Occurrence of moulds and ochratoxin A in dried fruits and vegetables from the Serbian market

SANDRA BULUT – GORDANA DIMIĆ – SONJA SMOLE MOŽINA – MIHAEL ČARGO – IRENA RAKIĆ – JELENA ŽIVANČEV – IGOR ANTIĆ – SUNČICA KOCIĆ-TANACKOV

Summary

Various dried fruits and vegetables (raspberries, cherries, grapes, blueberries, apricots, cranberries, figs, plums, pears, apples, tomatoes, mixed vegetables), with and without surface disinfection, were analysed for the presence of moulds and ochratoxin A. Dried figs were the only ones in which moulds were not detected. In other samples, contamination by moulds was reduced after surface disinfection, while internalized contamination remained. Species observed in dried fruits were from the genera *Aspergillus*, *Cladosporium*, *Emericella*, *Eupenicillium*, *Eurotium*, *Monilia*, *Mucor*, *Penicillium*, *Rhizopus*, *Talaromyces*, *Trichoderma* and *Xeromyces*, and in vegetables were *Aspergillus*, *Penicillium* and *Rhizopus*. Among the dominant genera and species in dried fruits were *Penicillium*, which represented 24.5 % of all isolates, *Aspergillus* (22.3 %) and *Rhizopus* (19.2 %), with *P. glabrum*, *A. niger* and *Rh. oryzae* as the most common species. The dominant genera and species in dried vegetables were *Penicillium* (6.4 %) and *Rhizopus* (5.3 %), with *P. glabrum*, *Rh. microspores* and *Rh. oligosporus*, as the most common species. Potential ochratoxin A producers found in this study were *A. niger* and *P. verrucosum*. However, the results of immunochemical analysis by ELISA indicated that the content of ochratoxin A in all samples was lower than 0.1 $\mu g \cdot kg^{-1}$.

Keywords

mould; ochratoxin A; contamination; dried fruits; dried vegetables

Fresh fruits and vegetables, due to the high water content, are susceptible to microbiological spoilage. However, pH of fruits ranges from 2.5 to 4.5 and this low pH protects them from bacteria [1, 2]. Therefore, the most common fruit spoilage agents are yeasts and in particular moulds, which are more tolerant to acidic environment [1]. Regarding vegetables, many of them have higher, near-neutral pH, ranging from 5.1 to 6.8. Therefore, they may be susceptible to bacterial spoilage, but moulds represent also important spoilage agents of vegetables [1, 3].

One of the most common methods to protect food from microbiological spoilage and increase its shelf life, especially fruits and vegetables, is drying. Dried fruits and vegetables have a significant nutritional value and economic importance due to large production [4]. Drying presents one of the oldest ways of preserving food. Reducing water activity value slows down the growth and reproduction of microorganisms, while reducing it further may completely prevent their growth and reproduction. With a reduction of water activity, the number of spoilage microorganisms also decreases, and when it comes to dried fruits and vegetables, this may restrict the spoilage flora to xerophilic and xerotolerant moulds [1].

Since contamination and spoilage by moulds is characteristic for dried fruits, examination for the presence of mycotoxins is of great importance. Barkai-Golan and Paster [5] reported that the presence of mycotoxins presents a bigger

Sandra Bulut, Gordana Dimić, Sunčica Kocić-Tanackov, Department of Food Preservation Engineering, Faculty of Technology Novi Sad, University of Novi Sad, Bulevar cara Lazara 1, 21000 Novi Sad, Serbia.
Sonja Smole Možina, Mihael Čargo, Department of Food Science and Technology, Biotechnical Faculty, University of Ljubljana, Jamnikarjeva 101, 1000 Ljubljana, Slovenia.
Irena Rakić, Department of Applied and Engineering Chemistry, Faculty of Technology Novi Sad, University of Novi Sad, Bulevar cara Lazara 1, 21000 Novi Sad, Serbia.
Jelena Živančev, Igor Antić, Department of Oil and Petrochemical Engineering, Faculty of Technology Novi Sad, University of Novi Sad, Bulevar cara Lazara 1, 21000 Novi Sad, Serbia.

Correspondence author:
Sunčica Kocić-Tanackov, e-mail: suncicat@uns.ac.rs

problem in dried fruits compared to fresh fruits. Mycotoxins are secondary metabolites of moulds belonging to genera *Aspergillus*, *Penicillium*, *Alternaria* and *Fusarium* [6]. The first three are the major contributors of mycotoxin production in fruits [7]. However, toxinogenic moulds characteristic for dried fruits and vegetables are usually *Aspergillus* spp. and *Penicillium* spp., probably due to the need for higher substrate moisture in the case of *Alternaria* spp. and *Fusarium* spp. The most common mycotoxins associated with dried fruits and vegetables are aflatoxins and ochratoxins. Products contaminated with those mycotoxins may cause health hazards and huge economic losses.

