Quality characteristics of processed cheese with avocado (*Persea americana*) seed powder

HOWIDA A. EL-SAYED – MERANDA A. TAWFEK – OSAMA A. IBRAHIM

Summary

Avocado seed powder is a rich source of vitamins, antioxidants and minerals, which could be incorporated in cheese processing. Hence, the aim of the present work was to incorporate it in processed cheese processing where total phenolic compounds (*TPC*), antioxidant activity and phenolic compounds profile were determined. Moreover, chemical, physical, rheological and organoleptic characteristics of the resulted cheese were investigated. Results showed that avocado seed powder incorporation in processed cheese increased *TPC* (17–33 $g\cdot kg^{-1}$, expressed as gallic acid equivalents), antioxidant activity (by 32–42 %), proteins content (by 8–9 %), ash (by 5.9–6.8 %), fibre (0.2–0.3 %), fatty acids (butyric, caprylic, capric, lauric, myristic, myristoleic), minerals (Ca, Zn, Fe, Cu, Mn, P, Mg) and vitamins (A, C, E, B complex). Acidity, melting index and oil separation decreased as avocado seed powder level increased. Thus, avocado seed powder as a functional ingredient could be used in the production of novel spread-type processed cheese with good chemical, rheological and nutritional characteristics, without defects in organoleptic attributes during storage.

Keywords

avocado seeds; antioxidant; processed cheese; quality; shelf life

Foodstuffs enriched with nutraceuticals in the human diet lead to an improvement of various biochemical attributes related to the well-being of the individuals. The commercial success and preferential marketing of several foodstuffs are frequently based on highlighting their nutritional properties [1]. However, numerous medical studies recommend fruits consumption mainly due to their impact on prevention and treatment of various diseases [2].

Avocado (*Persea americana*) is a tropical and subtropical tree, which is grown mainly in Australia, Spain and South Africa, although it is native to southern Mexico. Avocado is health-promoting and can be used for hypercholesterolemia improvement, hypertension treatment as well as could be useful in cardiovascular health and diabetes II [3]. Avocado is considered a rich source of phenolic compounds with high antioxidant activity and antimicrobial activity. The lipophilic extract of avocado inhibits prostate cancer, induces apoptosis in breast cancer and prevents liver injury. Also, avocado fruit is considered a natural source of monounsaturated lipids and essential fatty acids [4].

Avocado seeds are useful in delaying the oxidation of model food systems such as emulsions of sunflower oil in water or meat burgers [5]. Avocado seeds were also proposed to be valorized as flour, which would be of special use in tropical countries, where crops such as wheat, barley, millet or rye do not lead to good quality flours and lead to higher costs for preparation of functional beverages and baked products [6].

Functional foods refer to foodstuffs or food ingredients that provide specific physiological useful impact and/or reduce the risk of chronic disease beyond basic nutritional benefits. Processed cheese is widely consumed in several forms including spreads, slices, as well as could be used in sandwiches or pizza. Processed cheese popularity is based on various factors such as diversity, easy customization as an ingredient, adaptability

Howida El-Sayed, Meranda Tawfek, Dairy Sciences and Technology Research Department, Food Technology Research Institute, Agricultural Research Centre, 9 Algamaa street, P. O. 12619, Giza, Egypt.
Osama Ibrahim, Dairy Science Department, Industries and Nutrition Research Institute, National Research Centre, 33 Bohouth Street, P. O. 12622, Cairo, Egypt.

Correspondence author:
Osama Ibrahim, e-mail: osama_nrc@hotmail.com

to fast foods or attractive packaging into suitable forms [7].

Thus, the objective of the present work was to prepare a novel processed cheese supplemented with avocado seed powder (ASP) as a functional food ingredient. Hence, total phenolic compounds and antioxidant activity were determined, together with identification of phenolic compounds of the product that were determined using high-performance liquid chromatography (HPLC). Moreover, chemical, physical, rheological and organoleptic attributes of the resulted processed cheese were determined.

MATERIALS AND METHODS

Materials

Fresh fully ripened avocado fruits and processed cheese ingredients were purchased from the local market in Cairo, Egypt. Commercial melting emulsifying salt Joha S9s recommended for spreadable processed cheese making was obtained from ICL Food Specialties (Landenburg, Germany). Gallic acid, 2,2-diphenyl-1-picryl-hydrazyl (DPPH) and Folin-Ciocalteu phenol reagent were obtained from Sigma Aldrich (St. Louis, Missouri, USA). All other chemicals used were of analytical grade.

Preparation of avocado seed powder

The avocado seeds were washed and air-dried at room temperature (25 ± 2 °C) in the laboratory for 3 days. After that, they were dried at 50 °C for 4 h using oven (Agilent Technologies, Santa Clara, California, USA), ground using a laboratory mill and then sieved through 60 mesh sieve (250 μm). The avocado seed powder (ASP) was packed in polyethylene bags and stored at 5 °C until used for a maximum of one month.

Chemical analysis of avocado seed powder

The proximate composition of ASP for moisture (AOAC 945.43), fat (AOAC 948.22), protein and water soluble nitrogen (AOAC 984.13) as well as for ash (AOAC 950.49) contents was determined [8]. Crude fibre was determined according to ISO 15884:2002 [9]. Total carbohydrates (TC) were calculated by difference using the Eq. 1.

$$TC = 100 - (W + P + F + Fi + A) \quad (1)$$

where W is of water, P is protein, F is fat, Fi is fibre and A is ash.

The chemical composition of the resulted ASP is presented in Tab. 1.

Preparation of methanolic extracts

An amount of 100 g of ASP or processed cheese was extracted using a mixture of methanol and water (80 : 20 by volume) during 12 h with stirring (8 Hz) in a dark place at 25 °C. The solution was then filtered through Whatman filter paper No. 1 (Whatman, Maidstone, United Kingdom) and the solvent was reduced in volume by a rotary evaporator at a temperature of 35 °C. The concentrated extract was stored in a brown glass vial at –20 °C for further analysis for a maximum of one month.

Total phenolics content

The total phenolics content of ASP and processed cheese extracts were evaluated using Folin-Ciocalteu reagent in accordance with the modified method described by Lafka et al. [10]. Total phenolics content was calculated based on a calibration curve prepared with gallic acid as a standard and expressed as grams of gallic acid equivalents (GAE) per kilogram.

Identification of phenolic compounds

Phenolic compounds of ASP methanolic

Tab. 1. Chemical composition of ingredients and avocado seed powder.

