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THE LEVEL OF 3-MCPD FATTY ACID ESTERS IN VEGETABLE OILS AND MARGARINES COLLECTED FROM TURKISH MARKET

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ABSTRACT
The aim of the present research was to investigate the content of 3-MCPD esters in vegetable oils (hazelnut, sunflower, rapeseed and corn oil) and margarines sold out in Turkish local markets. A series of 27 margarines and vegetable edible oil collected from markets were analyzed in the study according to DGF C VI 18 (10) method. Among vegetable oils, corn oil presented highest amount of 3-MCPD esters (2.49 mg/kg) followed by rapeseed oil (0.95 mg/kg), sunflower oil (0.79 mg/kg) and hazelnut oil (0.45 mg/kg), respectively. Highest amount of 3-MCPD esters was detected for margarines (4.54 mg/kg) among analyzed samples. The results demonstrate that 3-MCPD esters can occur at different levels in various vegetable oils and margarines available on Turkish markets.
Keywords: 3-MCPD esters, margarine, market survey, vegetable oil

TÜRK PİYASASINDAN TOPLANAN BİTKİSEL YAĞLAR VE MARGARİNLERDE 3-MCPD YAĞ ASİDİ ESTERLERİNİN DÜZEYİ

ÖZ
Çalışmanın amacı Türkiye piyasasında satışa sunulan bitkisel yağlar (fındık yağı, ayçiçek yağı, kanola yağı ve mısır yağı) ve margarinlerde 3-MCPD ester içeriklerinin belirlenmesidir. Çalışmada marketlerden alınan toplam 27 adet margarin ve bitkisel yağ DGF C VI 18 (10) metoduna göre analiz edilmiştir. Bitkisel yağlar arasında en yüksek 3-MCPD ester miktarı (2.49 mg/kg) mısır yağında tespit edilmiş olup, bunu sırasıyla kanola yağı (0.95 mg/kg), ayçiçek yağı (0.79 mg/kg) ve fındık yağı (0.45 mg/kg) izlemiştir. Analiz edilen örnekler arasında en yüksek 3-MCPD ester miktarı margarinlerde (4.54 mg/kg) saptanmıştır. Çalışma sonuçları Türkiye piyasasında yer alan çeşitli bitkisel yağlar ve margarinlerde farklı miktarlarda 3-MCPD esterlerinin bulunabileceğini göstermektedir.
Anahtar kelimeler: 3-MCPD esterleri, bitkisel yağ, margarin, pazar araştırması

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INTRODUCTION

3-Monochloropropane-1,2-diol (3-MCPD), the representer of chlorinated chloropropanols (Hamlet et al., 2002) is a food processing contaminant which may be formed during processing of various fat containing matrices at elevated temperatures in the presence of chloride containing components (Ermacora and Hrncirik, 2013; Karl et al., 2016). 3-MCPD belongs the group of chloropropanols which consists of 5 main compounds including 2-monochloropropane- 1,3-diol (2-MCPD), 3-monochloropropane-1-ol, 1,3-dichloropropane-2-ol (1,3-DCP), 2,3-dichloropropane-1-ol (2,3-DCP) and 3-monochloropropane-1,2-diol (3-MCPD) (Lee and Khor, 2015). The studies have shown that the chloropropanols, specially chloropropanediols, can occur both in free and ester (bound) forms (Jedrkiewicz et al., 2016). The free form of 3-MCPD was firstly discovered by Velíšek et al. (1978) in acid-hydrolysed vegetable proteins and its ester form was initially discovered by Davidek et al. (1980). In following years, 3-MCPD was detected in its free form in many heat treated food products including coffee (Doležal et al., 2005), cheese (Crews et al., 2001), malt-based food products (Hamlet et al., 2002), toast, biscuits and bread crust (Hamlet and Sadd, 2004; Breitling-Utzmann et al., 2005; Weißhaar, 2011). The maximum daily tolerable limit was defined by Scientific Committee on Food in 2001, as 2 μg/kg body weight for free 3-MCPD (Karl et al., 2016).

