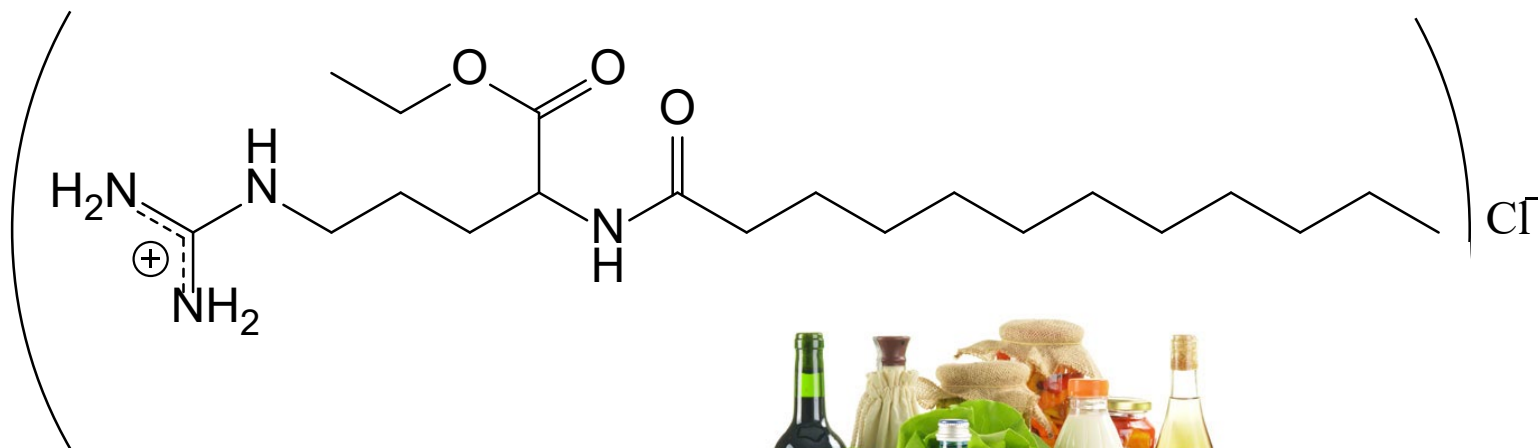


Determination of Lauric Arginate Ethyl Ester in Food



Dr. Boris Yiu-Tung WONG
Chemist, Additives, Contaminants and Composition Section
Government Laboratory, Hong Kong
05 Nov 2024

Lauric Arginate Ethyl Ester (1)

What is Lauric Arginate Ethyl Ester (LAE)月桂酰精氨酸乙酯?

- An amino acid based cationic surfactant
- Derived from arginine, ethanol and lauroyl chloride.
- Exceptional antimicrobial efficacy
- Low toxicity
- Hydrolyzed quickly to lauric acid and L-arginine in vivo.



Lauric Arginate Ethyl Ester (2)

Codex General Standard for Food Additives (GSFA) Online Database

The "Codex General Standard for Food Additives" (GSFA, Codex STAN 192-1995) sets forth the conditions under which permitted food additives may be used in all foods, whether or not they have previously been standardized by Codex. The Preamble of the GSFA contains additional information for interpreting the data. Users are encouraged to consult the Preamble when using this database.

CODEX ALIMENTARIUS
INTERNATIONAL FOOD STANDARDS



Food and Agriculture
Organization of
the United Nations



World Health
Organization

E-mail: codex@fao.org - www.codexalimentarius.org

GENERAL STANDARD FOR FOOD ADDITIVES

CXS 192-1995

Adopted in 1995. Revision 1997, 1999, 2001, 2003, 2004, 2005, 2006, 2007, 2008, 2009, 2010, 2011, 2012, 2013, 2014, 2015, 2016, 2017, 2018, 2019, 2021, 2023



Lauric Arginate Ethyl Ester (3)

Page 174 – 175 of the latest version of "Codex General Standard for Food Additives" (GSFA, Codex STAN 192-1995)

CXS 192-1995 Table
One

174

FoodCatNo	FoodCategory	MaxLevel	Notes	Year Adopted
12.2.2	Seasonings and condiments	GMP	477	2023