Ochratoxins are secondary metabolites produced by three main mould species: *A. ochraceus*, *P. verrucosum*, and *Aspergillus* section Nigri, especially *A. carbonarius* [4]. Several different ochratoxins exist, but ochratoxin A is the most common, important and widespread [2]. It was shown to cause various adverse effects on human health (carcinogenic, nephrotoxic, immunosuppressive, teratogenic and genotoxic). However, few mycotoxins in dried fruits are regulated by Serbian and EU regulations [8, 9]. The presence of ochratoxin A in dried vine fruit (currants, raisins and sultanas) is limited by these regulations to 10 $\mu g \cdot kg^{-1}$, while there is no regulation in EU and Serbia for other dried fruits.

In this work, various dried fruits and vegetables purchased at local markets and in health-promoting food stores in Novi Sad, Serbia, were examined for the presence of moulds and ochratoxin A.

MATERIALS AND METHODS

Samples

Twenty-six samples of 12 dried fruits and vegetables were analysed, namely, raspberries, cherries, grapes, blueberries, apricots, cranberries, figs, plums, pears, apples, tomatoes, mixed vegetables, were purchased at local markets and health-promoting food stores in Novi Sad, Serbia. Ten incremental samples were collected to obtain an aggregate sample of 1 kg total weight, for every sample. Samples were properly labeled, sealed and kept in clean polyethylene bags in refrigerator until use (4 °C, up to 5 days).

Chemicals

Sabouraud maltose agar and Czapek's agar were obtained from HiMedia (Mumbai, India). Sodium hypochlorite, sodium bicarbonate and chloramphenicol were obtained from Sigma-Aldrich (St. Louis, Missouri, USA). HCl was obtained from Alfapanon (Bački Petrovac, Serbia) and dichloromethane from PanReac AppliChem (ITW Reagents, Barcelona, Spain). All other reagents used in this study were of analytical grade.

Isolation and quantification of moulds

Isolation and quantification of moulds in samples was done under aseptic conditions, based on two different methods, depending on the sample. Samples were analysed without surface disinfection and with surface disinfection achieved by immersion to 0.3% sodium hypochlorite for 2 min with subsequent washing with sterile distilled water. The latter approach allowed quantification of internal contamination.

Direct plating method

In direct plating method, samples were placed directly on solidified agar media for detecting, enumerating and isolating fungi [3]. From each sample, 6 to 9 whole or 1/4 dried fruits were directly put on the surface of Sabouraud maltose agar. Antibiotic chloramphenicol was added to agar to inhibit the growth of bacteria. Dried fruits that were examined in this way were raspberries, cherries, grapes, blueberries, cranberries (all whole fruits), apricots and figs (quarters of fruits from both). Plates were incubated for 5–7 days at 25 °C, the colonies were counted and results were expressed as mean values in colony forming units per gram.

Dilution method

The dilution method was used for examination of cut-sliced fruits and vegetables, such as dried plums, pears, apples, tomatoes and mixed vegetables. An amount of 10 g of sample was chopped and then mixed with 90 ml of 0.8% NaCl in an Erlenmeyer flask, homogenized for 15 min, at 3 Hz with the homogenizer (Unimax 1010, Heidolph, Schwabach, Germany). Series of decimal dilutions were prepared and one millilitre was transferred into Petri dishes, into which Sabouraud maltose agar with chloramphenicol was poured. Plates were incubated for 5–7 days at 25 °C. The colonies were counted and expressed as mean values in colony forming units per gram.

Identification of moulds

Based on their macromorphological properties, moulds were transmitted to an appropriate agar to obtain pure culture. Colonies assumed to belong to groups Zygomycetes or Dematiaceae were subcultured on Sabouraud maltose agar. Colonies assumed to belong to groups Asco-

mycetes or Deuteromycetes were subcultured on Czapek's agar. The plates were incubated at 25 °C for 7–10 days and then, identification of moulds was done based on their microscopic characteristics and macroscopic characteristics of the colonies. Macroscopic characteristics implied the colony diameter, colour, texture, pigmentation and reverse colour of colony. Under microscope, morphological characteristics, namely, metules, phialides, conidia and their length, diameter, size and shape were observed. Identification was done based on criteria described by KLICH [10], SAMSON et al. [11], SAMSON and FRISVAD [12], PITT and HOCKING [3].

