

Animal feed and feeding

UDC 639.111.4:636.085.19

doi: 10.15389/agrobiology.2018.4.779eng

doi: 10.15389/agrobiology.2018.4.779rus

MYCOTOXINS DIFFUSION IN FEEDS OF SUMMER PASTURING RATION OF *Rangifer tarandus* IN ARCTIC ZONES OF RUSSIA

E.A. YILDIRIM¹, L.A. ILINA¹, K.A. LAISHEV¹, V.A. FILIPPOVA¹, A.V. DUBROWIN¹,

T.P. DUNYASHEV¹, G.Yu. LAPTEV¹, I.N. NIKONOV², A.A. YUZHAKOV³,
T.M. ROMANENKO⁴, Yu.P. VYLKO⁴

¹JSC «Biotrof+», 19 korp. 1, Zagreb'skii bulv., St. Petersburg, 192284 Russia, e-mail ilina@biotrof.ru (✉ corresponding author), deniz@biotrof.ru, dumova@biotrof.ru, dubrowin.a.v@yandex.ru, timur@biotrof.ru, laptev@biotrof.ru;

²All-Russian Research Veterinary Institute of Poultry Science — Branch of the Federal Scientific Center All-Russian Research and Technological Poultry Institute RAS, Federal Agency of Scientific Organizations, 48, ul. Chernikova, St. Petersburg—Lomonosov, 198412 Russia, e-mail ilnikonov@yandex.ru;

³Northwest Center for Interdisciplinary Research of Food Security Problems, Federal Agency of Scientific Organizations, 7, sh. Podbel'skogo, St. Petersburg—Pushkin, 196608 Russia, e-mail layshev@mail.ru, alyuzhakov@yandex.ru;

⁴Laverov Federal Center for Integrated Arctic Research (FCIARctic) RAS, Naryan-Mar Agro-Experimental Station, Federal Agency of Scientific Organizations, 1a, ul. Rybnikov, Naryan-Mar, Nenets AO, 166004 Russia, e-mail nmshos@atnet.ru, vylko.yury@yandex.ru

ORCID:

Yildirim E.A. orcid.org/0000-0002-5846-5105

Ilina L.A. orcid.org/0000-0003-2789-4844

Laishev K.A. orcid.org/0000-0003-2490-6942

Filippova V.A. orcid.org/0000-0001-8789-9837

Dubrowin A.V. orcid.org/0000-0001-8424-4114

Dunyashev T.P. orcid.org/0000-0002-3918-0948

The authors declare no conflict of interests

Acknowledgements:

Supported financially by Russian Science Foundation, grant № 17-76-20026 “Rumen microbiocenosis of *Rangifer tarandus* as a basis for developing promising biotechnologies for reindeer herding in Russia's Arctic regions”