LAURIC ARGINATE ETHYL ESTER

INS 243 Lauric arginate ethyl ester Functional Class: Preservative

FoodCatNo	FoodCategory	MaxLevel	Notes	Year Adopted
01.6.1	Unripened cheese	200 mg/kg	XS221, XS262, XS273, XS275	2023
01.6.2.1	Ripened cheese, includes rind	200 mg/kg	XS274, XS276, XS277	2023
01.6.3	Whey cheese	200 mg/kg		2011
01.6.4	Processed cheese	200 mg/kg		2011
01.6.5	Cheese analogues	200 mg/kg		2011
01.7	Dairy-based desserts (e.g. pudding, fruit or flavoured yoghurt)	200 mg/kg	170	2011
02.2.2	Fat spreads, dairy fat spreads and blended spreads	200 mg/kg	XS253 & XS256	2023
04.1.2.2	Dried fruit	200 mg/kg		2011
04.1.2.11	Fruit fillings for pastries	200 mg/kg		2011
04.2.1.2	Surface-treated fresh vegetables (including mushrooms and fungi, roots and tubers, pulses and legumes, and aloe vera), seaweeds, and nuts and seeds	200 mg/kg		2011
04.2.1.3	Peeled, cut or shredded fresh vegetables (including mushrooms and fungi, roots and tubers, pulses and legumes, and aloe vera), seaweeds, and nuts and seeds	200 mg/kg		2011
04.2.2.3	Vegetables (including mushrooms and fungi, roots and tubers, pulses and legumes, and aloe vera), and seaweeds in vinegar, oil, brine, or soybean sauce	200 mg/kg		2011
05.1.3	Cocoa-based spreads, including fillings	200 mg/kg	XS86	2016
05.3	Chewing gum	225 mg/kg		2011
06.5	Cereal and starch based desserts (e.g. rice pudding, tapioca pudding)	200 mg/kg		2011
08.2.1	Non-heat treated processed meat, poultry, and game products in whole pieces or cuts	200 mg/kg		2016
08.2.2	Heat-treated processed meat, poultry, and game products in whole pieces or cuts	200 mg/kg	396	2019
08.2.3	Frozen processed meat, poultry, and game products in whole pieces or cuts	200 mg/kg	3 & 374	2016
08.3.1	Non-heat treated processed comminuted meat, poultry, and game products	315 mg/kg		2016
08.3.2	Heat-treated processed comminuted meat, poultry, and game products	200 mg/kg	377	2019
08.3.3	Frozen processed comminuted meat, poultry, and game products	315 mg/kg	3 & 374	2016
09.2.4.1	Cooked fish and fish products	200 mg/kg		2018
09.2.4.2	Cooked mollusks, crustaceans, and echinoderms	200 mg/kg		2018

CXS 192-1995 Table
One

175

FoodCatNo	FoodCategory	MaxLevel	Notes	Year Adopted
09.2.4.3	Fried fish and fish products, including mollusks, crustaceans, and echinoderms	200 mg/kg	419	2018
09.2.5	Smoked, dried, fermented, and/or salted fish and fish products, including mollusks, crustaceans, and echinoderms	200 mg/kg	333, XS189, XS222 & XS236	2021
09.3.1	Fish and fish products, including mollusks, crustaceans, and echinoderms, marinated and/or in jelly	200 mg/kg		2018
09.3.2	Fish and fish products, including mollusks, crustaceans, and echinoderms, pickled and/or in brine	200 mg/kg		2018
09.3.3	Salmon substitutes, caviar, and other fish roe products	200 mg/kg	XS291	2018
09.3.4	Semi-preserved fish and fish products, including mollusks, crustaceans, and echinoderms (e.g. fish paste), excluding products of food categories 09.3.1 - 09.3.3	200 mg/kg		2018
10.2	Egg products	200 mg/kg		2011
10.4	Egg-based desserts (e.g. custard)	200 mg/kg		2011
12.2.2	Seasonings and condiments	200 mg/kg		2011
12.5	Soups and broths	200 mg/kg	XS117	2015
12.6.1	Emulsified sauces and dips (e.g. mayonnaise, salad dressing, onion dip)	200 mg/kg		2011
12.6.2	Non-emulsified sauces (e.g. ketchup, cheese sauce, cream sauce, brown gravy)	200 mg/kg	XS306	2023
12.7	Salads (e.g. macaroni salad, potato salad) and sandwich spreads excluding cocoa- and nut-based spreads of food categories 04.2.2.5 and 05.1.3	200 mg/kg		2011
14.1.4.1	Carbonated water-based flavoured drinks	50 mg/kg		2011
14.1.4.2	Non-carbonated water-based flavoured drinks, including punches and ades	50 mg/kg		2011
14.1.4.3	Concentrates (liquid or solid) for water-based flavoured drinks	50 mg/kg	127	2011

LECITHIN

INS 322(i) Lecithin Functional Class: Antioxidant, Emulsifier, Flour treatment agent

FoodCatNo	FoodCategory	MaxLevel	Notes	Year Adopted
01.1.2	Other fluid milk (plain)	GMP	410	2018
01.2.2	Rennet milk (plain)	GMP		2013
01.4.1	Pasteurized cream (plain)	GMP	236	2013
01.4.2	Sterilized and UHT creams, whipping and whipped creams, and reduced fat creams (plain)	GMP		2013
01.8.2	Dried whey and whey products, excluding whey cheeses	GMP	XS331	2023
02.1.1	Butter oil, anhydrous milkfat, ghee	GMP	507	2021
02.1.2	Vegetable oils and fats	GMP	511, 519, XS33 & XS325R	2023
02.1.3	Lard, tallow, fish oil, and other animal fats	GMP		2018
04.1.1.2	Surface-treated fresh fruit	GMP	454	2021

* 3 different MPLs in selected food: 50, 200 & 315 mg/kg



Lauric Arginate Ethyl Ester (4)

SYNONYMS

Lauric arginate ethyl ester; lauramide arginine ethyl ester; ethyl-N^α-lauroyl-L-arginate·HCl; LAE; INS No. 243

DEFINITION

Ethyl lauroyl arginate is synthesized by esterifying arginine with ethanol, followed by reacting the ester with lauroyl chloride. The resultant ethyl lauroyl arginate is recovered as the hydrochloride salt, which is filtered and dried.

