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Recyclable mono materials for packaging of fresh chicken fillets: New design for recycling in circular economy

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The focus on sustainability and circular economy is leading to a need for development of new food packaging concepts, including recyclable materials that ideally consist of a single material in a monolayer system. This research was focused on the possibility of replacing complex multilayered material [amorphous polyethylene terephthalate/polyethylene (APET/PE)] with simple recyclable mono material [high-density polyethylene (HDPE)] for packaging of chicken fillets in modified atmosphere packaging (CO_2/N_2: 60%/40%). Bacterial growth measured as total viable count (TVC), lactic acid bacteria and *Enterobacteriaceae*, *Brochothrix thermosphacta* and *Escherichia coli* for chicken fillets packed in HDPE mono materials was compared with chicken fillets packed in APET/PE.

TVC increased during the storage period (24 days) with high level of TVC count (7 $\log_{10}$ CFU/g) recorded at Days 19–20 of storage in both HDPE and APET/PE material. No significant differences were recorded in off-odour between chicken stored in APET/PE compared with HDPE in CO_2/N_2 atmosphere during the storage period (samples were regarded as acceptable on the 24th day of storage). The drip loss increased in all samples during storage, and no significant differences between samples stored in different materials were recorded.

Significant differences in bacterial growth were recorded between samples with different gas volume to product volume (G/P) ratio (Day 17), implying that higher G/P ratio is resulting in lower TVC count. The lowest G/P ratio caused the highest drip loss, whereas addition of CO_2 emitter reduced the drip loss to some extent.

This research is very encouraging as it provides new insight into the use of monolayer materials as well as the importance of design for recycling in circular economy.

KEYWORDS
active packaging, circular economy, design for recycling, food packaging, recyclable, shelf life

1 | INTRODUCTION

Food waste reduction is one of the global targets of the United Nations development goal 12 to ensure responsible consumption and production patterns.[1] Food packaging with its all noteworthy functions is of high importance for sustainable development and food waste reduction.[2] On the other hand, food packaging also presents an environmental challenge, relying on non-renewable resources and use of non-recyclable materials.[3,4] Close to 40% of plastic used in Europe are used for packaging, whereas less than 40% of packaging materials

are recycled.[5] In this manner, it is very challenging to find optimal food packaging solutions for reduction of food waste.[6] Maintaining the shelf life and the quality and safety of food substrate while reducing food packaging waste is a task to be solved.[6–8]

Packaging innovations moved towards development of advanced materials with good mechanical, thermophysical and barrier properties.[9] In the last decades, the industry has focused on optimization in packaging processes and reducing thickness, but this has often resulted in increased use of complex structures including polyamide (PA). PA has significantly higher environmental impact per kilogramme than polyolefins.[10] In most cases, these materials were developed as complex structures to provide specific requirements of diverse food substrates, for example, laminated structures with multiple layers containing different materials types.[11] One of the main reasons for using complex structure is their gas barrier properties, which are needed in combination with modified atmosphere packaging (MAP) and of importance related to product shelf life (specific for long storage time). Recycling of these multilayered materials is very often not practical or possible; thus, the materials are ending in landfill and/or being incinerated. Research and development nowadays are moving to packaging solutions for sustainable development like recyclable mono materials. However, these materials are rarely suitable for packaging of perishable and very sensitive food substrates.[12]

Improvements in material properties for simpler mono materials can be compensated by design of packaging itself [e.g., by introducing packaging conditions through a modified atmosphere or by using active packaging (AP) solutions]. AP is an innovative way for shelf life extension and/or improvement of food packaging conditions. These systems are roughly divided in two groups, scavengers and emitters, and their activity is designed in accordance with specific food substrate and its properties.[13]

Chicken meat is consumed worldwide because of its good nutritional quality, high-quality proteins and low total fat content.[14,15] However, chicken has a relatively short shelf life under refrigerator conditions and thus considered as a highly perishable food product.[14,16–18] New techniques in prolonging fresh chicken shelf life under refrigerating conditions have been extensively researched. MAP has been recognized as a very efficient non-thermal technique for preservation of fresh products. Optimal combination of gases inside of packaging is preventing spoilage and extending the product shelf life.[14,15,17–23] Common gas mixture for packaging of fresh chicken fillets in Norway consists of CO_2 (50%–70%) and N_2 (50%–30%),[23] whereas high content of oxygen combined with CO_2 (50%–75% O_2/50%–25% CO_2) is often applied for meat and also poultry in many countries in Europe. However, it is very common to have some amount of residual oxygen related to vacuum time/flushing/production speed, as well as to the size/volume of the packages and flexibility of the material. One possibility to exclude residual oxygen from the package is addition of oxygen scavengers resulting in extended shelf life.[14] High amounts of CO_2 can, on the other hand, be dissolved in the meat product and cause collapse of the packaging. Minimal concentration of CO_2 should be ~25%; however, it is well known that higher amounts of CO_2 will result in shelf life extension of chicken meat.[21,24] Besides concentration of CO_2 and food substrate, dissolution of CO_2 depends on the quantity of the gas introduced into the package. For that reason, gas volume to product volume (G/P) ratio in the package is also very important for the effective MAP.[25] Due to this fact, besides oxygen scavengers, CO_2 emitters can be used as AP solution for extended shelf life of packed chicken fillets.

The aim of this research was to identify possible substitution of complex multilayered amorphous polyethylene terephthalate/polyethylene (APET/PE) (laminate) with high-density polyethylene (HDPE) mono material without jeopardizing the quality of packed food. Evaluation of the effect of mono materials on food quality and shelf life was conducted through packaging of fresh chicken fillets. MAP and AP solutions (O_2 absorbers and CO_2 emitters) were used to overcome withdrawals in material characteristic to keep initial food quality and safety.

2 | MATERIALS AND METHODS

2.1 | Packaging

Chicken breast fillets were obtained from a commercial processing plant (Nortura, Hærland, Norway), and the packaging was performed within 24 h after slaughtering. This study was performed with HDPE trays (Berry Bebo, Kristiansand, Norway) and APET/PE trays based on APET/PE (Multipet) 550 μm sheet manufactured by Wipak (Nastola, Finland) and thermoformed by JiHå Plast AB (Karlskoga, Sweden). HDPE was chosen as material to evaluate the potential for using material with low oxygen barrier properties both in relation to the effect on the food quality and shelf life and the efficiency of the oxygen scavenger.

All trays were sealed with the same top web, Biaxer 65 XX AFM (Wipak, Nastola, Finland), consisting of biaxial-oriented polyethylene terephthalate/polyethylene/ethylene vinyl alcohol/polyethylene (BOPET/PE/EVOH/PE) with 65-μm thickness and an oxygen transmission rate (OTR) of 5 cm^3/m^2*d*bar (23°C at 50% RH) (stated in data sheet). OTR for the APET/PE web before forming was 7 cm^3/m^2*d*bar (23°C at 50% RH). Information about the packaging materials including the OTR used for the chicken breast fillets are given in Table 1.