Component	Ras cheese base		Avocado seed powder
	Mature	Fresh	
Dry matter [%]	65.8 ± 0.1	54.4 ± 1.5	97.4 ± 0.4
Fat [%]	34.8 ± 0.1	24.8 ± 1.2	7.0 ± 1.3
Protein [%]	25.5 ± 0.1	22.3 ± 0.9	5.0 ± 0.1
Carbohydrate [%]	0.1 ± 0.0	1.6 ± 0.0	84.1 ± 1.0
Fibre [%]	ND	ND	4.5 ± 0.1
Ash [%]	5.4 ± 0.0	5.8 ± 1.6	1.3 ± 0.2
Water soluble nitrogen/Total nitrogen	25.5 ± 0.1	23.1 ± 0.8	5.3 ± 0.0
Total phenolics content [g·kg^{-1}]	ND	ND	38.2 ± 1.1
Antioxidant activity [%]	ND	ND	53.3 ± 1.3

All parameters are represented as mean of replicates ± standard error (n = 4).
Total phenolics content is expressed as grams of gallic acid equivalents per kilogram. ND – not determined.

extract were identified and quantified by HPLC using an Agilent 1260 series instrument (Agilent Technologies). The separation was carried out using Zorbax Eclipse C18 column (4.6 mm × 250 mm, 5 μm particle size; Agilent Technologies). The mobile phase consisted of water (A) and 0.2 $g \cdot l^{-1}$ trifluoroacetic acid in acetonitrile (B) at a flow rate of 1 $ml \cdot min^{-1}$. The mobile phase was programmed consecutively in a linear gradient as follows: 0 min (80 % A); 0–5 min (80 % A); 5-8 min (40 % A); 8–12 min (50 % A); 12–14 min (80 % A) and 14–16 min (80 % A). The detection was carried out spectrophotometrically at 280 nm. The injection volume was 10 μl and the column temperature was maintained at 35 °C. Phenolic compounds of each sample were identified by comparing their relative retention time with those of the standard mixture chromatogram. The concentration of an individual phenolic compound was calculated based on the peak area, then converted to milligrams of the phenolic compound per kilogram of ASP.

Antioxidant activity

The antioxidant activity of ASP and processed cheese extracts were determined using DPPH radical-scavenging method as described by MATTHUS [11]. The radical-scavenging activity of the tested samples was expressed as a percentage inhibition of DPPH (I), calculated according to the following formula:

$$I = \frac{A - A_0}{A} \times 100 \tag{2}$$

where A is absorbance at 515 nm of the control sample and A_0 is the final absorbance of the test sample at 515 nm.

Manufacture of processed cheese

Processed cheese was manufactured according to the method of MEYER [12]. In the present study, processed cheese was prepared from fresh and mature Ras cheese as the most popular Egyptian hard cheese, which is similar to Greek "Cephalotyre" cheese. Samples were prepared with ASP at the level of 10 g, 17 g or 34 g per kilogram of formula blend and marked A, B and C, respectively. Tab. 1 presents data on the chemical composition of the processed cheese. The formulation ingredients that were used in the processed cheeses manufacture are presented in Tab. 2. The resulted processed cheese samples were filed into suitable glass jars, air-tightly closed and stored at 5 °C until analysis for a maximum of 3 months when fresh, and after 1, 2 and 3 months. Two replicates of the whole experiment and parameters analysis were done.

Tab. 2. Blend formulas of spread-type processed cheese with avocado seed powder.

Ingredient	Spread-type processed cheese treatment			
	Control	ASP10	ASP17	ASP34
ASP [$g \cdot kg^{-1}$]	0	10	17	34
Mature cheese [%]	12.0	11.3	10.8	9.6
Fresh cheese [%]	50.3	49.3	48.6	47.0
Emulsifying salt [%]	2.5	2.5	2.5	2.5
Water [%]	35.2	36.0	36.4	37.5

ASP – avocado seed powder.

Chemical composition of cheese

Processed cheese and their ingredients were chemically analyzed for moisture (AOAC 926.08), fat (AOAC 933.05), and protein (AOAC 991.20-23) contents according to AOAC [13]. Titratable acidity, ash and water soluble nitrogen contents were determined as described by LING [14], while crude fibre were determined according to standard ISO 15884:2002 [9]. Total volatile fatty acid (*TVFA*) was determined according to KOSIKOWSKI and MISTRY [15]. The pH values were measured using a digital pH-meter (Hanna Instruments, Woonsocket, Rhode Island, USA). Total carbohydrates were calculated by difference. Fatty acid profile of processed cheese were extracted as described in standards ISO 14156:2001 [16] and determined according to standard ISO 15884:2002 [9] using a gas chromatography (GC) system. Vitamins content of processed cheese were determined for vitamin A [16], vitamin B complex [17], vitamin C [18] and vitamin E [19]. Minerals content of processed cheese was determined as described by HANKINSON [20] using atomic absorption spectrophotometer No. 3300 (Perkin Elmer, Waltham, Massachusetts, USA).

Physical properties of cheese

Physical properties of processed cheese during 3 months of storage were performed as follow: oil separation index was estimated by the method of THOMAS et al. [21], melting quality was measured using the melt ability test apparatus as described by GUNASEKARAN and AK [22], and penetration value was measured using pentameter (Cochler, Bohemia, New York, USA) as described by BOURNE [23].

Colour parameters of processed cheese during storage for 3 months were performed using model D2s A-2 colorimeter (Hunter, Reston, Virginia, USA) tri-stimulus values of the colour namely L^*, a^* and b^* as described HUNTER and HAROLD [24].

Textural profile analysis (TPA) were performed according to Lobato-Calleros et al. [25] using a Texture ProTM texture analyzer (TMS-Pro, Sterling, Virginia, USA), equipped with 113.4 kg load cell and connected to a computer programmed with Food Technology Corporation's Texture Lab Pro software (TMS-Pro). The texture profile parameters including hardness, cohesiveness, gumminess, chewiness, adhesiveness and springiness were investigated.

Organoleptic evaluation of cheese

Organoleptic attributes of spread-type processed cheese during 3 months of storage were evaluated as described by Meyer [12]. Cheese samples were evaluated by means of 10 panelists from the staff of Dairy Sciences and Technology Research Department (Food Technology Research Institute, Giza, Egypt), with maximum score points of 40 points for flavour, body and texture (40 points) together with cheese appearance (20 points).