Chemical name

Ethyl-N^α-dodecanoyl-L-arginate·HCl

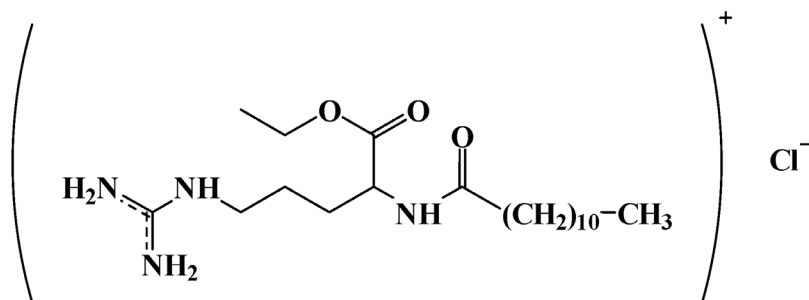
C.A.S. number

60372-77-2

Chemical formula

C₂₀H₄₁N₄O₃Cl

Structural formula



Formula weight

421.02

Source: Joint FAO/WHO Expert Committee on Food Additives (JECFA)



Background Information (1)

Amendment of Cap. 132 BD Preservatives in Food Regulation

- To update the definitions of “preservative” and “antioxidant”;
- To update the list of permitted preservatives / antioxidants; and
- To update / stipulate the maximum permitted levels (MPLs) of the permitted preservatives and antioxidants in specified food categories.



Background Information (2)

Consultation Document

Proposed Amendments to the Preservatives in Food Regulation (Cap.132BD)

May 2023

2.6 We also propose to add another 4 food additives, which are currently not permitted for use under the existing Cap. 132BD. Among them, benzoyl peroxide, lauric arginate ethyl ester and stearyl citrate are listed in the Codex GSFA. Rosemary extract has been evaluated by JECFA to ensure its safety and it is currently permitted for use as an antioxidant in the Mainland, Australia, New Zealand, the EU and Singapore.



環境及生態局
Environment and Ecology Bureau



食物環境衛生署
Food and Environmental
Hygiene Department



食物安全中心
Centre for Food Safety

Background Information (3)

The Government of the Hong Kong Special Administrative Region
Press Releases



| **Font Size:** | **Sitemap**

Preservatives in Food (Amendment) Regulation 2024 ▾ GO

Preservatives in Food (Amendment) Regulation 2024 gazetted

The Government gazetted today (October 10) the Preservatives in Food (Amendment) Regulation 2024 (Amendment Regulation) to update the food safety standards in relation to preservatives and antioxidants in food.

The objective of the Amendment Regulation is to keep local food safety standards on par with international standards, thereby enhancing consumer protection and facilitating the food trade. The Amendment Regulation has kept the standards of the Codex Alimentarius Commission (Codex) as the backbone, supplemented with those adopted by the Mainland and by Hong Kong's other major food-trading partners.

A Government spokesman said, "A four-month public consultation was conducted from May 29 to September 30 last year, to collect views on the proposed amendments to the Preservatives in Food Regulation (Cap. 132BD). Respondents and stakeholders generally welcomed and supported the proposed amendments. Taking into account the comments received during the public consultation, we have formulated the updated proposals, as reflected in the Amendment Regulation."

The Amendment Regulation updates the definitions of "preservative" and "antioxidant" with reference to the corresponding definitions adopted by Codex. The list of permitted preservatives/antioxidants has also been updated, which comprises 58 preservatives/antioxidants after the legislative amendments. The number of "additive-food" pairs with specified maximum permitted levels has increased from around 900 to around 2 000.

"We consider that the Amendment Regulation should be implemented as soon as possible. However, we acknowledge the importance of providing sufficient time for the stakeholders to adjust to the new food safety standards, and for the local testing laboratories to establish the capability for performing relevant tests. We have also taken into consideration the longer shelf life of processed and pre-packaged foods that may contain preservatives or antioxidants. On balance, we propose a transitional period of 24 months, during which it would be legal for any single food item to comply wholly with the requirements of either the existing or the amended Cap. 132BD," the spokesman added.

The Amendment Regulation will be tabled at the Legislative Council on October 16 for negative vetting.