Liquid absorber (absorber type MP-2501-70, Faerch, Denmark) or a CO_2 emitter (emitter type XPC-40-170-080130-70-037, produced by McAirlaid's Vliesstoffe GmbH, Berlingerode, Germany, and delivered from Bewi Tommen Gram, Trondheim, Norway) was added into the trays. The producer of the emitter was given all information about the packaging concept (volume of tray, G/P ratio and gas composition) in order to provide optimal capacity of the emitters. However, the same capacity of the CO_2 emitters was provided and used for all packages independent of tray volume and gas/product ratio. An oxygen scavenger (scavenger type ZPT, Mitsubishi Gas Chemical, Tokyo, Japan) was combined with a liquid absorber or a CO_2 emitter for some of the samples. The type of O_2

TABLE 1 List of selected packaging materials with relevant information: Polymer type, tray volume, dimensions, weight (n = 5) and thickness (n = 5) and OTR (n = 4)

Tray no.	Tray material	Tray volume	Tray dimension (bottom) (top)	Tray height	Tray weight	Tray thickness		OTR packages
		(ml)	(L*W) (mm)	(H) (mm)	(g)	Bottom (μm)	Wall (μm)	23°C, 100% internal RH, 50% RH outside (ml O_2/pkg*day)
1	HDPE	600	180*118 194*132	25	23.0 ± 0.5	700 ± 28	592 ± 45	4.1 ± 0.1
2	HDPE	1200	176*114 194*132	50	35.3 ± 0.4	884 ± 61	715 ± 66	3.7 ± 0.2
3	APET/PE	1200	176*114 194*132	50	21.9 ± 0.0	377 ± 4	310 ± 14	0.8 ± 0.3

Abbreviations: APET/PE, amorphous polyethylene terephthalate/polyethylene; HDPE, high-density polyethylene; OTR, oxygen transmission rate.

scavenger was selected according to product information from the producer and if the scavenger was suitable for food contact and complies with European regulations. The O_2 scavenger should not be in direct contact with the drip loss from the product; thus, the scavenger was placed in a small cup of aluminium inside the trays. Prior to the shelf life experiment of chicken breast fillets, a pre-experiment for evaluation of oxygen scavengers was performed. The pre-evaluation consisted of gas composition (oxygen level) evaluation of empty packages and evaluation of packages with oxygen scavengers intended to be used in the shelf life experiment.

Packaging of chicken fillets was performed on a tray sealing machine, Multivac T200 (Multivac, Wolfertschwenden, Germany). The most commonly used gas composition for meat in Norway, 60% CO_2/40% N_2, was applied. In addition, the often-applied modified atmosphere (MA) for meat packaging in Europe (25% CO_2/75% O_2) was added for some samples of HDPE and APET/PE (both 1200 ml) for comparison. Both gases were supplied as food grade pre-mixtures (AGA, Oslo, Norway). Three different G/P ratios (1.0, 1.5 and 2.6) were obtained using trays with different volumes (600 or 1200 ml) and/or different product weight (320–500 g). The G/P ratio 1.5 was selected based on the G/P ratio currently used by the producer. All samples were stored in the dark at 4 ± 0.8°C with sampling time after 8, 13, 17, 21 and 24 days. Triplicate analyses were carried out.

Tray material, tray volume, G/P ratio, product weight, gas composition and the use of liquid absorber and AP [O_2 scavenger (scav.) and/or CO_2 emitter (emit)] applied in the experiment are given in Table 2.

TABLE 2 Experimental design: Overview of packaging material, volumes, G/P ratio, gas composition and active packaging used for the experiment

Sample no.	Tray material	Tray volume (ml)	G/P ratio	Product weight (g)	Packaging atmosphere	Liquid absorber	Active packaging
1	HDPE	600	1.0	320	60%CO_2/40%N_2	Yes	None
2	HDPE	600	1.0	320	60%CO_2/40%N_2	Yes	O_2 scavenger
3	HDPE	600	1.0	320	60%CO_2/40%N_2	No	O_2 scavenger + CO_2 emitter
4	HDPE	1200	1.5	500	60%CO_2/40%N_2	Yes	None
5	HDPE	1200	1.5	500	60%CO_2/40%N_2	Yes	O_2 scavenger
6	HDPE	1200	1.5	500	60%CO_2/40%N_2	No	O_2 scavenger + CO_2 emitter
7	HDPE	1200	2.6	350	60%CO_2/40%N_2	Yes	None
8	HDPE	1200	2.6	350	60%CO_2/40%N_2	Yes	O_2 scavenger
9	HDPE	1200	2.6	350	60%CO_2/40%N_2	No	O_2 scavenger + CO_2 emitter
10	APET/PE	1200	1.5	500	60%CO_2/40%N_2	Yes	None
11	HDPE	1200	1.5	500	75%O_2/25%CO_2	Yes	None
12	APET/PE	1200	1.5	500	75%O_2/25%CO_2	Yes	None

Abbreviations: APET/PE, amorphous polyethylene terephthalate/polyethylene; HDPE, high-density polyethylene.

2.2 | Headspace gas composition

The headspace atmosphere of packages was analysed for CO_2 and O_2 levels immediately after packaging and at each sampling time to check for leakage. The content of $\%CO_2$ and $\%O_2$ was determined using a CheckMate 9900 O_2/CO_2 analyser (PBI Dansensor, Ringsted, Denmark). Gas was sampled from the packages using a needle through self-sealing patches on the packages. In addition, 10 packages with chicken fillet were followed throughout the storage time for gas evaluation. The gas composition of those packages was measured at Days 0, 3, 6, 8, 10, 13, 15, 17, 21 and 24 after packaging to follow the effect of AP. At Days 0, 3, 10, 17 and 24, all 10 packages were evaluated. For the rest of the days, only five of them were evaluated to ensure enough headspace and to avoid leakages throughout the storage time. In addition, pre-evaluation of AP solutions activity (scavengers and emitters) was performed.

2.3 | Drip loss

Drip loss was determined by initially weighing the chicken fillets and the package with liquid absorber /CO_2 emitter /O_2 scavenger and calculating the increase in weight of the packages (including the absorber/CO_2 emitter/O_2 scavenger) at each sampling time. Results are given as the percentage (%) of initial product weight and refer to the corresponding drip loss from the sample.

2.4 | Off-odour evaluation

Off-odour evaluation of the chicken fillets was performed by a lab panel of three to six experienced assessors at sampling days 8, 13, 17, 21 and 24 of storage. The panel was trained in using the actual method. Prior to the experiment and evaluation, the assessors were trained and agreement in freshness was discussed (meaning highest intensity of fresh odour). The samples were ranked on a scale of 1 to 5: 5 indicates a fresh product, 3 indicates alterations of the product, but still acceptable, whereas below 3 indicates an unacceptable product (similarly as performed at the producer). Fresh chicken fillet (stored at −20°C and thawed for 24 h at 4°C the day before off-odour evaluation) was used as a reference. The samples, including two fresh fillets, were served in random order, coded with a three-digit number.