Statistical analysis

The results average values were analysed by SAS software (SAS Institute, Cary, North Carolina, USA) using ANOVA procedure for analysis of variance. The results were expressed as mean ± standard error and the differences between means were tested for significance using Duncan's multiple range at $p \leq 0.05$.

RESULTS AND DISCUSSION

Chemical characterization

It is well-known that genetic (different avocado cultivars), environmental factors and cultivation conditions (different tree or soil management) can have impact on ASP composition and phenolic components content [2]. Results of chemical analysis of ASP are presented in Tab. 1. According to Akasha et al. [26], ASP may be considered a good emulsifying ingredient due to the carbohydrates content. This is in line with Alqattani et al. [27] who reported that date pit powder contains polysaccharides associated with proteins, which enhance its functional properties as well as raise their capability as an emulsifier. Also, Di Stefano et al. [2] reported the total phenolics content of green (1.7–58.8 $mg\cdot kg^{-1}$) and ripe (6.4–28.3 $mg\cdot kg^{-1}$) avocado fresh fruits. In our study, the total phenolics content of ASP extract was 38.2 $g\cdot kg^{-1}$ and antioxidant activity was 53.3 % (Tab. 1). Tab. 3 shows the content of identified phenolic compounds in ASP quantified using HPLC. It could be noted that gallic acid (226.3 $mg\cdot kg^{-1}$) was the dominant phenolic constituent found in the methanolic extract of ASP, followed by kaempferol (121.0 $mg\cdot kg^{-1}$). In previous studies, phenolic compounds gentisic acid, 4-hydroxybenzoic acid, gallic acid, chlorogenic acid, isoramnetin, coffeic acid, ferulic acid, narirutin, *p*-coumaric acid, taxifolin and epicatechin were found in samples [2].

Physico-chemical analysis of cheese

Data on the chemical characterization of spread-type processed cheese containing ASP are given in Tab. 4. It could be noted that the total solids content including protein, ash and fat of ASP-enriched processed cheese were higher than those of control cheese with significantly ($p \leq 0.05$) higher values of fibre content, which was mainly due to ASP ingredients (Tab. 1). However, fibre is a useful component correlated to gut and cardiovascular health [28]. Mohamed and Hussein [29] reported similar findings for processed cheese with cress (*Lepidium sativum*) seed.

Also, Tab. 4 shows that the total phenolic compounds (expressed as GAE) of spread-type processed cheese containing ASP ranged from 17.6 $g\cdot kg^{-1}$ to 33.0 $g\cdot kg^{-1}$. Its antioxidant activity was 32.9–42.4 %. It could be mainly due to phenolic compounds in ASP as well as due to their antioxidant activity (Tab. 1, Tab. 3). However, the addition of a plant material rich in phenolic components, such as red radish roots nanopowder or jalapeno red pepper, to cheese increased their total phenolics content and antioxidant activity compared to plain cheese [30, 31].

Tab. 3. Phenolic compounds content in avocado seed powder.

Phenolic compounds	Content [$mg\cdot kg^{-1}$]
Gallic acid	226.3
Kaempferol	121.0
Catechin	92.6
Chlorogenic acid	75.4
Cinnamic acid	24.8
Methyl gallate	12.9
Pyro catechol	12.2
Syringic acid	4.6
Naringenin	9.0
Caffeic acid	2.5
Ellagic acid	ND
Coumaric acid	ND
Vanillin	ND
Ferulic acid	ND
Daidzein	ND
Querectin	ND
Hesperetin	ND

ND – not detected.

Tab. 4. Chemical characteristics of cheese samples with avocado seed powder.

Parameter	Processed cheese			
	Control	ASP10	ASP17	ASP34
Dry matter [%]	41.8 ± 0.0 a	41.8 ± 0.0 a	41.9 ± 0.0 a	42.1 ± 0.0 a
Fat [%]	21.5 ± 0.0 a	21.9 ± 0.0 a	22.5 ± 0.0 a	22.5 ± 0.0 a
Protein [%]	7.8 ± 0.0 a	8.3 ± 0.0 a	8.9 ± 0.1 a	9.2 ± 0.0 a
Fibre [%]	0.0 ± 0.0 d	0.1 ± 0.0 c	0.2 ± 0.0 b	0.3 ± 0.0 a
Ash [%]	5.6 ± 0.0 a	5.9 ± 0.1 a	6.5 ± 0.0 a	6.8 ± 0.0 a
Total phenolics content [g·kg^{-1}]	10.9 ± 0.6 d	17.6 ± 1.1 c	25.9 ± 2.3 b	33.0 ± 0.9 a
Antioxidant activity [%]	12.7 ± 1.3 c	32.9 ± 0.7 b	33.5 ± 1.7 b	42.4 ± 1.2 a

All parameters are presented as mean of replicates ± standard error (n = 4). Means with different superscript letters in the same row are significantly different at $p \leq 0.05$.
Control, ASP10, ASP17, ASP34 – processed cheese with avocado seed powder at the level of 0 g·kg^{-1}, 10 g·kg^{-1}, 17 g·kg^{-1} and 34 g·kg^{-1} formula blend, respectively. Total phenolics content is expressed as grams of gallic acid equivalents per kilogram.

Water soluble nitrogen of processed cheese is presented in Fig. 1. The results showed that water soluble nitrogen of ASP-containing processed cheese were higher than plain cheese, which could be attributed to protein solubilization that resulted from the activity of the contained thermostable protease and polyphosphate hydrolysis in the added emulsifying salts [29]. Also, water soluble nitrogen gradually increased in both control and ASP-containing processed cheese with the storage period progressed.

Data on total volatile fatty acids (TVFA) of spread-type processed cheese are given in Fig. 2. The results showed that TVFA of ASP-containing processed cheese were higher than plain cheese, which could be attributed to the ASP fat content (Tab. 1). Also, TVFA gradually increased in both control and ASP-processed cheese with the storage period progressed.

Tab. 5 shows the minerals content of spread-type processed cheese containing ASP. It could be noted that the addition of ASP increased the mineral content including Ca, Zn, Fe, Cu, Mn, P and Mg. Also, control processed cheese had the highest Na and K contents in comparison with ASP-containing processed cheese. In contrast, Mohamed and Hussein [29] reported that the addition of cress seeds to processed cheese increased their Fe, K, P and Zn contents compared to plain cheese.