Ends/Thursday, October 10, 2024
Issued at HKT 10:00

NNNN

[Archives](#) [Yesterday's Press Releases](#)

[Back to Index Page](#) [Back to top](#)

[Today's Press Releases](#)



Analytical methods for detection of LAE (1)

- LAE in packaging materials
- Migration of LAE into simulants and chicken muscles
- For chicken muscles, extraction with ACN
- Non-targeted screening by UPLC-QTOF (including LAE)



Contents lists available at [SciVerse ScienceDirect](#)

Food and Chemical Toxicology

journal homepage: www.elsevier.com/locate/foodchemtox



Migrants determination and bioaccessibility study of ethyl lauroyl arginate (LAE) from a LAE based antimicrobial food packaging material

M. Aznar^a, J. Gómez-Estaca^b, D. Vélez^b, V. Devesa^b, C. Nerín^{a,*}

^a Analytical Chemistry Department, GUIA Group, I3A, EINA, University of Zaragoza, M^o de Luna 3, 50018 Zaragoza, Spain

^b Institute of Agrochemistry and Food Technology (IATA-CSIC), Avda. Agustín Escardino 7, 46980 Paterna, Valencia, Spain

Ref.: M. Aznar et. al. *Food and Chemical Toxicology*, 2013, **56**, 363 – 370.



Analytical methods for detection of LAE (2)

- Ultrasonic extraction with 9:1 ACN/Water
- Extraction: 1 x 10mL then 2 x 5mL
- Filter through 0.22 μm PTFE filter.
- LC-MS/MS analysis

第8卷 第4期
2017年4月

食品安全质量检测学报
Journal of Food Safety and Quality

Vol. 8 No. 4
Apr., 2017

液相色谱-串联质谱法测定肉鱼禽产品中的防腐剂 月桂酰精氨酸乙酯盐酸盐

赵艳菊*, 杨晋青, 聂磊, 杨保刚, 周耀斌

(上海市质量监督检验技术研究院, 上海 200233)

摘要: **目的** 建立液相色谱串联质谱法测定肉鱼禽产品中的防腐剂月桂酰精氨酸乙酯盐酸盐(ethyl lauroyl arginine hydrochloride, LAE)的分析方法。**方法** 通过超声波溶剂提取法提取样品中的 LAE, 通过色谱柱筛选确定 Waters ACQUITY UPLC®BEH C₁₈(2.1 mm × 100 mm, 1.7 μm)色谱柱, 流动相为 0.1%甲酸水溶液和乙腈, 梯度洗脱, 然后经质谱法检测, 以外标法定量。**结果** 最佳提取条件为: 提取溶剂为水:乙腈=1:9(V:V), 提取时间为 5 min, 提取次数为 3 次。LAE 的线性范围为 0~20 $\mu\text{g/L}$, 线性相关系数为 0.9990。方法的检出限为 0.3 $\mu\text{g/kg}$, 方法的定量限为 1.0 $\mu\text{g/kg}$; 通过加标回收率实验, 确定方法的加标回收率范围为 82.5%~105.0%。符合 GB27404-2008 标准规定。**结论** 本方法简单、便捷, 适用于肉鱼禽产品中 LAE 含量的测定。

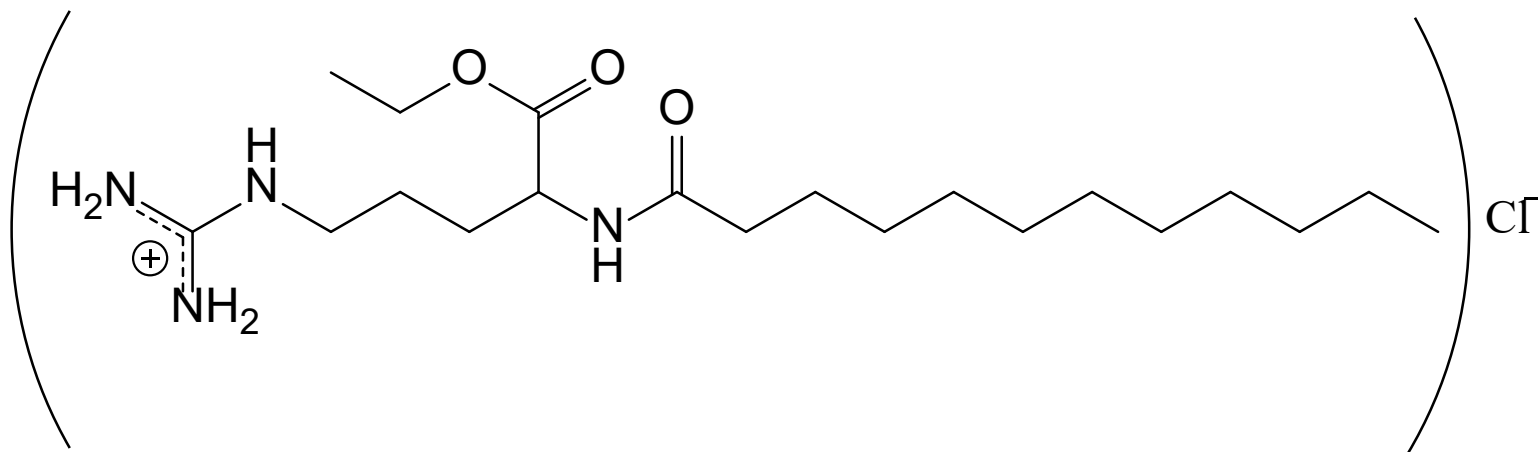
关键词: 月桂酰精氨酸乙酯盐酸盐; 液相色谱串联质谱法; 溶剂提取

Ref.: Y.J. Zhao et al. Journal of Food Safety and Quality, 2017, 8, 1455-1459 .

