ANNALS OF "DUNAREA DE JOS" UNIVERSITY OF GALATI
MATHEMATICS, PHYSICS, THEORETICAL MECHANICS
FASCICLE II, YEAR XVI (XLVII) 2024, No. 2
DOI: https://doi.org/10.35219/ann-ugal-math-phys-mec.2024.2.06

Novel foods - overview, legislation and methods of analysis

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Abstract
Novel foods have recently gained increasing popularity. Given their diversity and nutritional values, which are comparable to commonly consumed foods, novel foods had to be regulated both in terms of the procedural stages required for market introduction at the European Union level, implicitly in Romania, and the requirements they must meet for safe consumption. The analysis methods used for their characterization are generally classic, standardized ones or can be developed "in-house", as is the case for methods developed for the determination of mycotoxins.
The method of analysis used for the determination of mycotoxins involves high performance liquid chromatography coupled with a mass spectrometer (LCMSMS) after the prior extraction of mycotoxins using QuEChERS extraction. The novelty of recent years is the inclusion on the list of novel foods approved at the European Union level of laboratory-grown meat and insects, such as flour beetle larvae available in frozen, paste, or dried form, and house crickets, available in frozen, powdered or desiccated form.

Keywords: novel foods, insects, LCMSMS, QuEChERS, extraction

1. INTRODUCTION

New foods, known as novel foods, represent, according to EU Regulation No. 2283/2015, "any food that was not used for human consumption to a significant extent in the territory of the Union before 15 May 1997, regardless of the date of accession of the Member States to the Union". This category of novel foods includes 10 types of foods that vary from foods with modified structures and nanomaterials to foods obtained through new technologies and food supplements.

Also/Additionally, according to the same EU Regulation No. 2283/2015, this category of novel foods also includes traditional foods from third countries "for which there is a history of safe food use", such as insects or various oilseeds (e.g. Bambara peanuts). Regulation (EU) No. 2470/2017 establishes maximum permitted limits both for microbiological criteria and for various physico-chemical parameters, pesticide residues, contaminants.

The introduction of new foods, such as traditional foods commonly consumed in different parts of the world, into the European Union market has been achieved in small steps since 1997, experiencing an accelerated growth in recent times, especially due to the constantly adapting culinary habits influenced by intercultural exchanges generated by migration, tourism and, last but not least, as a challenge for the younger generation. Thus, from the moment of the appearance of EU Regulation no. 2470 on December 20, 2017 until 05.09.2024, this regulation has undergone 150 amendments, each introducing new foods classified as novel foods.

In our country, the notion of novel food has begun to be increasingly prominent in consumer culture, largely due to recent media coverage of insect consumption, namely entomophagy, as well as that of the production of meat grown in a laboratory using different cell substrates and, last but not least, the media coverage of the consumption of various plants that are not commonly consumed in our country. However, the consumption of various oleaginous fruits or different parts of plants newly introduced in EU Reg. 2470/2017 has not received as much media attention as entomophagy or the consumption of meat grown in a laboratory.

Also, on the European Union market, food products such as pasteurized milk treated with UV, bread treated with UV, products that also belong to the novel food category, have begun to be present, more and more often on store shelves. UV treatment plays a beneficial role by enhancing the final amount of vitamin D3 found in these products through the conversion of cholecalciferol into vitamin D3, as specified in EU Regulation 2470/2017.

The food supplement market has recently experienced a particular boom, especially in the online environment. The food supplements provided for by Reg. EU 2470/2017 are very different, ranging from various extracts (fermented black bean extract, powdered cranberry extract, Echinacea pupurea extract from cell cultures, etc.) to hydrolysates (egg membrane hydrolysate), amino acids (L-alanyl-Lglutamine), cetylated fatty acids, different oils (Krill oil), and plants (Aloe Vera, Noni, etc.).

2. EXPERIMENTAL

In order to be placed on the market, novel foods must comply with legislative regulations both in terms of the procedural stages of market placement and legislative regulations regarding various requirements for physicochemical parameters., microbiological parameters, contaminants, etc.

Legislative regulations

The legislation governing novel foods is represented, at the EU level, by: Reg. EU no. 2283/2015 "on novel foods" and Reg. EU no. 2470/2017 "establishing the Union list of novel foods in accordance with Regulation (EU) 2015/2283 of the European Parliament and of the Council on novel foods". It is important to note that novel foods are subject to the same rules on food safety established in Reg EC no. 178/2002, Reg. 852/2004, Reg. No. 853/2004, Reg. No. 1924/2006 and last but not least in Reg EU no. 1169/2011.