Data on the vitamins content of the spread-type processed cheese containing ASP are presented in Tab. 6. It could be noticed that the addition of ASP increased the vitamins content of cheese in comparison with plain cheese including thiamine (B1), pyridoxine (B6), riboflavin (B2), niacin (B3), cobalamin (B12), folic acid (B9), retinol (A), tocopherol (E) and ascorbic acid (C). This could be mainly due to the vitamins content of ASP. However, the avocado fruit contains unsaturated fatty acids, carbohydrates, quantities of vitamins C, B, E, and A and had been recognized

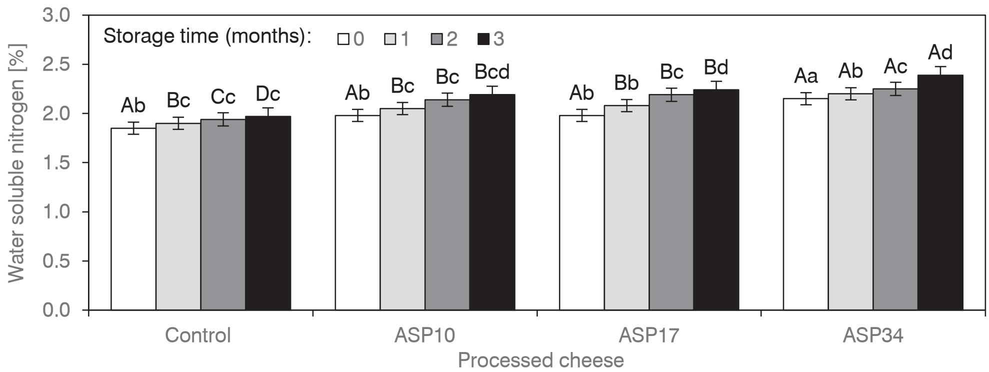

Fig. 1. Water soluble nitrogen content of cheese samples with avocado seed powder during storage.

Cheese samples were stored at 5 ± 1 °C. Means with different capital letters during the storage period for each treatment and different small letters between the treatments are significantly different at $p \leq 0.05$ (n = 4).
Control, ASP10, ASP17, ASP34 – processed cheese with avocado seed powder at the level of 0 g·kg^{-1}, 10 g·kg^{-1}, 17 g·kg^{-1} and 34 g·kg^{-1} formula blend, respectively.

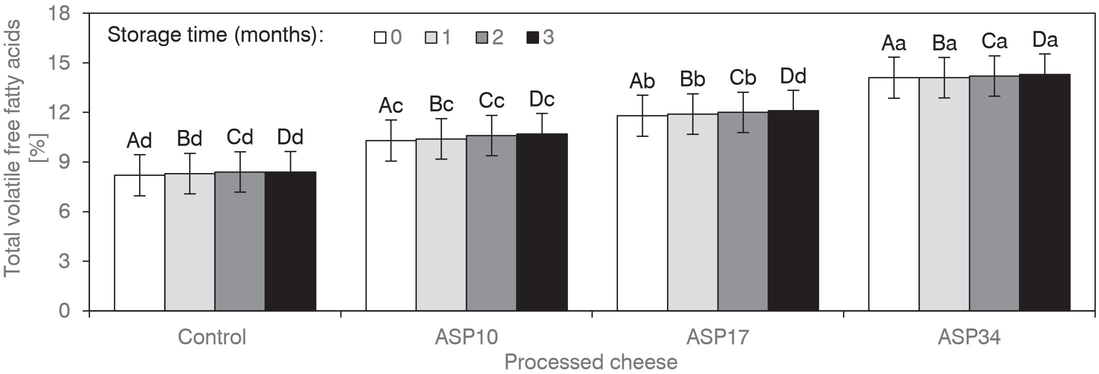

Fig. 2. Total volatile free fatty acids content of cheese samples with avocado seed powder during storage.

Cheese samples were stored at 5 ± 1 °C. Means with different capital letters during the storage period for each treatment and different small letters between the treatments are significantly different at $p \leq 0.05$ ($n = 4$).
Control, ASP10, ASP17, ASP34 – processed cheese with avocado seed powder at the level of 0 g·kg^{-1}, 10 g·kg^{-1}, 17 g·kg^{-1} and 34 g·kg^{-1} formula blend, respectively.

Tab. 5. Minerals content of cheese samples with avocado seed powder.

Mineral [mg·kg^{-1}]	Processed cheese			
	Control	ASP10	ASP17	ASP34
Na	1 403.6 ± 0.0 ^{a}	1 370.9 ± 0.0 ^{b}	1 231.3 ± 0.0 ^{c}	1 098.6 ± 4.0 ^{d}
K	45.8 ± 0.0 ^{a}	44.4 ± 0.0 ^{a}	39.2 ± 2.0 ^{b}	33.3 ± 2.0 ^{c}
Ca	238.4 ± 0.0 ^{d}	343.5 ± 0.0 ^{c}	362.9 ± 0.0 ^{b}	384.0 ± 0.0 ^{a}
Zn	1.2 ± 0.0 ^{d}	1.5 ± 0.0 ^{c}	1.5 ± 0.0 ^{b}	1.6 ± 0.0 ^{a}
Fe	0.8 ± 0.0 ^{d}	0.9 ± 0.1 ^{c}	1.0 ± 0.0 ^{b}	1.4 ± 0.0 ^{a}
Cu	0.0 ± 0.0 ^{a}	0.0 ± 0.0 ^{a}	0.0 ± 0.0 ^{a}	0.0 ± 0.0 ^{a}
Mn	0.0 ± 0.0 ^{a}	0.0 ± 0.0 ^{a}	0.1 ± 0.0 ^{a}	0.1 ± 0.0 ^{a}
P	76.2 ± 0.0 ^{d}	90.2 ± 0.0 ^{c}	90.9 ± 0.0 ^{b}	95.5 ± 0.0 ^{a}
Mg	4.9 ± 0.0 ^{b}	5.0 ± 0.0 ^{b}	5.2 ± 0.0 ^{a}	5.3 ± 0.0 ^{a}

All parameters are represented as mean of replicates ± standard error ($n = 2$). Means with different superscript letters in the same row are significantly different at $p \leq 0.05$.
Control, ASP10, ASP17, ASP34 – processed cheese with avocado seed powder at the level of 0 g·kg^{-1}, 10 g·kg^{-1}, 17 g·kg^{-1} and 34 g·kg^{-1} formula blend, respectively.

with a relatively higher nutritional value among other fruits [3].

