



Cite this: *Phys. Chem. Chem. Phys.*,
2025, 27, 18942

Investigating dose rate effects and reactive species formation in irradiated multilayer films – part 2 PE/EVOH/PE†

Blanche Krieguer,^{id}*^{abc} Samuel Dorey,*^a Nicolas Ludwig,^{id}^d Florent Kuntz,^d
Sylvain R. A. Marque,^{id}*^b Nathalie Dupuy,^{id}*^c and Fabien Girard^c

This study investigates the impact of gamma rays, X-rays, and electron beam irradiation on PE/EVOH/PE multilayer films focusing on how dose rate influences polymer modifications and the formation of reactive species like peroxides and methionine sulfoxide from methionine solution, which can affect protein stability. Using advanced analytical techniques such as electron spin resonance (ESR) and high-performance liquid chromatography (HPLC), the study measures radical formation and methionine oxidation. Results indicate that post-irradiation ageing and contact time with methionine significantly affect methionine sulfoxide levels, while the impact of dose rate varies by irradiation technology. Oxidation of methionine solution in contact with irradiated film remains similar whatever gamma, X-ray and e-beam technologies are used to irradiate the film.

Received 29th October 2024,
Accepted 6th June 2025

DOI: 10.1039/d4cp04153d

rsc.li/pccp

Introduction

Following our previous study on ethylene vinyl acetate/ethylene vinyl alcohol/ethylene vinyl acetate (EVA/EVOH/EVA) films,¹ this study investigates the impacts of irradiation technologies on polyethylene/polyethylene polyvinyl alcohol/polyethylene (PE/EVOH/PE) multilayer films, essential in the biotechnological and biopharmaceutical sectors. These films are used for storing and transporting biopharmaceutical products and require sterilization, typically through gamma radiation.^{2–5} The increasing production of biopharmaceutical products and concerns about the future capacity of gamma radiation sterilization have led to the investigation of alternative methods, including electron beam (e-beam) and X-ray irradiation.^{6–12}

Electron-beam irradiation uses an electron accelerator to convert electricity into a radiation beam, which can be used directly or to produce X-ray *via* the Bremsstrahlung effect.^{13,14} Both X-ray and gamma rays are photons, but gamma rays come from radioactive sources. Their similar energy deposition patterns allow for an easy transition from gamma-ray to X-ray

technologies, with X-ray potentially offering better dose uniformity.¹² E-beams, however, have less penetration due to stronger interactions with matter. One of the key differences among X-ray, e-beam, and gamma-ray technologies is the dose rate, which significantly influences biological and polymer systems. Low-dose-rate (LDR) exposures can enhance cellular repair, while high-dose-rate (HDR) exposures often cause more severe damage. In polymers, dose rate affects properties like cross-linking and oxidation, impacting their application suitability.¹⁵

A multiscale approach is essential to understand the role of dose rate in ionizing radiation exposure, especially in sterilized polymer systems used in biopharmaceuticals. Recent studies have focused on the effects of irradiation on multilayer films, revealing changes in physical, mechanical, and chemical properties.^{16–29} Additionally, irradiation influences the formation of transient species like hydrogen peroxide, which can affect protein oxidation and function.^{30,31} Methionine residue oxidation, for example, can alter protein stability and efficacy, impacting therapeutic proteins and antibodies.^{32,33}

This study aims to compare the effects of X-ray, e-beam, and gamma irradiation on PE/EVOH/PE films, focusing on polymer modifications and interactions with biopharmaceutical solutions. We investigate the generation of reactive species, such as peroxides and peracids, through methionine oxidation, using high-performance liquid chromatography (HPLC) and electron spin resonance (ESR) to assess the impacts of each technology. This research extends the observations from part 1,¹ providing deeper insights into the role of dose rate and irradiation technology in polymer applications.

^a Sartorius Stedim FMT S.A.S, Z.I. Les paluds, Avenue de Jouques CS91051, 13781, Aubagne Cedex, France

^b Aix Marseille University, CNRS, ICR, Case 551, 13397 Marseille, France.
E-mail: samuel.dorey@sartorius.com, sylvain.marque@univ-amu.fr

^c Aix Marseille University, Avignon University, CNRS, IRD, IMBE, Marseille, France.
E-mail: nathalie.dupuy@amu.fr

^d Aerial, 250 Rue Laurent Fries, 67400 Illkirch, France

† Electronic supplementary information (ESI) available. See DOI: <https://doi.org/10.1039/d4cp04153d>



Materials and methods

Samples

PE single-use plastics bags investigated are made from a multi-layer film, namely the S80 film, composed of one layer of EVOH sandwiched between two layers of PE, with a total thickness of approximately 360 μm (Fig. 1). Sample bags are provided by Sartorius stedim FMT S.A.S., Aubagne, France.