In our country, the introduction of a novel food on the market is regulated by GD No. 590/2018 "on establishing the institutional framework and measures for the implementation of Regulation (EU) 2015/2283 of the European Parliament and of the Council of 25 November 2015 on novel foods, amending Regulation (EU) No. 1169/2011 of the European Parliament and of the Council and repealing Regulation (EC) No. 258/97 of the European Parliament and of the Council and Commission Regulation (EC) No. 1852/2001 as well as Commission Implementing Regulation (EU) 2018/456 on the procedural steps of the consultation process for the establishment of novel food status in accordance with Regulation (EU) 2015/2283 of the European Parliament and of the Council on novel foods".

Regulation UE 2470/2017

Reg. EU 2470/2017 "establishing the Union list of novel foods in accordance with Regulation (EU) 2015/2283 of the European Parliament and of the Council on novel foods" is a complex regulation that presents two interwoven approaches, being structured on two tables.

The first table regulates the authorized novel foods and provides the following information: the name of the authorized novel food, the conditions under which the novel food may be used, the specific category of food in which the novel food may be found, the maximum quantity in which it may be found, labelling requirements, and other relevant requirements.

Novel foods are subject to the same labelling requirements as those set out in Reg. EU no. 1169/2011. Specifically, the name of the novel food must be specified, and additional information may be required regarding consumption by persons from vulnerable groups. Importantly, there must also be a warning regarding the allergic potential associated withthe consumption of these types of food. At the same time, nutritional and health claims must comply with Reg. EC no. 1924/2006.

The second table includes specifications regarding the same novel food and provides the following information: the name of the authorized novel food and specifications regarding various microbiological, physico-chemical parameters, contaminants, etc.

EU Reg. 2470/2017 regulates all aspects of authorized novel foods, with a wide scope of applicability, and encompasses both products of animal origin (e.g. insects, uv-treated milk, etc.), products of non-animal origin (e.g. Bambara peanuts, Noni, uv-treated bread, etc.), various extracts/powders used in industry, food supplements.

From the point of view of official control, what needs to be specified is the importance of the correct classification of the food placed on the market and the compliance with the requirements of EU Reg. 2470/2017. Thus, a comparison must be made with other EU regulations in force such as the updated EU Regulation 915/2023, which governs contaminants in strict categories of food and sets maximum permissible limits for them. The reason why this differentiation is important is that EU Reg. 2470/2017 contains many more specifications than Reg. EU 915/2023, which regulates the maximum permitted limits for contaminants, or other regulations applicable to food products, for example for microbiological criteria such as EU Regulation no. 2073/2005)

The growing interest in entomophagy within the European Union, including in our country, determines intense discussions due to the novelty of the subject in this area of the Earth. The characterization of novel food represented by different types of insects, according to EU Reg. no. 2470/2017, is much more complex both in terms of the requirements for its use and in terms of the requirements for other products of plant origin.

Methods of analysis

The analytical techniques used to determine the physicochemical and microbiological characteristics of insects include classical standardized methods for microbiological determinations or for determining heavy metal contamination. For mycotoxin determinations, laboratory-developed methods are generally based on "in-house" methods coupled with QuEChERS - type extraction *"Fast Easy Cheap Efficient Robust Safe"*.

Development of a method for determining mycotoxins in house crickets

In an authorized veterinary and food safety laboratory, a method for the simultaneous determination of aflatoxin B1 and ochratoxin A contamination in house crickets was developed in the framework of research for a doctoral thesis using QuEChERS extraction and determination by high-performance liquid chromatography coupled with mass spectrometry (LCMSMS).

Thus, QuEChERS extraction involves vigorous shaking of the sample with a small mixture of different organic solvents, such as acetonitrile, methanol and inorganic phases such as magnesium sulfate and sodium chloride, laboratory consumables. [13,14]

The adoption of QuEChERS extraction methods orients analytical techniques towards the desideratum of Green Analytical Chemistry, which is interdependent on the principles of green sample preparation [1]

Innovation in Romania

The laboratory study was conducted on live insects, namely house crickets, and monitored for 14 days the assess the transfer rate of mycotoxins (aflatoxin B1 and ochratoxin A) from contaminated feed at a level of 2 µg/kg aflatoxin B1 and 32 µg/kg ochratoxin A in the insects studied. The house crickets were purchased online (Fig. 1).

Fig. 1 Acheta domestica – delivery form

The insects were analyzed at the beginning of the study to determine the initial contamination levels and were subsequently monitored at 7-day intervals. 6 replicates were performed for each analytical series, with the number of replicates chosen according to EU Regulation No. 2021/808.

The house crickets were fed ad libitum with contaminated feed and were kept under optimal conditions of temperature, light and humidity.

The analytical technique used for the determination of mycotoxins was represented by LCMSMS and involved two steps: extraction of mycotoxins from the sample and chromatographic determination of mycotoxins.