Received November 9, 2017

Laptev G.Yu. orcid.org/0000-0002-8795-6659

Nikonov I.N. orcid.org/0000-0001-9495-0178

Yuzhakov A.A. orcid.org/0000-0002-0633-4074

Romanenko T.M. orcid.org/0000-0003-0034-7453

Vylko Yu.P. orcid.org/0000-0002-6168-8262

Abstract

In the summer and autumn, the food base of *Rangifer tarandus* consists of up to 300 species of higher plants, of which lichens account for about 15 %. It is shown that in the tissues of some higher plants and in the soil under lichens there are micromycetes capable of producing mycotoxins. In the present work, we are the first to estimate the content of mycotoxins for the components of summer reindeer rations, i.e. *Salix borealis*, *Vaccinium uliginosum*, *Betula nana*, and *B. pendula*. The aim of the study was to analyze the distribution of mycotoxins in the components of the summer diet of reindeer. Samples of genera *Cladonia* and *Nephroma* lichens, higher plants of the species *Salix borealis*, *Vaccinium uliginosum*, *Betula nana*, *B. pendula*, and mixtures of perennial grasses were collected in early August 2017 in the pastures of the Harp town of the Yamalo-Nenets Autonomous District, the Nelmin-Nos town of Nenets Autonomous District and the Pushnoy town of the Murmansk region. Aflatoxins (AFLA), ochratoxin A (OTA), T-2 toxin (T-2), zearalenone (ZEN), deoxynivalenol (DON) were detected and measured in the samples using ELISA test. During the mycotoxicological evaluation of the summer food ration components of reindeer, we found multiple contaminations with the mycotoxins. The samples of *Embryophyta* representatives revealed a greater number of toxic metabolites compared to samples of the genera *Cladonia* and *Nephroma* lichens. Practically, in all samples of higher plants, the presence of mycotoxins T-2, ZEN and DON produced by *Fusarium* pathogens which affects plants during vegetation, as well as AFLA and OTA metabolites of micromycetes *Aspergillus* sp. and *Penicillium* sp. which previously were considered not adapted for growth and reproduction in plant tissues during the growing season. However, AFLA and OTA were the least represented on virtually all samples. It is of interest that OTA was not detected in any of the lichen samples assayed. In most of the samples, DON *Fusarium* toxins dominated with accumulation in lichens up to 0.15 mg/kg and in *Embryophyta* samples up to 33.8 mg/kg, as well as ZEN at the amount of up to 0.1227 mg/kg and 2.543 mg/kg, respectively. Mycotoxin contamination of the samples of genus *Cladonia* practically did not have regional differences, whereas in the mixture of perennial grasses and in *V. uliginosum* mycotoxin contamination varied to a large extent depending on the place of plant growth. The mycotoxins are found

in concentrations that may pose a threat to animal health.

Keywords: *Rangifer tarandus*, feed, lichens, mycotoxins, ELISA

Reindeer herding as a source of food, fell, velvet antlers and endocrine-enzyme products plays a leading role for the regions of the Arctic zone of the Russian Federation. The diet of *Rangifer tarandus* consists predominantly of scanty, low-nutritious vegetable fodder. In the winter-spring period, the share of lichens in the diet increases to 70% and only 30% falls on the representatives of *Embryophyta*. In the summer-autumn period, the basis of the animals' food supply is composed of up to 300 species of higher plants (the representatives of the *Salicaceae* and *Poaceae* families, *Betula nana* species, perennial grasses, etc.), the lichens are not more than 15% [1].

It is shown that the tissues of some higher plants and soil under lichens contain the micromycetes which are able to produce mycotoxins. Thus, T.Yu. Tolpysheva [1] discovered the presence of significant amounts of micromycetes of the *Penicillium* genus in soils under lichens. It is known that some members of this genus can produce ochratoxin A [2]). A.A. Burkin et al. [3] have found various mycotoxins (alternariol, sterigmatocystin, mycophenolic acid, citrinin, cyclopiazonic acid and others) in the lichens which belong to 20 genera of the *Cladoniaceae*, *Nephromataceae*, *Parmeliaceae*, *Umbilicariaceae*, *Peltigeraceae* and *Teloschistaceae* families. The presence of micromycetes, which are the producers of mycotoxins, in the tissues of tree and shrub species growing in the Arctic zone of Russia has not been noted before. Nevertheless, the fact of occurrence of toxigenic micromycetes in perennial grasses which also constitute the basis of the reindeer diet is well studied. A.S. Orina et al. [4] have found in the tissues of perennial grasses a significant amount of micromycetes of the *Fusarium* genus. It is known that *F. sporotrichioides* and *F. langsethiae* species are capable of producing T-2 toxin, *F. culmorum* and *F. graminearum* produce zearalenone, *F. culmorum* and *F. graminearum* produce deoxynivalenol [5].

The reindeer physiology (as opposed to cattle) including the state of rumen microflora underwent much less anthropogenic influence. According to a number of researchers [6, 7], the anaerobic microorganisms of reindeer rumen are able to detoxify usnic acid [8] and mycotoxins [1, 9] from lichens. However, the emergence of intoxication in reindeer due to eating lichens has been demonstrated [10].

