Regulations relating to mycotoxins in almonds in European context

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Abstract

Mycotoxins are secondary metabolites produced by several species of fungi and having a toxic effect on humans and farm animals. In particular, almonds, a rich source of nutrients and phytochemicals, can be contaminated by aflatoxins, one of the most important mycotoxins, mainly produced by Aspergillus flavus and Aspergillus parasiticus. The reference regulations in this field are continuously improved and updated worldwide. This paper reports the current state of the European regulations on aflatoxins in almonds concerning the limits, and the procedures for performing official controls and for import.

Introduction

Mycotoxins are secondary metabolites produced by fungal species, mainly in the genera Aspergillus, Penicillium and Fusarium. Mycotoxins have a toxic effect on humans and farm animals. In recent years, health authorities have dedicated much attention to food contamination by mycotoxins. Important sources of contamination are cereals, nuts, dried fruits, spices, dairy products, coffee, wine, and meat products (1). The risks arising from the ingestion of mycotoxins in contaminated foods, and the huge economic losses that this causes, have led many countries to impose control measures to protect the health of consumers and overcome the associated economic problems. In particular, almonds (Amygdalus communis L.) are a rich source of nutrients and phytochemicals, and can be contaminated by aflatoxins (AFs), one of the main groups of mycotoxins produced in nature. The contamination of almonds is influenced by food integrity, especially at the level of the shell: almonds damaged by insects are contaminated with higher concentrations of AFs than almonds that are not damaged (2, 3). Moderate and regular consumption (~30 g daily) of almonds and nuts generally has health-promoting effects. Almonds are rich in monounsaturated and polyunsaturated fatty acids, and phytosterols, and have a low glycemic index, thereby reducing some risk

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factors related to diabetes and cardiovascular disease (4-7). Almonds also have anti-inflammatory and antioxidant activities owing to their α-tocopherol and polyphenol content, including flavonoids, anti-cancer dietary components, and hepatoprotective, neuroprotective and cholesterol-lowering properties (8-11). The consumption of almonds is also associated with an increase in bone mineral density (12), and their oils have been shown to have antibacterial and antifungal activities (13).

The aim of this study was to present the current state of the European regulations on mycotoxins (particularly AFs) in almonds, by consulting the Official Journal of the European Union.

**Regulations**

Countries regulating the presence of mycotoxins have significantly increased over the years. The supervision and control of mycotoxins have become an important objective for manufacturers, regulators and researchers worldwide. Therefore, regulations are continuously updated, based on scientific evidence provided by international organizations such as the United Nations Joint Expert Committee on Food Additives (JECFA) and European Food Safety Authority (EFSA).

Determination of the maximum tolerable concentration of mycotoxins in foods (as well as other carcinogens of natural origin) takes into account the ALARA principle (as low as reasonably achievable) (14). This prevention policy aims to minimize the risk by maintaining exposure to the lowest mycotoxin levels that are reasonably achievable.

**Aflatoxins**

AFs are secondary metabolites that are mainly produced by *Aspergillus flavus* and *Aspergillus parasiticus*, and are chemically related to difuranocumarins. Among the 17 AFs known, only five are considered to be relevant for both distribution and toxicity: B1, B2, G1, G2 and M1. AF exposure through the food chain can cause aflatoxicosis in humans and farm animals. Acute exposure episodes may include symptoms as haemorrhage, acute liver damage, oedema, alteration in digestion, absorption and/or metabolism of nutrients, and may result in death (15). Chronic aflatoxicosis, due to exposure to low and persistent doses of AFs, results in cancer and immune suppression.

On September 23, 1994 the Scientific Committee on Food (SCF), a technical body of the European Commission, expressed the opinion that AFs are genotoxic carcinogens (16). On the basis of this evaluation, some limits on the total content of AFs (B1, B2, G1 and G2) in food were established over the following years, with particular reference to the presence of AFB1. B1 is one of the most toxic and carcinogenic AFs in the liver, and is classified in group 1 (substance definitely carcinogenic to humans) by the International Agency for Research on Cancer (IARC).

Regulation (EC) No. 1881/2006 (14) set the levels for mycotoxin contaminants, declaring that foods exceeding these limits cannot be marketed. Regulation (EC) No. 1881/2006 also imposed an important ban on products that have undergone detoxification. Foodstuffs containing mycotoxins should not be deliberately detoxified by chemical treatment. Consequently, until 2010, foodstuff export was strict and difficult. Recently, the European Community planned changes to the legislation concerning AF limits, nuts sampling strategy and import.

On January 25, 2007 EFSA published its opinion on the potential increase in consumer health risk posed by a possible increase in the existing maximum levels for AFs in almonds, hazelnuts, pistachios, and derived products (17). The Scientific Panel on Contaminants in the Food chain (CONTAM) concluded
that changing the maximum levels for total
AFs in almonds and derived products from
4 to 8 or 10 μg/kg would have minor effects
on the estimates of dietary exposure and
cancer risk. CONTAM assumed that the
intake of AFs from nuts represents only a
small percentage of total dietary exposure
to these mycotoxins, and concluded that
exposure to AFs from all sources should
be as low as reasonably achievable, so
it is important to reduce the number of
highly contaminated foods reaching the
market (18). Regulation (EU) No. 165/2010
established the maximum level for total
AFs in almonds and derived products, as
defined by the Codex Alimentarius (the
joint FAO/WHO commission to develop
rules and international dietary guidelines),
modifying the previous Regulation (EC) No.
1881/2006. Additionally, while Regulation
(EC) No. 1881/2006 refers to nuts without a
clear distinction among the different species
of nuts, Regulation (EU) No. 165/2010
introduces for the first time the term almond,
associated with pistachio and apricot kernels
(Table 1).