Samples irradiation

Gamma rays. PE bags were irradiated with gamma rays from ^{60}Co at Ionisos in Dagneux, France, at an average dose rate of 2 kGy h^{-1} . A first process needed several days, accumulating the dose over multiple runs with unknown rest times in the irradiation bunker, while the second was completed in a single run. Additionally, PE bags were irradiated with gamma rays from ^{60}Co at Steris in the USA, with an average dose rate of 8 kGy h^{-1} (multistep process).

X-ray. PE bags were irradiated using a 7 MeV Rhodotron at Steris in Däniken, Switzerland, with an average dose rate of 80 kGy h^{-1} and a maximum power of 560 kW. Additionally, PE bags were irradiated at Aerial-CRT in Strasbourg, France, using the Feerix facility based on a Rhodotron. A 7 MeV X-ray beam was generated by converting a 7 MeV electron beam in a tantalum target. The average dose rate was 13 kGy h^{-1} .

e-beam. PE bags were irradiated with a 10 MeV Rhodotron at Ionisos in Chaumesnil, France, at a dose rate of 18 000 kGy h^{-1} with a power source of 28 kW. Additionally, PE bags underwent e-beam irradiation at Aerial-CRT using the same Feerix[®] facility as the X-ray treatment. A 10 MeV vertical electron beam was used, with an average dose rate of 18 000 kGy h^{-1} (5 kGy s^{-1}), and the irradiation was performed in 50 kGy increments to prevent critical temperature increases in the samples.

Dosimetry. The targeted doses were approximately 30, 50, 70, and 100 kGy , depending on the irradiation technology, with a dose accuracy target of $\pm 10\%$. There were two phases of irradiation. Table 1 summarizes the dosimetry for the ESR study and Table 2 for the HPLC study. To accurately measure the absorbed dose, alanine pellets with ESR spectroscopy (Magnetech MS5000 ESR, Bruker) were used, along with AerEDE[®] dosimetry software (Aerial, France). Dosimetry readings

Table 1 Effective doses on samples irradiated by gamma, X-ray and e-beam for ESR analysis

Received doses (kGy, $\pm 10\%$)	Target doses (kGy)		50	100
	Gamma	Ionisos, France (1.9 kGy h^{-1})		51
X-ray	Aerial, France (13 kGy h^{-1})		50	99
e-beam	Aerial, France (18 000 kGy h^{-1})		55	109

Table 2 Effective doses on samples irradiated by gamma, X-ray and e-beam for HPLC analysis

Received doses (kGy, $\pm 10\%$)	Target doses (kGy)			
	Gamma	X-ray	e-beam	
Gamma	Ionisos, France (1.9 kGy h^{-1})		29	59
	Ionisos, France (2.4 kGy h^{-1})		32	52
	Steris, USA (8 kGy h^{-1})		58	54
X-ray	Aerial, France (13 kGy h^{-1})		26	51
	Steris, Switzerland (80 kGy h^{-1})		—	—
e-beam	Ionisos, France (18 000 kGy h^{-1})		25	52
	Aerial, France (18 000 kGy h^{-1})		25	52

were traceable to an international standard. Dosimeters were placed inside the sample box to ensure dose uniformity.

Analytical methods

The analytical methods, including ESR, HPLC, and equivalency analysis are described in part 1.¹

Results and discussion

Electron spin resonance

Fig. 2 displays ESR signal of radical species in PE/EVOH/PE multilayer film nine days after irradiation by gamma, X-ray and e-beam at 50 kGy and 100 kGy .

A three-peak signal is observed, and the radical species generated in the PE/EVOH/PE multilayer film are identified as hydroxyalkyl radicals in EVOH, similar to those in the EVA/EVOH/EVA multilayer film.^{1,20,34}

Fig. 3 shows small difference in concentration of radical species generated one day after X-ray and e-beam irradiation. Over a longer period, there are no significant differences between the three irradiation technologies, meaning a likely similar process of decay.

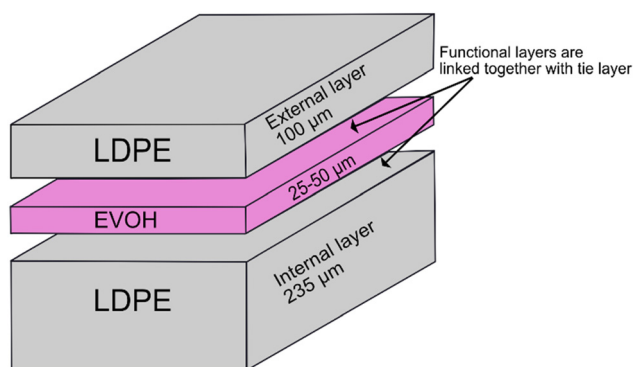


Fig. 1 Representation of PE/EVOH/PE multilayer film (S80 film), low density polyethylene (LDPE) is noted as PE all along the text.

