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# TOXIN PRODUCING MICROMYCETES ON IMPORTED PRODUCTS OF PLANT ORIGIN

Albinas Lugauskas, Vita Raudonienė, Laima Šveistytė

Institute of Botany, Biodeterioration Reseach Laboratory, Vilnius, Lithuania

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**Abstract:** Recently the food of people is profusely supplemented with vegetables and fruit imported from various regions. Investigations on the mycological state of imported foodstuffs revealed that the marketed vegetables, fresh, dried and frozen fruit are contaminated with propagules of various micromycetes. The obtained results allow the conclusion that vegetables and fruit can become a good substrate for mycotoxin producing micromycetes. The micromycetes develop on everyday products and can become the cause of slow toxicoses, which are characterized by a diversity of symptoms and are difficult to diagnose. Therefore, contamination of food products with micromycetes of the *Penicillium*, *Aspergillus*, *Fusarium*, *Alternaria*, *Paecilomyces*, *Trichotecium*, *Rhizopus* genera should receive particular attention. It should be noted that a strain growing on a particular type of vegetable or fruit could synthesize and excrete different toxic secondary metabolites.

**Address for correspondence:** Prof. Dr. Habil. Albinas Lugauskas, Institute of Botany, Biodeterioration Reseach Laboratory, Žaliųjų ežerų 49, LT-08406 Vilnius, Lithuania. E-mail: lugauskas@botanika.lt

**Key words: fungi, contamination, toxin, vegetable, fruit, products, imported.**

## INTRODUCTION

Aiming to enrich and add variety to nutrition, as well as for economic reasons, many raw materials and products of plant origin are imported. They are widely used by both city and countryside inhabitants. Vegetables, fruit, berries, seeds and grains are imported. Sellers according to their biological properties, geographical zones of cultivation, ripening time and degree of ripeness, classify these products. The imported vegetative vegetables are subdivided into tuberous: potato (*Solanum* L.), sweet potato (*Ipomea* L.), sunroot (*Helianthus tuberosus* L.); rootstock: carrot (*Daucus* L.), parsnip (*Pastinaca* L.), celery (*Apium* L.), parsley (*Petroselinum* Hill), turnip (*Brassica rapa* L. ssp. *rapa*), radish (*Raphanus* L.), garden radish (*Raphanus sativus* L.), garden beet (*Beta vulgaris* subsp.), onion (*Allium* L.), common garlic (*Allium sativum* L.); spices: dill (*Anethum* L.), lovage (*Levisticum* Hill), basil (*Ocimum* L.), fennel (*Foeniculum* Mill.); dessert: globe artichoke (*Cynara* L.); generative vegetables including pulses: field bean (*Vicia faba* L.), pea (*Pisum* L.), bean (*Phaseolus* L.) and cereals – maize (Zea L.); fruit vegetables: tomato (*Lycopersicon* Mill.), eggplant (*Solanum melongena* L.), paprika (*Capsicum* L.); gourd: gourd (*Cucurbita* L.), watermelon (*Citrullus* Schrad. ex Eckl. et Zeyh.), yellow melon (*Cucumis melo* L.), vegetable marrow (*Cucurbita pepo* convar. *giromontiina* Greb.), pattypan (*Cucurbita pepo* convar. *patissonina* Greb.), and cucumber (*Cucumis* L.).

The imported fruit can be grouped into pomes: apples (*Malus* Mill.), pears (*Pyrus* L.), flowering quince (*Chaenomeles* Lindl.), whitebeam (*Sorbus* L.); drupes: apricots (*Armeniaca* Scop.), peach (*Persica* Mill.), plums (*Prunus* L.), cherries (*Cerasus* Mill.), cornel (*Cornus* L.); berries: grape (*Vitis* L.), hagrose (*Rosa* L.), currant (*Ribes* L.), cranberry (*Oxycoccus* Hill), cowberry (*Vaccinium*

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*vitis-idaea* L.), black whortleberry (*Vaccinium myrtillus* L.) and compound berries: blackberry (*Rubus idaeus* L.), cloudberry (*Rubus chamaemorus* L.), and European dewberry (*Rubus caesius* L.).

A variety of nuts and drupes are brought: hazel (*Corylus* L.), walnut (*Juglans regia* L.), buckeye (*Aesculus* L.), cedar (*Cedrus* Trew), almonds (*Amygdalus* L.), and pistachios (*Pistacia* L.). Fruit of tropic and subtropical regions comprise a large part of the imported foodstuffs: orange (*Citrus sinensis* (L.) Osbeck), mandarin (*Citrus reticulata* Blanco), grapefruit (*Citrus paradisi* Macfad.), lemon (*Citrus* L.), banana (*Musa* L.), pineapple (*Ananas* Mill.), mango (*Mangifera* L.), fig (*Ficus carica* L.), date (*Phoenix dactylifera* L.), and persimmon (*Diospyros* L.).

The variety of the imported foodstuffs of plant origin and conditions of their growth, harvesting, processing, packing, transportation, and finally marketing predetermine the micromycete quantity and species diversity functioning on them. Some authors [6, 11] distinguish micromycetes, which damage the products of plant origin and cause the so-called "market diseases". *Botrytis cinerea, Rhizopus stolonifer, Colletotrichum lindemuthianum, C. coccodes*, *Alternaria alternata*, *Penicillium digitatum*, *P. expansum*, *P. italicum*, *Sclerotinia sclerotiorum, Monilinia fructigena, Mucor piriformis*, *M. racemosus*, as well as some other micromycete species of the *Phytophtora*, *Bremia*, *Venturia*, *Fusarium*, *Diplodia*, *Phomopsis*, *Trichotecium*, *Cladosporium* genera are ascribed to the causing agents. Very often in damaged products of plant origin communities of different micromycete species and other microorganisms function [14, 37, 43].

Vegetables, fruit, sometimes even seeds damaged by micromycetes turn into a watery mass. On the skin of fresh fruit brownish or grey spots appear; mycelium of a fungus penetrates into deeper tissues. Some micromycetes cause dry rots. Then the part of a vegetable or fruit damaged by fungi gradually dries, becomes corky, resembling wrinkled skin. The fruit value is reduced and causes considerable economic losses. This is one of the negative points of the micromycete action. The other point is that of their hazard, i.e., as the products damaged by micromycetes are marketed, micromycetes are free to spread and cause real risk to human health. If vegetables, fruit or berries are damaged by micromycetes producing and excreting toxic secondary metabolites, these products can be the cause of chronic mycotoxicoses of people and animals. According to some researchers [7, 10, 19, 25, 26, 32], the following micromycetes, frequently occurring on vegetables and fruit, should be particularly mentioned: *Penicillium expansum*, *P. crustosum*, *P. italicum*, *P. variabile*, *Fusarium sporotrichiodes*, *F. poae*, *F. moniliforme*, *Aspergillus fumigatus*, *A. niger*, *Alternaria alternata*, and *Bipolaris sorokiniana*.

Many products of plant origin are brought dried or processed. They are on sale in various shops, market places. Some researchers state that the majority of micromycetes detected on these products are able to synthesize mycotoxins harmful to human health [7, 9, 13, 14, 20, 21, 26, 27, 30, 35, 37, 38, 40, 43]. The majority are micromycetes belonging to the genera *Aspergillus*, *Penicillium*, *Paecilomyces*, *Alternaria*. Toxin producing micromycetes of the above-mentioned genera were detected by Armenian scientists [45] in various herbal preparations, pills, capsules, granules, solutions, ointments and herbal tea mixtures.