2.5 | Appearance of package

At the time of packaging, the top web in packages with CO_2 is often almost flat or sometimes more gas is added, resulting in slightly convex packages. CO_2 is dissolved into the product and followed by a reduction in the CO_2 content in the headspace, leading to underpressure in rigid packages.[26] The appearance of the packages and top web of the samples stored in the trays was evaluated by using a scale of 0 to 6 where 0 was defined as packages with extremely underpressure (concave), 3 was neutral and 6 was defined as packages with extremely overpressure (convex).[24]

2.6 | Microbiological analyses

Microbiological analyses were performed on chicken breast fillets at the time of packaging (Day 0) and at each sampling time. A piece of 3×3 cm^2 and 1-cm depth (approximately 10 g) was cut, weighed, diluted 1:10 with peptone water and macerated for 1 min. Around 100 μl of an appropriate 10-fold dilution was spread out on an agar plate either by using a sterile L-shaped rod (when expected bacterial number to be between log 2 and log 5 CFU/g) or by using Whitley automatic spiral plater (WASP) (Don Whitley Scientific Ltd., West Yorkshire, UK; when expected bacterial number to be more than log 5 CFU/g) on the following agar plates:

- Plate count agar (PCA; Oxoid CM 0463) for total viable counts (TVCs); spread plate, incubation at 30°C for 72 h, both aerobic and anaerobic conditions (method according to NMKL no. 86).
- Man–Rogosa–Sharke (MRS) agar (Oxoid CM 361) for lactic acid bacteria (LAB); spread plate, incubation at 25°C for 2–5 days, aerobic condition (method according to NMKL no. 140).
- Streptomycin thallous acetate actidione (STAA) agar base (CM 0881 with selective supplement SR 0151E, Oxoid, Hampshire, England) for determination of *Brochothrix thermosphacta*; spread plate, incubation at 25°C for 48 h, aerobic condition (method according to NMKL no. 141).
- Chromagar (Day 0), brilliance *Escherichia coli*/coliform medium (Oxoid Microbiology Product, Thermo Fisher Scientific, Corporate UK) was applied for determination of *(E. coli)*; spread plate, incubation at 37°C for 18–24 h, aerobic condition.
- Violet red bile glucose agar (VRBGA) (Oxoid CM 1082, Hampshire, UK) for *Enterobacteriaceae*; incubation at 37°C for 24 h, semi-aerobic conditions, cells embedded in pour plate agar with sterile overlay (method according to NMKL no. 144). A 1000-ul appropriate dilution was transferred to a sterile Petri dish.

Microbial counts are expressed as average colony forming units per gram (CFU)/g.

2.7 | Statistical analyses

All statistical analyses were performed in Minitab 18 (ref). One-way ANOVA was performed for all responses (bacterial counts, off-odour, drip loss and appearance) at each sampling time. In addition, evaluation of significant differences was performed using general linear model (GLM) ANOVA for one set of the samples including the nine samples stored in HDPE with different G/P ratio (G/P), AP (A) and all storage time (T) (8, 13, 17, 21 and 24 days). The model included the main effects (G/P, A and T) and their interactions.

3 | RESULTS AND DISCUSSION

3.1 | Potential for substitute complex materials with recyclable mono materials

The aim of this study was to evaluate the possibilities to replace non-recyclable complex multilayered materials with recyclable mono materials and if AP solutions (O_2 scavengers and CO_2 emitter) are needed to obtain similar shelf life. In our former work,[12] complex materials have been successfully replaced with mono materials for packaging of hotdogs (cooked processed sausages). However, hotdogs are processed products and less perishable than fresh meat.

In the pre-experiment, trays with oxygen scavengers had different levels of oxygen right after packaging (0.0, 0.6 and 1.1% O_2). The scavengers removed the oxygen from all trays and the oxygen level was close to zero throughout the entire experiment (10–14 days).

Evaluation of gas composition and activity of AP (oxygen scavengers and CO_2 emitters) was performed in empty trays and trays with chicken fillets. The oxygen scavengers were active and absorbed all the oxygen the entire test period for both empty packaging and packaging with product.

The content of oxygen in trays with chicken was below 0.3% for all samples (Table 3).

CO_2 level was reduced in all samples during storage time. The difference in OTR between the trays was not reflected in the O_2 or CO_2 concentrations during storage. In the samples with CO_2/N_2 atmosphere, the residual amount of O_2 after packaging was very low (below 0.02) as given in Table 3. In samples with oxygen scavenger and for samples with lower G/P ratio (1.0 and 1.5), the CO_2 level decreased below 40% after 8 days followed by a further reduction. Even in samples with CO_2 emitters, the level of CO_2 was reduced to 42%–49%. This reduction of CO_2 indicates that the capacity of the emitters was not completely optimized. Gas composition test confirmed activity of oxygen scavengers and CO_2 emitters through the whole storage time.

Poultry are perishable food products with relatively short shelf life; thus, technologies aiming at shelf life extension, increased transport efficiency and economic and environmental impact are of interest to the industry.[18] Microbial growth is the main reason for deterioration of fresh product such as chicken and main indicator of food safety. TVC (aerobic and anaerobic), LAB and *Enterobacteriaceae* count for chicken breast fillets packed in HDPE mono materials, compared with reference APET/PE, are presented in Table 4. For comparison reason, both materials with high oxygen gas composition were also packed and stored in same conditions during storage period (4°C; 24 days).

Initial TVC (aerobic) of chicken breast fillets was 2.3 ± 0.4 $\log_{10}$ CFU/g, similar levels as reported by others.[15,20] TVC increased during the whole storage period (24 days) for all samples as expected. Development of TVC until it reached relatively high level of bacteria (7 $\log_{10}$ CFU/g[26]) for APET/PE was at Days 19–20, and similar results were detected when chicken was stored in HDPE. This is profoundly different to what was reported by Patsias et al.[27] where 7 $\log_{10}$ CFU/g of TVC was reached after 10–12 days in CO_2/N_2 atmosphere. However, in their study, the initial level of TVC was 4.3–4.6 compared with our 2.3 $\log_{10}$ CFU/g, in addition to only 30% CO_2 in the gas, compared with the presented study.

On the 13th day of storage, significant difference was recorded between chicken stored in APET/PE in CO_2/N_2 and chicken stored in high oxygen (high TVC count on high oxygen). Within the 17th and 21st day of storage, no significant difference between samples was recorded, whereas on the last storage day, significantly lower level of TVC was recorded for chicken stored in APET/PE in CO_2/N_2 compared with chicken stored in HDPE in high oxygen. As can be seen, the differences were more related to the differences in gas composition than material type and will not be discussed further. However, no significant difference between samples packed in different material types and same gas composition was recorded.

TVC (anaerobic) initial value was 2.1 ± 0.1 $\log_{10}$ CFU/g. As given in Table 4, the growth of TVC in anaerobic incubation condition was similar to growth of TVC at aerobic incubation. No significant difference between listed samples was recorded ($p > 0.05$) (Table 4). TVC (anaerobic) for chicken stored in APET/PE reached 7 $\log_{10}$ CFU/g on Days 17–18, whereas in HDPE, similar level of bacteria was reached after 19–20 days. Our results are comparable with results reported by Holck et al.[24] for chicken stored in 60%CO_2/40%N_2 with G/P ratio 1.4 and 2.5. They also reported that increase of the partial pressure of CO_2 in the gas resulted in progressively reduction in the growth rate of TVC.