The mycological profile of dried fruits and vegetables collected in this study included isolation of 94 mould strains. The share (S) of certain genus and species of moulds was calculated according Eq. 1:

$$S = \frac{A}{B} \times 100 \quad (1)$$

where A is number of isolates of a genus or species and B is total number of isolates of all genera or species.

Immunochemical analysis

Ochratoxin A in analysed samples was detected using enzyme-linked immunosorbent assay (ELISA). Samples were ground by A11 basic beater (model A11BS000, IKA-Werke, Staufen, Germany) that reduces soft, medium-hard, and brittle materials with a Mohs' hardness of up to 6. The used granularity of the feeding material was not greater than 10 mm in diameter with the appropriate amount of feed material placed into the container. The applied blending time was 45 s, at 40 Hz. An amount of 5 g of finely ground sample was mixed with 15 ml of 1 mol·l^{-1} HCl and 30 ml of dichloromethane, which was then shaken for 15 min on a low-speed shaker at 7 Hz (VELP Scientifica, Usmate Velate, Italy). After separating the liquid from the solid phase, 5 ml of the lower dichloromethane phase was transferred to a tube and 5 ml of sodium bicarbonate (0.13 mol·l^{-1}) was added and shaken for 15 min with a low-speed shaker at 7 Hz. The sample was then centrifuged at 2 200 ×g for 15 min. A volume of 150 μl of the upper aqueous phase was taken and diluted with 350 μl of sodium bicarbonate (0.13 mol·l^{-1}). For determination was done using l'screen Ochratoxin ELISA kit (Tecna, Trieste, Italy) and the analyses were performed according to the test kit instructions. Absorbance at 450 nm was read using a microplate reader Multiskan EX (Thermo Fisher Scientific, Waltham, Massachusetts, USA).

RESULTS AND DISCUSSION

Quantitative data on moulds isolated from dried fruits and vegetables are presented in Tab. 1. By direct plating method, total numbers of moulds in samples without surface disinfection ranged from 0 CFU·g^{-1} (apricots, figs) to 0.62 CFU·g^{-1} (grapes), and from 0 CFU·g^{-1} (blueberries, figs) to 0.14 CFU·g^{-1} (cranberries) in samples with surface disinfection. By dilution method, total numbers of moulds in samples without surface disinfection ranged from 10 CFU·g^{-1} (mixed vegetables) to 170 CFU·g^{-1} (apples), and from 5 CFU·g^{-1} (mixed vegetables) to 90 CFU·g^{-1} (apples) in samples with surface disinfection. In almost all samples, the decrease of total numbers of moulds after surface disinfection was observed. Relatively high numbers of moulds after surface disinfection were recorded for dried apricots, which was probably due to sample heterogenity. Mould contamination after surface disinfection was reduced by values from 47 % (apples) to 100 % (blueberries). Dried figs were the only ones in which the presence of moulds was not detected. These results suggest that surface is the main site of contamination by moulds in most dried fruits.

ABBAS et al. [13] analysed the presence of moulds in 84 dried fruit and nut samples without surface disinfection (walnut, almond, apricot kernel, cashew nut, coconut, pine nut, peanut, pistachio, prunes, dry dates, fig, mulberry, raisins, apricot) from Karachi, Pakistan, by using dilution

Tab. 1. Total counts of moulds before and after surface disinfection of dried fruits and vegetables.

Dried fruit/vegetable	Total counts of moulds [CFU·g^{-1}]	
	Before surface disinfection	After surface disinfection
Direct-plating method		
Raspberries	0.15	0.07
Cherries	0.42	0.09
Grapes	0.62	0.11
Blueberries	0.10	0.00
Apricots	0.00	0.02
Cranberries	0.37	0.14
Figs	0.00	0.00
Dilution method		
Plums	60	30
Pears	85	10
Apples	170	90
Tomatoes	55	10
Mixed vegetables	10	5

Tab. 2. Share of genera and species of moulds found in dried fruits and vegetables.

