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Liquid Chromatography−Electrospray Ionization−Tandem Mass Spectrometry Method for the Determination of Epoxidized Soybean Oil in Food Products

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Epoxidized soybean oil (ESBO) is widely used as a plasticizer and stabilizer in such polymers [poly(vinyl chloride) in particular] commonly adopted for manufacturing of gaskets of the lids for glass jars and plastic films for food packaging. Human exposure to ESBO and its derivatives is likely to occur over a lifetime with a significant variation according to life stage. A reversed phase liquid chromatography interfaced with electrospray ion trap tandem mass spectrometry method for the determination of ESBO in foods was developed. A simple sample treatment procedure entailing the use of an extraction step with dichloromethane without any further cleanup was proved. Chromatographic separation was performed using two C18 columns with an aqueous acetic acid−acetone−acetonitrile mixture as the mobile phase under gradient conditions. The method was validated in terms of detection limits (4 mg kg^{-1}), quantitation limits, linearity (established over 2 orders of magnitude), recovery (good mean recoveries, higher than 90% for all of the signals detected), precision (RSD% < 8), and trueness. The applicability of the method to the determination of ESBO in different food matrices (in particular those rich in edible oil) was demonstrated, and the performances were compared to those reachable by the commonly well-known gas chromatography−mass spectrometry procedure.

KEYWORDS: Epoxidized soybean oil; LC-MS/MS; gaskets; food matrices; sauces

INTRODUCTION

ESBO (epoxidized soybean oil; see **Figure 1**) is a well-known plasticizer (prepared by epoxidation of soybean oil with an active oxygen compound like a peroxide or a peracid) used in poly(vinyl chloride) (PVC)-lined metallic closure gaskets: with this technical solution, it is possible to form an airtight seal, which prevents microbiological contamination in traditionally sealed jars and bottles adopted to contain food commodities, and to provide easy opening at the same time. Furthermore, ESBO acts as a stabilizer and hydrogen chloride scavenger in relation with the typical PVC aging phenomena.

Food jars are commonly closed with a metal cap coated with a gasket made of PVC mixed with up to 40−50% plasticizers (ESBO, epoxidized linseed oil, and other different substances) in direct contact with the food, and in these formulations, ESBO can usually be present up to 35% (*1*). There is thus a potential for the migration of ESBO and its derivatives into the food during both sterilization and storage. Probably the major part of the migration occurs during the packaging and sterilizing procedure, when the food is warm and under high pressure; then, the migration can continue by occasional contact during transport and storage.

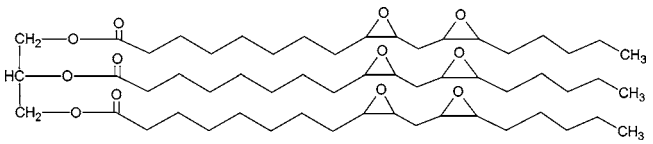

Figure 1. Structure of $(LLL)_{ox}$, one of the defined chemical species in ESBO (from linoleic in sn-1, sn-2, and sn-3). See also ref *2* for a more complete explanation about the complex composition of ESBO.

The recent growing interest in ESBO is derived from a request of the investigation on the infants exposure evaluation that the European Food Safety Authority (EFSA) made in 2004: in particular, the EFSA recommended the development of a specific migration limit (SML) for ESBO in baby foods (*2*, *3*). The toxicity of ESBO was assessed for the first time and extensively evaluated in 1988 (*4*). Samples showed mild skin- and eye-irritating properties (*5*). Then, ESBO presented no evidence of carcinogeneticity or genotoxicity; ESBO also showed very low acute toxicity in rats (LD_{50} > 5 g/kg bw), and no indication of mutagenicity or adverse effects was seen. After that, the Scientific Committee for Food of the European Union proposed a tolerable daily intake of 1 mg kg^{-1} body weight, resulting in a maximum tolerated migration (for adults of 60 kg) corresponding to the overall migration; for this reason,

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ESBO was authorized in Europe as an additive without a SML in foods (*6*, *7*).