In summary, the steps can be described as follows: 5 grams of the sample are homogenized with 20 ml of extraction solvent water/acetonitrile/acetic acid (50/49/1 v/v/v), vigorous homogenization for 30 minutes, 6 grams of magnesium sulfate and 1 gram of sodium chloride are added to the thus homogenized sample, vigorous stirring for 3 minutes, centrifugation, the expressed supernatant is filtered and diluted part by part (1/1 v/v) with methanol, and thus the sample is prepared to be analyzed by the liquid chromatographic technique (Fig 2).

Fig .2 LCMSMS equipment

The results obtained are shown for house crickets in Table 1.

Table 1. Mycotoxin levels found in house crickets

Amount of mycotoxin added to feed	Values found after 7 days	% Recovery at 7 days	Values found after 14 days	% Recovery at 14 days
Aflatoxin B1 2 µg/kg	0.36 µg/kg	18.41%	0.34 µg/kg	17.00 %
Ochratoxin A 32 µg/kg	8.08 µg/kg	25.27%	7.67 µg/kg	23.98 %

According to EU Reg. 2023/915, the maximum permitted limits for cereals intended for human consumption, for the studied contaminants, are 2 µg/kg for aflatoxin B1 and 3 µg/kg for ochratoxin A; the values are summarized in Table 2.

Table 2. Maximum permitted limits for cereals according to EU Reg. No. 2023/915

Mycotoxin	Cereals
Aflatoxin B1	2 µg/kg
Ochratoxin A	3 µg/kg

According to EU Reg 2470/2017, the requirements for aflatoxin B1 and ochratoxin A in house crickets are given in Table 3.

Table No. 3. Mycotoxin requirements in house crickets according to EU Reg. 2470/2017

Contaminant	Requirements according EU Reg 2470/2017
Aflatoxin B1	≤ 2 µg/kg
Ochratoxin A	≤ 1 µg/kg

3. RESULTS AND DISCUSSION

After the dynamic analysis of the results obtained, a relatively uniform response of house crickets to feeding with aflatoxin B1 and ochratoxin A-contaminated feed is observed in the sense that: at day 7 of the study, an accumulation of mycotoxins was observed, aflatoxin B1 increased by 18.41% and ochratoxin A increased by 25.27%; at day 14 of the study, a decrease in the concentration of mycotoxins analyzed in the insects studied was observed, aflatoxin B1 is found in a percentage of 17.00% and ochratoxin A is found in a percentage of 23.98%. By simple mathematical calculation of decrease, it is found that aflatoxin B1 in house crickets decreased by 1.41% at day 14, compared to day 7, and ochratoxin A decreased by 1.29% at day 14 , compared to day 7 (Fig. 2).

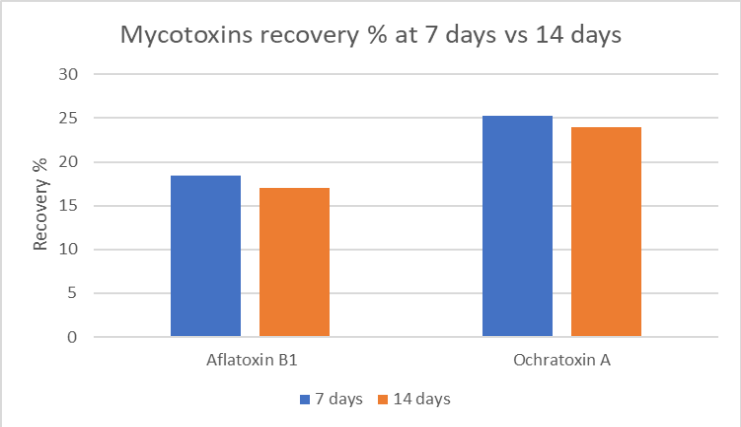

Fig.2. Mycotoxins recovery % at 7 days vs 14 days

4. CONCLUSIONS

The analytical technique used, namely the simultaneous determination of mycotoxins such as aflatoxin b1 and ochratoxin a by LCMSMS, is a technique that proves to be reliable, with low reagent consumption and obtaining repeatable and reproducible results that demonstrate the suitability of the method for its intended purpose.

Data processing demonstrates that the mobility factor, respectively the ratio between the contaminant found in the studied insects and the amount of mycotoxin in the feed is sub-unitary, so that it can be concluded that the insects do not have the ability to fix the mycotoxins studied.

The results of the "in vivo" study were surprising and open the way for new research. potentially conducted over a longer period and with different types of contaminants for a possible future integration of products not complying with the maximum permitted limits into the feed of different types of insects and thus reducing food waste.

Novel foods, regardless of their type, guide both the authorities and the final consumer towards sustainability and a circular economy.

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