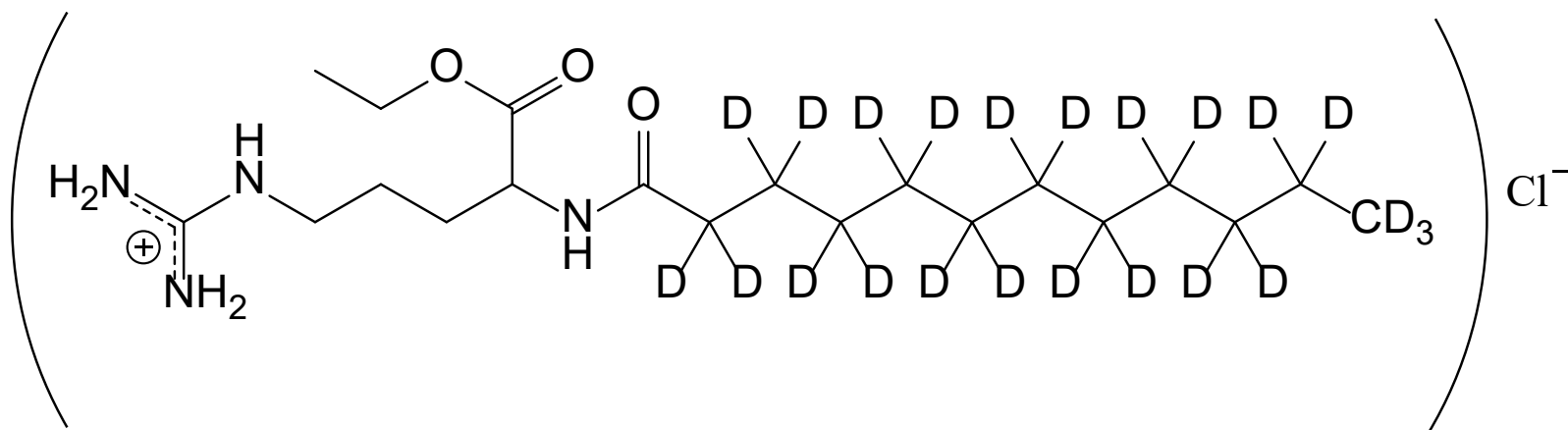


Our proposed LC-MS/MS method (for LAE in food) (1)