In a recent study (Wöhrlin et al., 2015), it was shown that 3-MCPD esters may occur even in infant formula. In the report of The German Federal Institute for Risk Assessment (BfR), it was reported that 3-MCPD was fully cleaved from its esters in gastrointestinal tract (Bockisch et al., 1993). From this point of view, it has been declared that all threshold levels for the compounds in free form are valid for the ester form. Furthermore, in the studies of Abraham et al. (2013) and Appel et al. (2013), the relative bioavailability of 3-MCPD esters was reported as 86 %.

It was in the literature reported that 3-MCPD is hazardous for human health (Cho et al., 2008; MacMahon et al., 2013). Until today, 3-MCPD was detected in various foods and food ingredients, especially in refined vegetable oils (Zelinková et al., 2006; Weißhaar, 2008; Weißhaar and Perz, 2010; Schilter et al., 2011).

Oil refining is an essential process to ensure oil quality and safety (Hrncirik and van Duijn, 2011). During oil refining, not only free fatty acids are removed, but also undesired flavour and color components and minor contaminants such as pesticides and polyaromatic hydrocarbons are removed (Hrncirik and van Duijn, 2011). However, thermal treatment of oils may trigger the occurrence of some undesired components including *trans* fatty acids, acylglycerol polymers (Beljaars et al., 1994), cyclic fatty acid esters (Destaillats and Angers, 2005) and 3-MCPD esters (Hrncirik and van Duijn, 2011; Craft et al., 2012). There are two main thermal treatments where oils are subjected to high temperatures; deep fat frying and deodorization (Destaillats et al., 2012). Deodorization is attributed as a pure thermal process of lipids without the addition of other food constituents (Destaillats et al., 2012). During refining of edible oils, deodorization is characteristically conducted at temperatures between 180°C and 270°C under 3-7 mbar vacuum with the introduction of injected stripping steam into the process (Dijkstra and Segers, 2007).

It has been in the literature reported that chlorination of acylglycerols may occur during deodorization step of oils and this situation results in the formation of 3-MCPD esters (Zelinková et al., 2006; Franke et al., 2009).

In refined oils, the presence of 3-MCPD esters was primarily reported by Zelinková et al. (2006). In their study, the contents of 3-MCPD esters were found up to 2.46 mg/kg for virgin oils. Since almost no bound 3-MCPD was found in unrefined oils, the major problem was reported as the refining process in terms of the occurrence of 3-MCPD esters (Weißhaar, 2008). Jędrkiewicz et al. (2016) reported that 3-MCPD ester levels of refined rapeseed, sunflower and olive oils in the Polish markets were between 0.26-0.30 mg/kg.

On the other hand, the lipid fractions in margarines available on Polish markets contained 1.3-7.3 mg/kg of bound 3-MCPD. Becalski et al. (2015) stated that walnut, grape seed, rice bran and palm oil shortenings were found to have the highest level of 3-MCPD esters in the Canadian market. MacMahon et al. (2013) investigated the content of 3-MCPD esters present in edible oils in the United States and they reported that the amount of 3-MCPD esters ranged from 0.005 to 7.2 mg/kg in 94 refined edible oils.

There are various reports in literature analysing the contents of 3-MCPD esters in vegetable oils and different products, however to the best of our knowledge there is no information considering the amounts of 3-MCPD esters in vegetable oils and margarines in Turkish local markets. The current data may be useful to regulate maximum daily tolerable limit of 3-MCPD and other compounds belonging to the group of chloropropanols.

MATERIALS AND METHODS

Materials and Chemicals

The table margarine and vegetable oil samples were collected from local markets in Aydın, Turkey. The samples were kept at 4°C until being analyzed. There were a totally of 5 corn oil, 3 sunflower oil, 3 hazelnut oil, 2 rapeseed oil and 14 margarine samples.