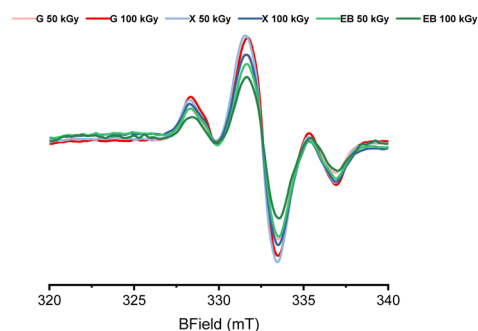


Fig. 2 ESR signal of PE/EVOH/PE multilayer film nine days after gamma (red line), X-ray (blue line) and e-beam (green line).



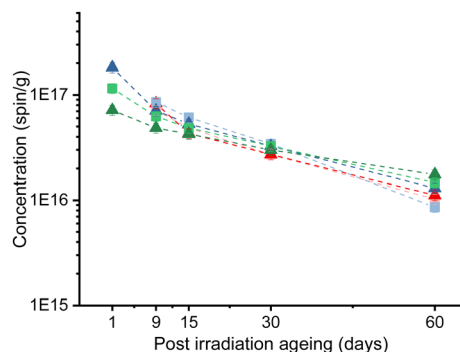


Fig. 3 Monitoring of the radicals concentration by ESR as concentration (spin per g) vs. ageing post irradiation (days). Squares correspond to sample irradiated at 50 kGy and triangle correspond to sample irradiated at 100 kGy. Red for gamma, blue for X-ray and green for e-beam. Dotted line is for better reading.

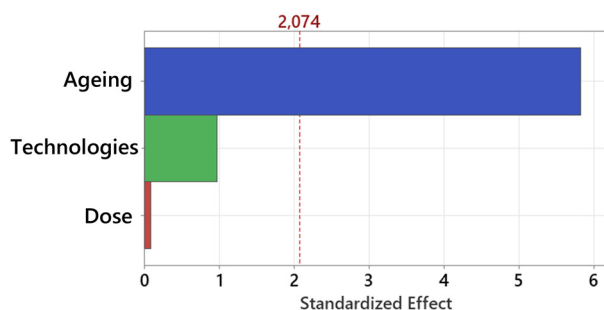


Fig. 4 Pareto analysis of effects on radical concentration: ageing post irradiation (blue); dose (red) and irradiation technologies (gamma, X-ray and e-beam) (green). The dotted lines correspond to the significance threshold given by the software MODDE for 95% confidence with p -value < 0.05.

Regression model (Fig. 4) shows that only post-irradiation ageing significantly affects the radical concentration, which decreases after irradiation. Equivalent amount of radical species are generated in samples irradiated by the three technologies, regardless of the dose. The equations and coefficients of the regression models obtained for each sample are reported in the ESI.†

Oxidation assay by high performance liquid chromatography

Comparison of irradiation technology. As in part 1,¹ the multilayer films were irradiated in different irradiation facilities with varying dose rates for each irradiation technology. The quantity of methionine sulfoxide formed by the oxidation caused by methionine solution in contact with the film, formed was measured three and six months after irradiation, with the concentrations presented in Fig. 5. The concentration of methionine sulfoxide obtained in contact with the PE/EVOH/PE film is very low compared to that with the EVA/EVOH/EVA film.^{1,27,35}

Regression models were developed to assess whether the dose rate, the dose, ageing post irradiation, methionine contact time and dose rate can have a significant effect on the