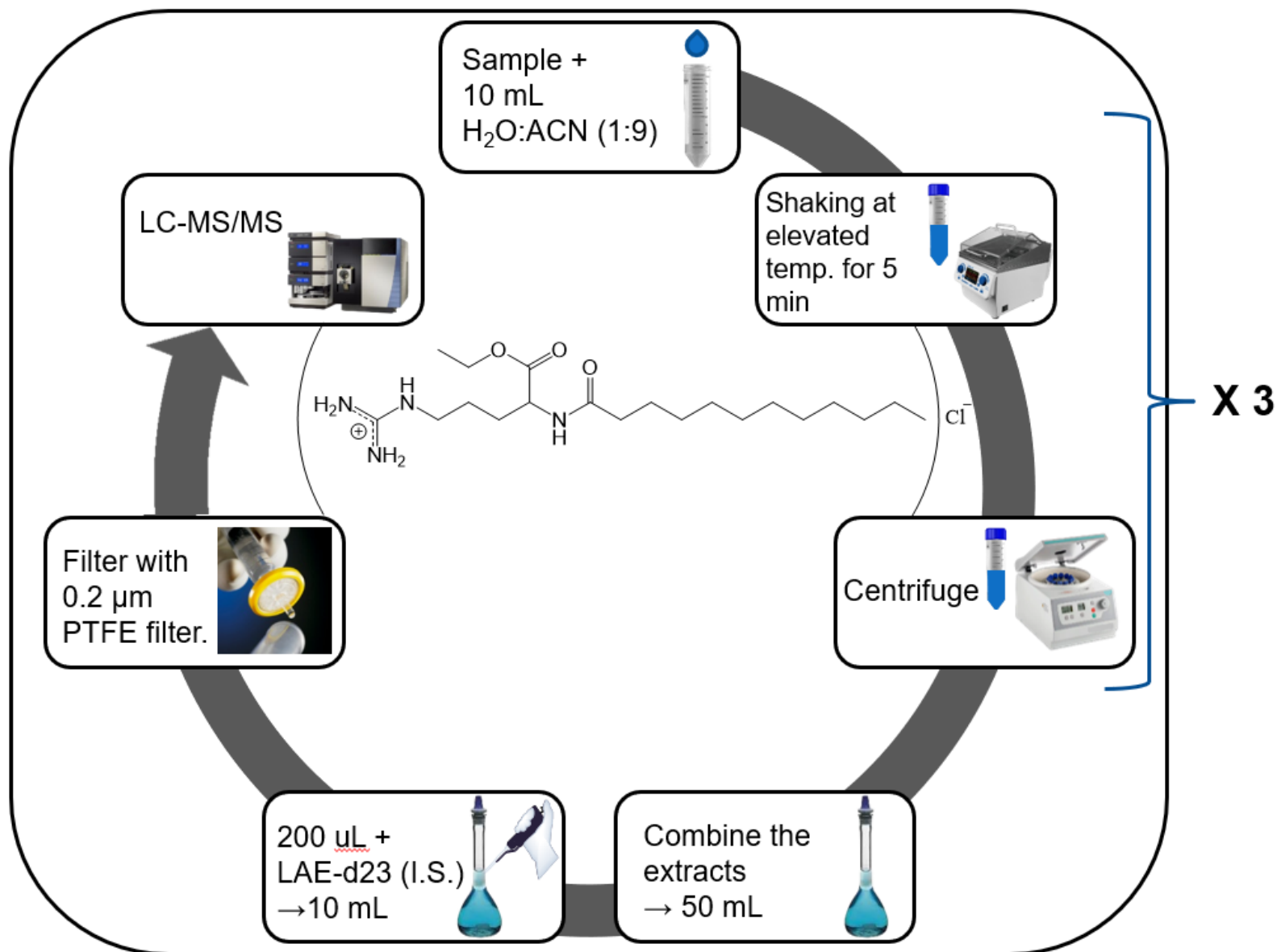
Measurand: LAE



Measurand: LAE-d₂₃



Schematic of our proposed LC-MS/MS method (for LAE in food)



Ref.: Manuscript on “Development of an LC-MS/MS method for the detection of LAE in food” is under preparation.



Our proposed LC-MS/MS method (for LAE in food) (2)

LC conditions:

Column:	Acquity UPLC BEH C18 (2.1 mm x 100 mm, 1.7 μ m) or equivalent
Mobile phase:	Solvent A: 0.1% formic acid in water
	Solvent B: ACN
Flow rate:	0.3 mL/min
Injection volume:	5 μ L

MS/MS conditions:

Compound	Precursor Ion (m/z)	Product Ion (m/z)
LAE	385	186
	385	326
LAE-d23	408	187
	408	391

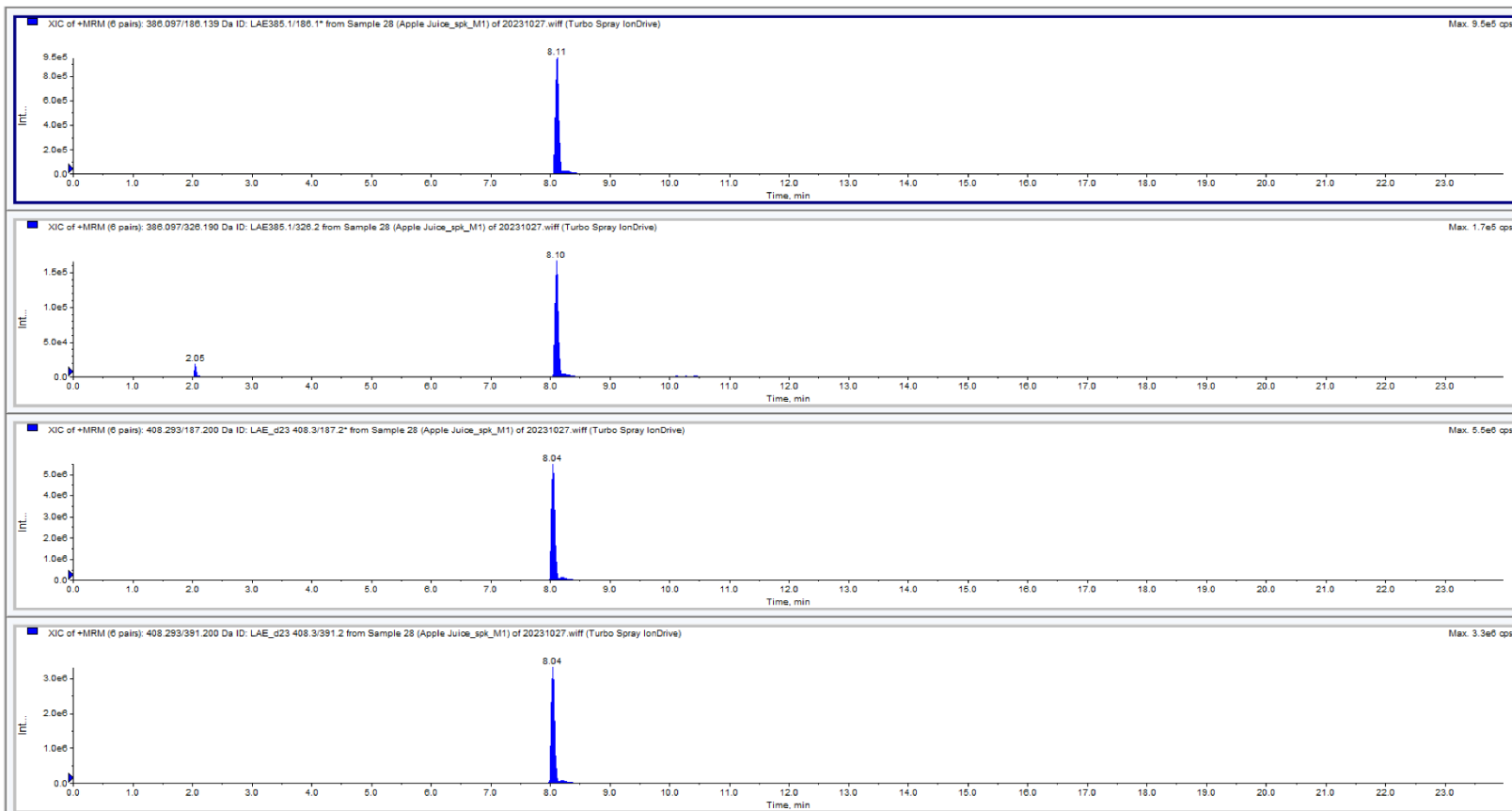
Ref.: Manuscript on “Development of an LC-MS/MS method for the detection of LAE in food” is under preparation.