Vice versa, derivatives of ESBO, such as chlorohydrins (formed by the interaction of ESBO with HCl eliminated from PVC degradation processes) and also cyclic compounds, are at the moment under investigation to better understand their real migration in food, chemical structures, and toxicological concerns (*8*, *9*). Most recent surveys (*10*, *11*) have shown that samples of baby food collected from the market can contain ESBO above the legally permitted overall migration level of 60 mg kg^{-1} and in some cases over 100 mg kg^{-1}. Previous scientific works suggested that the natural occurrence of diepoxidized fatty acids and triglycerides is negligible in relation to the level arising from migration (*12*, *13*). The analytical method developed was in fact accordingly based on measurement of the diepoxidized fatty acid component (linoleic acid); thus, levels of ESBO recorded in foods using this method can be assumed to originate primarily from ESBO migration. In this way, the most well-known gaschromatographic procedure for ESBO analysis is the one set up by Castle et al. in 1988 (*14*): lipids extraction, transmethylation in alkaline conditions, conversion into fatty acid methyl esters, and a final derivatization of the epoxide groups to form stable 1,3-dioxolane compounds easily detectable due to their characteristic MS fragmentation. This method works very well but needs a diepoxy methyl-eicosadienoate commercially unavailable internal standard (only very recently, some chemical suppliers are considering the possibility of including it in their catalog), two-stage derivatization, and double evaporation of the sample to dryness, which made it quite time-consuming and not so user-friendly; the limit of detection (LOD) could vary significantly with the nature of the food matrix (in relationship with the state of dispersion of fat rather than the fat content itself) and is in general around 5 mg kg^{-1}. At the time of writing of this paper, an interesting modified method, based on an on-line transfer of the normal phase liquid chromatography (LC) extract eluted toward a gas chromatograph equipped with a flame-ionization detector, was published (*15*).

The purpose of the present work was to develop a LC−electrospray ion trap−tandem mass spectrometry (LC-ESI-ion trap-MS/MS) method for ESBO determination in complex food matrices.

A single-laboratory validation procedure was executed including evaluation on detection limits, quantitation limits, linearity, accuracy, recovery, and selectivity. To our knowledge, the work reported here is the first LC-MS method dedicated to the analysis of ESBO in food products. The real application was demonstrated by a survey on Italian retail samples and a parallel data comparison with the traditional gas chromatography−mass spectrometry (GC-MS) procedure carried out on various food sauce products.

MATERIALS AND METHODS

Chemicals and Consumables. Different ESBO commercial products kindly donated from producing caps companies were analyzed; no high differences in the compositions were evident. After comparison, the substance Epoxol D65S (Faci, Genova, Italy) was chosen as a reference. Dichloromethane, acetonitrile, methanol, and acetone were high-performance liquid chromatography (HPLC) gradient grade solvents and were supplied by Merck (Darmstadt, Germany). Acetic acid was also an HPLC gradient grade reagent obtained from BDH VWR International Ltd. (Poole, England). Ultrapure water was used throughout the experiments (MilliQ system, Millipore, Bedford, MA). Syringe filters (0.45 μm) were made of hydrophilic polypropylene (GHP Acrodisc, Pall Gelman Laboratory, Ann Arbor, MI); glass vials with septum screw caps were supplied by Agilent Technologies (Willimgton, DE).

Experimental GC-MS Analysis. The entire contents of each jar were removed and homogenized. Each homogenate was then extracted and treated by transmethylation with subsequent derivatization of the epoxide groups to form dioxolane derivatives analyzed by GC-MS technique following the Castle et al. procedure (*12*). GC-MS analysis was carried out on a GC HRGC 5300 Mega Series equipped with a split/splitless injection port and a QMD 1000 mass detector (Carlo Erba Instruments, Milan, Italy).

Experimental LC-MS Analysis. *Sample Preparation.* On some occasions, for food products in edible oil, analyzing only the oil by referring the concentration to the whole product was useful by the admitted assumption that the whole ESBO was present in the oil phase. Anyway, a good homogenization of the sample was the first step of sample preparation, especially because ESBO concentrated near the stopper in the upper portion of the food. Therefore, after the homogenization of the content of at least three jars (which also included the food adhering to the lid in order to obtain an average result) with UltraTurrax (IKA Werke, Janke & Kunkel GmbH & Co KG, Staufen, Germany), an accurately weighed subsample (2 g) was further treated in a vortex mixer with dichloromethane (20 mL) for 1 min. The mixture was centrifuged at 3000 rpm for 10 min, and the clear supernatant was quantitatively transferred (3 mL) into a vial. It was then filtered through a syringe filter before LC-MS analysis.