LAB and *Enterobacteriaceae* count followed the same trend as TVC. According to Vihavainen et al.,[28] storage in anaerobic and CO_2-rich atmosphere often leads to LAB being the dominant flora in the first part of the storage time, and *Pseudomonas* and *Enterobacteriaceae* may constitute a substantial part in last phase of the storage period.[23] This is in accordance to the results in our study.

B. thermosphacta represent common spoilage bacteria for chicken breast fillets.[20,23] It produces metabolites influencing on product off-odour and flavour, thus influencing the product shelf life. Generally, when oxygen is removed, growth of spoilage bacteria like *Pseudomonas* is inhibited, whereas *Enterobacteriaceae* and *B. thermosphacta* may dominate in 50%CO_2/50%N_2.[29] According to the gas composition in the packages in our study, the level of oxygen was below 0.3% through the whole storage time. In this study, counts of *B. thermosphacta* and *Escherichia coli* were under the limit of detection for all combinations and will not be discussed further.

According to one-way ANOVA, there were no significant differences in counts of TVC between samples packed in different material types during the whole storage period. Chicken stored in APET/PE trays with oxygen-rich atmosphere had significant higher level of TVC (aerobic) compared with storage in CO_2/N_2 atmosphere on the 13th day of storage. This was also the case for level of LAB. Similar counts in LAB in the two gas atmospheres could be expected as bacteria such as *Lactobacillus* may grow in both aerobic and anaerobic conditions.[30] According to Liang et al.,[31] *Pseudomonas* sp, *Brochothrix* sp and *Carnobacterium* sp comprise the dominant spoilage bacteria in fresh chicken breast under aerobic conditions stored at 4°C. In our study,

TABLE 3 Gas composition (O_2 and CO_2) in packages with chicken fillets stored in different G/P ratio (1.0, 1.5 and 2.6) and different gas compositions (60%/40% CO_2/N_2 and 75% /25% O_2/CO_2) with no active packaging (AP), with oxygen scavenger (scav.) and oxygen scavenger combined with CO2 emitter (emitter) during the storage period (0–24 days)

		HDPE G/P 1.0 60%/40% CO_2/N_2			HDPE G/P 1.5 60%/40% CO_2/N_2			HDPE G/P 2.6 60%/40% CO_2/N_2			APET/PE G/P 1.5 60%/40% CO_2/N_2			HDPE G/P 1.5 75%/25% O_2/CO_2	APET/PE G/P 1.5 75%/25% O_2/CO_2
	Storage days	No AP	Scav.	Scav. + emitter	No AP	Scav	Scav. + emitter	No AP	Scav.	Scav. + emitter	No AP	Scav.	Scav. + emitter	No AP	No AP
O_2%	0	0.02 ± 0.00	0.02 ± 0.01	0.01 ± 0.00	0.01 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.01 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.01 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	72.78 ± 0.00	72.98 ± 0.00
	8	0.17 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.02 ± 0.02	0.00 ± 0.00	0.00 ± 0.00	0.06 ± 0.02	0.00 ± 0.00	0.00 ± 0.00	0.01 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	75.70 ± 1.45	75.00 ± 0.98
	13	0.23 ± 0.03	0.00 ± 0.00	0.00 ± 0.00	0.03 ± 0.03	0.00 ± 0.00	0.00 ± 0.00	0.08 ± 0.01	0.00 ± 0.00	0.00 ± 0.00	0.01 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	72.17 ± 1.51	71.63 ± 0.81
	17	0.10 ± 0.01	0.00 ± 0.00	0.00 ± 0.00	0.02 ± 0.02	0.00 ± 0.00	0.00 ± 0.00	0.07 ± 0.02	0.00 ± 0.00	0.00 ± 0.00	0.02 ± 0.02	0.00 ± 0.00	0.00 ± 0.00	71.50 ± 0.53	70.57 ± 0.76
	21	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.05 ± 0.05	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	66.33 ± 4.51	66.13 ± 1.45
	24	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	56.03 ± 2.92	60.30 ± 2.46
CO_2%	0	60.09 ± 0.39	59.39 ± 0.67	60.45 ± 0.29	59.93 ± 0.24	60.16 ± 0.41	60.65 ± 0.25	60.69 ± 0.23	60.40 ± 0.18	60.84 ± 0.18	59.98 ± 0.30	60.38 ± 0.35	60.91 ± 0.18	22.36 ± 0.09	22.58 ± 0.00
	8	31.07 ± 1.02	24.67 ± 1.07	48.90 ± 1.54	34.90 ± 0.26	31.07 ± 1.16	42.17 ± 0.15	42.93 ± 0.35	41.33 ± 0.93	47.80 ± 1.44	35.07 ± 1.20	32.57 ± 0.71	46.97 ± 2.25	19.63 ± 1.21	20.67 ± 0.86
	13	27.20 ± 1.30	22.80 ± 1.20	47.83 ± 0.64	34.20 ± 0.10	30.93 ± 1.12	41.73 ± 0.90	41.97 ± 0.21	39.90 ± 0.95	48.73 ± 0.61	35.60 ± 0.92	33.77 ± 0.64	44.90 ± 1.13	22.03 ± 1.33	22.93 ± 0.58
	17	26.77 ± 0.35	22.67 ± 0.60	44.43 ± 1.26	33.10 ± 1.04	30.50 ± 0.26	42.97 ± 1.27	41.40 ± 0.56	38.17 ± 1.87	49.10 ± 1.28	35.27 ± 0.50	32.43 ± 0.99	45.67 ± 0.80	22.60 ± 0.35	24.40 ± 0.52
	21	26.97 ± 1.10	22.87 ± 0.15	43.33 ± 1.86	33.37 ± 1.00	29.87 ± 0.42	42.30 ± 1.35	39.67 ± 1.10	37.60 ± 0.44	48.80 ± 2.10	34.83 ± 1.19	32.87 ± 1.12	46.43 ± 0.97	26.77 ± 3.73	28.43 ± 1.17
	24	28.57 ± 0.83	25.77 ± 0.45	42.47 ± 2.64	33.87 ± 0.67	30.53 ± 0.90	42.23 ± 1.15	39.60 ± 0.10	37.50 ± 0.78	49.07 ± 1.36	36.30 ± 0.56	34.07 ± 1.25	45.47 ± 1.91	35.77 ± 2.33	31.90 ± 2.42

Abbreviations: APET/PE, amorphous polyethylene terephthalate/polyethylene; HDPE, high-density polyethylene.