Toluene, *tert*-butyl methyl ether (tBME), methanol, hexane, ethyl acetate, diethyl ether, *iso*-octane, sodium methoxide, sodium chloride, sodium bromide, sodium sulphate, phenyl boronic acid and 3-chloro-1,2-propane-1,1,2,3,3-d5-diol were purchased from Sigma (St-Louis, USA).

Methods

Determination of 3-MCPD ester contents

The concentrations of 3-MCPD esters were determined according to the DGF Standard Method C-VI 18 (10) (DGF, 2009). In this indirect method, after addition of internal standard and *tert*-butyl methyl ether, the esters were cleaved under alkali conditions using sodium methoxide. The reaction was then stopped either with sodium chloride (Assay A) or sodium bromide (Assay B). The non-desired, non-polar compounds were then removed using *n*-hexane and the resulting solution was extracted with the mixture of diethyl ether and ethyl acetate. Finally it was derivatized with phenylboronic acid and injected to GC-MS (GC-MS 2010, Shimadzu, Kyoto, Japan) system. Analysis of target compounds was performed according to the method of Cheng et al. (2016). The chromatographic separation was achieved using a capillary column (Restek Rxi-5 ms column, 30 m × 0.25 mm i.d. × 0.25 μm film thickness) and the injector was run in splitless mode. Helium was the carrier gas with a constant flow rate of 1.18 mL/min. The oven temperature programme was set as follows: 80°C raised to 155°C with a heating rate of 5°C/min and then it was raised to 300°C with 60°C/min. Quantification of the results was carried out by monitoring characteristic ions at m/z 150 for 3-MCPD-d5 and m/z 147 for 3-MCPD.

RESULTS AND DISCUSSION

The ester-linked 3-MCPD contents of corn, sunflower, hazelnut and rapeseed oils belonging to different brands were presented in Table 1. Among analyzed samples of vegetable oils the content of 3-MCPD esters ranged between 0.51-2.49 mg/kg for corn oil belonging to different brands. Weißhaar (2011) reported that the median 3-MCPD ester content as 1.7 mg/kg in refined corn oils sold out in German markets. Haines et al. (2011) and Kuhlmann (2011), on the other side, reported lower 3-MCPD ester contents for refined/processed corn oils.

The sunflower oils contained 0.12-0.79 mg/kg of bound 3-MCPD. The results are in good agreement with the results (0.6 mg/kg) of Raznim et al. (2012) in terms of 3-MCPD esters and with the results of Kuhlmann (2011), who reported that refined sunflower oil contained 0.1-2.1 mg/kg of bound 3-MCPD. However Zelinková et al. (2006) reported lower content of 3-MCPD esters for refined sunflower oils in Hungarian (<300 μg/kg) and French markets (<100 μg/kg).

The rapeseed oils in local markets in Turkey contained 0.42-0.95 mg/kg of 3-MCPD esters.

Similar 3-MCPD ester contents (0.6 mg/kg) for retailed rapeseed oils was obtained by Raznim et al. (2012). Lower content of 3-MCPD esters in refined rapeseed oil was reported by Kuhlmann (2011), who showed that refined rapeseed oils contained from 0.1 to 1.0 mg/kg of bound 3-MCPD.

Table 1. The amount of 3-MCPD esters (mg/kg) in vegetable oils sold out in local markets of Turkey (C: Corn oil, S: Sunflower oil, H: Hazelnut oil, R: Rapeseed oil)

Oil	3-MCPD Esters (mg/kg)
C1	0.83±0.17
C2	0.57±0.17
C3	2.49±0.36
C4	0.51±0.06
C5	1.49±0.22
S1	0.12±0.02
S2	0.79±0.18
S3	0.61±0.02
H1	0.24±0.01
H2	0.45±0.13
H3	0.26±0.04
R1	0.95±0.20
R2	0.42±0.08