Calibration and Quantification. "ESBO free" pesto and tomato-based sauces were appropriately produced in pilot plants and packed without the use of gaskets (the absence of ESBO had been in any case verified apart); they were then used as blanks to calculate matrix-matched LODs and limits of quantitation (LOQs) and to generate calibration curves directly obtained in the matrix itself. Working solutions of ESBO (1000 ppm) were obtained by dissolving 100 mg of ESBO commercial product in 100 mL of dichloromethane. These solutions were considered stable at 4 °C for at least 1 month. The method used for the quantitative analysis of ESBO was different according to the availability of uncontaminated food matrices: If an "ESBO-free" matrix was effectively available, the calibration curve was created on it, preparing spiked samples at opportune concentrations (in our case, in a range from 10 to 100 mg kg^{-1}, spiking 2 g subsamples with 20−200 μL of a 1000 mg kg^{-1} ESBO standard solution in dichloromethane) and carrying out the extraction in an analogous way applied for unknown samples. When this was not possible, the selected procedure executed on each single unknown sample was the standard addition method at at least three different levels (in general 30, 60, and 90 mg kg^{-1} or opportunely chosen according to the probable concentration expected; see **Figure 3**). For both, the quantification approaches were based on the areas of a series of principal peaks that altogether represented approximately 90% of the ESBO composition. Quantification of the greater possible number of components was considered necessary, taking into account potentially different migration levels of each single ESBO mixture component from the gasket toward the food matrix, because of a possible unequal content in the various commercial caps or a different migration ability in relation to the chemical nature of the food matrix involved.

LC. The pump used for reversed phase (RP) HPLC was a Surveyor LC Pump (Thermo Finnigan, San Jose, CA), a dual piston quaternary pump with a built-in vacuum degasser. The chromatographic separation was performed using two Synergy 4μ Hydro-RP 80A, 150 mm × 2.00 mm (Phenomenex, Torrance, CA) columns in series with a flow rate of 0.4 mL/min and a gradient solvent program based on 15 mM acetic acid in water (15 mM AcOH), acetonitrile (AcN), and acetone (see **Table 1**). Injections (10 μL) were made using a Surveyor Autosampler (Thermo Finnigan).

Mass Spectrometry. The ESI experiments were carried out in a Ion-Trap LCQ Advantage (Thermo Finnigan) mass spectrometer in positive scan mode with a capillary temperature of 350 °C and a sheat gas flow rate set to 40 units; the spray voltage and the capillary voltage were kept at 4500 and 45 V, respectively. Selected reaction monitoring (SRM) experiments were performed using a maximum injection time