Genus	Genus share [%]		Species	Species share [%]	
	Dried fruits	Dried vegetables		Dried fruits	Dried vegetables
Aspergillus	22.3	2.1	*A. niger*	17.0	1.1
			A. niveus	1.1	0.0
			A. penicilloides	2.1	0.0
			A. ustus	1.1	0.0
			A. versicolor	1.1	1.1
Cladosporium	1.1	0.0	*C. sphaerospermum*	1.1	0.0
Emericella	3.2	0.0	*E. nidulans*	1.1	0.0
			E. rugulosa	3.2	0.0
Eupenicillium	5.3	0.0	*Eu. javanicum*	1.1	0.0
			Eu. alutaceum	4.3	0.0
Eurotium	1.1	0.0	*E. rubrum*	1.1	0.0
Monilia	2.1	0.0	*M. sitophilia*	2.1	0.0
Mucor	3.2	0.0	*M. racemosus*	3.2	0.0
Penicillium	24.5	6.4	*P. aurantiogriseum*	0.0	1.1
			P. citreonigrum	1.1	0.0
			P. corylophilum	2.1	1.1
			P. decumbens	1.1	1.1
			P. fellutanum	1.1	0.0
			P. glabrum	11.7	2.1
			P. nalgiovense	2.1	0.0
			P. oxalicum	0.0	1.1
			P. purpurogenum	1.1	0.0
			P. restrictum	1.1	0.0
			P. simplicissimum	1.1	0.0
			P. thomii	1.1	0.0
			P. verrucosum	1.1	0.0
Rhizopus	19.2	5.3	*Rh. microsporus*	5.3	2.1
			Rh. oligosporus	3.2	2.1
			Rh. oryzae	7.5	1.1
			Rh. stolonifer	3.2	0.0
Talaromyces	1.1	0.0	*T. trachyspermus*	1.1	0.0
Trichoderma	2.1	0.0	*T. harzianum*	2.1	0.0
Xeromyces	1.1	0.0	*X. bisporus*	1.1	0.0

method for mycological analysis. According to the obtained results, among the dried fruit samples, raisins and apricots showed the highest contamination by moulds (60 CFU·g^{-1}). Total numbers of moulds in dried plums (prunes) were reported to be 30 CFU·g^{-1}, much lower compared to results obtained in this research (60 CFU·g^{-1}).

We found in dried fruits genera *Aspergillus*, *Cladosporium*, *Emericella*, *Eupenicillium*, *Eurotium*, *Monilia*, *Mucor*, *Penicillium*, *Rhizopus*, *Talaromyces*, *Trichoderma* and *Xeromyces*, while genera found in dried vegetables were *Aspergillus*, *Penicillium* and *Rhizopus* (Tab. 2). The dominant mould genus in dried fruits was *Penicillium*, which represented 24.5 % of all isolates, with *P. glabrum* as the most common species (11.7 %). The dominant genus in dried vegetables was *Penicillium* (6.4 %) with *P. glabrum* (2.1 %) as the most common species. *Penicillium* and *Aspergillus* species are widespread in nature and among the most common airborne moulds, being frequent food contaminants [7]. These results are in line

Tab. 3. Share of genera and species of moulds in dried fruits and vegetables before surface disinfection.

Sample	Genus	Share [%]	Species	Share [%]
Raspberries	*Aspergillus*	100.0	*A. niger*	100.0
Cherries	*Aspergillus*	70.0	*A. niger*	40.0
			A. penicilloides	20.0
			A. niveus	10.0
	Emericella	10.0	*E. rugulosa*	10.0
	Penicillium	10.0	*P. purpurogenum*	10.0
	Rhizopus	10.0	*Rh. oryzae*	10.0
Grapes	*Aspergillus*	66.7	*A. niger*	66.7
	Penicillium	16.7	*P. corylophilum*	16.7
	Rhizopus	16.7	*Rh. stolonifer*	16.7
Blueberries	*Aspergillus*	50.0	*A. ustus*	50.0
	Penicillium	50.0	*P. simplicissimum*	50.0
Apricots	–	–	–	–
Cranberries	*Aspergillus*	60.0	*A. niger*	60.0
	Penicillium	40.0	*P. glabrum*	20.0
			P. verrucosum	20.0
Plums	*Aspergillus*	14.3	*A. niger*	14.3
	Penicillium	28.6	*P. glabrum*	28.6
	Rhizopus	14.3	*Rh. oligosporus*	14.3
	Trichoderma	28.6	*T. harzianum*	28.6
	Monilia	14.3	*M. sitophilia*	14.3
Pears	*Rhizopus*	72.7	*Rh. microspores*	36.4
			Rh. oligosporus	18.2
			Rh. oryzae	9.1
			Rh. stolonifer	9.1
	Talaromyces	9.1	*T. trachyspermus*	9.1
	Monilia	9.1	*M. sitophilia*	9.1
	Xeromyces	9.1	*X. bisporus*	9.1
Apples	*Aspergillus*	9.1	*A. versicolor*	9.1
	Cladosporium	9.1	*C. sphaerospermum*	9.1
	Eupenicillium	9.1	*Eu. alutaceum*	9.1
	Penicillium	36.4	*P. glabrum*	18.2
			P. nalgiovense	9.1
			P. corylophilum	9.1
	Rhizopus	27.3	*Rh. oryzae*	27.3
	Mucor	9.1	*M. racemosus*	9.1
Tomatoes	*Aspergillus*	33.3	*A. niger*	16.7
			A. versicolor	16.7
	Penicillium	66.7	*P. oxalicum*	16.7
			P. corylophilum	16.7
			P. glabrum	16.7
			P. decumbens	16.7
Mixed vegetables	*Penicillium*	25.0	*P. glabrum*	25.0
	Rhizopus	75.0	*Rh. oryzae*	25.0
			Rh. oligosporus	25.0
			Rh. microsporus	25.0