Fig. 3 shows that titratable acidity with opposite trend of pH values (data not shown) of spread-type processed cheese during storage for 3 months. The titratable acidity of ASP-containing processed cheese treatments was higher than control cheese during their storage period. Also, titratable acidity gradually increased in both control and ASP-containing processed cheese with the storage period prolonged, which might be correlated to the emulsifying salts hydrolysis and their interaction with proteins [26].

Fatty acids profile of cheese

The fatty acids proportions of ASP-containing processed cheese were higher than of plain cheese for butyric acid (C4:0), caprylic acid (C8:0), capric acid (C10:0), lauric acid (C12:0), myristic (C14:0) and myristoleic acids (C14:1). This could be due to the fatty acid content of ASP (Tab. 7). However, avocado fruit is considered a natural source of monounsaturated lipids and essential fatty acids such as linoleic and linolenic acid [3]. Foglietta et al. [32] reported that capric, caproic and caprylic acids can be used to decrease both body weight and body fat. Also, palmitioleic, steari, and oleic fatty acid contents of control cheese were higher than ASP-containing processed cheese. Moreover, ASP-containing processed cheese with the lowest level of ASP recorded the highest proportion of various fatty acids including butyric acid, laueic acid, myristic acid, myristoleic acid, pentadecanoic acid, linoleic acid, linolenic acid and the conjugated linolenic acid (CLA). However, the consumption of *trans* vaccenic acid could provide positive effects on human health beyond those linked to CLA regarding cardiovascular diseases, inflammation, cancer, as well as immune functions. Also, intake of dairy products which contained pentadecanoic acid could be used to improve the insulin sensitivity and reduce the risk of diabetes type 2 [33].

Tab. 6. Vitamins content of cheese samples with avocado seed powder.

Vitamin	Processed cheese			
	Control	ASP10	ASP17	ASP34
Thiamine (B1) [mg·kg^{-1}]	0.4 ± 0.0^{d}	1.1 ± 0.0^{c}	2.9 ± 0.0^{b}	3.8 ± 0.4^{a}
Riboflavin (B2) [mg·kg^{-1}]	0.2 ± 0.0^{d}	1.2 ± 0.0^{c}	1.9 ± 0.0^{b}	2.7 ± 0.0^{a}
Niacin (B3) [mg·kg^{-1}]	0.9 ± 0.0^{d}	16 ± 0.1^{c}	36.3 ± 0.0^{b}	63.7 ± 0.1^{a}
Pyridoxine (B6) [mg·kg^{-1}]	1.5 ± 0.0^{d}	3.5 ± 0.4^{c}	6.0 ± 0.0^{b}	16.8 ± 0.0^{a}
Folic acid (B9) [mg·kg^{-1}]	20.1 ± 0.5^{d}	80.1 ± 0.0^{c}	152.6 ± 0.0^{b}	308.6 ± 0.0^{a}
Cobalamin (B12) [μg·kg^{-1}]	44.5 ± 0.0^{d}	69.9 ± 0.0^{c}	122.9 ± 0.0^{b}	225.0 ± 0.0^{a}
Retinol (A) [μg·kg^{-1}]	10.9 ± 0.0^{d}	71.9 ± 0.0^{c}	155.5 ± 0.0^{b}	310.1 ± 0.0^{a}
Ascorbic acid (C) [mg·kg^{-1}]	49.5 ± 0.0^{d}	102.2 ± 0.0^{c}	205.0 ± 0.0^{b}	401.0 ± 0.0^{a}
Tocopherol (E) [mg·kg^{-1}]	0.1 ± 0.0^{d}	2.0 ± 0.0^{c}	4.5 ± 0.0^{b}	10.1 ± 0.0^{a}

All parameters are represented as mean of replicates ± standard error ($n = 2$). Means with different superscript letters in the same row are significantly different at $p \leq 0.05$. Content is expressed on dry weight basis.
Control, ASP10, ASP17, ASP34 – processed cheese with avocado seed powder at the level of 0 g·kg^{-1}, 10 g·kg^{-1}, 17 g·kg^{-1} and 34 g·kg^{-1} formula blend, respectively.

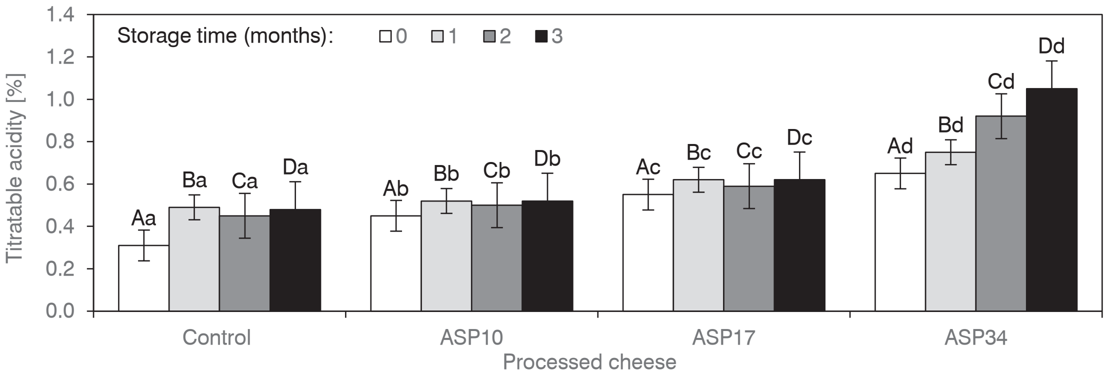

Fig. 3. Titratable acidity of cheese samples with avocado seed powder during storage.

Cheese samples were stored at 5 ± 1 °C. Means with different capital letters during the storage period for each treatment and different small letters between the treatments are significantly different at $p \leq 0.05$ ($n = 4$).
Control, ASP10, ASP17, ASP34 – processed cheese with avocado seed powder at the level of 0 g·kg^{-1}, 10 g·kg^{-1}, 17 g·kg^{-1} and 34 g·kg^{-1} formula blend, respectively.

Tab. 7. Fatty acids profile of cheese samples with avocado seed powder.