TABLE 4 Microbial count given as log CFU/g for APET/PE and HDPE with G/P 1.5 samples and different gas compositions (60%/40% CO_2/N_2 and 75%/25% O_2/CO_2) during the storage period (0–24 days)

	TVC (aerobic) (log CFU/g)				TVC (anaerobic) (log CFU/g)				LAB (log CFU/g)				*Enterobacteriaceae* (log CFU/g)			
	60%/40% CO_2/N_2		75% /25% O_2/CO_2		60%/40% CO_2/N_2		75% /25% O_2/CO_2		60%/40% CO_2/N_2		75% /25% O_2/CO_2		60%/40% CO_2/N_2		75% /25% O_2/CO_2	
	APET/PE	HDPE	APET/PE	HDPE	APET/PE	HDPE	APET/PE	HDPE	APET/PE	HDPE	APET/PE	HDPE	APET/PE	HDPE	APET/PE	HDPE
0	2.3 ± 0.4	2.3 ± 0.4	2.3 ± 0.4	2.3 ± 0.4	2.1 ± 0.1	2.1 ± 0.1	2.1 ± 0.1	2.1 ± 0.1	2.0 ± 0.0	2.0 ± 0.0	2.0 ± 0.0	2.0 ± 0.0	1.2 ± 0.2	1.2 ± 0.2	1.2 ± 0.2	1.2 ± 0.2
8	**4.2^{B} ± 0.1**	4.6 ± 0.6	4.9 ± 0.1	**5.3^{A} ± 0.4**	4.2 ± 0.1	4.6 ± 0.6	4.9 ± 0.0	5.3 ± 0.6	4.0 ± 0.4	4.5 ± 0.5	4.3 ± 0.8	4.8 ± 0.6	2.4 ± 0.2	3.1 ± 0.7	2.3 ± 0.2	3.1 ± 0.6
13	**5.7^{B} ± 0.5**	6.5 ± 0.5	**6.9^{A} ± 0.0**	6.3 ± 0.5	5.7 ± 0.5	6.3 ± 0.6	6.7 ± 0.1	6.2 ± 0.4	**5.5^{B} ± 0.1**	6.2 ± 0.4	**6.5^{A} ± 0.0**	6.0 ± 0.4	4.0 ± 0.7	3.9 ± 1.0	4.0 ± 0.3	3.0 ± 0.9
17	6.4 ± 0.2	6.6 ± 0.3	7.1 ± 0.3	6.9 ± 0.3	6.4 ± 0.3	6.8 ± 0.5	6.9 ± 0.8	6.8 ± 0.2	5.7 ± 0.5	5.8 ± 0.9	6.8 ± 0.7	6.3 ± 0.7	4.6 ± 0.7	5.1 ± 0.6	4.6 ± 0.2	4.1 ± 0.5
21	7.3 ± 0.5	7.1 ± 0.5	7.4 ± 0.1	7.0 ± 0.3	7.3 ± 0.5	7.1 ± 0.5	7.3 ± 0.5	6.9 ± 0.3	6.3 ± 0.4	6.4 ± 0.6	6.6 ± 0.4	5.9 ± 0.4	6.0 ± 0.6	5.7 ± 0.7	5.0 ± 0.3	4.6 ± 0.9
24	**6.8^{B} ± 0.6**	7.3 ± 0.3	7.8 ± 0.4	**8.2^{A} ± 0.2**	6.8 ± 0.4	7.4 ± 0.3	7.3 ± 0.5	7.2 ± 1.7	6.5 ± 0.3	6.6 ± 0.6	6.6 ± 0.8	7.4 ± 0.1	5.9 ± 0.5	6.3 ± 0.4	5.8 ± 0.1	6.5 ± 0.6

Note. Statistical analysis of variance (one-way ANOVA) has been performed for each analysis and within each sampling time. Means that are statistically different ($p > 0.05$) are presented bold with letters. Samples with different letters belonging to different Tukey Groups and are significantly different.

Abbreviations: APET/PE, amorphous polyethylene terephthalate/polyethylene; HDPE, high-density polyethylene; LAB, lactic acid bacteria; TVC, total viable count.

LAB and *Enterobacteriaceae* constituted a major part of the spoilage bacteria, but as described above, counts of *B. thermosphacta* were under the limit of detection (<2) also for chicken stored under aerobic conditions in oxygen-rich gas atmosphere.

In relation to microbiology, off-odour is also of great importance, as many volatile substances are produced by microbiological metabolism, causing unpleasant off-odour. MAP can have positive impact in this manner, depending on gas composition. In particular, CO_2 inhibits growth of bacteria that can make off-flavours in chicken breast fillets.[23] Influence of different materials and gas compositions on off-odour of packed chicken breast fillets is presented in Table 5.

The off-odour was evaluated with decreasing score during storage. It must be noted that there were no significant differences in off-odour between chicken stored in APET/PE and in HDPE in CO_2/N_2 atmosphere during the whole storage period. On the 24th day, chicken stored in APET/PE and HDPE had similar score (3 and 3.1, respectively), indicating alteration of product but still acceptable. This result is in accordance with the microbiology results (TVC aerobic count). In addition, samples packed in high oxygen were unacceptable in off-odour (1.7) for both materials (Table 5.) as well as high level of TVC for these samples after 24 days of storage (7.8 log CFU/g APET/PE and 8.2 for HDPE) (Table 4.)

Chicken fillets, with high content of nutrients and water, are prone to drip loss during storage. In addition to possibly having an economical effect, visible liquid due to drip loss is undesirable and could lead to rejection by the consumers as drip loss may give an unpleasant appearance. The drip loss increased somewhat in all samples during storage. In the first part of storage time, apparently lower drip loss was measured for chicken stored in HDPE compared with APET/PE in CO_2/N_2 atmosphere (1.6 and 2.1, respectively). However, according to one-way ANOVA, no significant differences between samples stored in different materials or different gas composition were recorded. The drip loss in our study was in the same range as reported by others.[24,32] It has been suggested by Patsias et al.[27] that the water-holding capacity is decreased with CO_2 in the headspace due to CO_2 dissolution. The effect of gas composition/available CO_2 on drip loss of chicken has also reported by Holck et al.,[24] showing that underpressure due to the solubility of CO_2 in the meat is also of importance to the drip loss and not only the amount of CO_2.

In addition, appearance of the package is very important for the consumers. At the packaging day (Day 0), the appearance of packed chicken fillets was defined as neutral (scored with 3). The appearance of APET/PE (score 1.5) trays was significantly different than HDPE trays (score 2.0) for chicken stored in CO_2/N_2 atmosphere in the beginning of the experiment (storage days 8 and 13), implying that samples packed in APET/PE had more underpressure than HDPE samples. At the end of the storage time (Day 24), no significant differences were recorded. However, values were 1.5 for APET/PE and 1.8 for HDPE samples, implying underpressure in packages to certain extent due to dissolved CO_2 in product, which is also in relation to the measured drip loss. Increased CO_2 content can result in increased CO_2 dissolved in the product.

All presented data are showing that the similar quality and shelf life can be obtained with mono material (HDPE) as APET/PE and AP is not needed to prolong the shelf life for the selected G/P ratio.

3.2 | Evaluation of the impact of G/P ratio on the effect of AP and mono material

Further, this research study was also focused on different G/P ratios and their potential effect on the shelf life. Possible reduction in G/P ratio can potentially increase transport efficacy, whereas increase of G/P ratio can increase product shelf life. Additionally, influence of added AP on product shelf life was studied in relation to different G/P ratio and material type.