On the processed products of plant origin and in premises where they are processed, micromycetes of *Aspergillus flavus* group, ascribed to *Aspergillus oryzae* species, are frequent and, according to [44], these fungi are closely related to the aflatoxigenic species *Aspergillus flavus* and *A. parasiticus*. Although these food fungi have never been shown to produce aflatoxin, they contain homologues of several aflatoxin biosynthesis pathway genes. Deletions and other genetic defects have led to the silencing of the aflatoxin pathway in *Aspergillus oryzae*. This information is very important as under the impact of certain environmental factors, micromycetes widely spread in grain and other food processing premises where production technologies are related with alterations in temperature or other physical factors; conditions favourable for aflatoxin production may form, especially when in communities where closely related fungal species able to synthesize these toxins occur. Crossbreed processes are possible [1, 12, 15, 41].

The aim of the research was to determine micromycetes on the imported and widely marketed vegetables, fruit, berries, seeds, grains and other foodstuffs of plant origin; to identify their impact upon the quality of food products and potential risk to the health of consumers.

## MATERIALS AND METHODS

**Investigated premises.** During the period of 2002–2004 samples of the imported vegetables, fruit, berries, seeds, grains and their products were taken in two wholesale centres where many trading companies supply imported goods. Part of the imported goods were packed in places of their growth or in storehouses of the countries of their origin and, therefore, the origin of their fungal infection is ascribable to the ecological niche of that environment. Another part of their production was during transportation when they were in contact with the ambient air, transportation vehicle and got into new storehouses with particular mycological conditions. The imported products were stored, distributed, transported and marketed in new environment. During these processes the possibilities for the products to become contaminated with micromycetes present in the premises occurred. As in large wholesale centres there is an intense movement of people and vehicles, micromycete propagules can easily get on the products from the outdoor air and environment. The surface of the majority of dried fruits and vegetables is sticky. This helps micromycete propagules attach to the surface of products and start their functioning. In both wholesale centres the imported raw material of plant origin and products (vegetables, fruits, berries, seeds, processed and unprocessed grain) were on sale.

**Isolation of micromycetes.** In the above-mentioned premises, 223 samples of vegetables, fruits, berries and other foodstuff of plant origin were taken. They were analyzed according to the methods described by [17, 23, 24, 25, 29, 30, 36]. When visual observation allowed the presumption of the contamination by one infection agent, the plating method was employed; in case of possible mixed infections the method of diluting was applied. In the first case, a piece of infected product, cut of with sterile scalpel, was placed onto a Petri dish containing malt extract agar medium with chloramphenicol (50 mg/l). In the second case, 1 g of product was taken and placed in 10 ml of sterile water, shaken for 15 min and a series of dilution series were carried out. 1 ml of suspension was drawn into 9 cm diam. Petri dish and poured over with 15 ml (48ºC) of the same malt extract agar medium enriched with antibiotic. The dishes were kept for 4 days in a thermostat at a temperature of 28ºC, and for the next 4 days at a temperature of 20ºC; the regime of light and dark was changed every 12 hours. Pure micromycete cultures were isolated, cultivated in standard Czapek agar, standard malt, and corn extract media at a temperature of 28ºC for 5–7 days and identified according to the manuals [8, 12, 16, 17, 22, 28, 31, 35]. Detection frequency (%) of each identified species was calculated.

**Evaluation of the micromycete toxicity.** Ability of micromycetes to synthesize and excrete secondary metabolite (mycotoxins) was tested applying described methods by [12, 39]. Micromycetes were cultivated on standard Czapek, Czapek yeast extract agar (CYA) and yeast extract-sucrose agar (YES) media for 7–14 days at a temperature of 28ºC. Significant changes in the colour of the fungal colony and abundant excretion of pigment into CYA and YES media, comparing with the growth in Czapek media, in the authors opinion, allow the supposition that the investigated strain can be a potential producer of mycotoxins. The authors indicate that the above-mentioned media particularly induce the synthesis of mycotoxins by micromycetes of the *Aspergillus*, *Penicillium*, *Fusarium* and *Alternaria genera*.

Primary selection of micromycetes according to their ability to synthesize toxic metabolites was performed employing the method of thin layer chromatography (ISO 8178-2:1999). The following systems of solvents were used in the research: chloroform-metanol (98:2), chloroform-metanol (8,5:1,5), chloroform-metanol (95:5), toluol-etilacetat-formic acid (5:4:1), benzol-acetic acid (9:1), toluol-acetone-metanol (5:3:2). Micromycetes were cultivated for 20 days in agar CYA and YES media.

Determination of toxins in samples of vegetables, fruits, berries, seeds, grain and their products was performed by the ELISA method [3, 36, 39]. Extraction of mycotoxins and tests were performed according to manufacturers instructions. The VERATOX®, Alotox (total), VERATOX®DOH5/5, VERATOX®-Ochratoxin A, Aflatoxin, T-2 toxin, zearalenone and RIDACHREEN® Ochratoxin A test kits (R-Biopharm AG, Germany) were used for the analysis.

**Statistical analysis.** The obtained results were processed using Microsoft Excel 2000, Statistica 5.1 and Primer 5 programs.

## RESULTS

The research was carried out in large supermarkets where already sorted products of higher quality, less damaged or contaminated with microorganism propagules are brought. In properly equipped and well maintained wholesale premises the conditions for micromycete development are not favourable: low relative humidity, temperature not exceeding 20ºC, and a sufficient ventilation system. In these premises, intense movement of people and vehicles, packaging, unloading and loading takes place. As a result, the air in these premises abounds in dust and micromycete propagules. It was noticed that in such conditions micromycetes of the *Aspergillus*, *Penicillium*, *Mucor*, *Rhizomucor*, *Rhizopus* genera can spread quicker than other fungi, are able to contaminate and damage a wide range of products of plant origin, and remain viable under rather extreme environmental conditions: humidity shortage, low air temperature, the influence of ultraviolet radiation, and other physical factors. Analysis of micromycete species detected on foodstuffs stored in various premises and determination of the ratio between the number of the detected micromycete species and the amount of the tested products (p) revealed that in specialized vegetable storehouses, where various vegetables are not just preserved but also sorted, packed, and loaded into containers, this ratio is significantly ($p<0.05$) higher than the average. This ratio is even higher in non-specialized vegetable storehouses. T-test showed the number of micromycete species per sample. In this case, the average, is the number obtained after dividing the number of micromycete species detected in all investigated premises by the number of samples. In specialized and non-specialized vegetable and fruit storehouses a higher relative number of micromycete species per one type of product was determined comparing with premises for short-termed storing, sorting, and wholesale marketing. However, in specialized storehouses for onions and apples and in wholesale premises the average number of micromycete species per sample was significantly lower ($p<0.05$) than the average number of micromycete species per one sample in all investigated premises. In premises where products are not just stored but also sorted and sold the diversity of micromycete species was evidently higher. This diversity is constantly enriched with micromycetes brought together with the imported vegetables, fruits, berries, seeds, grain. Under such conditions, the most