There are no any publications concerning the containing of toxic metabolites of micromycetes in higher plants being the components of summer pasture diets of reindeers both in domestic and foreign literature. The study of this problem is of considerable scientific interest.

In this work, we for the first time discovered the presence of mycotoxins in such components of summer pasture diets of reindeers as *Salix borealis*, *Vaccinium uliginosum*, *Betula nana* and *B. pendula*.

The objective of these researches was to analyze the prevalence of mycotoxins in the components of the summer diet reindeer of *Rangifer tarandus* using enzyme-linked immunosorbent assay.

Techniques. The samples of reindeer summer diets were collected at the beginning of August 2017 in the territory of tundra and forest-tundra pastures located in the Kharp township (Priural'sky district, Yamalo-Nenets Autonomous Okrug), Nelmin-Nos village (Nenets Autonomous Okrug) and Pushnoy township (Kola district, Murmansk region). The lichens of the *Cladonia* and *Nephroma* genera, higher plants *Salix borealis* (northern willow), *Salix Polar* (polar willow), *Vaccinium uliginosum* (blueberry), *Betula nana* (dwarf birch) and *Betula pendula* (drooping birch) species as well as the mix of perennial grasses have been sampled in 3 replications. The samples have been cleared from the remains of other plants, soil and bark.

The content of aflatoxins (AFLA), ochratoxin A (OTA), T-2 toxin (T-2),

zearalenone (ZEN) and deoxynivalenol (DON) in the samples was investigated using the of enzyme-linked immunosorbent assay (AgraQuant test system, Romer Labs, Inc., Austria) according to manufacturer's recommendations. The mycotoxins, with the exception of DON, were extracted with 70% methanol, DON with distilled water. The solutions of five analyzed mycotoxins of known concentrations were used as the standards. In analysis for ZEN and T-2, 10% hydrochloric acid was used as the stopping solution, for AFLA, OTA and DON the stopping solution was 10% phosphoric acid. The optical density (OD) was measured at $\lambda=450\text{nm}$ (a Stat Fax 303+ microstrip photometer, Awareness Technology, Inc., USA).

The calculation of sample mean (M) values) and the error probability, with a rejection of the null hypothesis p , and standard error of means $\pm\text{SEM}$ was made with Microsoft Excel 2010 software.

Results. The multicomponent samples repeating the composition of the averaged summer pasture diet of reindeers have been formed (Table 1).

1. The composition of the multicomponent samples repeating the average summer pasture diet of reindeer *Rangifer tarandus* (Arctic zone of Russia, 2017)

Component	Percentage of the component in the total diet		
	Kharp township (tundra)	Pushnoy township (forest tundra)	Nelmin-Nos village (tundra)
<i>Cladonia</i>	5	10	10
<i>Nephroma</i>	5	—	—
<i>Salix borealis</i>	5	20	20
<i>Salix polaris</i>	15	—	—
<i>Vaccinium uliginosum</i>	10	—	5
<i>Betula nana</i>	25	20	15
<i>Betula pendula</i>	5	20	20
Mix of perennial herbs	30	30	30

N o t e. Dashes mean that the component has not been represented in the total diet.

Almost in the all investigated samples (both lichens and higher plants) we have detected the presence of mycotoxins (Table 2). To date, the maximum permissible concentration (MPC) of mycotoxins for reindeer feed is not established in Russia. In the Unified Veterinary (Veterinary and Sanitary) Requirements for the products subject to the veterinary control (supervision) (approved by the Decision of the Commission of the Eurasian Economic Community No. 137 dated June 18, 2010) the MPC established for oats, wheat and barley cereals are 0.004 mg/kg for AFLA, 0.005 mg/kg for OTA, 0.06 mg/kg for T-2, 0.1 mg/kg for ZEN, and 1 mg/kg for DON. In most investigated samples we detected the excess of aflatoxins, ochratoxin A, T-2 toxin, zearalenone and DON compared to the above MPC values. The multiple contamination of all samples with mycotoxins (at least with three ones) has been observed that enhanced their toxic impact on animals [4].