with current knowledge in the field as many literature data reported *Aspergillus* and *Penicillium* as the most common moulds in dried fruits and vegetables, with *A. niger* as the most frequently isolated from dried fruits [13–16]. The large presence of *A. niger* in dried fruits and vegetables is not surprising as this mould is able to grow in a wide range of temperatures, low water activity and it is adapted to a hot and humid environment. Furthermore, during drying, sugars in the fruits become more concentrated, leading to even more selective pressure in favour of black aspergilli [2, 5, 17].

In Tab. 3, shares of genera and species in dried fruits and vegetables before surface disinfection are presented. In Tab. 4, shares of genera and species in dried fruits and vegetables after surface disinfection are presented. The latter data illustrate the levels of internal contamination.

The most common genera isolated from dried fruits and vegetables before surface disinfection were *Aspergillus*, *Penicillium* and *Rhizopus*. In dried raspberries, cherries, grapes and cranberries the dominant genus was *Aspergillus* with the share of 100.0 %, 70.0 %, 66.7 % and 60.0 %, respectively. Isolated species of the genus *Aspergillus* were *A. niger* (dried cherries, grapes and cranberries), *A. penicilloides*, *A. niveus* (dried cherries), and *A. ustus* (dried blueberries). *Penicillium* spp. were presented in the highest percentage in dried tomatoes, cranberries and blueberries, with the share of 66.7 %, 40.0 % and 50.0 %, respectively. Isolated *Penicillium* spp. were *P. oxalicum*, *P. corylophilum*,

Tab. 4. Share of genera and species of moulds in dried fruits and vegetables after surface disinfection.

Sample	Genus	Share [%]	Species	Share [%]
Raspberries	*Penicillium*	100.0	*P. decumbens*	100.0
Cherries	*Aspergillus*	33.3	*A. niger*	33.3
	Eurotium	33.3	*E. rubrum*	33.3
	Penicillium	33.3	*P. glabrum*	33.3
Grapes	*Penicillium*	100.0	*P. glabrum*	100.0
Blueberries	–	–	–	–
Apricots	*Penicillium*	100.0	*P. glabrum*	100.0
Cranberries	*Penicillium*	75.0	*P. glabrum*	25.0
			P. citreonigrum	25.0
			P. thomii	25.0
	Eupenicillium	25.0	*Eu. alutaceum*	25.0
Plums	*Penicillium*	33.3	*P. restrictum*	33.3
	Emericella	33.3	*E. nidulans*	33.3
	Rhizopus	33.3	*Rh. microsporus*	33.3
Pears	*Aspergillus*	20.0	*A. niger*	20.0
	Emericella	20.0	*E. nidulans*	20.0
	Eupenicillium	20.0	*Eu. alutaceum*	20.0
	Penicillium	40.0	*P. glabrum*	20.0
			P. fellutanum	20.0
Apples	*Eupenicillium*	22.2	*Eu. alutaceum*	11.1
			Eu. javanicum	11.1
	Penicillium	22.2	*P. glabrum*	11.1
			P. nalgiovense	11.1
	Rhizopus	33.3	*Rh. oryzae*	22.2
			Rh. stolonifer	11.1
	Mucor	22.2	*M. racemosus*	22.2
Tomatoes	*Penicillium*	100.0	*P. aurantiogriseum*	100.0
Mixed vegetables	*Rhizopus*	100.0	*Rh. oligosporus*	50.0
			Rh. microsporus	50.0

Tab. 5. Share of toxinogenic moulds isolated from dried fruits and vegetables and their toxins [3, 10–12].