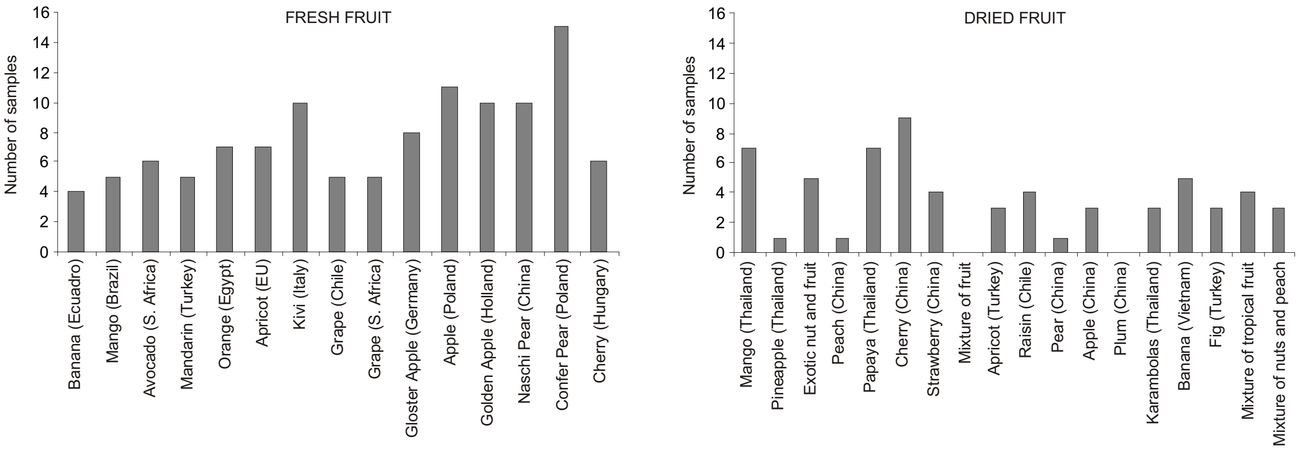


**Figure 1.** Number of micromycete species isolated from the imported fruit.

frequent were fungi of *Penicillium granulatum* (detection frequency 14.3–20.0%), *P. italicum* (9.5%), *P. chrysogenum*, *P. digitatum* (7.9% each) species. In a shop where potatoes and various vegetables are sold, *Acremonium strictum*, *Verticillium alboatrum* (detection frequency 15% each) and *Cladosporium cladosporioides*, *C. cucumerinum* (10% each) were frequent. Micromycetes of the last-mentioned species were recorded on potatoes (*C. cladosporioides*) and dill (*C. cucumerinum*) imported from Spain.

According to statistical analysis by the LSD test, genera and species of micromycetes significantly dominating on products of plant origin in the investigated premises are presented in Tables 1 and 2.

The majority of micromycetes recorded in the investigated premises were ascribed to the *Penicillium*

**Table 1.** Dominant micromycete genera (according to the LSD test): statistically significant differences are bold-typed

| Micromycete genus | Genera distribution, % | Significance of differences in detection frequency of micromycete genera (p) | | | | |
|---|---|---|---|---|---|---|
| | | *Penicillium* | *Aspergillus* | *Fusarium* | *Mucor* | *Rhizopus* |
| *Penicillium* | 45.4 | | | | | |
| *Aspergillus* | 10.8 | 0.000 | | | | |
| *Fusarium* | 3.6 | 0.000 | 0.042 | | | |
| *Mucor* | 3.5 | 0.000 | 0.040 | 0.984 | | |
| *Rhizopus* | 3.2 | 0.000 | 0.033 | 0.922 | 0.937 | |
| *Acremonium* | 3.0 | 0.000 | 0.027 | 0.860 | 0.875 | 0.937 |
| *Cladosporium* | 2.4 | 0.000 | 0.018 | 0.739 | 0.754 | 0.814 |
| *Sclerotinia* | 2.2 | 0.000 | 0.016 | 0.695 | 0.710 | 0.769 |

**Table 2.** Dominant micromycete species (according to the LSD test): statistically significant differences are bold-typed

| Micromycete species | Detection frequency, % | Significance of differences in detection frequency of micromycete species (p) | | | | |
|---|---|---|---|---|---|---|
| | | *Penicillium expansum* | *Aspergillus niger* | *Penicillium granulatum* | *Penicillium claviforme* | *Penicillium spinulosum* |
| *Penicillium expansum* | 13.2 | | | | | |
| *Aspergillus niger* | 12.8 | 0.8973 | | | | |
| *Penicillium granulatum* | 8.8 | 0.1218 | 0.1560 | | | |
| *Penicillium claviforme* | 6.0 | 0.0101 | 0.0145 | 0.3019 | | |
| *Penicillium spinulosum* | 5.6 | 0.0069 | 0.0101 | 0.2456 | 0.8973 | |
| *Penicillium italicum* | 5.4 | 0.0057 | 0.0084 | 0.2203 | 0.8464 | 0.9485 |
| *Sclerotinia sclerotiorum* | 5.1 | 0.0038 | 0.0057 | 0.1756 | 0.7469 | 0.8464 |
| *Penicillium chrysogenum* | 4.9 | 0.0031 | 0.0047 | 0.1560 | 0.6985 | 0.7962 |
| *Penicillium biforme* | 4.5 | 0.0020 | 0.0031 | 0.1218 | 0.6056 | 0.6985 |
| *Rhizopus oryzae* | 4.2 | 0.0013 | 0.0020 | 0.0937 | 0.5186 | 0.6056 |
| *Penicillium digitatum* | 4.0 | 0.0011 | 0.0017 | 0.0818 | 0.4777 | 0.5613 |
| *Rhizopus stolonifer* | 4.0 | 0.0011 | 0.0017 | 0.0818 | 0.4777 | 0.5613 |
| *Botrytis cinerea* | 3.4 | 0.0005 | 0.0008 | 0.0533 | 0.3663 | 0.4387 |
| *Alternaria alternata* | 3.2 | 0.0004 | 0.0007 | 0.0458 | 0.3331 | 0.4015 |
| *Acremonium strictum* | 3.1 | 0.0003 | 0.0005 | 0.0393 | 0.3019 | 0.3663 |

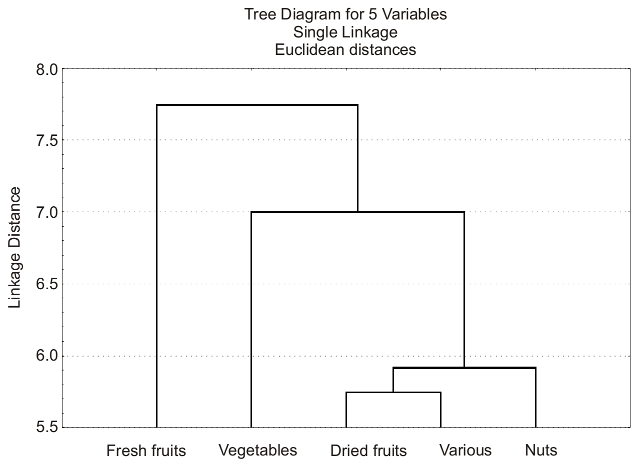


**Figure 2.** Micromycetes species diversity on the products of plant origin.

(45.4%), *Aspergillus* (10.8%), *Fusarium* (3.6%), *Mucor* (3.5%), *Rhizopus* (3.2%), *Acremonium* (3.0%), *Cladosporium* (2.4%), and *Sclerotinia* (2.2%) genera. Less frequent were fungi of the *Alternaria*, *Botrytis*, *Phoma*, *Rhizoctonia*, *Verticillium* genera; agents of plant diseases able to develop on both living and dead parts of plants are ascribed to these genera.