Fatty acid [%]	Processed cheese			
	Control	ASP10	ASP17	ASP34
Butyric acid (C4:0)	8.6	11.9	11.9	8.7
Caproic acid (C6:0)	1.6	1.4	2.0	1.6
Caprylic acid (C8:0)	0.9	1.3	1.3	1.8
Capric acid (C10:0)	2.1	2.7	2.9	2.5
Lauric acid (C12:0)	2.5	3.6	3.3	2.8
Myristic (C14:0)	10.2	11.8	10.9	10.3
Myristoleic acid (C14:1)	0.9	1.2	1.0	0.9
Myristolinoleic acid (C14:2)	0.8	0.9	0.8	0.8
Pentadecanoic acid (C15:0)	1.6	1.9	1.6	1.6
Palmitic (C16:0)	28.9	25.4	25.9	29.3
Palmitioleic (C16:1)	2.5	2.4	2.3	2.3
Stearic (C18:0)	10.7	8.8	9.8	9.9
Oleic (C18:1)	24.6	20.1	22.3	23.3
Linoleic acid (C18:2)	1.9	2.4	1.8	2.3
Linolenic acid (C18:3)	0.5	0.7	0.5	0.7
Conjugated linolenic acid (C18:3, n6) (CLA)	1.3	1.3	1.3	1.2

Relative content of fatty acids is given as percentage of peak area.
Control, ASP10, ASP17, ASP34 – processed cheese with avocado seed powder at the level of 0 g·kg^{-1}, 10 g·kg^{-1}, 17 g·kg^{-1} and 34 g·kg^{-1} formula blend, respectively.

Physical properties of cheese

The penetrometer reading of ASP-containing processed cheese were higher than of control cheese during three months of storage, as presented in Fig. 4A. It might be due to the altered contents of protein, fat and calcium, which affect the cheese matrix. Also, Fig. 4A shows a tendency of a decrease in the penetrometer values either in ASP-containing and control processed cheese samples with the storage period prolonged. This may be due to the interaction between ASP and protein network as well as the chemical changes during storage. These results are similar with those reported by MOHAMED et al. [34].

The oil separation percentage (Fig. 4B) of ASP-containing processed cheese were lower than those of control processed cheese during the storage period, which could be due to the increase in cheese acidity and water soluble nitrogen during the storage period. However, processed cheese oil

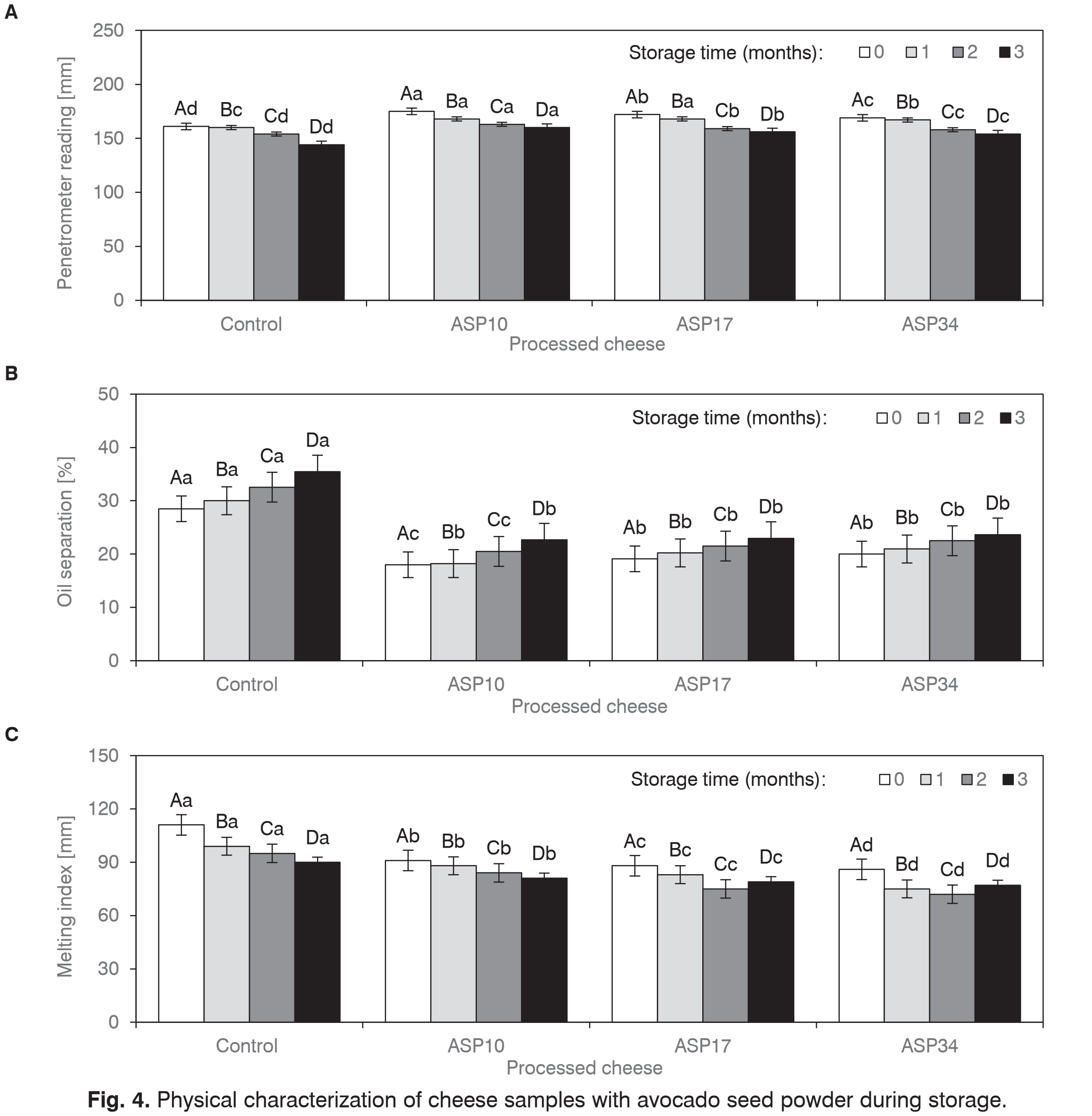

Fig. 4. Physical characterization of cheese samples with avocado seed powder during storage.

A – penetrometer reading, B – oil separation, C – melting index.
Cheese samples were stored at 5 ± 1 °C. Means with different capital letters during the storage period for each treatment and different small letters between the treatments are significantly different at $p \leq 0.05$ ($n = 4$).
Control, ASP10, ASP17, ASP34 – processed cheese with avocado seed powder at the level of 0 g·kg^{-1}, 10 g·kg^{-1}, 17 g·kg^{-1} and 34 g·kg^{-1} formula blend, respectively.

separation depends on the state of milk protein and fat in cheese emulsion that can be affected by type and amount of the raw materials in the base formula, acidity, cooking time and temperature [26]. MOHAMED and HUSSEIN [29] observed gradual decrease in processed cheese oil separation with the increasing content of cress seeds. Also, Fig. 4B shows that the oil separation percentage increased with progressing storage period.