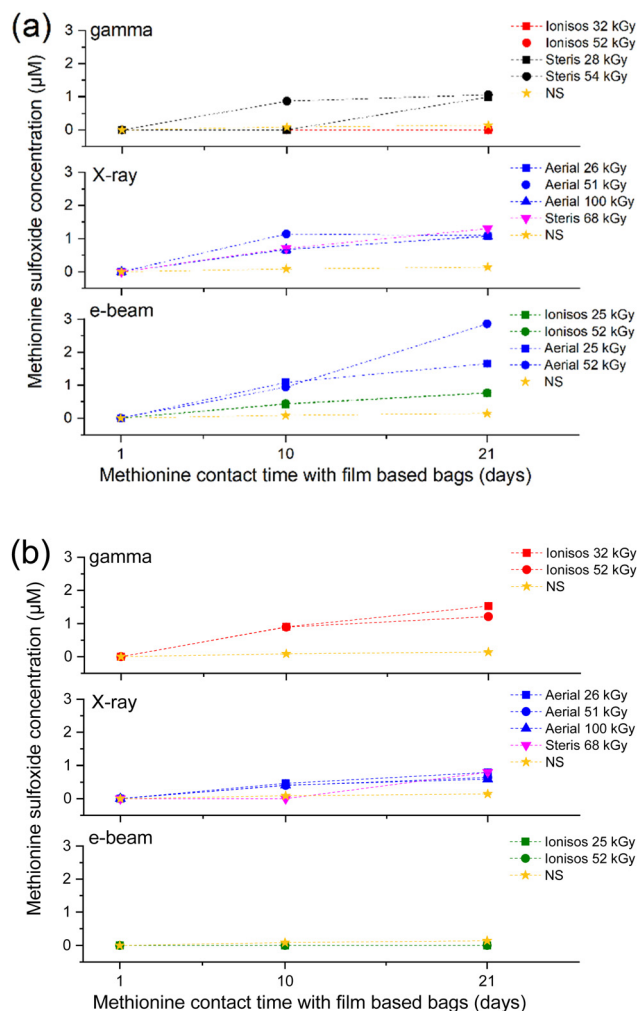


Fig. 5 Bags filled with methionine solution (50 μ M) (a) Three months after irradiation. (b) Six months after irradiation. Methionine sulfoxide concentration (μ M) in stored solution for 0, 1, 10 and 21 days and analyzed by HPLC. Irradiation doses were 30 kGy (square), 50 kGy (circle), 70 kGy (down triangle) and 100 kGy (up triangle). Non irradiated sample (NS) are in yellow stars. Irradiations at Ionisos, Dagneux, France in red (2 kGy h^{-1}), at Steris, USA, in black (8 kGy h^{-1}), at Aerial, France in blue (13 kGy h^{-1} for X-ray and $18\,000\text{ kGy h}^{-1}$ for e-beam), at Steris, Daniken, Switzerland in pink (80 kGy h^{-1}) and at Ionisos, Chausmenil, France in green ($18\,000\text{ kGy h}^{-1}$). Dotted line is for better reading.

methionine sulfoxide concentration generated in the multilayer film. The equation model is given as (1).

$$\begin{aligned} \text{Methionine sulfoxide concentration} = & 0.132 + 0.00102 \cdot \text{Dose} \\ & - 0.0464 \cdot \text{Ageing post irradiation} + 0.04487 \cdot \text{Methionine} \\ & \text{contact time} + 0.000002 \cdot \text{Dose rate} \end{aligned} \quad (1)$$

The regression models results, shown in Fig. 6, reveal that only the contact time with the methionine solution significantly affects methionine oxidation, with a coefficient of 0.045 indicating a slight increase in methionine sulfoxide over time. This is in accordance with the results observed in Fig. 5. The dose rate, influenced by irradiation technology, does not significantly impact methionine oxidation. Despite varying dose rates



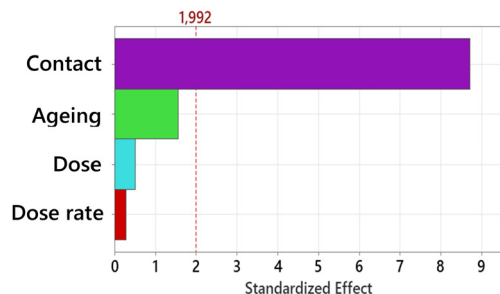


Fig. 6 Pareto analysis of effects on methionine sulfoxide concentration: dose (kGy) (blue), ageing post irradiation (green), methionine contact time (days) (purple) and dose rate (kGy h⁻¹) (red). The dotted lines correspond to the significance threshold given by the software MODDE for 95% confidence with p -value < 0.05.

among technologies—gamma rays (2 to 8 kGy h⁻¹), X-ray (13 to 80 kGy h⁻¹), and electron beams (up to 18 000 kGy h⁻¹)—the irradiation itself does not seem affect the production of methionine sulfoxide in multilayer films.

