ETHYL LAUROYL ARGINATE
Chemical and Technical Assessment

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1. Summary

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 hydrochloride salt and is a white, solid product which is filtered off and dried. The active ingredient is the hydrochloride salt of ethyl-N^{α}-lauroyl-L-arginate (ethyl-N^{α}-dodecanoyl-L-arginate·HCl, CAS number 60372-77-2) and its molecular weight is 421.02. The content of ethyl-N^{α}-lauroyl-L-arginate is between 85-95%. Ethyl lauroyl arginate is a white powder and its solubility in water at 20ºC is greater than 247 g/kg. It is stable for more than 2 years at room temperature when protected in a closed container. This substance is intended to be used as preservative for its anti-microbial activity. The synonyms are lauric arginate ethyl ester, lauramide arginine ethyl ester, LAE and INS No. 243.

Ethyl lauroyl arginate had not been evaluated by the Committee. At 39th CCFAC (2007) it was proposed to be evaluated for safety by the Committee.

2. Description

Ethyl lauroyl arginate is composed of lauric acid, L-arginine HCl and ethanol. It is synthesised by the Spanish company Laboratorios Miret, S.A. (LAMIRSA).

The active ingredient of ethyl lauroyl arginate is the hydrochloride salt of an N-fatty acyl-substituted amino acid ethyl ester, ethyl-N^{α}-lauroyl-L-arginate HCl. Its chemical name is ethyl-N^{α}-dodecanoyl-L-arginate HCl (CAS number: 60372-77-2) and its molecular weight is 421.02. Ethyl lauroyl arginate contains between 85-95% of this active ingredient. Structure formula is shown in Figure 1.

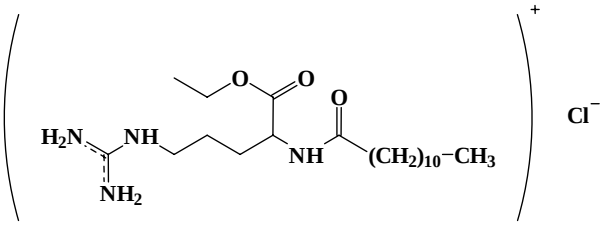

Figure 1. Ethyl-N^{α}-lauroyl-L-arginate HCl

Ethyl lauroyl arginate is a white powder and its solubility in water at 20ºC is greater than 247 g/kg. Commercial products are formulated its 20-25% solutions in appropriate food-grade solvents.

Ethyl-N^{α}-lauroyl-L-arginate HCl present in ethyl lauroyl arginate is stable for more than 2 years at room temperature when protected in a closed container. The aqueous stability of ethyl lauroyl arginate has been evaluated under acid conditions and at varying temperatures. The acids employed to evaluate the stability were phosphoric, citric, tartaric, malic and fumaric acids and the temperatures were 4, 25 and 50ºC. The results indicate that the stability of ethyl lauroyl arginate decreases with increasing temperature and reducing pH. In general, the strong inorganic acids affected stability more than the organic acids studied.

Ethyl lauroyl arginate has been evaluated for food safety as antimicrobial in food by the U.S. Food and Drug Administration (FDA) in 2005, and as a food preservative by the European Food Safety Authority (EFSA) in 2007.

The synonyms are lauric arginate ethyl ester, lauramide arginine ethyl ester, LAE and INS No. 243.

3. Manufacturing

The method of manufacturing is described in European Patent No.1294678. The starting materials employed for the manufacture of ethyl lauroyl arginate are L-arginine HCl (CAS 1119-34-2), ethanol (CAS 64-17-5), thionyl chloride (CAS 7719-09-7), sodium hydroxide (CAS 1310-73-2), lauroyl chloride (CAS 112-16-3) and deionised water (CAS 7732-18-5). LAMIRSA's patented production process consists of two steps followed by a filtration process.