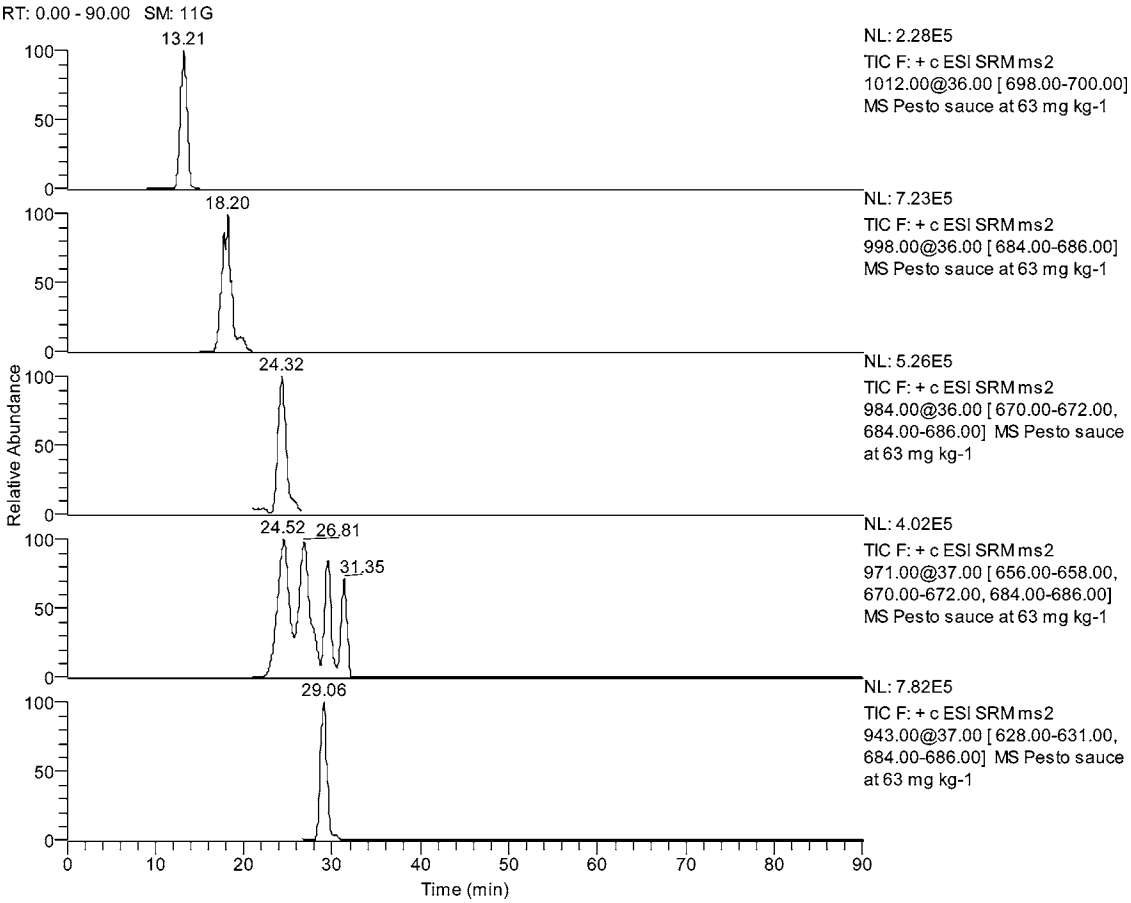

Figure 2. LC-ESI-MS/MS ESBO chromatograms of selected ionic fragments of ESBO mixture components in the extract of a pesto sample with a concentration close to 60 mg kg^{-1}.

Table 1. Chromatographic Gradient Solvent Program

time (min)	AcOH 15 mM (%)	AcN (%)	acetone (%)
0.0	10	85	5
4.0	10	85	5
8.0	2	88	10
12.0	2	88	10
35.0	2	10	88
55.0	2	10	88
65.0	10	85	5
80.0	10	85	5

of 250 ms and three microscans. All parts of the equipment and data processing were performed by the computer software Xcalibur (Thermo Finnigan).

RESULTS AND DISCUSSION

The method set up started with a study of ESBO commercial products by repeated infusions of 1 ppm standard solutions in dichloromethane to evaluate all of the detectable characteristic signals and to optimize operative parameter conditions in terms of capillary potential, flux, eluant composition, and so on. The optimized final method consists of the quantification of the five more intense and significant signals, which all together represent approximately 90% of the ESBO mixture. In particular, in this way, $[M + Na]^+$ clusters for the different epoxidized tryacylglycerols can be observed (**Table 2**). The percentage contribution of each of these peaks has been evaluated according to their signal intensity and also by injections under isocratic conditions (15 mM AcOH:AcN:acetone = 2:88:10). All of the signals have been acquired in SRM mode considering the transitions as reported on **Table 3** (see also **Figure 2**). It has to be evidenced that in the fragmentation pattern of each single component, there is the loss of a fragment corresponding to the diepoxidized linoleic chain (*m*/*z* 313). Finally, analytical quality assurance measures were employed for ESBO, which involved inclusion

Table 2. ESBO Mixture Components Signals Detected by LC-MS

signal	*m*/*z*	attributions[a]	formula	% in the mixture[b]
$[M_I + Na]^+$	1012	**(LnLL)ox**, (LnLnO)ox	$C_{57}H_{96}O_{13}$	14
$[M_{II} + Na]^+$	998	**(LLL)ox**, (LnLO)ox, (LnLnS)ox	$C_{57}H_{98}O_{12}$	34
$[M_{III} + Na]^+$	984	**(OLL)ox**, (LnOO)ox, (LnLS)ox	$C_{57}H_{100}O_{11}$	25
$[M_{IV} + Na]^+$	971	**(LOO)ox**, (LLS)ox, (LnOS)ox	$C_{57}H_{102}O_{10}$	14
$[M_V + Na]^+$	943	(SOO)ox, (SSL)ox	$C_{57}H_{106}O_8$	13

[a] Symbols used refer to the triacylglycerol structure: Ln, linolenic chain; L, linoleic chain; O, oleic chain; and S, stearic chain. With the acronym "ox", it is intended that all of the triacylglycerol double bonds have been oxidized. Attributions reported in bold style are considered the most probable taking into account the medium composition of natural soybean oil. [b] These percentages have been calculated neglecting ESBO mixture minor constituent (which altogether represent approximately 10% of the total).