Moreover, Fig. 4C shows that the values of melting index of ASP-containing processed cheese were lower than those of plain cheese during the storage period, while the values of melting index of all cheese samples decreased with storage period prolonged. It could be mainly due to the fatty acids profile of the resulted cheese (Tab. 7), as well as the chemical changes occurring in the resulted cheese including acidity, protein state, emulsifying salts and the slight decrease in moisture content [26].

Colour attributes of cheese

Data on colour of ASP-containing and control cheese samples are presented in Tab. 8. ASP-containing processed cheese had higher values of colour parameter a^* while lower values of colour parameters L^* and b^* compared to control cheese.

Tab. 8. Colour attributes of cheese samples with avocado seed powder.

Colour parameter	Processed cheese			
	Control	ASP10	ASP17	ASP34
L^*	72.3	70.2	68.3	66.9
a^*	11.1	12.5	12.7	12.6
b^*	32.4	28.9	27.9	29.6

Control, ASP10, ASP17, ASP34 – processed cheese with avocado seed powder at the level of 0 g·kg^{-1}, 10 g·kg^{-1}, 17 g·kg^{-1} and 34 g·kg^{-1} formula blend, respectively.
L^* – value represents darkness from black (0) to white (100), a^* – value represents colour ranging from red (0) to green (100), b^* – value represents colour ranging from yellow (0) to blue (100).

This reflected the degree of orange colour due to ASP added.

Texture profile of cheese

Tab. 9 shows TPA of processed cheese containing ASP compared to plain cheese. It could be noted that the addition of ASP increased the hardness of cheese, which was mainly due to the dry matter content (Tab. 4), as well as the presence of different protein types in the prepared ASP-containing cheese, which contained avocado proteins together with milk proteins. Similarly, higher hard-

Tab. 9. Textural profile of cheese samples with avocado seed powder during storage.

	Storage (months)	Processed cheese			
		Control	ASP10	ASP17	ASP34
Hardness [N]	0	26.4 ± 0.1 ^{Dd}	31.6 ± 0.2 ^{Dc}	35.9 ± 0.4 ^{Db}	36.5 ± 0.4 ^{Da}
	1	26.9 ± 0.1 ^{Cd}	32.8 ± 0.2 ^{Cc}	36.7 ± 0.4 ^{Cb}	37.8 ± 0.4 ^{Ca}
	2	29.5 ± 0.6 ^{Bd}	35.6 ± 0.7 ^{Bc}	39.6 ± 0.9 ^{Bb}	40.1 ± 0.5 ^{Ba}
	3	32.2 ± 0.1 ^{Ab}	40.2 ± 0.2 ^{Aa}	44.4 ± 0.2 ^{Aa}	44.9 ± 0.3 ^{Aa}
Springiness [mm]	0	6.1 ± 0.0 ^{Da}	5.2 ± 0.0 ^{Db}	4.9 ± 0.0 ^{Dc}	4.8 ± 0.0 ^{Dc}
	1	6.4 ± 0.0 ^{Ca}	5.6 ± 0.0 ^{Cb}	5.2 ± 0.0 ^{Cb}	5.1 ± 0.0 ^{Cb}
	2	6.7 ± 0.1 ^{Ba}	6.0 ± 0.1 ^{Ba}	5.5 ± 0.1 ^{Bb}	5.4 ± 0.1 ^{Bb}
	3	6.9 ± 0.0 ^{Aa}	6.4 ± 0.0 ^{Aa}	5.9 ± 0.0 ^{Ab}	5.8 ± 0.0 ^{Ab}
Gumminess [N]	0	11.4 ± 0.0 ^{Da}	9.8 ± 0.0 ^{Db}	9.3 ± 0.0 ^{Db}	9.1 ± 0.0 ^{Db}
	1	12.1 ± 0.0 ^{Ca}	10.5 ± 0.0 ^{Cb}	10.3 ± 0.0 ^{Cb}	10.2 ± 0.0 ^{Cb}
	2	14.5 ± 0.2 ^{Ba}	12.5 ± 0.5 ^{Bb}	12.3 ± 0.2 ^{Bb}	12.0 ± 0.3 ^{Bb}
	3	16.7 ± 0.0 ^{Aa}	15.3 ± 0.0 ^{Ab}	15.5 ± 0.0 ^{Ab}	15.3 ± 0.0 ^{Ab}
Cohesiveness	0	0.3 ± 0.0 ^{Dc}	0.3 ± 0.0 ^{Dc}	0.3 ± 0.0 ^{Db}	0.4 ± 0.0 ^{Da}
	1	0.3 ± 0.0 ^{Cc}	0.3 ± 0.0 ^{Cc}	0.3 ± 0.0 ^{Cb}	0.5 ± 0.0 ^{Ca}
	2	0.3 ± 0.0 ^{Bb}	0.3 ± 0.0 ^{Bb}	0.4 ± 0.0 ^{Bb}	0.5 ± 0.0 ^{Ba}
	3	0.3 ± 0.0 ^{Ab}	0.4 ± 0.0 ^{Ab}	0.4 ± 0.0 ^{Ab}	0.5 ± 0.0 ^{Aa}
Chewiness [N·mm^{-1}]	0	68.9 ± 0.0 ^{Da}	50.9 ± 2.3 ^{Db}	45.8 ± 0.0 ^{Dc}	43.8 ± 0.0 ^{Dc}
	1	76.9 ± 0.0 ^{Ca}	58.7 ± 2.3 ^{Cb}	53.2 ± 0.0 ^{Cb}	52.1 ± 0.0 ^{Cb}
	2	97.1 ± 1.2 ^{Ba}	75.0 ± 1.3 ^{Bb}	67.8 ± 0.8 ^{Bc}	65.4 ± 0.8 ^{Bc}
	3	116.9 ± 0.0 ^{Aa}	97.0 ± 0.0 ^{Ab}	92.2 ± 0.0 ^{Ab}	89.0 ± 0.1 ^{Ac}

All parameters are presented as mean of replicates ± standard error (n = 2). Means with different superscript letters in the same row are significantly different at $p \leq 0.05$. Content is expressed on dry weight basis.
Control, ASP10, ASP17, ASP34 – processed cheese with avocado seed powder at the level of 0 g·kg^{-1}, 10 g·kg^{-1}, 17 g·kg^{-1} and 34 g·kg^{-1} formula blend, respectively.

ness values were observed for processed cheese incorporated with date seed powder [26]. Moreover, other TPA of ASP-containing processed cheese treatments were lower than of plain cheese, which could be due to the moisture and fat contents. All TPA of processed cheese increased as the storage period progressed. The obtained findings coincided with those mentioned by Darwish et al. [35].