- The first step of production concerns the esterification of the carboxyl group of L-arginine HCl with ethanol, using thionyl chloride as an esterification agent and exploiting the heat of reaction in order to carry out this reaction. The final product obtained in this step is the ethyl arginate 2HCl.
- The second step of production involves the condensation of lauroyl chloride with the α-amino group of ethyl arginate 2HCl in an aqueous medium.
- The last step in the production process of ethyl lauroyl arginate is the filtration of the reaction mixture through a press filter to separate out the final product obtained via the reactions described above.

Once the filtration process is finished, the resulting product is a white solid with a content of water from 11.5 to 18.6% and a content of the active ingredient between 71% and 81%. This product is stored at refrigeration temperatures inside a stainless steel container and it may be dried to produce ethyl lauroyl arginate with a content of the active ingredient between 85 and 95% and a water content <5%. Ethyl lauroyl arginate is stored at refrigeration temperatures and inside a stainless steel holding tank.

The manufacturing process of ethyl lauroyl arginate has been validated, according to GMP, from the product obtained at the end of the first part of the synthesis, ethyl arginate 2HCl, to the production of the product before being dried which has a content of active ingredient 71-81%. The equipment involved in this validation process was the reactor of the second process of synthesis and the filter press.

4. Characterization

4.1 Physicochemical characterization

The physicochemical properties of ethyl lauroyl arginate are shown Table 1. They are tested by the EEC methods for the Determination of Physicochemical Properties, Directive 92/69/EEC, Part A, Methods A1-A17 and the OECD Guidelines for the Testing of Chemicals.

Table 1. Physicochemical Properties

EEC Method	OECD Method	Test	Result
A1	102	Melting temperature	50.5 to 58.0°C
A2	103	Boiling temperature	Decomposes from 107°C
A3	109	Relative density $\left(D_4^{20}\right)$	1.11
A4	104	Vapour pressure	5.45×10^{-4} Pa at 25°C
A5	115	Surface tension	25.43 mN/m for a 1 g/l aqueous solution at 19°C
A6	105	Water solubility	Greater than 247 g/Kg at 20°C
A8	107	Partition coefficient	1.43 at 20°C
A10	-	Flammability (solids)	Not highly flammable
A14	-	Explosive properties	Not explosive
A16	-	Relative self-ignition temperature for solids	Does not self ignite
A17	-	Oxidising properties	Not oxidising
-	110	Particle size distribution	0% below 10 μm
-	-	Soil adsorption coefficient	$K_{oc} = 58.0$

Ethyl lauroyl arginate is soluble up to 20% in propylene glycol, glycerin and ethanol. The pH of 1 % aqueous solution is the range of 3.64 to 4.25 on 4 batches.

4.2. Chemical characterization

The chemical characterizations of 6 batches of ethyl lauroyl arginate are shown at Table 2.

Ethyl-N^{α}-lauroyl-L-arginate HCl is the active ingredient of ethyl lauroyl arginate and its contents are 88.1-90.3%.

Possible impurities are residual materials, by-products and others. Lauric acid, L-arginine HCl and ethanol are materials and their residual amounts are 2.3-3.0%, 0.1-0.4% and <0.2%, respectively. N^{α}-Lauroyl-L-arginine, ethyl laurate and ethyl arginate 2HCl are by-products and their contents are 1.5-2.3%, 1.0-2.0%, <0.01-0.4%, respectively. Other three minor by-products (P2, P3 and P4) share the structure of ethyl-N^{α}-lauroyl-L-arginate HCl but have an additional lauroyl group. Impurities P2 and P4 should be in a ratio smaller than 1% and 0.5%, respectively. P3 is 0.064% in a batch.

Water contents are 0.9-4.1%. Ashes heated at 700° are mainly NaCl and their amounts are 0.7-1.5%.

Arsenic, cadmium, lead and mercury are not detected.