The content of mycotoxins largely varied depending on the species. In the samples of higher plants we detected the higher content of toxic metabolites than in *Cladonia* and *Nephroma* lichens. Also, the higher content of mycotoxins in comparison with that for lichens in its pure form has also been found in the multicomponent mixes similar to averaged summer diets.

In the almost all samples of higher plants, we have detected the presence of T-2, ZEN, and DON produced by fusarium pathogens [11] which affects vegetating plants and of AFLA and OTA which are the metabolites of *Aspergillus* sp. and *Penicillium* sp. [12]. Nevertheless, in our experiments, AFLA and OTA were the least represented almost in the all samples. The presence of ochratoxin A was not detected in any of the investigated samples of lichens. Almost in all samples, the DON fusariotoxins produced by *F. culmorum* and *F. graminearum* dominated and accumulated in lichens up to 0.15 mg/kg and in *Embryophyta* up to 33.8 mg/kg, ZEN content was up to 0.1227 mg/kg and 2.543 mg/kg, respectively ($p \leq 0.05$).

The contamination of lichens of *Cladonia* genus by mycotoxins has no geo-

2. The average content (mg/kg) of mycotoxins in the components of the summer pasture diet of the *Rangifer tarandus* reindeers (Arctic zone of Russia, 2017)

Mycotoxin	<i>Cladonia</i>	<i>Nephroma</i>	<i>Vaccinium uliginosum</i>	<i>Salix borealis</i>	Mix of perennial herbs	<i>Betula pendula</i>	<i>B. nana</i>	Mix of the diet components
Kharptownship (Priuralsky district, Yamalo-Nenets Autonomous Okrug)								
AFLA	0.005±0.0002*	0.003±0.0006	0.123±0.0190	0.129±0.0100	0.011±0.0047	0.111±0.0054*	—	0.089±0.0041**
OTA	< l. r. d.	< l. r. d.	0.0371±0.00186*	0.0968±0.00800	0.0007±0.00003*	0.0895±0.00640	—	0.041±0.0036
T-2	0.0385±0.00200*	0.0179±0.00083*	1.9690±0.09300*	1.0210±0.10000	0.0004±0.00008	0.4050±0.04700	—	0.1058±0.009
ZEN	0.04±0.002	0.12±0.009	2.54±0.110*	2.44±0.220	0.12±0.008	1.94±0.150	—	0.52±0.021*
DON	0.003±0.0003	0.150±0.0075*	10.300±0.9000	9.810±0.8700	1.550±0.07500*	< l. r. d.	—	1.700±0.0750*
Pushnoy township (Kola district, Murmansk region)								
AFLA	0.003±0.0001*	—	—	0.067±0.0084	0.009±0.0004*	0.111±0.0080	0.107±0.0200	0.057±0.0021*
OTA	< l. r. d.	—	—	0.0307±0.00140*	< l. r. d.	0.0601±0.00690	0.0540±0.00270*	0.0195±0.00310
T-2	0.0098±0.00400	—	—	0.1770±0.03000	< l. r. d.	0.1484±0.00720*	0.1387±0.01200	0.0288±0.00110*
ZEN	0.09±0.004*	—	—	0.56±0.040	0.13±0.008	1483.00±97.600	0.90±0.039*	0.49±0.047
DON	< l. r. d.	—	—	2.790±0.1200*	0.350±0.0370	1.060±0.0890	2.080±0.4800	2.510±0.2400
Nelmin-Nos village (Nenets Autonomous Okrug)								
AFLA	0.0046±0.00058	—	0.005±0.0200	0.129±0.0060*	0.009±0.0003*	—	0.1351±0.0230	0.099±0.0120
OTA	< l. r. d.	—	< l. r. d.	0.1119±0.00970	< l. r. d.	—	0.1017±0.00800	0.0755±0.00310*
T-2	0.0064±0.00100	—	< l. r. d.	0.7615±0.06400	0.0548±0.00210*	—	0.8188±0.03200*	0.4154±0.89000
ZEN	0.09±0.012	—	0.49±0.021*	1.25±0.058*	0.19±0.0094*	—	1.70±0.170	0.79±0.035*
DON	0.020±0.0010*	—	0.130±0.0090	2.600±0.3400	1.020±0.2200	—	33.800±1.7000*	1.530±0.4300