Organoleptic properties of cheese

The organoleptic attributes of processed cheese are presented in Fig. 5. The results show that the addition of ASP during processed cheese production improved all sensorial attributes without significant ($p < 0.05$) differences including flavour (Fig. 5A), body and texture (Fig. 5B) as well as appearance (Fig. 5C). This was reflected by an increase in their total score (Fig. 5D) compared to plain processed cheese without any defects appearing during the storage period of 3 months. Flavour attributes coincided with the fatty acids content of ASP-containing processed cheese in comparison with plain cheese (Tab. 7). However, Mohamed and Hussein [29] reported that the cress seeds had peppery, tangy flavour and aroma, which improved the processed cheese flavour. Also, the queen cakes as avocado baked products produced using 150 g·kg^{-1} avocado seed powder had acceptable organoleptic properties that were comparable with control, making it a good formulation to obtain an acceptable product [5]. Regarding to body and texture of ASP-containing

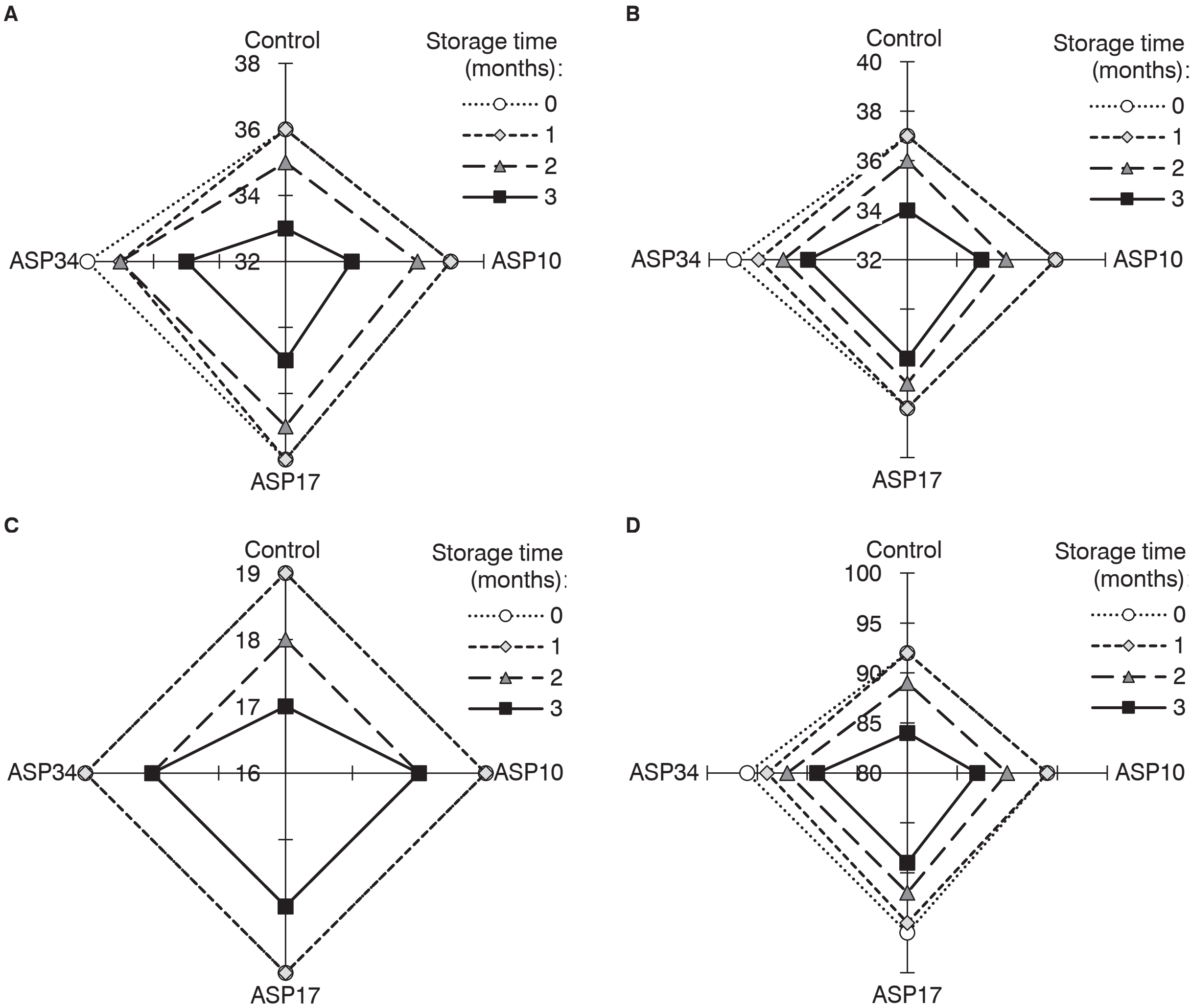

Fig. 5. Organoleptic properties of cheese samples with avocado seed powder during storage.

A – flavour, B – body and texture, C – appearance, D – total score.
Cheese samples were stored at 5 ± 1 °C.
Control, ASP10, ASP17, ASP34 – processed cheese with avocado seed powder at the level of 0 g·kg^{-1}, 10 g·kg^{-1}, 17 g·kg^{-1} and 34 g·kg^{-1} formula blend, respectively.

processed cheese, their higher protein content could improve the physico-chemical properties compared to plain processed cheese (Tab. 4). These results were confirmed by TPA of cheese as presented in Tab. 9 [26]. In addition, the appearance of ASP-containing processed cheese scored higher than the plain cheese. This could be attributed to ASP colour, which improved the cheese appearance as well as consumer overall acceptability. Colour attributes (Tab. 8) confirmed these findings.

CONCLUSIONS

ASP could be used in the processing of spread-type processed cheese as an excellent functional ingredient at the level of 34 $g\cdot kg^{-1}$. This gained good overall acceptability with physico-chemical and textural characteristics without any defects appearing during 3 months of storage. Hence, ASP as a novel by-product from avocado may improve texture and could increase fibre content as well as biological activity of processed cheese.

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