Representative LC-MS/MS Chromatograms (1)

~ 50 mg/kg

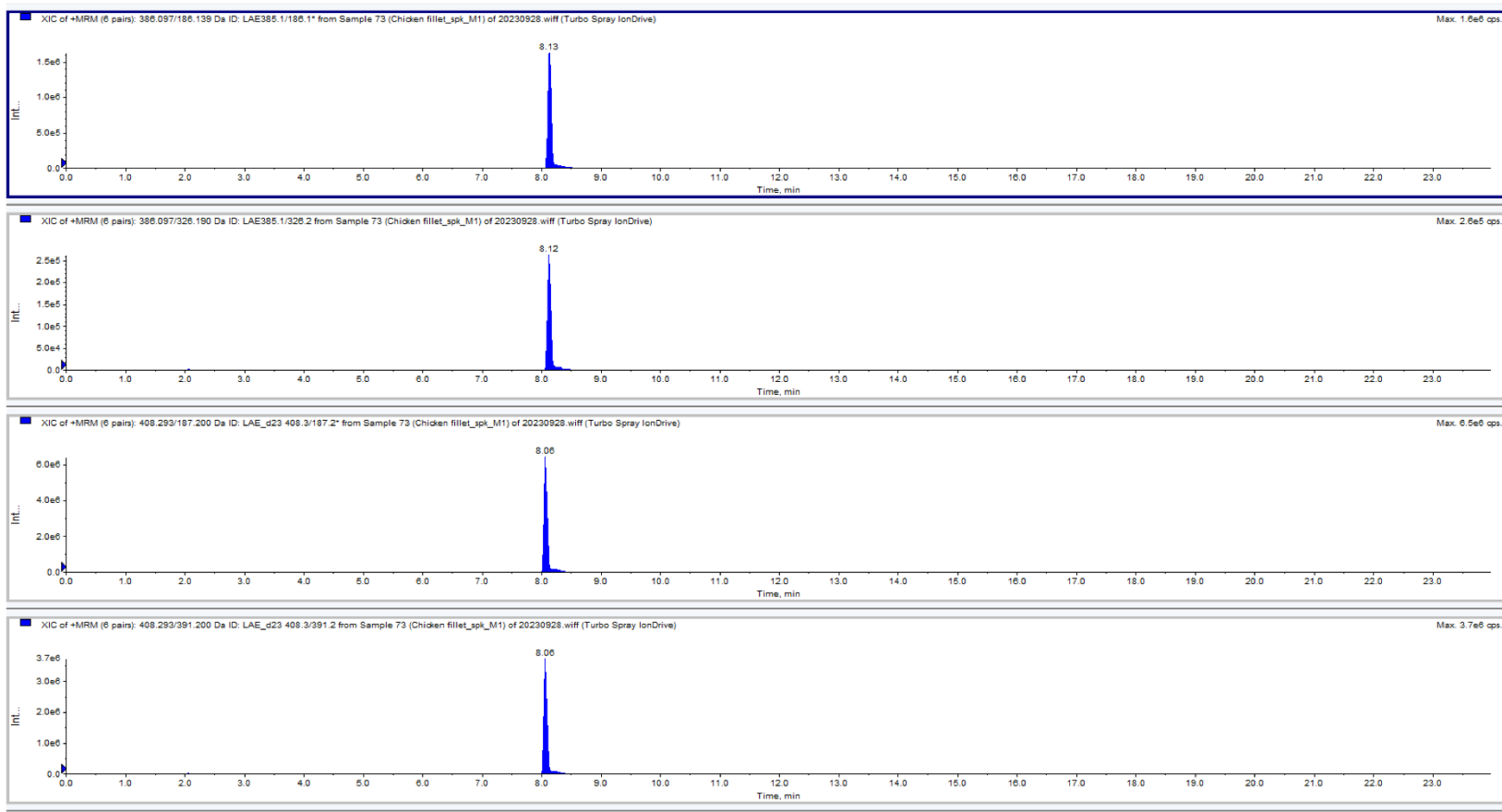
Liquid Sample (Apple Juice)