Mould species	Toxins	Species share [%]
Aspergillus niger	Naphto-γ-pyrones, malformins, ochratoxin A	18.1
Aspergillus versicolor	Sterigmatocystin, nidulotoxin	2.1
Emericella nidulans	Sterigmatocystin, emestrin	2.1
Emericella rugulosa	Sterigmatocystin	3.2
Penicillium aurantiogriseum	Penicillic acid, verrucosidin, nephrotoxic glyopeptides, anacine, auranthin, aurantiamin	1.1
Penicillium citreonigrum	Citreoviridin	3.2
Penicillium glabrum	Citromycetin	13.8
Penicillium nalgiovense	Penicillin	2.1
Penicillium oxalicum	Secalonic acid D	1.1
P. simplicissimum	Verruculogen, fumitremorgen B, penicillic acid, viridicatumtoxin	1.1
P. verrucosum	Ochratoxin A, citrinin, verrucolone, verrucines	1.1

P. decumbens (dried tomatoes), *P. glabrum* (dried tomatoes and cranberries), *P. verrucosum* (dried cranberries), and *P. simplicissimum* (dried blueberries). In dried raspberries, the only species that was identified before surface disinfection was *A. niger*, while *P. decumbens* was the only identified specie after surface disinfection. This means that *A. niger* contaminated the surface and *P. decumbens* the internal tissue of raspberries. *Rhizopus* spp. were the most presented in dried mixed vegetables (75.0 %), and in dried pears (72.7 %), with isolated species *Rh. microspores*, *Rh. oligosporus*, *Rh. oryzae* (dried mixed vegetables and pears) and *Rh. stolonifer* (dried pears). *E. rugulosa* was isolated only from dried cherries, *X. bisporus* and *T. trachyspermus* from dried pears, *C. sphaerospermum* and *M. racemosus* from dried apples, *T. harzianum* from dried plums, while *M. sitophilia* was found in dried pears and plums.

After surface disinfection, only in dried blueberries no moulds were found, while in other samples, species from the genera *Aspergillus*, *Emericella*, *Eupenicillium*, *Eurotium*, *Mucor, Penicillium* and *Rhizopus* were isolated.

During the growth on fruits and vegetables, several *Aspergillus*, *Penicillium* and other species can produce various mycotoxins that are harmful to humans and animals. Barkai-Golan and Paster [5] suggested that dried fruits represent a bigger problem when it comes to mycotoxins compared to fresh fruits, so it can be assumed that the drying process is of great importance. Fruits or vegetables must be of high quality and the process of drying must start immediately after harvest [5, 18]. Major mycotoxins associated with toxigenic species of the genera *Aspergillus*, *Penicillium* and *Emericella* [3, 10–12] isolated from dried fruits and vegetables in this study, and the share of these moulds in the total mycopopulation are shown in Tab. 5. According to the results, toxinogenic moulds that produce aflatoxins were not identified, while *A. niger*, a potential producer of ochratoxin A, was present in the largest percentage, with a share of 18.1 % in the total mycopopulation. *A. niger* has been confirmed as a producer of ochratoxin A, but to a much lesser extent compared to well established producers of this mycotoxin *A. ochraceus* and *A. carbonarius* [5, 16, 19]. *P. verrucosum*, with the share of 1.1 % in the total mycopopulation, is considered the most important ochratoxin A producer of the genus *Penicillium* [3]. The possibility of ochratoxin A presence in the samples was examined but the results showed that the content of ochratoxin A in all samples of dried fruits and vegetables was lower than 0.1 $\mu g \cdot kg^{-1}$ (data not shown).

The presence of ochratoxin A was reported in many dried fruits, such as figs in the range of 0.37–7.86 $\mu g \cdot kg^{-1}$ [20], 0.87–24.37 $\mu g \cdot kg^{-1}$ [21], and 0.1–30.0 $\mu g \cdot kg^{-1}$ [16], sultanas in the range of 0.51–58.04 $\mu g \cdot kg^{-1}$ [21], white and black sultanas in the range of 0.1–5.0 $\mu g \cdot kg^{-1}$ and from 0.1 $\mu g \cdot kg^{-1}$ to > 30 $\mu g \cdot kg^{-1}$ [16], plums in levels up to 5.0 $\mu g \cdot kg^{-1}$ [16, 22], dates in levels up to 5.0 $\mu g \cdot kg^{-1}$ [16]. However, contamination of products with potentially toxinogenic moulds does not always imply the presence of their toxins [14]. It is known that the most common factors that influence mycotoxin production are temperature, moisture content or water activity, food substrate and genetic potential of the mould [14, 23]. Further such factors are harvesting and drying conditions [2] and environ-

mental stress conditions such as insect infestation, drought, cultivar susceptibility, mechanical damage, nutritional deficiencies, and unseasonable temperature, rainfall or humidity [6].