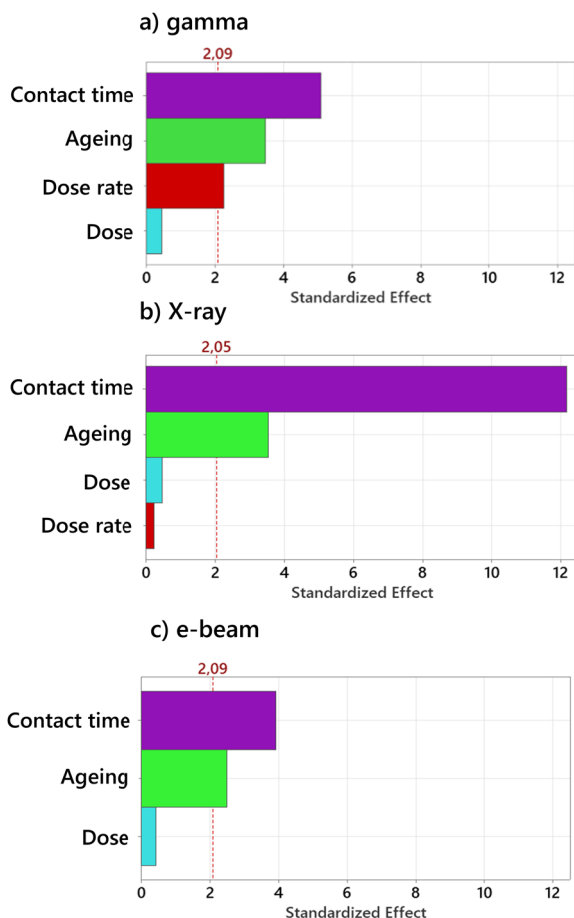


Fig. 7 multilayer film irradiated (a) by gamma (b) by X-ray (c) by e-beam. Pareto analysis of effects on methionine sulfoxide concentration: dose (kGy) (blue), ageing post irradiation (green), methionine contact time (days) (purple) and dose rate (kGy h⁻¹) (red). The dotted lines correspond to the significance threshold given by the software MODDE for 95% confidence with p -value < 0.05.

Variation of dose rate

A multiparametric regression model was performed for each irradiation technology to evaluate whether variations in dose rate within the same technology impact methionine sulfoxide concentration. For e-beam technology, the dose rate is similar for all irradiation sites, making dose rate comparison for this technology impossible. The gamma equation model is given in (2) and the X-ray equation model is given in (3).

$$\begin{aligned} \text{Methionine sulfoxide concentration} = & -1.095 + 0.00252 \cdot \text{Dose} \\ & + 0.1875 \cdot \text{Ageing post irradiation} + 0.03953 \cdot \text{Methionine} \\ & \text{contact time} + 0.0610 \cdot \text{Dose rate} \end{aligned} \quad (2)$$

$$\begin{aligned} \text{Methionine sulfoxide concentration} = & -0.384 - 0.00057 \cdot \text{Dose} \\ & - 0.0750 \cdot \text{Ageing post irradiation} + 0.04569 \cdot \text{Methionine} \\ & \text{contact time} - 0.00028 \cdot \text{Dose rate} \end{aligned} \quad (3)$$

The results from the regression models, depicted in Fig. 7, indicate that both the contact time with the methionine solution and the ageing post irradiation significantly influence methionine oxidation for the three irradiation technologies.

The regression model indicates that, for gamma irradiation (Fig. 7a), the dose rate significantly impacts methionine oxidation (p -value = 0.035). In contrast, for X-ray irradiation (Fig. 7b), the dose rate does not have a significant effect on methionine oxidation (p -value = 0.805).

Single-run and multiple-runs gamma irradiation processes

To complete the study, an HPLC analysis was performed to compare two gamma irradiation processes using the same irradiator at Ionisos in France, maintaining a constant dose rate of 2 kGy h⁻¹. The single run consisted in irradiating continuously samples until reaching the desired dose. The multiple-runs consisted in irradiating samples over multiple

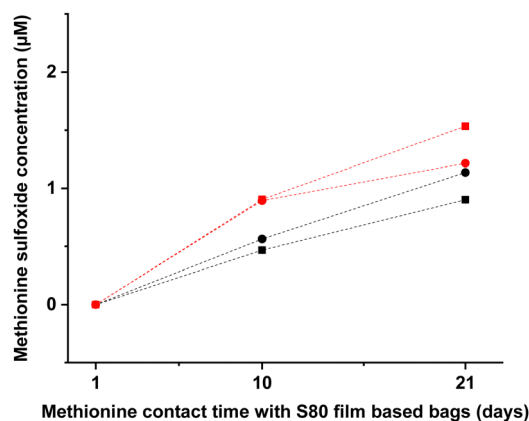


Fig. 8 Methionine sulfoxide concentration (µM) in stored solution for 1, 10 and 21 days, 6 months after irradiation and analyzed by HPLC. Irradiation doses were 30 kGy (square), 50 kGy (circle). The irradiation process conducted in a single run is represented in red, while the process conducted in multiple runs is represented in black. Dotted line is for better reading.