Representative LC-MS/MS Chromatograms (2)

~ 300 mg/kg

Solid Sample (Chicken Fillet)



Method Performance

Sample	Sample Type
Cereal	Carbohydrate-rich Food
Clam chowder soup	Carbohydrate-rich Food
Tomato sauce	Carbohydrate-rich Food
Dried fruit	Dietary fibre-rich food
Mixed salad	Dietary fibre-rich food
Red amaranth (紅莧菜)	Dietary fibre-rich food
Cheese	Fat/Oil-rich Food
Frozen comminuted meat (Sausage)	Fat/Oil-rich Food
Frozen salmon	Fat/Oil-rich Food
Apple juice	Food composed mainly of water
Coconut water	Food composed mainly of water
Coke	Food composed mainly of water
Ginger Ale soda	Food composed mainly of water
Chewing gum	Others
Hazelnut chocolate spread	Others
Italian mixed herbs	Others
Beef	Protein-rich Food
Chicken fillet	Protein-rich Food
Cured meat	Protein-rich Food
Dried meat	Protein-rich Food
Egg	Protein-rich Food
Pork	Protein-rich Food

Solid samples

Liquid samples

Solid samples



Recovery and Precision data (1)

Summary

Sample type	Spike level (mg/kg)	Average Recovery (%)	RSD(%)
Liquid samples	5	96	2.0
Liquid samples	50	99	3.9
Liquid samples	75	100	2.7
Solid samples	20	95	5.3
Solid samples	30	97	6.4
Solid samples	200	99	4.0
Solid samples	300	100	4.0
Solid samples	475	101	3.7



Recovery and Precision data (2)

Summary

- The spike levels are corresponding to:
- ~ 0.1, 1 and 1.5 times the Maximum Permitted Level

MPL (mg/kg)	Spike level 1	Spike level 2	Spike level 3
50	5	50	75
200	20	200	300
315	30	300	475

- Spike recoveries are within 80% - 120%
- RSDs are $\leq 15\%$



Identification Criteria

L 180/84

EN

Official Journal of the European Union

21.5.2021

COMMISSION IMPLEMENTING REGULATION (EU) 2021/808

of 22 March 2021

on the performance of analytical methods for residues of pharmacologically active substances used in food-producing animals and on the interpretation of results as well as on the methods to be used for sampling and repealing Decisions 2002/657/EC and 98/179/EC

(Text with EEA relevance)

Relative Retention Time (RRT)

➤ $\pm 1\%$

Relative Intensities

➤ $\pm 40\%$



Matrix effects (1)

LC-MS/MS analysis often suffers from matrix effects (MEs), especially when analysis is performed with **electrospray ionization source (ESI)**. MEs are caused by the alternation of ionization efficiency of target analytes in the presence of co-eluting compounds in the sample matrix.

$$\text{ME (\%)} = \left[\left(\frac{m_{\text{matrix}}}{m_{\text{Solvent}}} \right) - 1 \right] \times 100$$

Slope of Cal. Curve
in sample matrix

Slope of Cal. Curve
in solvent



Matrix effects (2)

Implication

- $\frac{m_{matrix}}{m_{solvent}} > 1 \rightarrow$ Matrix enhancement
- $\frac{m_{matrix}}{m_{solvent}} < 1 \rightarrow$ Matrix suppression
- If ME% $\geq \pm 20$, need to be addressed in calibration (SANTE 11312/2021)

**ANALYTICAL QUALITY CONTROL AND
METHOD VALIDATION PROCEDURES FOR
PESTICIDE RESIDUES ANALYSIS
IN FOOD AND FEED
SANTE 11312/2021**

Supersedes Document No. SANTE/2019/12682. Implemented by 01/01/2022



Matrix effects (3)

		MEs (%)	
Sample	Matrix type	Low Cal. Range	High Cal. Range
Apple	Food composed mainly of water	-4.62	2.73
Meat Floss	Protein-rich Food	-3.04	7.21
Cereal	Carbohydrate-rich Food	1.61	4.14
Cheese	Fat/Oil-rich Food	0.92	0.10
Red Amaranth	Dietary fibre-rich food	-4.77	-3.02

ME% in all matrices $\leq \pm 20\%$



Thank you!