Selection of AP solutions was in accordance with their activity. Oxygen scavengers are often used to remove residual oxygen to maintain oxygen-free atmosphere during storage, thus preventing the growth of aerobic microorganisms, discoloration and off-flavour of the product,[15] whereas CO_2 emitters are usually used to increase CO_2 level and inhibit bacterial growth. Influence of different G/P ratios and AP on TVC count (aerobic and anaerobic), LAB and *Enterobacteriaceae* count is presented in Figures 1, 2, 3 and 4 respectively.

GLM ANOVA for the set of samples including all HDPE samples with different G/P ratio (G/P), AP (A) and storage time (T) (8, 13, 17, 21 and 24 days) is presented in Table 6. The model included the main effects (G/P, A and T) and their interactions. The table includes responses where significant effects were obtained (no significant effects were detected for counts of *B. thermosphacta*). Storage time has the most effect for TVC and LAB (81.88% and 75.47%, respectively) followed by G/P ratio (3.31% and 2.39%, respectively). For *E. coli* and *Enterobacteriaceae*, only storage time had significant effect.

According to one-way ANOVA, no significant differences between the samples were observed in the first part of storage time. Significant higher level of TVC (aerobic) was measured for chicken stored in HDPE with G/P ratio 1.0 with O_2 scav. (7.51 $\log_{10}$ CFU/g) compared with G/P ratio 1.5 (6.26 $\log_{10}$ CFU/g). Similar results and significance between G/P 1.0 and G/P 1.5 were observed for TVC (anaerobic). On the 21st day of storage, chicken stored in HDPE with G/P 1.0 reached high level of TVC (7.29 $\log_{10}$ CFU/g for aerobic and 7.59 $\log_{10}$ CFU/g for anaerobic), whereas TVC count for G/P 2.6 was below 7 $\log_{10}$ CFU/g (6.61 for aerobic and 6.06 for anaerobic). Despite the one-way ANOVA recording no significant difference, these results are implying that higher G/P ratio is resulting in lower TVC count. Levels of most TVC, LAB and *Enterobacteriaceae* did not show any significant difference between samples with different G/P ratio and AP solutions.

Influence of different G/P ratios and addition of Active packaging (AP) (oxygen scavengers and CO_2 emitters) on drip loss, off-odour and appearance of the package is presented in Figures 5, 6 and 7, respectively. According to the GLM, all main factors (G/P ratio), AP (A) and all storage time (T) had significant effect on the off-odour, drip loss and appearance (Table 6). For the drip loss, the

TABLE 5 Off-odour, drip loss and appearance for chicken stored in APET/PE and HDPE trays with G/P 1.5 during the storage period (0–24 days)

	Off-odour (scores 1–5)				Drip loss (wt.%)				Appearance (scores 0–6)			
	60%/40% CO_2/N_2		75%/25% O_2/CO_2		60%/40% CO_2/N_2		75%/25% O_2/CO_2		60%/40% CO_2/N_2		75%/25% O_2/CO_2	
Days	APET/PE	HDPE	APET	HDPE	APET/PE	HDPE	APET	HDPE	APET/PE	HDPE	APET	HDPE
0	5.0 ± 0.0	5.0 ± 0.0	5.0 ± 0.0	5.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	3.0 ± 0.0	3.0 ± 0.0	3.0 ± 0.0	3.0 ± 0.0
8	5.0 ± 0.0	5.0 ± 0.0	4.9 ± 0.1	5.0 ± 0.0	2.1 ± 0.4	1.6 ± 0.5	1.3 ± 0.1	1.3 ± 0.5	**1.5^{C} ± 0.0**	**2.0^{B} ± 0.0**	**2.2^{B} ± 0.3**	**2.5^{A} ± 0.0**
13	**4.9^{A} ± 0.1**	5.0 ± 0.0	3.8 ± 0.4	**3.3^{B} ± 0.8**	2.6 ± 0.7	2.6 ± 0.0	1.8 ± 0.5	2.4 ± 0.6	**1.5^{A} ± 0.0**	**2.0^{B} ± 0.0**	**2.0^{B} ± 0.0**	**2.0^{B} ± 0.0**
17	3.8 ± 0.7	4.0 ± 0.6	3.2 ± 0.4	2.9 ± 0.4	2.8 ± 0.7	2.6 ± 0.3	2.3 ± 0.3	2.5 ± 0.7	1.5 ± 0.0	1.8 ± 0.3	1.7 ± 0.3	1.5 ± 0.0
21	**3.5^{A} ± 0.2**	2.9 ± 1.0	2.4 ± 0.1	**2.0^{B} ± 0.2**	2.5 ± 0.5	2.8 ± 0.1	2.6 ± 0.8	2.6 ± 0.6	**1.5^{B} ± 0.0**	1.8 ± 0.3	**2.0^{A} ± 0.0**	1.7 ± 0.3
24	**3.0^{A} ± 0.4**	**3.1^{A} ± 0.2**	**1.7^{B} ± 0.2**	**1.7^{B} ± 0.1**	2.9 ± 0.6	2.9 ± 0.5	2.8 ± 0.1	2.7 ± 0.2	1.5 ± 0.0	1.8 ± 0.3	1.5 ± 0.0	1.5 ± 0.0

Note. Statistical analysis of variance (one-way ANOVA) has been performed for each analysis and within each sampling time. Means that are statistically different ($p > 0.05$) are presented bold with letters. Samples with different letters belonging to different Tukey Groups and are significantly different.

Abbreviations: APET/PE, amorphous polyethylene terephthalate/polyethylene; HDPE, high-density polyethylene.

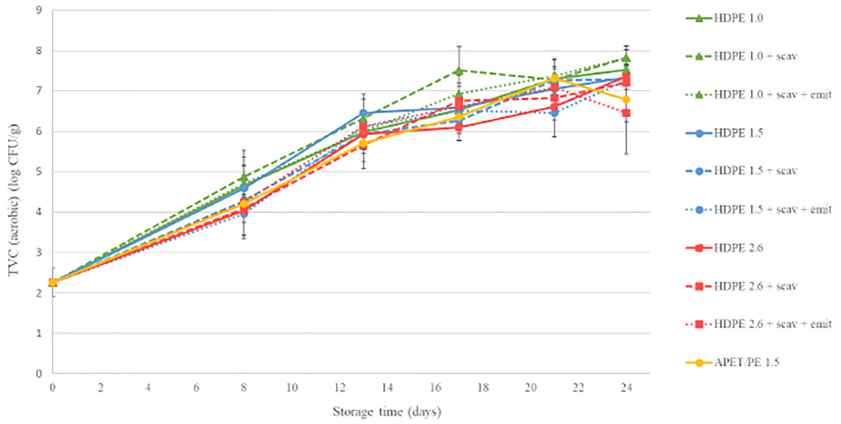

FIGURE 1 Influence of different G/P ratios and active packaging on total viable counts (TVCs) of bacteria (aerobic)