Table 3. MS/MS Parameters for Detection of ESBO

ESBO component	parent ion *m*/*z*	fragments monitored *m*/*z*	normalized collision energy %
M_I	1012	699	36
M_{II}	998	685	36
M_{III}	984	685, 671	36
M_{IV}	971	685, 671, 658	37
M_V	943	685, 630	37

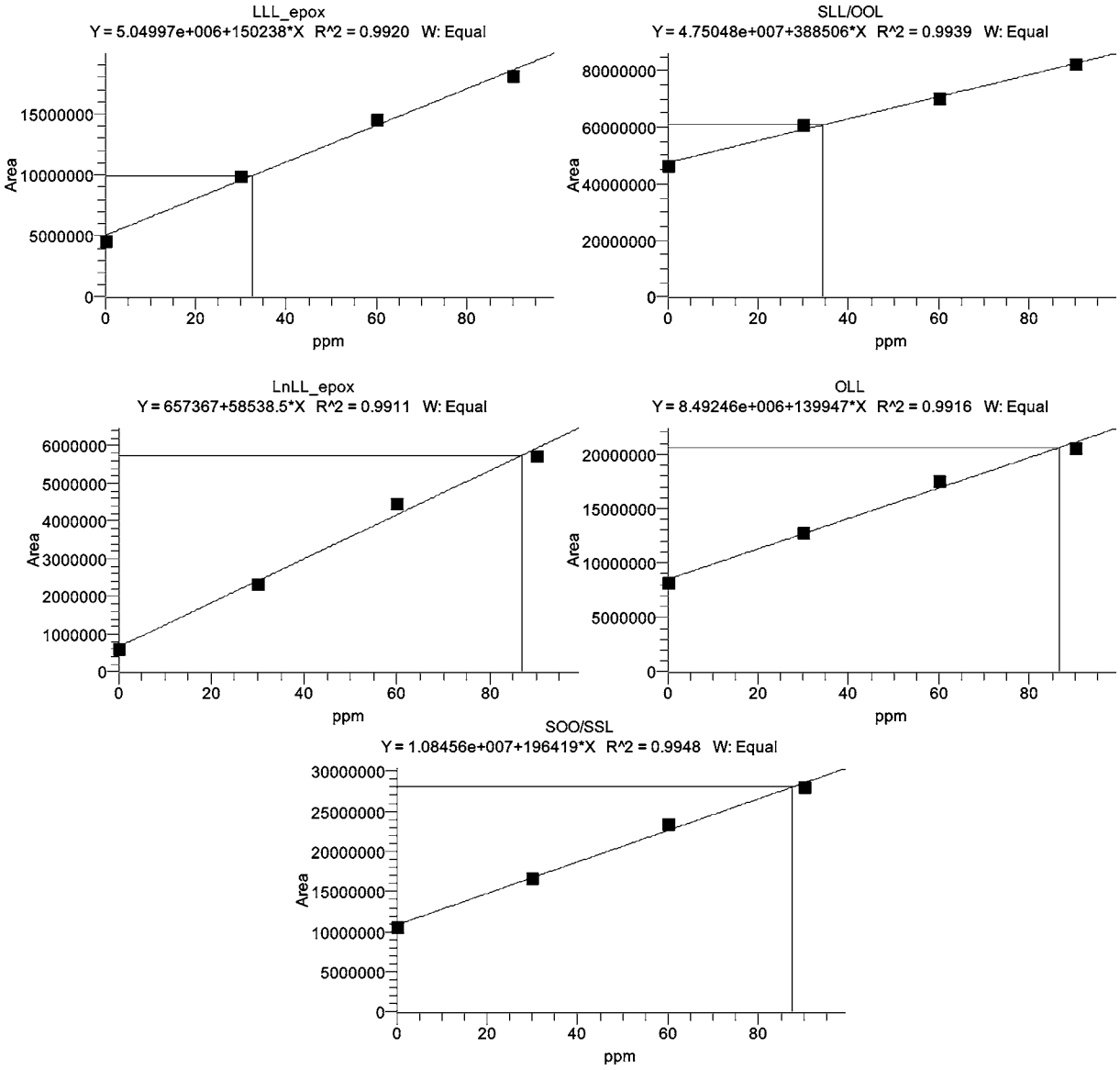

Figure 3. Application of the standard addition method on a tomato–cheese-based sauce (calibration curves obtained for each ESBO mixture component signals).

of 10 samples in each analysis batch, duplicate samples (prepared from an ESBO-free food matrix when it was possible) spiked at 30 and 60 mg kg^{-1}, and a reagent blank.

Method Validation. *Selectivity.* **Figure 4** shows the chromatograms of a pesto sauce extract (with few ppm ESBO content) and the same extract spiked with ESBO standard solution up to a final concentration of 60 mg kg^{-1}, demonstrating that there are no significant variations in shape and retention time of the peaks and therefore that the method is compatible with a complex food matrix very enriched in edible oil or with a high fat content.