Table 2. Chemical characterizations of ethyl lauroyl arginate batches

Chemical name	Content (%)						
	3036*	5733*	7446*	8302*	8411*	10234*	Ave.±SD
Ethyl-N^{α}-lauroyl-L-arginate HCl	89.4	90.3	88.2	88.1	88.5	88.2	88.8±0.9
N^{α}-Lauroyl-L-arginine	1.5	2.1	1.9	2.3	2.2	1.6	1.9±0.3
Lauric acid	2.7	3.0	2.7	2.3	2.6	2.5	2.6±0.2
Ethyl laurate	1.5	2.0	1.0	1.5	1.1	1.4	1.4±0.4
L-Arginine HCl	0.3	0.3	0.1	0.4	0.3	0.4	0.3±0.1
Ethyl arginate 2HCl	0.3	0.4	<0.1	<0.01	<0.01	<0.1	0.2±0.1
Ethanol	<0.2	<0.2	<0.2	<0.2	<0.2	<0.2	<0.2
Water	4.1	0.9	3.7	2.9	2.6	2.8	2.8±1.1
Ashes (700°, mainly NaCl)	0.7	0.9	1.5	0.9	0.8	0.8	0.9±0.3
Arsenic (As)	<3	<3	<3	<3	<3	<3	<3
Cadmium (Cd)	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1	<0.1
Lead (Pb)	<0.5	<0.5	<0.5	<0.5	<0.5	<0.5	<0.5
Mercury (Hg)	<0.5	<0.5	<0.5	<0.5	<0.5	<0.5	<0.5

*: Batch No. of ethyl lauroyl arginate produced by LAMIRSA.

5. Functional uses

Ethyl lauroyl arginate is used as preservative. Ethyl-N^{α}-lauroyl-L-arginate HCl which is a cationic surfactant has a wide spectrum of activity against Gram positive and negative bacteria, yeasts and moulds. It acts on cell membranes and the cytoplasm, and inhibits the growth of populations but in no case is cell lysis observed under the conditions studied. It can be used wide range of foods.

6. Reaction and fate in foods

The extent of hydrolysis of ethyl lauroyl arginate under various conditions was determined by measurement of the percentage of ethyl-N^{α}-lauroyl-L-arginate HCl recovered in each sample. In 24 out of 33 samples no hydrolysis process took place. Only 9 samples showed interaction with the components of the sample. In 4 of these 9 samples, it was hydrolysed to N^{α}-lauroyl-L-arginine which is the main metabolite. In the remainder of the samples, in which it was combined with nitrite, meat or soya proteins or ovo-albumin or lacto-albumin, more extensive hydrolysis occurred. In spite of this, no formation of nitrosamines was observed.

The stability of ethyl-N^{α}-lauroyl-L-arginate HCl was also evaluated in eight different food matrices. Five of these matrices were examples of processed foods and the rest were examples of fresh foods. It was found to be stable throughout the duration of the study in all processed food matrices and only in the fresh food matrices was observed its decrease.

7. References

Center for Food Safety and Applied Nutrition, U.S. Food and Drug Administration (2005) Agency Response Letter GRAS Notice No. GRN 000164, http://cfsan.fda.gov/~rdb/opa-g164.html

Scientific Committee on Consumer Products, European Commission (2005) Opinion on Ethyl Lauroyl Arginate, COLIPA No P95, SCCP/0837/04.

European Food Safety Authority, 2007. Opinion of the Scientific Panel on Food Additives, Flavourings, Processing Aids and Materials in Contact with Food on a request from the Commission related to an application on the use of ethyl lauroyl arginate as a food additive, The EFSA Journal, 511, 1-27.

LAMIRSA, 2008. LAE Lauric Arginate "an innovative molecule for preservation", http://www.mirenat.net/

European Patent No. 1294678 held by Lamirsa. Abbreviated description: Process for the preparation of cationic surfactant products composed of an esterified amino acid and a fatty acid linked by an amide bond to the amino group of the amino acid.