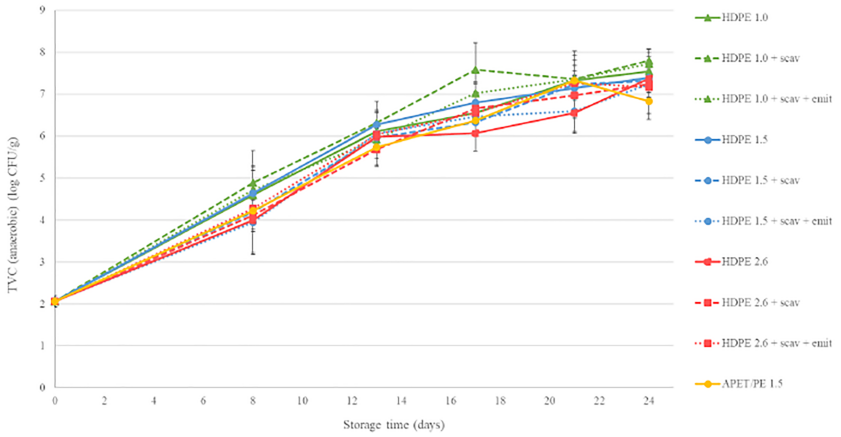

FIGURE 2 Influence of different G/P ratios and active packaging on total viable counts (TVCs) of bacteria (anerobic)

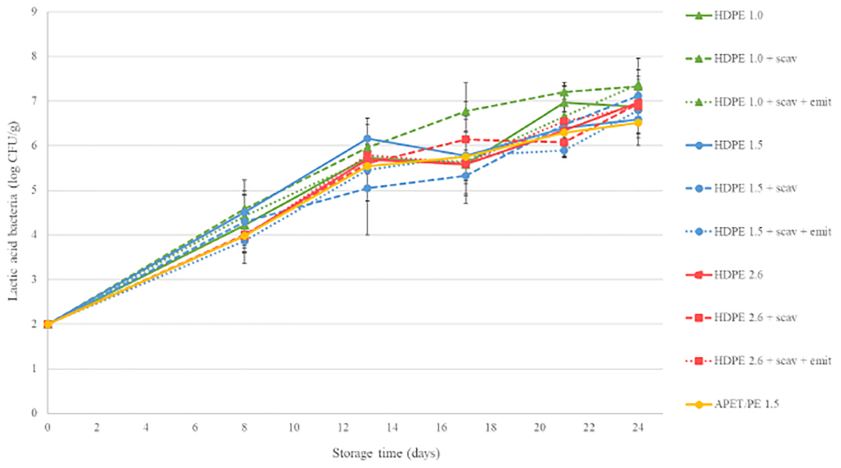

FIGURE 3 Influence of different G/P ratios and active packaging on counts of lactic acid bacteria (LAB)

G/P ratio had the most effect (70.68%) followed by storage time (9.22%) and AP (7.78%) and with no interaction effects. Also, for the off-odour, G/P ratio had the most effect (40.93%), whereas AP had the second most effect (20.95%) and followed by storage time (12.26%). In addition, both interaction effects with G/P ratio [(G/P) xA and (G/P) xT] were significant. Storage time had the most effect for the off-odour (68.77%) as for the bacterial growth; in addition, both G/P ratio (6.25%) and AP (1.25%) had significant effect.

As can be seen in Figure 5, highest drip loss was recorded for chicken stored in HDPE with G/P ratio 1.0 on the 24th day of storage without any AP (no scavengers or emitters). With addition of O_2 scavengers, this value decreases (from 7.6 to 6.2), whereas addition of CO_2 emitters additionally decreased drip loss to 5.1. Significant

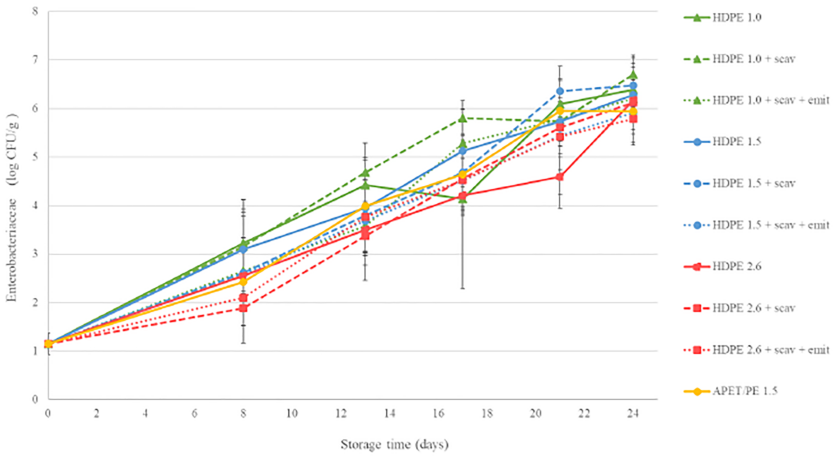

FIGURE 4 Influence of different G/P ratios and active packaging on *Enterobacteriaceae*

TABLE 6 Overview of ANOVA results for the responses, main effect and second-order interaction (general linear model) for gas volume to product volume ratio (G/P ratio1.0, 1.5 and 2.6); active packaging (A) and storage time (T)

		TVC (aerobic)	LAB	*Enterobacteriaceae*	*E. coli*	Off-odour	Drip loss	Appearance
Gas volume to product volume ratio	G/P	3.31*	2.29*	3.36	0.33	6.25*	70.68*	40.93*
Active packaging	A	0.18	0.39	0.66	0.02	1.47*	7.78*	20.95*
Storage time	T	81.88*	75.47*	73.43*	92.97*	68.77*	9.22*	12.26*
	(G/P)xA	0.94	1.49*	0.65	0.18	2.08*	0.53	3.83*
	(G/P)xT	0.77	0.77	0.35	1.31*	2.34	0.89	7,68*
	AxT	0.69	1.17	1.09	0.06	1.44	0.70	0.70
	Error	12.22	18.40	17.47	5.14	17.65	10.20	13.44
	R^2 (adj)	84.55%	76.73%	77.92%	93.51%	77.68%	87.10%	83.01%

Note. All samples stored at 4°C. The numbers are explained variances.
*Values marked with asterisks shows significant effects within 95% level.
Abbreviations: LAB, lactic acid bacteria; TVC, total viable count.

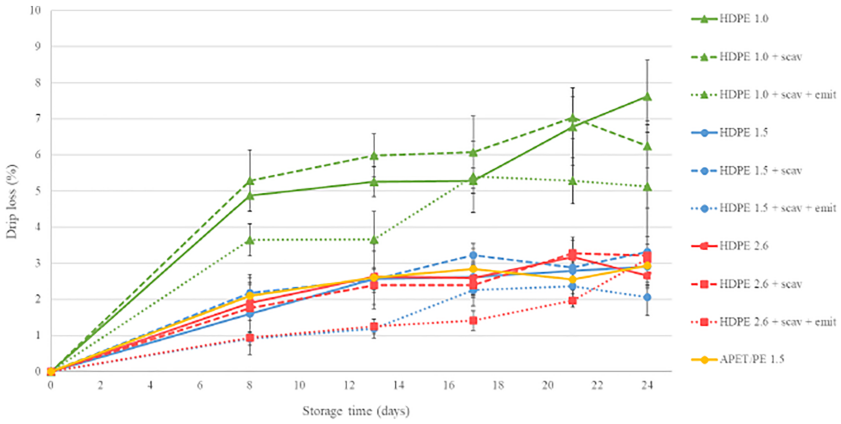

FIGURE 5 Influence of different G/P ratios and addition of active packaging (oxygen scavengers and CO_2 emitters) on drip loss

difference has been recorded between chicken fillets packed without AP and samples with both scavengers and emitters. These results show that there is an interaction between CO_2 emitter and drip loss, resulting in lower drip loss when CO_2 emitter is applied in MAP with high content of CO_2. This phenomenon can be explained by the fact that CO_2 is soluble in the meat, resulting in reduced level of CO_2 during storage (Table 3). This is followed by a decrease in the headspace volume and underpressure in the packages, specific in rigid packaging.[21,26] Adding a CO_2 emitter can compensate for the reduced headspace and underpressure and thus reduce the drip loss in the product, which is also shown by others.[24] These results show that using CO_2 emitter will contribute in reducing the drip loss. Significant