Linearity. For the establishment of linearity, five different concentrations (10, 20, 30, 60, and 100 mg kg^{-1}) were obtained by spiking an ESBO-free pesto matrix, executing at least three replicate injections. ESBO compound responses were found linear (**Table 4**) over the concentration range explored with correlation coefficients >0.99 in all cases. Mandel's fitting test was performed at a 95% confidence level to satisfy linearity requirements.

Accuracy. Accuracy was evaluated by calculating the percent recovery by the assay of known, spiked amounts of analyte to an ESBO-free pesto matrix: Nine determinations were performed over two concentration levels, 30 and 100 mg kg^{-1}. **Table 5** summarizes the good results, expressed as percent recovery and relative standard deviation (RSD%).

Precision. Precision was calculated in terms of intraday repeatability as RSD% at three concentration levels; six determinations of 15, 30, and 100 mg kg^{-1} concentrations (obtained by spiking an ESBO-free pesto matrix) were assessed over a short period of time by the same analyst with the same equipment (*16*). **Table 6** reported results of the determinations with the final RSD%, which confirm an acceptable repeatability of the method.

LOD and LOQ. The detection limit (*yd*) and quantitation limit (*yq*) were preliminarily calculated as signals based on the mean blank (*ybm*) and the standard deviation (*sb*) of the blank signals as follows:

$$yd = ybm + 2Tsb \quad yq = ybm + 10Tsb$$

where T is a constant of the *t*-Student's distribution (one-sided) depending on the confidence level and the degrees of freedom ($\upsilon = n - 1$, n = number of measurements). A total of 10 blanks measurements were performed to calculate *ybm* and *sb*. An

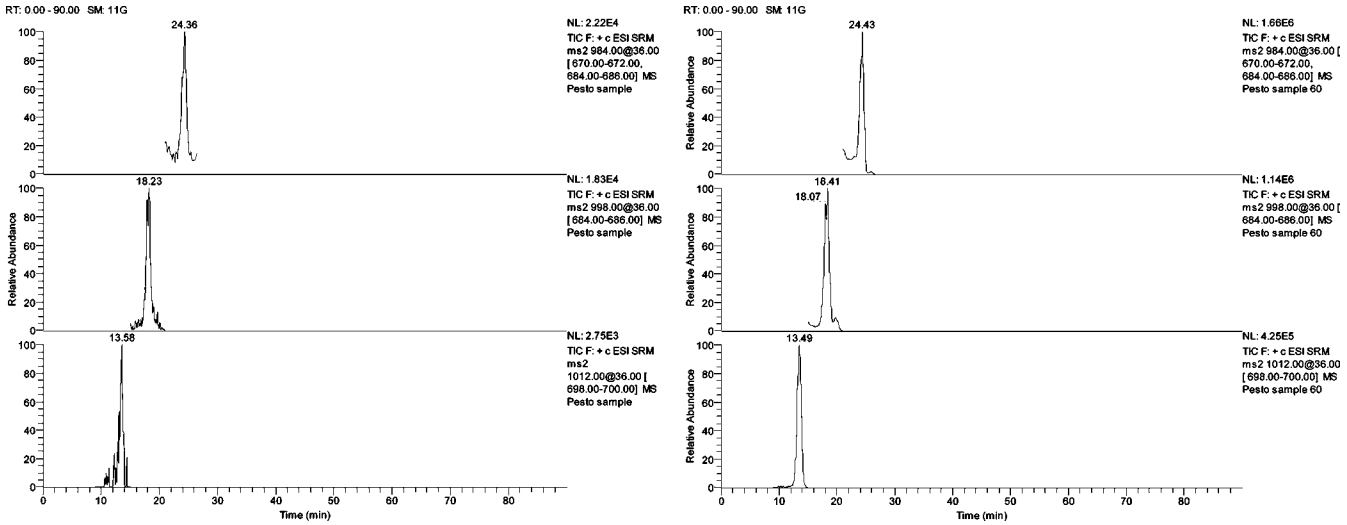

Figure 4. Example of three ESBO mixture component signals detected in SRM mode and related to a pesto sauce extract with a very low ESBO content (few ppm) (left) and the same extract spiked with ESBO standard solution up to reach a final concentration of 60 mg kg^{-1} (right).