The diversity of micromycete species in the investigated premises depended upon the variety of the analyzed food products, environmental conditions and protective measures applied. It was determined that certain micromycetes are frequent in particular premises or typical of a particular kind of foodstuffs, still others are not substrate-specific and can be detected in a variety of premises (Tab. 2).

In many premises, micromycetes of the following species were detected: *Acremonium strictum*, *Aspergillus niger*, *Botrytis cinerea*, *Fusarium moniliforme*, *Geotrichum candidum*, *Penicillium biforme*, *P. chrysogenum*, *P. claviforme*, *P. digitatum*, *P. expansum*, *P. funiculosum*, *P. granulatum*, *P. italicum*, *P. palitans*, *P. paxilli*, *P. spinulosum*, *Rhizopus oryzae*, *R. stolonifer*, and *Sclerotinia sclerotiorum*. These micromycetes are the least substrate-specific; they are widespread cosmopolitan micromycetes.

Many micromycete species were isolated from the imported fresh and dried fruits. On all imported fruit micromycetes of certain species were recorded. Data on the abundance and species diversity of micromycetes on particular fruits are presented in Fig. 1.

Data about dominant micromycetes on the imported and marketed fresh and dried fruits are presented in Table 3.

The cluster analysis (Fig. 2) shows the differences between the composition of micromycete species isolated from nuts and fresh fruit. Nevertheless, species diversity of prevailing micromycetes in the fresh fruits and vegetables was similar.

On fresh fruit, *Penicillium expansum*, *Sclerotinia sclerotiorum*, *Alternaria alternata*, *Penicillium italicum* as well as abundant, usually white, sometimes yellowish, reddish or dark-colored *Mycelia sterilia* dominated; a coating formed of *Mycelia sterilia* also prevailed on dried fruit. *Penicillium expansum*, *Aspergillus niger*, *Rhizopus oryzae*, *Penicillium italicum* dominated on dried fruit.

**Table 3.** Micromycetes dominating on the imported fruits, A – fresh fruits, B – dried fruits, detection frequency, %.

| Micromycete species | Isolated | A % | B % |
|---|---|---|---|
| *Alternaria alternata* (Fr.) Keissl. | Apricots, apples, pears | 40 | 0 |
| *Alternaria pluriseptata* (P. Karst et Kar.) Jorst. | Apples, pears | 20 | 0 |
| *Alternaria solani* Sorauer | Apples, pears | 27 | _ |
| *Aspergillus niger* Tiegh. | Mangos, exotic nut and fruit mix, raisins, tropic fruit mix | 0 | 22 |
| *Aspergillus oryzae* (Ahlb.) Cohn | Cherries, strawberries, raisins | 0 | 17 |
| *Sclerotinia sclerotiorum* (Lib.) de Bary | Mangos, banana, avocados, oranges, apples, grapes, kiwi fruits, pears, sweet cherries | 47 | 0 |
| *Penicillium brevicompactum* Dierckx | Mangos, apples, banana | 0 | 0 |
| *Penicillium daleae* K.M. Zalessky | Oranges, grapes, pears | 20 | 0 |
| *Penicillium expansum* Link | A. Mangos, avocados, oranges, kiwi fruits, grapes, apples, pears B. Mangos, pineapples, cherries | 67 | 27 |
| *Penicillium ialicum* Wehmer | Mangos, mandarins, oranges, apricots, kiwi fruits, apples | 40 | 22 |
| *Penicillium verrucosum* Dierckx | Apples, pears | 27 | 0 |
| *Rhizopus oryzae* Went ex Prins. Geerl. | Exotic nut and fruit mix, papayas, tropic fruit mix, nut and peach mix | 33 | 14 |
| *Monilinia fructigena* (Fr.) West. | Apples, pears | 20 | 0 |
| *Sclerotinia sclerotiorum* (Lib.) de Bary | Mangos, banana, avocados, oranges, grapes, kiwi fruits, apples, pears, sweet cherries | 47 | 0 |
| *Mycelia sterilia* | Banana, mangos, avocados, oranges, apricots, kiwi fruits, grapes, apples, pears, sweet cherriesMangos, exotic nut and fruit mix, papayas, cherries, strawberries, apricots, apples, carambolas, figs, tropic fruit mix, nut and peach mix | 40 | 61 |

These micromycetes are considered to be cosmopolitan and their substrate-specificity cannot be determined. Analysis of micromycetes on stone-fruits such as cherries, sweet cherries, plums, peaches, apricots, nectarines, etc. revealed *Ulocladium chartarum* (detection frequency 27.3%), *Penicillium italicum* (22.7%), *Aspergillus niger* (18.2%), *Alternaria alternata* (13.6%) being most frequent. However, their detection frequency often depended upon the country of origin of the sweet cherries, as well as the place and type of marketing. In sweet cherries imported from Hungary, *Ulocladium chartarum, Verticillium trifidum* (detection frequency 66.7%) prevailed; *Aspergillus niger*, *Penicillium italicum* micromycetes were more rare. In sweet cherries imported from Poland, *Geotrichum candidum* (66.7%) prevailed, *Absidia cylindroposra*, while *Penicillium granulatum* were less frequent. Sweet cherries of unknown origin sold in the market place were mostly contaminated with *Aspergillus niger*, *Geotrichum fermentans*, and *Sclerotinia sclerotiorum*

Plums imported from Hungary were damaged by *Acremonium roseolum*, *Alternaria alternata, Ulocladium chartarum, Penicillium expansum* (detection frequency <50%); *Aureobasidium prunicola*, *Pleospora infectoria* were less frequent.

Only *Paecilomyces javanicus* micromycetes were isolated from apricots imported from Spain. More severely damaged were apricots imported from France; *Cladosporium tenuissimum* developed on them forming dark delineated spots on fruit skin while *Ulocladium chartarum* covered the fruit with a dark coating. Inner tissues of apricots were damaged by *Penicillium corylophilum* and *P. damascenum*. Apricots brought from Hungary were contaminated with micromycetes of *Cladosporium*

**Table 4.** Aflatoxins detected in various imported dry fruit on sale, mg/kg$^{-1}$.

| Sample | Country | Importer | Aflatoxin B1 |
|---|---|---|---|
| Dried papaya | Thailand | JSC “Arimeksas” | 0.002 |
| Dried banana | Vietnam | JSC “Tiekėjų gildija” | 0.00275 |
| Banana chips | Philippines | JSC “Arimeksas” | 0 |

*cladosporioides* and *Penicillium italicum* species. On peaches imported from Spain, *Botrytis cinerea*, *Rhizopus stolonifer, Penicillium granulatum* and *P. italicum* (<50%) prevailed. Peaches brought from Greece were contaminated with *Aspergillus candidus, Eurotium niveoglaucum* and sometimes also with *Ascochyta pruni* and *Aureobasidium prunicola.*

It should be noted that the majority of micromycetes recorded of fruits are able to synthesize and excrete toxic secondary metabolites, which can remain during fruit drying when under the impact of high temperature the producer itself perishes.