CONCLUSIONS

Results of this study showed that among all analysed dried fruits and vegetables (raspberries, cherries, grapes, blueberries, apricots, cranberries, figs, plums, pears, apples, tomatoes, mixed vegetables), only dried figs did not show the presence of moulds.

In direct-plating method, the total numbers of moulds ranged from absence (apricots) to 0.62 CFU·g^{-1} (grapes) and, after surface disinfection, the percentage of reduction was up to 100 % (dried blueberries). In dilution method, the total numbers of moulds ranged from 10 CFU·g^{-1} (mixed vegetables) to 170 CFU·g^{-1} (apples) and the reduction after surface disinfection was up to 82 % (dried tomatoes).

Mycopopulation genera found in dried fruits were *Aspergillus*, *Cladosporium*, *Emericella*, *Eupenicillium*, *Eurotium*, *Monilia*, *Mucor*, *Penicillium*, *Rhizopus*, *Talaromyces*, *Trichoderma* and *Xeromyces*, while genera found in dried vegetables were *Aspergillus*, *Penicillium* and *Rhizopus*. The dominant mould genera in dried fruits were *Penicillium*, *Aspergillus* and *Rhizopus*, with *A. niger*, *P. glabrum* and *Rh. oryzae* as the most common species. The dominant mould genera in dried vegetables were *Penicillium* and *Rhizopus*, with *P. glabrum*, *Rh. microspores* and *Rh. oligosporus*, as the most common species. Potential ochratoxin A producers found in this study were *A. niger* and *P. verrucosum*. However, the results of immunochemical analysis indicated that the content of ochratoxin A in all samples was lower than 0.1 μg·kg^{-1} (limit of detection). Further research should include examination for the presence of some other mycotoxin characteristic for dried fruits and vegetables, such as aflatoxins.

Acknowledgements

This work was supported by the Provincial Secretariat for Higher Education and Scientific Research, Republic of Serbia, Autonomous Province of Vojvodina (Project No. 142-451-2623/2021-01) and by the Project "Mycotoxigenic moulds and mycotoxins in raw materials and products intended for human consumption", supported by Matica Srpska, Novi Sad, Serbia.

REFERENCES

1. Adams, M. R. – Moss, M. O.: Food Microbiology. 3rd ed. Cambridge : RSC Publishing, 2008. ISBN: 978-0-85404-284-5. DOI: 10.1039/9781847557940.
2. Drusch, S. – Ragab, W.: Mycotoxins in fruits, fruit juices, and dried fruits. Journal of Food Protection, *66*, 2003, pp. 1514–1527. DOI: 10.4315/0362-028x-66.8.1514.
3. Pitt, J. I. – Hocking, A. D.: Fungi and food spoilage. London : Blackie Academic, 2009. ISBN: 978-0-387-92207-2. DOI: 10.1007/978-0-387-92207-2.
4. Karaca, H. – Velioglu, Y. S. – Nas, S.: Mycotoxins: contamination of dried fruits and degradation by ozone. Toxin Reviews, *29*, 2010, pp. 51–59. DOI: 10.3109/15569543.2010.485714.
5. Barkai-Golan, R. – Paster, N.: Mycotoxins in fruits and vegetables. London : Academic Press, 2008. ISBN: 978-0-12-374126-4. DOI: 10.1016/B978-0-12-374126-4.X0001-0.
6. Fernández-Cruz, M. L. – Mansilla, M. L. – Tadeo, J. L.: Mycotoxins in fruits and their processed products: Analysis, occurrence and health implications. Journal of Advanced Research, *1*, 2010, pp. 113–122. DOI: 10.1016/j.jare.2010.03.002.
7. Barkai-Golan, R. – Paster, N.: Mouldy fruits and vegetables as a source of mycotoxins: part 1. World Mycotoxin Journal, *1*, 2008, pp. 147–159. DOI: 10.3920/WMJ2008.x018.
8. Commission regulation (EC) No 1881/2006 of 19 December 2006 setting maximum levels for certain contaminants in foodstuffs. Official Journal of the European Union, *49*, 2006, L364, pp. 5–24. ISSN: 1725-2555. <https://eur-lex.europa.eu/legal-content/EN/ALL/?uri=celex%3A32006R1881>
9. Pravilnik o maksimalnim koncentracijama određenih kontaminenata u hrani. (Rulebook on maximum concentrations of certain contaminants in food.). Službeni glasnik Republike Srbije, *75*, 2019, 81, pp. 17–32. ISSN: 0353-8389. <https://www.pravno-informacioni-sistem.rs/SlGlasnikPortal/eli/rep/sgrs/ministarstva/pravilnik/2019/81/1/reg> In Serbian.
10. Klich, A. M.: Identification of common *Aspergillus* species. Utrecht : Centraalbureau vor Schimmelcultures, 2002. ISBN: 9789070351465.
11. Samson, A. R. – Hoekstra, S. E. – Frisvad, C. J.: Introduction to food and airborne fungi. Utrecht : Centraalbureau vor Schimmelcultures, 2004. ISBN: 9789070351526.
12. Samson, A. R. – Frisvad, C. J.: *Penicillium* subgenus *Penicillium*: new taxonomic schemes, mycotoxins and other extrolites. Utrecht : Centraalbureau vor Schimmelcultures, 2004. ISBN: 90-70351-53-6.
13. Abbas, M. – Naz, S. A. – Shafique, M. – Jabeen, N. – Abbas, S.: Fungal contamination in dried fruits and nuts: a possible source of mycoses and mycotoxicoses. Pakistan Journal of Botany, *51*, 2019, pp. 1523–1529. DOI: 10.30848/PJB2019-4(31).
14. Alhussaini, M. S.: Mycobiota and mycotoxins of nuts and some dried fruits from Saudi Arabia. Journal of American Science, *8*, 2012, pp. 525–534. DOI: 10.7537/marsjas081212.73.