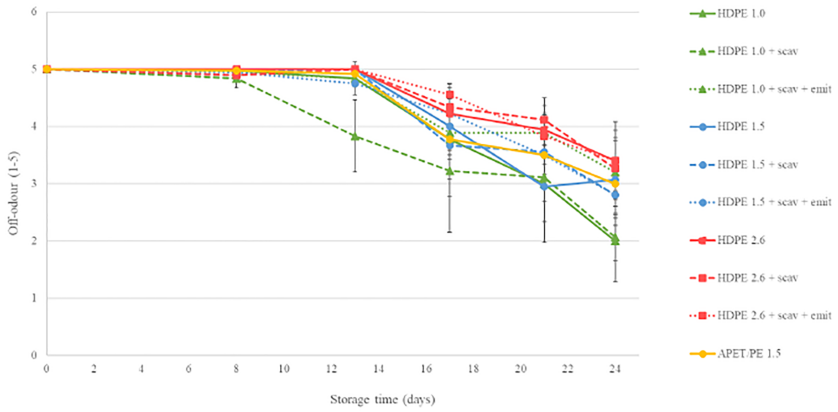

FIGURE 6 Influence of different G/P ratios and addition of active packaging (oxygen scavengers and CO_2 emitters) on off-odour

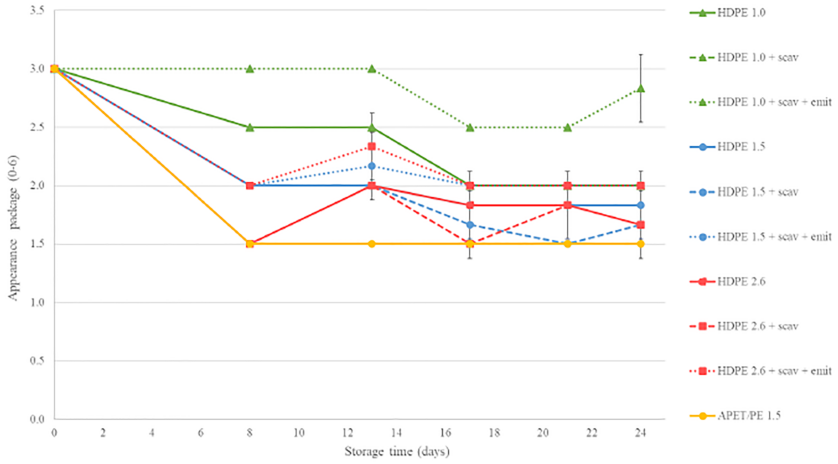

FIGURE 7 Influence of different G/P ratios and addition of active packaging (oxygen scavengers and CO_2 emitters) on appearance of the package

difference in drip loss between chicken fillets packed in trays with G/P 1.0 and all other samples has also been detected ($p < 0.05$), implying that lowest G/P ratio is causing higher drip loss. Addition of oxygen scavengers had no influence on drip loss within the same G/P ratio; nevertheless, drip loss has been reduced to some extent for all samples with addition of CO_2 emitters.

The off-odour of the packed chicken fillets is affected by initial gas composition (Figure 6). For all samples packed with gas composition CO_2/N_2, unacceptable off-odour level was reached at the end of the storage period (between 21–24 days). For samples with G/P ratio 1.0, addition of AP (both scavengers and emitters) had significantly higher score (above 3) and was regarded as acceptable. Addition of AP solutions to samples with higher G/P ratio did not influence the off-odour, and all samples were acceptable (scored with 3). It is interesting to point out that samples packed in high oxygen level reached unacceptable off-odour on Day 17 (2.4 for HDPE samples) and on Day 21 (1.7 for APET/PE samples).

Addition of AP affected off-odour of samples with the lowest G/P ratio (1.0), whereas other samples were not affected by the addition of the AP. This is implying that in the case of lower G/P ratio, shelf life can be maintained and/or prolonged with addition of AP solutions. However, for higher G/P ratios (in our case, 1.5 and 2.6), addition of AP did not have any influence on the chicken fillet shelf life.

Both G/P ratio and AP solutions are influencing the appearance of the package (Figure 7). Increase in G/P ratio is causing decrease in appearance score (on 8th day of storage, the scores were 1.5 for 2.6 G/P, 2 for 1.5 G/P and 2.5 for 1 G/P). Similar trend is followed during storage time. Addition of oxygen scavengers had no influence on appearance, whereas systems with both oxygen scavengers and CO_2 emitters had positive influence on the appearance (scores were above 2). Appearance of the package is related to the drip loss, as packages with scores less than 2 are undepressed and can cause additional drip loss (Figure 7). If we take this into account, it is obvious that addition of emitters has positive influence on both appearance and drip loss (less drip loss and higher score for the appearance) within the same G/P ratio for samples (Figure 7).

4 | CONCLUSION

The possibility of packaging chicken fillets in recyclable mono materials (HDPE) instead of complex multilayered materials (APET/PE) as a replacement for more sustainable packaging system was studied. All

samples packed in HDPE showed acceptable level of bacteria and acceptable off-odour for chicken fillets up to 19 days without addition of any AP solutions. For some attributes, storage in HDPE was even a better choice than APET/PE (appearance). Chicken fillets packed in HDPE with lowest G/P (1.0) ratio showed higher drip loss compared with higher G/P ratios without any AP. Addition of oxygen scavengers had no influence on drip loss within same G/P ratio; nevertheless, drip loss has been reduced to some extent for all samples with addition of CO_2 emitters. Although the influence of AP was not so pronounced within same G/P ratios, it is obvious that AP solutions are of interest in combinations with G/P ratios, gas compositions and selection of materials.

This study showed that the recyclable mono materials can be used for packaging of fresh chicken fillets without jeopardizing the shelf life. Outcome of this research presents a step forward in design for recycling, increase in recycling rates and less food packaging waste.

However, as food systems are very complex and diverse, this applies only to chicken fillets, and further research should be spreads on selection of diverse food systems to be packed in appropriate materials and selection of AP solutions. Moreover, further research should be also focused on recyclable top foil as well. Nevertheless, outcomes of this research are encouraging and shifting one step forward to EU sustainable goals and circular economy. Hopefully, this research will influence on use of more recyclable materials on Norwegian market, improve recyclability and use of recycled materials in diverse applications.

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