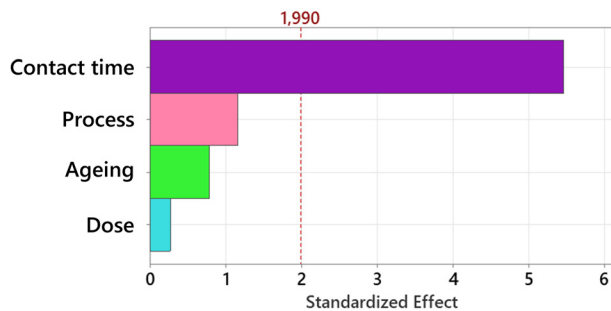


Fig. 9 Pareto analysis of significant effects on methionine sulfoxide concentration: dose (kGy) (blue), ageing post irradiation (green), methionine contact time (days) (purple) and different process of gamma irradiation (pink). The dotted lines correspond to the significance threshold given by the software MODDE for 9% confidence with p -value < 0.05.

runs within the irradiation bunker, with unknown rest times between runs.

Methionine sulfoxide concentration in the solution in contact with the bag was measured by HPLC in samples after 1, 2, 3, 6, 12, 24, and 36 months of irradiation at 30 kGy and 50 kGy (Fig. 8 and Fig. S1 in ESI†).

A regression model is developed to assess whether the irradiation process, post-irradiation ageing, contact time with the solution, and dose significantly impact the concentration of methionine sulfoxide formed.

The regression model (Fig. 9) reveals that the contact time with the solution and the multilayer film significantly influence methionine oxidation. Post-irradiation ageing, dose and processing have no significant effect on methionine oxidation in contact with gamma irradiated samples.

Conclusion

This study provides a comprehensive analysis of the effects of different irradiation technologies on PE/EVOH/PE multilayer films, crucial in the biotechnological and biopharmaceutical industries. ESR analysis highlights the generation of hydroxyalkyl radicals for gamma, X-ray, and electron beam irradiation, with similar decay kinetics observed in radical concentration post-irradiation. The study reveals that the dose rate does not significantly impact methionine sulfoxide formation, and the contact time with methionine solution is the main factor influencing oxidation.

A regression model of different gamma irradiation processes reveals that the concentration of oxidized methionine remains statistically equivalent whatever the processes.

These results highlight that the irradiation technologies have similar effects on PE/EVOH/PE films, as demonstrated with the analysis of doses up to 100 kGy.

The ESR study of the EVA/EVOH/EVA¹ and PE/EVOH/PE multilayer films revealed that hydroxyalkyl radicals are generated by irradiation in both films. Regardless of the technology used, the same radical is produced at equivalent concentrations for each material. The kinetics decays differ between the

two films. In the EVA/EVOH/EVA film, the radicals decay within nine days post-irradiation, whereas in the PE/EVOH/PE film, they persist for up to 60 days. This difference is ascribed to the thickness of the EVOH layer and to the –VOH contents. The thicker EVOH layer in the PE/EVOH/PE film likely favors the regeneration of radicals over time as previously reported.²⁰

The EVA/EVOH/EVA film generates more methionine sulfide in contact with the methionine solution than the PE/EVOH/PE film due to increased formation of peracids. In the EVA/EVOH/EVA film, hydroxyalkyl radicals react with oxygen to form peroxy radicals, which then oxidize methionine. The EVA/EVOH/EVA film produces more carboxylic acids, which react with hydrogen peroxide to form peracids, thereby increasing oxidation. In contrast, the PE/EVOH/PE film generates fewer carboxylic acids, limiting the formation of peracids and thus the oxidation of methionine.^{27,35}

For both films, the contact time of the methionine solution with the film is a significant factor. The dose rate to irradiate the EVA/EVOH/EVA film impacts the methionine oxidation *in fine*. It is not the case with the PE/EVOH/PE film. For both films, process has no significant effect on methionine oxidation in contact with gamma irradiated samples.

The EVA/EVOH/EVA film is impacted similarly by the three irradiation technologies (gamma, X-ray and e-beam). It is also the case for the PE/EVOH/PE film, with a different extent.

Conflicts of interest

There are no conflicts to declare.

Data availability

The data supporting this article have been included as part of the ESI.†

Acknowledgements

N. D., F. G. and S. R. A. M. are thankful to Aix-Marseille Université (AMU), Institut de Recherche pour le Développement (IRD) and Centre National pour la Recherche Scientifique (CNRS) for support.