In bananas imported from Ecuador, *Fusarium moniliforme*, *F. sporotrichioides* (detection frequency <50%), *Nectria haematococca*, *Acremonium charticola* (<30%) were recorded. Some of the mentioned fungi are characterized by the synthesis of toxic substances. For example, *Fusarium sporotrichioides* micromycetes excrete trichotecenes (deoxynivalenol, fusarin C, HT-2 toxin, T-2 toxin, zearalenon). These micromycetes can develop at low temperature, which is usually maintained in storehouses. *Fusarium moniliforme* fungi can produce various secondary metabolites; they are tolerant to an increased amount of NaCl in the medium, able to develop under condition of oxygen shortage, or even under anaerobic conditions. From bananas stored in low

**Table 5.** Mycotoxins in the imported beans with seasoning, mg/kg$^{-1}$

| Sample | Importer | Aflatoxins | Deoxy-nivalenol | T-2 toxin | Zearalenon | Ochratoxins |
|---|---|---|---|---|---|---|
| Roasted salted beans | JSC “Osterna ir Ko” | – | 0.08 | 0.1452 | 0 | 0.002 |
| Roasted beans with hot seasoning | JSC“Osterna ir Ko” | – | 0.03 | 0.0835 | 0.0118 | 0.003 |

**Table 7.** Mycotoxins in imported sunflower seeds, pumpkin seeds, poppy seeds and other products, mg/kg$^{-1}$

| Sample | Country | Importer | Aflatoxins | Deoxynivalenol | T-2 toxin | Zearalenon | Ochratoxins |
|---|---|---|---|---|---|---|---|
| Poppy seeds | Turkey | JSC “Arimeksas” | – | 0 | 0.0393 | 0.0112 | 0.0018 |
| Shelled pumpkin seeds | China | JSC “Arimeksas” | – | 0.04 | – | 0.0112 | – |
| Sesame | India | JSC “Arimeksas” | 0.0002 | – | – | – | – |
| Pumpkin seeds | Ukraine | Elčin Ariz | – | 0.04 | – | – | – |
| Roasted corn seeds | Holland | JSC “Arimeksas” | 0.0008 | 0.09 | 0.331 | 0.0116 | – |
| Sunflower seeds in shell | Hungary | JSC “Arimeksas” | 0 | – | – | 0.012 | 0.0026 |
| Desiccated coconut medium | Philippines | JSC “Arimeksas” | 0 | 0 | – | – | – |
| Shelled sunflower seeds | China | JSC “Arimeksas” | 0.0003 | – | – | 0.028 | 0.0033 |
| Exotic nut and fruit mix | Iran | | 0 | – | – | – | 0.0068 |
| Nut and raisin mix | Various countries | JSC “Arimeksas” | 0.0016 | – | – | – | – |

temperature, no aflatoxin producing micromycetes were isolated; however, analysis of dried bananas imported from Vietnam revealed traces of aflatoxins (Tab. 4).

Although the determined amounts of aflatoxin $B_1$ are small (0.00275 mg/kg$^{-1}$), if excessively used they can slowly accumulate in the organism and gradually influence functional abilities of cells.

We had no possibilities to determine the degree of contamination of fresh beans (*Phaseolus* L.) because only processed beans were brought from Chile. From samples of salted beans taken in wholesale centres, 13 micromycete species were isolated. Micromycetes of the *Penicillium* genus ascribed to *P. nigricans*, *P. expansum*, *P. brevicompactum*, *P. citrinoviride*, *P. commune*, *P. stoloniferum*, *P. paxilli* species dominated. *Fusarium nivale*, *F. moniliforme*, *Cylindrocarpon destructans*, *Trichoderma viride*, *Rhizopus oryzae*, *Mucor murorum* fungi were also recorded. Analysis of the contamination of these beans with mycotoxins revealed traces of deoxynivalenol, T-2 toxin and ochratoxin (Tab. 5).

Micromycetes of 8 species were isolated from roasted beans with lemon seasoning of unknown composition, which were brought from Chile. Among them, no producers of toxins of the trichotecenes group were recorded. However, sterile white mycelium, which could have been formed by some Fusarium species, comprised a large part (<60%) of the isolated micromycetes. In these beans, 0.03 mg/kg$^{-1}$ of deoxynivalenol, 0.0835 mg/kg$^{-1}$ of T-2 toxin, 0.0118 mg/kg$^{-1}$ of zearalenon and 0.003 mg/kg$^{-1}$ of ochratoxin were determined.

Such products should not be consumed. From the bean samples the following micromycetes were isolated: *Rhizopus oryzae*, *Mucor plumbeus*, *Aspergillus niger*, *Aspergillus ochraceus*, *Penicillium digitatum*, *P. citreoviride*, *P. expansum*, and *Mycelia sterilia*.

As the imported dried fruits were contaminated with the patulin producers such as *Penicillium expansum*, *P. claviforme*, *P. melinii*, *P. cyclopium*, *Aspergillus flavus*, and *A. terreus*, we have determined not just the degree of contamination with micromycetes, but also the amount (mg/kg$^{-1}$) of patulin in them (Tab. 6).

**Table 6.** Patulin in the imported dry fruit, mg/kg$^{-1}$.

| Sample | Country | Importer | Patulin |
|---|---|---|---|
| Dried fruit mix | Various | JSC “Tiekėjų gildija” | 0.012 |
| Dried cherries | China | JSC “Tiekėjų gildija” | 0.010 |
| Dried tropic fruit mix | Various | JSC “Tiekėjų gildija” | 0.010 |
| Raisins | Chile | JSC “Arimeksas” | 0.012 |
| Dried mangos | Thailand | JSC “Tiekėjų gildija” | 0.010 |
| Dried strawberries | China | JSC “Tiekėjų gildija” | 0.008 |

Micromycetes of the *Penicillium* genus are considered as the main producers of patulin [2, 3, 4, 5, 18, 33, 42].

Micromycetes producing toxic secondary metabolites were detected on the imported sunflower seeds, pumpkin seeds, poppy seeds and various mixes. The results obtained after analysis of the contamination of these products with the known mycotoxins are presented in Table 7.

Data presented in Table 7 indicate that traces of aflatoxin detected in sesame seeds imported from India, roasted maize seeds imported from Holland, shelled sunflower seeds imported from China, and in mixes of nuts and raisins imported from various countries.

On sesame seeds imported from India, *Aspergillus oryzae* (very close to *A. flavus* and *A. parasiticus*) dominated; *Aspergillus niger*, *Mucor hiemalis*, *Rhizomucor pusillus*, *Penicillium* sp. and *Mycelia sterilia* were also recorded. In roasted corn seeds imported from Holland no producers of aflatoxins were recorded; only *Aspergillus niger*, *Rhizopus oryzae* micromycetes and *Mycelia sterilia* were isolated. On shelled sunflower seeds brought from China only *Rhizopus oryzae* fungi and *Mycelia sterilia* were recorded. From the mixes of nuts and raisins *Rhizopus oryzae*, *Aspergillus niger*, *Penicillium digitatum*, *Monilinia* sp. and *Mycelia sterilia* were isolated. Therefore, no aflatoxin producers were recorded in the products, but the traces of mycotoxins were present. There could be a few explanations: either aflatoxin producers perish due to changing environmental conditions or under certain conditions aflatoxins can be produced by micromycetes of other species. This can also