The content of bound 3-MCPD in hazelnut oil was 0.24-0.45 mg/kg. Similar findings (<100 μg/kg) in terms of 3-MCPD ester contents were reported by Zelinková et al. (2006) for hazelnut oil from French origin. However higher amounts of bound 3-MCPD was reported by Kuhlmann (2011), who showed that in addition to palm oil, hazelnut oil contained unexpected high levels of 3-MCPD esters (19 mg/kg). Crews (2012) also reported high contents of ester-linked 3-MCPD (1.2-19 mg/kg) for refined hazelnut oil.

There were notable differences between the content of 3-MCPD esters of vegetable oil samples analyzed in the current work. The main precursors for the formation of 3-MCPD esters were reported in literature as mono- and diacylglycerols, total chlorine and chlorides (Franke et al., 2009). However the amount of critical precursors generally depend on oil type and quality (Craft and Destaillats, 2014). The differences in 3-MCPD ester contents of various oils may be attributed to the properties of raw material and deodorization conditions applied during refining process.

The margarines available on Turkish markets contained 0.57-4.54 mg/kg of 3-MCPD esters (Table 2). The highest level of 3-MCPD esters (4.54 mg/kg) on Turkish markets was lower than the findings of Jedrkiewicz et al. (2016) who reported that the lipid fractions of margarines available on Polish markets contained 1.3-7.3 mg/kg of bound 3-MCPD. The findings in the current study were also compatible to the results for margarines (0.4-4.5 mg/kg) reported by Weißhaar (2011) and German Federal Ministry of Food and Agriculture who stated that 200 margarine samples from German markets contained 224 μg/kg of 3-MCPD esters (Raczyk et al., 2017). Chung et al. (2013) also reported similar 3-MCPD ester content (93 μg/kg) for the margarines retailed in Hong Kong. Different studies in literature also highlighted that margarines are an abundant source of 3-MCPD esters (Bakhiya et al., 2011; Jędrkiewicz et al., 2016).

Table 2. The amount of 3-MCPD esters (mg/kg) in margarines sold out in local markets of Turkey (M: Margarine)

Oil	3-MCPD Esters (mg/kg)
M1	0.57±0.15
M2	0.58±0.05
M3	1.17±0.34
M4	0.84±0.17
M5	1.04±0.14
M6	1.93±0.02
M7	2.47±0.36
M8	1.35±0.42
M9	1.99±0.26
M10	1.04±0.15
M11	0.88±0.13
M12	1.52±0.28
M13	2.35±0.17
M14	4.54±0.97

In the present work, the 3-MCPD ester contents of margarines were in general higher than vegetable oils. This situation can be linked to the fact that almost all margarine samples tested in the current study contained palm oil which was reported to contain high amounts of 3-MCPD esters in literature (Raznim et al., 2012; Jedrkiewicz et al., 2016). Moreover the situation can also be linked to presence of salt in margarine formulations which may facilitate the formation of 3-MCPD esters (Becalski et al., 2015).

CONCLUSION

The objective of the current study was to determine the amounts of 3-MCPD esters in vegetable oils and margarines sold out in Turkish local markets.

Results of the study demonstrated that while some oils like sunflower, hazelnut and rapeseed oil showed contents of 3-MCPD esters below 1 mg/kg; corn oil and margarines contained much higher amounts of the esters which clearly show that the various types of oils had different qualifications for the formation. The high content of 3-MCPD esters in margarines was attributed to the lipid fraction of margarine formulations which mainly contain palm oil where 3-MCPD esters can occur in high concentrations considering the data presented in literature.

The present data can be used to determine the exposure estimations and risk assessments for 3-MCPD esters in foods. Further investigations should be made to determine the contents of bound 3-MCPD in various food products in order to develop novel analysis techniques and effective mitigation strategies.

Conflict of interest

The authors declare that there is no conflict of interest.

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