15. Saleh, I. – Al-Thani, R.: Fungal food spoilage of supermarkets' displayed fruits. Veterinary World, *12*, 2019, pp. 1877–1883. DOI: 10.14202/vetworld.2019.1877-1883.
16. Iamanaka, B. T. – Taniwaki, M. H. – Menezes, H. C. – Vicente, E. – Fungaro, M. H. P.: Incidence of toxigenic fungi and ochratoxin A in dried fruits sold in Brazil. Food Additives and Contaminants, *22*, 2005, pp. 1258–1263. DOI: 10.1080/02652030500260447.
17. Suleiman, M. S. – Nuntah, L. C. – Muhammad, H. L. – Mailafiya, S. C. – Makun, H. A. – Saidu, A. N. – Apeh, D. O. – Iheanacho, H. E.: Fungi and aflatoxin occurrence in fresh and dried vegetables marketed in Minna, Niger State, Nigeria. Journal of Plant Biochemistry and Physiology, *5*, 2017, pp. 176. DOI: 10.4172/2329-9029.1000176.
18. Magan, N. – Aldred, D.: Conditions of formation of ochratoxin A in drying, transport and in different commodities. Food Additives and Contaminants, *22*, 2005, pp. 10–16. DOI: 10.1080/02652030500412154.
19. Romero, S. M. – Comerio, R. M. – Larumbe, G. – Ritieni, A. – Vaamonde, G. – Fernández Pinto, V.: Toxigenic fungi isolated from dried vine fruits in Argentina. International Journal of Food Microbiology, *104*, 2005, pp. 43–49. DOI: 10.1016/j.ijfoodmicro.2005.04.001.
20. Bayman, P. – Baker, J. L. – Doster, M. A. – Michailides, T. J. – Mahony, N. E.: Ochratoxin production by the *Aspergillus ochraceus* group and *Aspergillus alliaceus*. Applied and Environmental Microbiology, *68*, 2002, pp. 2326–2329. DOI: 10.1128/AEM.68.5.2326-2329.2002.
21. Bircan, C.: Incidence of ochratoxin A in dried fruits and co-occurrence with aflatoxins in dried figs. Food and Chemical Toxicology, *47*, 2009, pp. 1996–2001. DOI: 10.1016/j.fct.2009.05.008.
22. Zohri, A. A. – Ardel-Gawad, K. M.: Survey of mycoflora and mycotoxins of some dried fruits in Egypt. Journal of Basic Microbiology, *33*, 1993, pp. 279–288. DOI: 10.1002/jobm.3620330413.
23. Tajkarimi, M. – Shojaee, M. H. – Yazdanpanah, H. – Ibrahim, S. A.: Aflatoxin in agricultural commodities and herbal medicine. In: Guevara-Gonzalez, R. G. (Ed.): Aflatoxins – biochemistry and molecular biology. London : IntechOpen, 2011, pp. 367–396. ISBN: 978-953-51-4422-9. DOI: 10.5772/23729.

Received 30 March 2022; 1st revised 17 August 2022; accepted 26 August 2022; published online 4 October 2022.