**Table 8.** Dominant micromycetes species on the imported nuts

| Micromycete species | Detection frequency, % | Product |
|---|---|---|
| *Aspergillus niger* Tiegh. | 62 | Pistachios, almonds, glazed hazel nuts, in-shell hazel nuts, in-shell walnuts, almonds in shell, peanuts with seasoning, cedar nuts in shell |
| *Geotrichum candidum* Link ex Pers. | 23 | In-shell hazel nuts, in-shell almonds, roasted and salted peanuts |
| *Mucor hiemalis* Wehmer | 23 | Almonds, in-shell cedar nuts, peanuts in flour and starch |
| *Penicillium digitatum* Sacc. | 31 | Pistachios, glazed hazel nuts, in-shell hazel nuts, in-shell almonds |
| *Rhizopus oryzae* Went ex Prins. Geerl. | 46 | Almonds, glazed hazel nuts, in-shell hazel nuts, Brazil nuts, peanuts with seasoning, peanuts in flour and starch |
| *Rhizopus stolonifer* (Ehrenb. ex Fr.) Vuill. | 38 | Cashew nuts, peanuts without seed-coating, in-shell walnuts, Brazil nuts, in-shell cedar nuts |
| *Mycelia sterilia* | 92 | Cashew nuts, pistachios, almonds, glazed hazel nuts, in-shell hazel nuts, in-shell walnuts, in-shell almonds, Brazil nuts, salted and roasted peanuts, peanuts with seasoning, in-shell cedar nuts, peanuts in flour and starch |

**Table 9.** Aflatoxins in various imported nuts (mg kg$^{-1}$)

| Sample | Country | Importer | Aflatoxins |
|---|---|---|---|
| Shelled hazel nuts | Turkey | JSC "Tiekėjų gildija" | 0.0016 |
| In-shell cedar nuts | Russia | JSC "Faktorius" | 0.0005 |
| Brazil nuts | Bolivia | "Alvas" A. Variakovo company | 0.0002 |
| In-shell hazel nuts | Turkey | JSC "Tiekėjų gildija" | 0 |
| Pistachios | Iran | JSC "Arimeksas" | 0 |
| In-shell almonds | USA | JSC "Tiekėjų gildija" | 0.0004 |
| In-shell walnuts | Ukraine | Uelčin Arizov | 0.0105 |
| Peanuts without seed-coating | China | JSC "Arimeksas" | 0 |
| Cashew nuts | India | JSC "Arimeksas" | 0.0005 |
| Almonds | USA | JSC "Arimeksas" | 0 |
| Peanuts with onion seasoning | China | Ž. Marčiauskas | 0 |

be said about deoxynivalenol, which was found in pumpkin seeds and roasted corn seeds. This mycotoxin is produced and excreted into the substratum by micromycetes of the *Fusarium* genus, which were not recorded on the studied foodstuffs. In poppy seeds and roasted corn seeds imported from Holland, T-2 toxin was recorded: 0.0393 mg/kg$^{-1}$ and 0.331 mg/kg$^{-1}$, respectively. Kormann (1990) states that this toxin is being synthesized and excreted into the medium by *Fusarium culmorum*, *F. incormatum*, *F. poae*, *F. solani*, *F. sporotrichioides*, *F. tricinctum*, *Trichoderma viride*. However, these micromycetes were not detected in either poppy seeds or in roasted corn seeds. Micromycetes of various *Fusarium* species are considered as the main producers of zearalenon. *F. avenaceum*, *F. culmorum*, *F. equisetii*, *F. lateritium*, *F. monilifomre*, *F. nivale*, *F. oxysporum*, *F. graminearum*, *F. sambucinnum*, *F. tricinctum* can synthesize and excrete this mycotoxin. In the tested products these fungi were not recorded. In the tested foodstuffs the producers of ochratoxins were more abundant; in poppy seeds and in-shell sunflower seeds micromycetes of *Penicillium viridicatum* group and in mixes of exotic nuts and fruits *Penicillium verrucosum* and *P. digitatum* were recorded.

Nuts are abundantly imported and marketed: pistachios, almonds, hazelnuts, walnuts, peanuts, cedar, cashew, and Brazil nuts. They are often contaminated with micromycetes of various species. The analysis revealed that on imported nuts, micromycete species presented in Table 8 could be considered as dominant.

There are no producers of aflatoxins among micromycete species prevailing on nuts [34]. However, tests of their contamination revealed traces of aflatoxins on shelled hazelnuts, in-shell cedar nuts, Brazil nuts, in-shell walnuts and cashew nuts (Tab. 9).

On in-shell cedar nuts brought from Russia, in-shell almonds from the USA, in-shell walnuts from Ukraine, *Aspergillus flavus*, producers of aflatoxins, were recorded. In other nut samples these fungi were not recorded at though traces of aflatoxins were found. Together with already mentioned fungi, other micromycetes were also recorded: *Aspergillus oryzae* on cashew nuts without seed-coats imported from China, *Trichoderma viride*, *Penicillium digitatum*, *P. viridicatum* on shelled hazel nuts from Turkey. High species diversity was registered on in-shell almonds imported from the USA (*Penicillium chrysogenum*, *P. expansum*, *P. variablile*, *P. digitatum*, *P. roquefortii*, and *P. crustosum*, *Aspergillus candidus*). The majority of these micromycetes are indicated as synthesizing mycotoxins [18].

From nuts on sale in large supermarkets some less frequent micromycete species were isolated. For example, *Aspergillus restrictus* micromycetes were frequent in supermarkets. It is difficult to determine from where and with what items they had been brought because they were isolated from pumpkin seeds imported from Hungary, pistachios from Iran, hazel nuts from Georgia, figs from Turkey, and raisins from Iran. *Rhizopus oryzae* micromycetes developed on nuts, seeds and dried fruit; their detection frequency reached 13.4%. Sometimes, in-shell hazel nuts and peanuts sold in supermarkets were contaminated with *Trichotecium roseum* micromycetes. Nuts imported from Iran were contaminated with *Chaetomium aureum* which is rare in Lithuania. In-shell hazel nuts, imported from Poland, were severely contaminated, mainly with *Penicillium* fungi, still other micromycetes were also intensively developing on them: *Trichotecium roseum* (detection frequency 80%), *Aspergillus niger* (60%), other fungi (*Hirsutella saussurei*, *Stachylidium veticillatum*) were less frequent. On hazel nuts imported from Turkey *Mucor circinellloides*, *Aspergillus niger* were frequent, while on those brought from Georgia - *Aspergillus restrictus*, *Gliocladium catenulatum* and *Sporotrichum pruinosum*. *Arthroderma tuberculatum* micromycetes were isolated from in-shell walnuts imported from Poland, although here, fungi of the *Penicillium* genus dominated. Sometimes, *Acremonium pincertoniae* fungi, which are characterized by pathogenic properties, were recorded on in-shell hazel nuts and isolated from watermelons imported from Russia and were sorted in the same premises.

On coconuts imported from the Ivory Coast, *Aspergillus candidum*, *A. itaconicus*, *A. niger*, *P. granulatum*, *P. tardum*, and *Rhizoctonia solani* were recorded. Their origin is most probably diverse. *A. itaconicus* and *P. granatense* could be brought from tropical regions because they had not been isolated earlier under local conditions. *Rhizoctonia solani* most probably found their way on coconuts from potatoes stored nearby.

Micromycetes of 65 species ascribed to 23 genera were isolated from the imported dry products of plant origin. Micromycetes of the *Penicillium* (40%) and *Aspergillus* (29%) genera prevailed. Specific, imported food products, functioning as substrata, condition high species diversity. Species of the *Rhizopus* genus constituted 7% of all isolated micromycete species, those of *Gliocladium*, *Trichotecium* - 3% each, *Mucor* - 2%. The majority of the identified micromycetes are known as producers of toxic secondary metabolites. Therefore, their presence on the food products decreases the quality of food and causes a real risk to the health of consumers.

