspect, focusing primarily on adhesive-free options. For lower pressures and/or storage times shorter than 35 days, EVOH-based multi-material films could also be feasible, regardless of the presence of adhesives. In this framework, it would be particularly interesting to study the feasibility of biodegradable or compostable materials other than PLA. Also, the assessment of recycled materials, such as rPET, would represent a challenging advancement, based on the possible enhanced migration of contaminants which, in normal conditions, does not occur. The application of these solutions could allow to further reduce the already low environmental impact of hyperbaric storage. Finally, it would be interesting to study if polymer compression during HS could be exploited to enhance packaging barrier properties during food storage: To the best of our knowledge, this is a completely unexplored research topic.

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CRediT authorship contribution statement

Federico Basso: Conceptualization, Data curation, Formal analysis, Investigation, Visualization, Writing – original draft; **Andrea Feroce**: Data curation, Validation, Formal analysis, Investigation, Visualization, Writing – original draft; **Lara Manzocco**: Resources, Supervision, Writing – review & editing; **Fabio Licciardello**: Methodology, Project administration, Writing – review & editing; **Maria Cristina Nicoli**: Funding acquisition, Resources, Writing – review & editing.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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