Note. AFLA — aflatoxins; OTA — ochratoxin A; T-2 — T-2 toxin; ZEN — zearalenone; DON — deoxynivalenol. Dashes mean that in this version the analysis for the mycotoxin content was not made; < l. r. d. — below the limit of reliable determination by the ELISA method.

* Statistically significant at $p \leq 0.05$

** Statistically significant at $p \leq 0.01$

graphical differences. The mycotoxins content in the samples of higher plants *V. uliginosum* and perennial grasses varied greatly depending on the place of growing. Thus, this index was the smallest in the samples of perennial grass mix from the forest-tundra pastures of the Pushnoy township compared to those grown in the tundra pastures of the Kharp township and Nelmin-Nos village. In *V. uliginosum* grown in the pasture of the Nelmin-Nos village the presence of OTA and T-2 was not detected, whereas in similar samples from Kharp township their content was 0.0371 and 1.969 mg/kg, respectively ($p \leq 0.05$). The content of AFLA, ZEN and DON in *V. uliginosum* samples from the Nelmin-Nos village was respectively 26.1, 5.2 and 79.2 times more than in the samples from the Kharp township. Summer meteorological conditions in 2017 in the Yamalo-Nenets, Nenets Autonomous Okrug and the Murmansk region were similar, thus the geographical differences in contamination by mycotoxins may be due to the soils dissimilarity in chemical composition and nutrients content. In addition, the described regional features may be related to the specificity of the structural organizations of epiphytic bacterial communities of higher plants. It has been reported that many bacteria including epiphytic perform the biodegradation of the secondary metabolites of micromycetes and have the antibiotic activity against the producers of these metabolites [13-15].

The presence of toxigenic micromycetes (*Fusarium sporotrichioides*, *F. langsethiae*, *F. verticillioides*, *F. culmorum* and *F. graminearum*), which are able to produce fusariotoxins, in higher plants is widely known [16, 17]. Before, it was assumed [18] that the representatives of the *Aspergillus* and *Penicillium* genera the producers of aflatoxins and ochratoxin A, are not adapted for growing and reproduction on plant during their growing. However in 2014, by quantitative PCR method, the micromycetes of the *Fusarium*, *Aspergillus* and *Penicillium* genera and their secondary metabolites, the aflatoxins, ochratoxin A, deoxynivalenol, zearalenone and T-2 toxin, have been detected in perennial herb mycobiota [19]). The presence of toxin-producing micromycetes in the mycobiota of trees and shrubs which are the components of the reindeer summer diets have not been detected before.

The issue about the sources of lichens contamination with mycotoxins is also almost unstudied. According to one of the hypotheses put forward by T.Yu. Tolpysheva [20], the contamination of lichens with mycotoxins occurs as the result of contact with the micromycetes *Aspergillus* sp., *Penicillium* sp. and *Fusarium* sp. from the soil under the lichens [21]. According to T.Yu. Tolpysheva [20], lichens are able to absorb the mycotoxins from soil solution. As the author explains, lichens are the poikilohydric organisms which passively absorb moisture. Lichens mostly grow on the soil as curtains in which their talli closely contact to keep the moisture longer. This activates the capillary rise of water with the dissolved substances from the soil up the lichens.

Earlier, some Russian [22-24] and foreign [25-27] researchers pointed out the high frequency of the occurrence of mycotoxins and their significant content in the growing fodder herbage for cattle, but the issue about the content of toxic metabolites in the leaves of woody and shrub plants is still unstudied. In foreign literature there are only a few instructions concerning the detection of mycotoxins in the leaves of woody cultivated plants [28].