## DISCUSSION AND CONCLUSIONS

Recently, food for human consumption has been profusely supplemented by vegetables and fruits imported from various regions. This allows people to consume more valuable and variable food and, therefore, better satisfy nutrition needs. However, investigations of the mycological state of the imported foodstuffs revealed that the marketed vegetables, fresh, dried and frozen fruit are contaminated with propagules of various micromycetes. Sometimes, micromycetes develop directly on the surface of vegetables and fruit or in their inner tissues. The research demonstrated that the imported fruit marketed in Lithuania were unequally contaminated with micromycete propagules. More severely contaminated were water-melons (Spain), potatoes (Italy), kiwi fruits (India), apples (Poland, Holland), and pears (China, Poland). The vast majority of micromycetes isolated from dried fruits were ascribed to the *Penicillium* Link (44%), *Aspergillus* Link and *Mucor* Mich. ex St.-Am. (19% each) genera. Micromycetes of the *Penicillium*, *Aspergillus* and *Mucor* genera also prevailed on the imported nuts and comprised 44%, 16% and 16%, respectively, of all the isolated fungi. A rather high number of micromycete species was recorded on seasoned and unseasoned roasted beans (Chile), in-shell sunflower seeds (Hungary), sesame seeds (India), and poppy seeds (Turkey).

On the imported vegetables, *Penicillium expansum*, *P. brevicompactum*, *Rhizopus oligosporus* prevailed. On fruit, *Alternaria alternata* (40%), *A. solani* (27%), *A. pluriseptata* (20 %) dominated. A rather large portion of fungi isolated from fruit is constituted of *Penicillium* fungi (*Penicillium expansum* (67 %), *P. italicum* (40%), P. *verrucossum* (27%), which are probably able to produce toxins. White, reddish and black *Mycelia sterilia* is abundant on fruit; it sometimes comprises up to 87% of the isolates.

On the imported dried fruits *Aspergillus niger* (22%), *A. oryzae* (17%), *Rhizopus oryzae* (22%) and some species of the *Penicillium* genus (*P. expasnsum*, *P. brevicompactum*) prevailed. On various imported nuts, *Aspergillus niger* (62%), *Rhizopus oryzae* (45%), *R. stolonifer* (38%), *Mucor hiemalis* (23%) and *Geotrichum candidum* (25%) prevailed; Mycelia sterilia forms up to 32% here. The already mentioned micromycetes prevailed on the imported nuts, sunflower seeds, pumpkin seeds, poppy seeds, etc.

In some imported and marketed fruit and vegetables, traces of toxins of aflatoxin group, produced by fungi of the *Aspergillus* genus, were registered. These toxins were recorded in shelled hazel nuts, in-shell cedar nuts, Brazil nuts, in-shell almonds, in-shell walnuts, cashew nuts, dried papaya and bananas. Deoxynivalenol, T-2 toxin and ochratoxin were determined in roasted and salted beans and in beans with seasoning.

Mycotoxins are also detected in the imported and marketed sunflower seeds, pumpkin seeds, poppy seeds and other products. In poppy seeds, the amounts of T-2 toxin, zearalenon and ochratoxins were 0.00393 $mg/kg^{-1}$, 0.0112 $mg/kg^{-1}$, 0.0018 $mg/kg^{-1}$, respectively. In shelled pumpkin seeds, 0.04 $mg/kg^{-1}$ of deoxynivalenol and 0.0112 $mg/kg^{-1}$ of zearalenon; in sesame seeds, 0.0002 $mg/kg^{-1}$ of aflatoxins; in pumpkin seeds, 0.04 $mg/kg^{-1}$ of deoxynivalenol; in roasted corn seeds, 0.0003 $mg/kg^{-1}$ of aflatoxins, 0.028 $mg/kg^{-1}$ of zearalenon, 0.0039 $mg/kg^{-1}$ of ochratoxins; in exotic nut and fruit mix, 0.0068 $mg/kg^{-1}$ of ochratoxins; in nut and raisin mix, 0,0016 $mg/kg^{-1}$ of aflatoxins were determined. Patulin was detected in some dried fruits: dried fruit mix (0.012 $mg/kg^{-1}$), dried cherries (0.010 $mg/kg^{-1}$), dried tropic fruit mix (0.010 $mg/kg^{-1}$), raisins (0.012 $mg/kg^{-1}$), dried mangos (0.010 $mg/kg^{-1}$) and dried strawberries (0.008 $mg/kg^{-1}$).

The obtained results allow the conclusion that vegetables and fruit can become a good substrate for mycotoxin producing micromycetes. The micromycetes develop on everyday products and can become the cause of slow toxicoses, which are characterized by a diversity of symptoms and difficult to diagnose. Therefore, contamination of food products with micromycetes of the *Penicillium*, *Aspergillus*, *Fusarium*, *Alternaria*, *Paecilomyces*, *Trichotecium* genera should receive particular attention. It should be noted that a strain growing on a particular type of vegetable or fruit could synthesize and excrete different toxic secondary metabolites.

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## REFERENCES

1. Adebajo LO, Popoola OJ: Mycoflora and mycotoxins in kola nuts during storage. *Afr J Biotechnol* 2003, **2(10)**, 365-368.
2. Beretta B, Gaiaschi A, Galli CL, Restani P: Patulin in apple-based foods: occurrence and safety evaluation. *Food Additives Contaminants* 2000, **17**, 399-406.
3. Chu FS: Recent Studies on immunoassays for mycotoxins. In: Beier RC, Stonker LH (Eds): *Immunoassays for resides analysis: Food safety*, 294-313. American Chemical Society, Washington 1996.
4. Ciegler A, Vesonder RF, Jackson LK: Production and biological activity of patulin and citrinin from *Penicillium expansum*. *Appl Environ Microbiol* 1977, **33**, 1004-1006.
5. De Sylos CM, Rodriguez-Amaya DB: Incidence of patulin in fruits and fruit juices marketed in Campinas, Brazil. *Food Additives Contaminants* 1999, **16**, 71-74.
6. Djakow JT, Ozereckovskaja OL, Dzavahija VG, Bagirova SF: *Obsčaja i molekularnaja fitopatologija*. Obsčestvo fitopatologov, Moskva 2001.
7. Dogbevi MK, Vachon C, Lacroix M: Physiochemical Properties of Dry Red Kidney Bean proteins and Natural Microflora as Affected by Gamma Irradiation. *J Food Science* 1999, **64(3)**, 540-542.
8. Domsch K, Gams W, Anderson T: *Compendium of Soil Fungi*. Vol. 1. London 1980.

9. Fapohunda SO, Ogunderx VW: Physiology of Fungi Associated with Fast Foods in Nigeria. *Intern Biodeter* 1990, **0265-3036**, 23-32.

10. Filtenborg O, Frisvad JC, Samson RA: Specific association of fungi to food and influence of physical environmental factors. **In:** Samson RA, Hoekstra ES (Eds): *Introduction to food and airborne fungi*. 6-th Edition, 306-320, Centralbureau voor Schimmelcultures, Utrecht, An Institute of the Royal Netherlands Academy of Arts and Sciences, The Netherlands 2000.