Notes and references

- B. Krieguer, S. Dorey, N. Dupuy, F. Girard, F. Kuntz, N. Ludwig and S. R. A. Marque, Investigating dose rate effects and reactivities formation in irradiated multilayer films – part 1 EVA/EVOH/E, *Phys. Chem. Chem. Phys.*, 2024, DOI: [10.1039/D4CP04152F](https://doi.org/10.1039/D4CP04152F).
- J. D. Vogel, The Maturation of Single-Use Applications, *BioProcess Int*, 2012, 10–19.



- 3 E. Mahajan, G. Lye and R. Eibl-Schindler, *Bringing polymer Science to Biotechnology Applications: A Single-Use Technology Conference Report*, BioProcess Int, 2018.
- 4 ISO 11137-1; *Sterilization of HealthCare Products-Radiation-Part 1: Requirements for Development, Validation and Routine control of a Sterilization Process for MedicalDevices*. ISO: Geneva, Switzerland, 2006.
- 5 *Guide to Irradiation and Sterilization Validation of Single-Use Bioprocess Systems*, BioProcess Int., 2008.
- 6 N. Dupuy, S. R. A. Marque, L. S. Fifield, M. Pharr, D. Staack, S. D. Pillai, L. Nichols, M. K. Murphy and S. Dorey, *Supplementing Gamma Sterilization with X-Ray and E-Beam Technologies*, *Bioprocess Tech.*, 2022, **20**, 24–28.
- 7 *BPAS Technical Guide, X-rays Sterilization of single-Use BioProcess Equipment, Part 1: Industry Need, Requirements&Risk Evaluation*, 2021.
- 8 P. M. Armenante and O. Akiti, in *Chemical Engineering in the Pharmaceutical Industry*, ed. D. J. Am Ende and M. T. Am Ende, John Wiley & Sons, Inc., Hoboken, NJ, USA, 2019, pp. 311–379.
- 9 D. Darwis, E. Erizal, B. Abbas, F. Nurlidar and D. P. Putra, *Radiation Processing of Polymers for Medical and Pharmaceutical Applications*, *Macromol. Symp.*, 2015, **353**, 15–23.
- 10 H. De Brouwer, *Comparison of the effects of x-ray and gamma irradiation on engineering thermoplastics*, *Radiat. Phys. Chem.*, 2022, **193**, 109999.
- 11 B. Croonenborghs, M. A. Smith and P. Strain, *X-ray versus gamma irradiation effects on polymers*, *Radiat. Phys. Chem.*, 2007, **76**, 1676–1678.
- 12 T. K. Kroc, *Monte Carlo Simulations Demonstrating Physics of Equivalency of Gamma, Electron- beam, and X-ray for Radiation Sterilization*, *Radiat. Phys. Chem.*, 2023, **204**, 110702.
- 13 G. Sadler, W. Chappas and D. E. Pierce, *Evaluation of e-beam, γ - and X-ray treatment on the chemistry and safety of polymers used with pre-packaged irradiated foods: a review*, *Food Addit. Contam.*, 2001, **18**, 475–501.
- 14 W. D. Loveland, D. J. Morrissey and G. T. Seaborg, *Modern Nuclear Chemistry*, Wiley, 1st edn, 2017.
- 15 D. Lowe, L. Roy, M. A. Tabocchini, W. Rühm, R. Wakeford, G. E. Woloschak and D. Laurier, *Radiation dose rate effects: what is new and what is needed?*, *Radiat. Environ. Biophys.*, 2022, **61**, 507–543.
- 16 F. Gaston, N. Dupuy, S. R. A. Marque, M. Barbaroux and S. Dorey, *Impact of γ -irradiation, ageing and their interactions on multilayer films followed by AComDim*, *Anal. Chim. Acta*, 2017, **981**, 11–23.
- 17 F. Gaston, N. Dupuy, S. R. A. Marque, M. Barbaroux and S. Dorey, *One year monitoring by FTIR of γ -irradiated multilayer film PE/EVOH/PE*, *Radiat. Phys. Chem.*, 2016, **125**, 115–121.
- 18 S. Dorey, F. Gaston, N. Dupuy, M. Barbaroux and S. R. A. Marque, *Reconciliation of pH, conductivity, total organic carbon with carboxylic acids detected by ion chromatography in solution after contact with multilayer films after γ -irradiation*, *Eur. J. Pharm. Sci.*, 2018, **117**, 216–226.
- 19 F. Gaston, N. Dupuy, S. R. A. Marque and S. Dorey, *Evaluation of multilayer film stability by Raman spectroscopy after gamma-irradiation sterilization process*, *Vib. Spectrosc.*, 2018, **96**, 52–59.