Table 4. Matrix-Matched Calibration Curves Established in Pesto Sauce Extracts Using the LC-ESI-MS/MS Method (Concentration Range, 10–100 mg kg^{-1})

ESBO component	Mandel's test[a] (F value)	$b_1 \pm s_{b1}$[b]
M_I	0.993	1.862 (±0.032)
M_{II}	0.996	5.359 (±0.029)
M_{III}	0.995	4.629 (±0.024)
M_{IV}	0.997	1.882 (±0.027)
M_V	0.994	2.797 (±0.016)

[a] Calibration function: $y = b_1x$. [b] Confidence level at 95%.

Table 5. Mean Recoveries and Relative Standard Deviation for ESBO Mixture Component Signals, Determined by Spiking an "ESBO Free" Pesto Sauce up to Reach Final Concentrations of 30 and 100 mg kg^{-1}

ESBO component	mean recovery (%)	RSD (%) (nine determinations)
	ESBO spiking level: 30 mg kg^{-1}	
M_I	95.8	7.9
M_{II}	91.3	6.6
M_{III}	90.8	7.1
M_{IV}	91.5	5.2
M_V	92.4	7.6
	ESBO spiking level: 100 mg kg^{-1}	
M_I	101.2	7
M_{II}	94.2	2.1
M_{III}	92.5	6.5
M_{IV}	97.8	2.6
M_V	93.4	6.7

extract of ESBO-free pesto sauce was used as the blank to determine matrix-matched LOD and LOQ. *yd* and *yq* were converted from the signal domain to the concentration domain to estimate the LOD and LOQ. In this way, under the optimized LC-MS/MS conditions and operating in SRM mode, LOD and LOQ of ESBO in pesto sauce matrices were at levels of 4 and 10 mg kg^{-1}, respectively.

Data Obtained on Real Samples. Working with the aim to demonstrate the effective application of this new method on real samples, 70 food sauces packed in glass jars (pesto and different kinds of tomato-based ones) were analyzed for their

Table 6. Repeatability of the LC-MS/MS Method Calculated on Pesto Sauce Matrix

ESBO component	RSD (%) at 15 mg kg^{-1}	at 30 mg kg^{-1}	at 100 mg kg^{-1}	overall
M_I	8.1	7.9	7.7	7.9
M_{II}	6.9	5.2	2.3	4.8
M_{III}	7.2	6.7	5.1	6.3
M_{IV}	5.3	4.5	3.4	4.4
M_V	6.2	5.8	4.4	5.5

Table 7. ESBO Concentration Data in Different Food Sauce Products (from Italian Retail Markets) Obtained by the Traditional GC-MS Procedure and Compared with Correspondent Measurements Using the Present LC-ESI-MS/MS Method

food sauce samples	ESBO concn (mg kg^{-1}) LC-MS	GC-MS
pesto sauce 1	45	58
pesto sauce 2	14	13
pesto sauce 4	78	64
pesto sauce 5	90	83
tomato–cheese sauce 1	49	67
tomato–cheese sauce 2	65	79
tomato–basil sauce 1	13	19
tomato–cheese–pepper sauce 1	20	26
tomato–meat sauce 1	16	10

ESBO contents. To give an example, results by the GC-MS procedure and the LC-ion trap-ESI-MS/MS method, obtained on some samples with a different matrix, covering a range between 10 and 100 mg kg^{-1}, are reported in **Table 7**.

In the complete survey, a significant variation in ESBO content was found, probably to be considered as a result of variable processing technologies, time/temperature contacts, storage/transport conditions, and obviously the nature and fat content/dispersion of the food matrix involved. In particular, analyzed sauces were in the order of pesto-based sauces (with mean values in general comprised between 50 and 80 mg kg^{-1}) ≫ tomato−cheese-based sauces (∼40−60 mg kg^{-1}) > tomato−basil-based sauces (∼15−30 mg kg^{-1}) ≫ tomato−chili/meat-based sauces (∼10−20 mg kg^{-1}).

In conclusion, there is a common interest between plastics producers, end users, and regulatory authorities in being able

to determine the levels of plasticizers that migrated into finished products and the mechanism of migration, to respect legislative requirements such as EEC positive lists of permitted additives. In this particular field, there is an actual growing interest in the control of ESBO, which is largely adopted as a plasticizer in PVC gaskets of glass jar caps.

A novel method for the detection of ESBO in complex food matrices by RP LC-ESI-ion trap-MS/MS was developed and validated for the first time. The application on real samples and a comparison between this method and the GC-MS one were made in a variety of food sauces (belonging to various categories different for fat or edible oil contents and states of dispersion), also confirming that the migration of ESBO can often exceed the permitted overall migration limit of 60 mg kg^{-1}.