Maximum levels adopted by the EU can
be subject to review to take into account
any advances in scientific and technical
knowledge and improvements in good
agricultural and manufacturing practices.
Member states and stakeholders annually
provide to the European Commission
information relating to any investigations
undertaken. The Commission makes these
results available to the member states.

### Table 1 - Maximum levels of AF in almonds [from Regulation (EU) No. 165/2010, modified]

<table>
<thead>
<tr>
<th>Description</th>
<th>B1</th>
<th>B1+B2+ G1+G2</th>
</tr>
</thead>
<tbody>
<tr>
<td>2.1.2. Almonds, pistachios and apricot kernels to be subjected to sorting, or other physical treatment, before human consumption or use as an ingredient in foodstuffs</td>
<td>12.0</td>
<td>15.0</td>
</tr>
<tr>
<td>2.1.6. Almonds, pistachios and apricot kernels intended for direct human consumption or use as an ingredient in foodstuffs</td>
<td>8.0</td>
<td>10.0</td>
</tr>
</tbody>
</table>

**Sampling and analysis**

Sampling plays a crucial role in
determination of the levels of mycotoxins, which are heterogeneous in foodstuffs.

The sampling procedure for the official control of mycotoxin levels in foodstuffs should be specified in the regulations in force, and should aim to obtain samples that are representative of the whole batch. It is also necessary that laboratories use methods of analysis with comparable levels of performance and that the general criteria of the analytical methods are fixed.

Regarding the official procedures for sampling and analytical methodology, Regulation (EC) No. 401/2006 (19) specifies the methods for sampling and analysis for the official control of the levels of mycotoxins in foodstuffs. However, because the Codex Alimentarius established a new sampling procedure for peanuts, almonds, hazelnuts and pistachios to be subjected to physical treatment and for direct human consumption, Regulation (EC) No. 401/2006 was later modified by Regulation (EU) No. 178/2010 (20) regarding groundnuts, oilseeds, nuts, apricot kernels, liquorice, and vegetable oil. It was further modified by the recent Regulation (EU) No. 519/2014 (21) regarding the sampling method for large/very large lots, or lots stored or transported in a way in which sampling throughout the lot is not feasible, for spices and food supplements.

Some authors (22) have pointed out that EU regulations state that “if it is
not possible to carry out the method of sampling described above because of the unacceptable commercial consequences resulting from damage to the lot (because of packaging forms, means of transport, etc) an alternative method of sampling may be applied provided that it is as representative as possible and is fully described and documented”, but the subsequent Regulation (EU) No. 178/2010 regarding almonds and nuts (20) still lacked a guide, as it does not indicate this method. Finally, Regulation No. 519/2014 (21) eliminated these doubts.

Import

The almond import refers to the recent Regulation (EU) No. 323/2014 amending Annexes I and II to Regulation (EC) No. 669/2009 implementing Regulation (EC) No. 882/2004 of the European Parliament and of the Council with regard to the increased level of official controls on imports of food of non-animal origin (23). Countries are subject to special conditions for the import of certain feeds and food, which are regulated by Regulation (EU) No. 884/2014 (24). The latter regulation repealed Regulation (EC) No. 1152/2009 (25), and imposed special conditions for AFs and governed the import of certain foods from developing countries (Brazil, China, Egypt, Iran, Turkey, Ghana, India and Nigeria). Because of favourable control results and the outcome of the Food and Veterinary Office inspection audit, the US is no longer subject to special conditions for the import of almonds. Removing special import conditions for US almonds means that a Voluntary Aflatoxin Sampling Plan (VASP) certificate is no longer a precondition for import into the EU. However, the Almond Board of California strongly advises its members to continue to use the VASP certificate. Moreover, the use of the health certificate is no longer required (26).

Conclusions

Despite the presence of mycotoxins (including AFs in almonds) in food appearing to be well regulated, it is impossible to entirely remove the risk of exposure, because they are natural contaminants. Given that almonds have health-promoting effects, appropriate strategies should be universally adopted, such as monitoring pre-harvest conditions, post-harvest storage, transport, and processing, to protect the health of the consumer.

Riassunto

Aspetti legislativi sulle micotossine nelle mandorle nel panorama europeo

Le micotossine sono metaboliti secondari prodotti da diverse specie fungine appartenenti ai generi Aspergillus, Penicillium e Fusarium ed hanno un effetto tossico sull’uomo e sugli animali da fattoria. Le mandorle, in particolare, sono una fonte ricca di nutrienti e sostanze fitochimiche, ma possono essere contaminate da aflatossine, una delle principali micotossine prodotte da Aspergillus flavus e Aspergillus parasiticus. Le normative di riferimento in questo ambito sono in continua evoluzione e suscitano un grande interesse in tutto il mondo.

Il presente lavoro riporta lo stato attuale della normativa europea sulle aflatossine nelle mandorle.

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