- 20 G. Audran, S. Dorey, N. Dupuy, F. Gaston and S. R. A. Marque, *Degradation of γ -irradiated polyethylene-ethylene vinyl alcohol-polyethylene multilayer films: An ESR study*, *Polym. Degrad. Stab.*, 2015, **122**, 169–179.
- 21 S. Dorey, F. Gaston, S. R. A. Marque, B. Bortolotti and N. Dupuy, *XPS analysis of PE and EVA samples irradiated at different γ -doses*, *Appl. Surf. Sci.*, 2018, **427**, 966–972.
- 22 S. Dorey, F. Gaston, N. Girard-Perier, N. Dupuy, S. R. A. Marque and L. Delaunay, *Generation of O₂-Permeation Barrier during the Gamma-Irradiation of Polyethylene/Ethylene-Vinyl Alcohol/Polyethylene Multilayer Film*, *Ind. Eng. Chem. Res.*, 2019, **58**, 14115–14123.
- 23 N. Girard-Perier, M. Claeys-Bruno, S. R. A. Marque, N. Dupuy, F. Gaston and S. Dorey, *Effects of X-ray, electron beam and gamma irradiation on PE/EVOH/PE multilayer film properties*, *Chem. Commun.*, 2021, **57**, 11049–11051.
- 24 N. Girard-Perier, F. Gaston, N. Dupuy, S. R. A. Marque, L. Delaunay and S. Dorey, *Study of the mechanical behavior of gamma-irradiated single-use bag seals*, *Food Packag. Shelf*, 2020, **26**, 100582.
- 25 N. Girard-Perier, M. Claeys-Bruno, S. R. A. Marque, N. Dupuy, F. Gaston and S. Dorey, *Effects of X-ray, electron beam and gamma irradiation on PE/EVOH/PE multilayer film properties*, *Chem. Commun.*, 2021, **57**, 11049–11051.
- 26 F. Gaston, N. Dupuy, N. Girard-Perier, S. R. A. Marque and S. Dorey, *Comprehensive investigation on physical and chemical properties of γ -irradiated multilayer PE/EVOH/PE film: A multiscale approach*, *Appl. Res.*, 2023, **2**, e202200065.
- 27 N. Girard-Perier, M. Claeys-Bruno, S. R. A. Marque, N. Dupuy, F. Gaston and S. Dorey, *Monitoring of peroxide in gamma irradiated PE/EVOH/PE multilayer film using methionine probe*, *Food Bioprod. Process*, 2022, **132**, 226–232.
- 28 N. Girard-Perier, S. Dorey, F. Gaston, F. Girard, S. R. A. Marque and N. Dupuy, *One-year ageing FTIR monitoring of PE/EVOH/PE film after gamma or electron beam irradiation*, *Polym. Degrad. Stab.*, 2022, **195**, 109790.
- 29 N. Girard-Perier, S. R. A. Marque, N. Dupuy, B. Krieguer and S. Dorey, *Gamma, E-Beam and X-ray Irradiations on PE/EVOH/PE Multilayer Film: An Industrial Point of View Regarding the Impact on Mechanical Properties*, *Polymers*, 2023, **15**, 2799.
- 30 T. Pędzinski, K. Grzyb, K. Skotnicki, P. Filipiak, K. Bobrowski, C. Chatgililoglu and B. Marciniak, *Radiation- and Photo-Induced Oxidation Pathways of Methionine in Model Peptide Backbone under Anoxic Conditions*, *Int. J. Mol. Sci.*, 2021, **22**, 4773.
- 31 V. Gold, *The IUPAC Compendium of Chemical Terminology: The Gold Book*, International Union of Pure and Applied Chemistry (IUPAC), Research Triangle Park, NC, 4th edn, 2019.



- 32 M. A. Rosenfeld, L. V. Yurina and A. D. Vasilyeva, Antioxidant role of methionine-containing intra- and extracellular proteins, *Biophys. Rev.*, 2023, **15**, 367–383.
- 33 D. Tavella, D. R. Ouellette, R. Garofalo, K. Zhu, J. Xu, E. O. Oloo, C. Negron and P. M. Ihnat, A novel method for in silico assessment of Methionine oxidation risk in monoclonal antibodies: Improvement over the 2-shell model, *PLoS One*, 2022, **17**, e0279689.
- 34 A. G. Davies, J. A. Howard and M. Lehnig, *Magnetic properties of free radicals- Organic O-, P-, Se-, Si-, Ge-, Sn-, Pb-, As-, Sb-Centered Radicals*, 1979, vol. 9.
- 35 N. Girard-Perier, M. Claeys-Bruno, S. R. A. Marque, N. Dupuy, F. Gaston and S. Dorey, Monitoring of Peroxide in Gamma Irradiated EVA Multilayer Film Using Methionine Probe, *Polymers*, 2020, **12**, 3024.