The new method demonstrates to be simple and comparably accurate, presenting an attractive alternative to the traditional GC-MS procedure: LC-MS/MS reduces the possibility of interference from other compounds present in the matrices; furthermore, another advantage is a reduction of time and costs for what concerns samples and calibration standards preparation steps. A future development, at the moment ongoing, is the possibility to adopt an ^{18}O-labeled-epoxidized triacylglycerol as an internal standard, appropriately synthesized by a controlled isotope labeling reaction.

LITERATURE CITED

(1) Simoneau, C. *Exposure Assessment of Epoxidised Soybean Oil in Baby Foods for Infants: Review of Data and Assessment*; European Commission, Joint Research Centre, Institute for Health and Consumer Protection: Ispra, Italy, 2004; Eur 21372 EN, 41 pp.

(2) Opinion of the Scientific Panel on Food Additives, Flavourings, Processing Aid and Materials in Contact with Food (AFC) on a request from the Commission related to the use of Epoxidised soybean oil in food contact materials" adopted on 26 May 2004. *EFSA J.* **2004**, *64*, 1−17. Available from the following web address: http://www.efsa.eu.int/science/afc/afc_opinions/467_en.html.

(3) MAFF. Food and Nutrient Intakes of British Infants Aged 6−12 Months; HMSO: London, United Kingdom, 1992.

(4) The British Industrial Biological Research Association (BIBRA). *Toxicity Profile on Epoxidised Soybean Oil*; BIBRA: Carlshalton, Surrey, United Kingdom, 1988.

(5) European Commission Scientific Committee for Food. *Compilation of the Evaluations of the Scientific Committee for Food on Certain Monomers and Additives Used in the Manufacturer of Plastic Materials Intended To Come into Contact with Foodstuffs until 21 March 1997*; Reports of the Scientific Committee for Food, 42nd series; European Commission: Luxembourg, SCF, 1999.

(6) European Union Commission Directive EU 90/128/EEC, currently repealed by EU Directive 2002/72/EC.

(7) Council of Europe (CoE) Resolution ResAP96/5, currently repealed by Framework Resolution ResAP(2004)1.

(8) Biedermann-Brem, S.; Biedermann, M.; Grob, K. Reaction of epoxidized soya bean oil (ESBO) with hydrogen chloride formed in PVC: investigation on model systems. *Mitt. Lebensmitteluntersuch. Hyg.* **2004**, *95*, 261−280.

(9) Piccinini, P.; Valzacchi, S.; Ezerskis, Z.; Simoneau, C. In *Level of Chlorohydrins from Epoxidised Soybean Oil in Various Baby Foods*, Proceedings of ILSI 3rd International Symposium on Food Packaging—Ensuring the Safety, Quality and Traceability of Foods, Barcelona, Spain, 17−19 November 2004.

(10) Hammarling, L.; Gustavsson, H.; Svensson, K.; Karsson, S.; Oskarsson, A. Migration of epoxidized soya bean oil from plasticized PVC gaskets into baby food. *Food Addit. Contam.* **1998**, *15*, 203−208.

(11) Fantoni, L.; Simoneau, C. European survey of contamination of homogenized baby food by epoxidized soybean oil migration from plasticized PVC gaskets. *Food Addit. Contam.* **2003**, *20*, 1087−1096.

(12) Castle, L.; Gilbert, J. Gas chromatographic-mass spectrometric determination of epoxidized soybean oil contamination of foods by migration from plastic packaging *J. Assoc. Off. Anal. Chem.* **1988**, *71*, 1183−1186.

(13) Castle, L.; Mayo, A.; Gilbert, J. Migration of epoxidised soya bean oil into foods from retail packaging materials and from plasticised PVC film used in the home. *Food Addit. Contam.* **1990**, *7*, 29−36.

(14) Castle, L.; Sharman, M.; Gilbert, J. Determination of high- and low- molecular mass plasticizers in strtch-type packaging films. *J. Chromatogr.* **1988a**, *437*, 274−280.

(15) Fankhauser-Noti, A.; Fiselier, K.; Biedermann-Brem, S.; Grob, K. Epoxidized soy bean oil migrating from the gaskets of lids into food packed in glass jars analysis by on-line liquid chromatography-gaschromatography. *J. Chromatogr. A* **2005**, 1082 (2), 214−219.

(16) ICH-Guidelines. Validation of Analytical Procedures: Methodology CPMP/ICH/281/95, Final (ICH Topic Q2B, Step 4).

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