11. Frazier CW, Westhoff D: *Food Microbiology*. 5th Edition. McGrow – Hill Book Company, New York 1988.

12. Frisvad JC: Fungal species and their specific production of mycotoxins. **In:** Samson RA, von Reenen-Hoekstra ES (Eds): *Introduction to Food-borne Fungi*. 6th Edition, 239-249. Centralbureau voor Schimmelcultures, Baor. Delff, Institute of the Royal Netherlands Academy of Arts and Sciences, The Netherlands 1988.

13. Hocking AD, Miscomble BF: Water relations of some *Zygomycetes* isolated from food. *Mycol Res* 1995, **99(9)**, 1113-1118.

14. Janchova D, Lukova L, Lebeda A, Pech E, Kutrova E: Dynamics of enzymes during pathogenesis of *Fusarium oxysporum* on pea seedlings. *Czech Mycol* 2000, **52**, 141-142.

15. Jemene M, Mateo R, Querol A, Huerta T, Hemandez E: Mycotoxins and mycotoxigenic moulds in nuts and sunflower seeds for human consumption. *Mycopathologia* 1991, **115**, 121-127.

16. Kozakiewicz Z: *Aspergillus* species on stored products. *Mycological papers* 1989, **161**, 1-188.

17. Kinderlerer JL: Czapek Cazein 50% Glucoze (CZC 50g): a new medium the identification of foodborne *Chrysosporium* spp. *Lett Appl Microbiol* 1995, **21**, 131-136.

18. Kormann K, Roth L vor, Frank H: *Giftpilze Pilzgifte Schimmelpilze: Mycotoxine Vorkommen. Inhaltsstaffe. Pilzalleguin. Nahrungsmittelvergftungen*. Landsberg am Lech: ecomed 1990.

19. Kudrjashova AA: *Mikrobiologicheskije osnovy sochranenja plodov i ovoschej*. Agropromizdat, Moskva 1986 .

20. L'vova LS, Orlova NJ, Omel'chenko VF: Gryby roda Penicillium i producenty ochratoksina A v zerne. *Prikladnaja biochimija i mikrobiologija* 1992, **28(6)**, 889-893.

21. Llewellyn C, Mooney RL, Cheatle TF, Flannigan B: Mycotoxin contamination of spices. An Update. *Inter Biodeter Biodegrad* 1992, **29**, 111-121.

22. Lugauskas A, Paškevičius A, Repečkienė J: *Patogeniški ir toksiški mikroorganizmai žmogaus aplinkoje*. Aldorija, Vilnius 2002.

23. Lugauskas A, Stakėnienė J: Mikromicetai paplitę ant sandėliuose ir prekyboje esančių vaisių ir uogų. *Ekologija* 2001, **1**, 3-11.

24. Lugauskas A, Stakėnienė J: Mikromicetai, išskirti iš sandėliuose ir prekyboje esančių daržovių. *Ekologija* 2001, **2**, 8-18.

25. Lugauskas A, Stakėnienė J: Toxin producing micromycetes on fruit, berries and vegetables. *Ann Agric Environ Med* 2002, **9**, 183-197.

26. Morin S, Magan N, Serra J, Ramos AJ, Canela R, Sonchis V: Fumonisin $B_1$ Production and Growth of *Fusarium moniliforme* and *Fusarium proliferatum* on Maize, Wheat, and Barley Grain. *J Food Science* 1999, **64(5)**, 921-924.

27. Naut MJR: Effect of *Rhizopus* and *Neurospora* spp. on growth of *Aspergillus flavus* and *A. parasiticus* and accumulation of aflatoxin $B_1$ in ground nut. *Mycol Res* 1989, **93(4)**, 518-523.

28. Nelson PE, Tauson TA, Marasas WF: *Fusarium species. An Illustrated manual for Identification*. The Pensylvania State University Press, London 1983.

29. Pitt JI: Editorial contributions to methods in food mycology. *Int J Food Microbiol* 1997, **35**, 99-101.

30. Rabie CJ, Lübben A, Marais GJ, Jonsen von Vouren H: Enumeration of fungi in barley. *Int J Food Minderol* 1997, **35**, 117-127.

31. Ramirez C: *Manual and Atlas of the Penicillia*. Amsterdam 1982.

32. Ramonytė D, Bašinskienė L: *Maisto toksikologija*. Technologija, Kaunas 2001.

33. Ross GU, Taniwaki MH, Sabino M, Vizoni T, Hirooka EY: Production of patulin in apples (*Malus domestica* Borkhausen), Gala and Fuji cultivars inoculated with *Penicillium* spp. *Ciencia e Tecnologia de Alimentos* 1998, **18(1)**, 63-67.

34. Sabino M, Milanez TV, Lamardo LC, Inomata EI, Zorzetto MA, Navas SA, Stofer M: Occurrence of aflatoxins in peanuts and peanut products consumed in the state of Sćo Paulo/Brazil from 1995 to 1997. *Revista de Microbilogia* 1999, **30**, 85-88.

35. Samson RA, Frisvad JC: *Penicillium subgenus Penicillium: new taxonomic schemes, mycotoxins and other extrolites. Studies in Mycology 49*. Centraalbureau voor Schimmel cultures, Utrecht. An institute of the Royal Netherlands Academy of Arts and Sciences, The Netherlands 2004.

36. Samson RA, Hocking AD, Pitt JL, King AD: *Modern Methods in Food Mycology*. Elsevier, Amsterdam 1992.

37. Samson RA, Hoekstra ES, Frisvad JC, Filtenborg O*: Introduction to food – and airborne fungi.* 6th Edition. Centralbureay voon Schimmelcultures, Utrecht, An Institute of the Royal Netherland Academy of Arts and Sciences. The Netherlands 2000.

38. Sharman M, MacDonald S, Sharkey AJ, Gilbert J: Sampling bulk consignments of dried figs for aflatoxin analysis. *Food Additives Contaminants* 1994, **11(1)**, 17-23.

39. Smith JE, Solomons GL (Eds): *Mycotoxin in Human Nutrition and Health. EC Directorate-General XII, Science Research and development Eur*. 16048 EN 1994.

40. Stakėnienė J, Lugauskas A, Levinskaitė L: Presence of potential toxin producing micromycetes on fruit and berries. *Bul Pol Acad Sci Biol Sci* 2001, **49(4)**, 413-420.

41. Strom G, Blomquist G: Airborne spores from mouldy citrus fruit – a potential occupational health hazard. *Ann Occup Hyg* 1986, **30**, 455-460.

42. Viñas I, Dadon J, Sanchis V: Citrinin-producing capacity of *Penicillium expansum* strains from apple packinghouses of Lerida (Spain). *Int J Food Microbiol* 1993, **19**, 153-156.

43. Warling PW: Incidence and detection of thermotolerant and thermophilic fungi from maize with particular reference to *Thermoascus* species. *Int J Food Microbiol* 1997, **35**, 137-145.

44. Wei DL, Jang SC: Production of aflatoxins by strains of the Aspergillus flavus group maintained in ATGC. *Mycopathologia* 1986, **94**, 19-24.

45. Zakarjan AA, Osipjan LL: Toksikologičeskaja charakteristika mikromicetov – kontaminantov nesteril'nych rastitel'nych lekarstvenych preparatov (rody Aspergillus i Penicillium). *Mikologija i fitopatologija* 1996, **30(5-6)**, 51-54.