Thus, in mycotoxicological assessment of the base components of the reindeer summer fodder, the lichens and higher plants growing in tundra and forest-tundra pastures of three regions of the Arctic zone of the Russian Federation, we have found the multiple contamination with aflatoxins, ochratoxin A, zearalenone, T-2 toxin and deoxynivalenol. Our findings indicate that the problem of contamination of higher plants by mycotoxins is much more acute com-

pared to that of lichens. The mycotoxin contamination of *Cladonia* genus almost has no regional differences, whereas the content of mycotoxins in the mix of perennial herbs and in *V. uliginosum* varies greatly depending on the place of growing. The detected mycotoxins are present in the concentrations which may pose a threat to animals.

REFERENCES

1. Chikalev A.I., Yuldashbaev Yu.A., Rodionov G.V. *Olenevodstvo* [Reindeer herding]. Moscow, 2015 (in Russ.).
2. Bennett J.W., Klich M. Mycotoxins. *Clin. Microbiol. Rev.*, 2003, 16(3): 497-516 (doi: 10.1128/CMR.16.3.497-516.2003).
3. Burkin A.A., Kononenko G.P. *Prikladnaya biokhimiya i mikrobiologiya*, 2013, 5(49): 522-530 (doi: 10.7868/S0555109913050036) (in Russ.).
4. Orina A.S., Gavrilova O.P., Gagkaeva T.Yu. *Zashchita i karantin rastenii*, 2017, 6: 25-27 (in Russ.).
5. Diaz D. *Mikotoksiny i mikotoksikozy* [Mycotoxins and mycotoxicoses]. Moscow, 2006 (in Russ.).
6. Palo R.T. Usnic acid, a secondary metabolite of lichens and its effect on in vitro digestibility in reindeer. *Rangifer*, 1993, 13: 39-43.
7. Sundset M.A., Edwards J.E., Cheng Y.F., Senosiain R.S., Fraile M.N., Northwood K.S., Praesteng K.E., Glad T., Mathiesen S.D., Wright A.D. Rumen microbial diversity in Svalbard reindeer, with particular emphasis on methanogenic archaea. *FEMS Microbiol. Ecol.*, 2009, 70(3): 553-562 (doi: 10.1111/j.1574-6941.2009.00750.x).
8. Luzina O.A., Salakhutdinov N.F. *Bioorganicheskaya khimiya*, 2016, 3(42), 2016: 276 (doi: 10.7868/S0132342316030106) (in Russ.).
9. Burkin A.A., Kononenko G.P. *Izvestiya Rossiiskoi akademii nauk. Seriya Biologicheskaya*, 2014, 3: 228 (doi: 10.7868/S0002332914030047) (in Russ.).
10. Kingsbury J.M. *Poisonous plants of the United States and Canada*. NY, 1964.
11. Kononenko G.P., Burkin A.A. V sbornike: *Uspekhi meditsinskoi mikologii* [Advances in medical mycology]. Moscow, 2003: 141-144 (in Russ.).
12. Burkin A.A. V sbornike: *Uspekhi meditsinskoi mikologii* [Advances in medical mycology]. Moscow, 2003: 122-124 (in Russ.).
13. Hernandez-Mendoza A., Garcia H.S., Steele J.L. Screening of *Lactobacillus casei* strains for their ability to bind aflatoxin B1. *Food Chem. Toxicol.*, 2009, 47(6): 1064-1068 (doi: 10.1016/j.fct.2009.01.042).
14. Čvek D., Markov K., Frece J., Friganović M., Duraković L., Delaš F. Adhesion of zearalenone to the surface of lactic acid bacteria cells. *Croatian Journal for Food Technology, Biotechnology & Nutrition*, 2012, 7(Special Issue): 49-52.
15. Shi L., Liang Z., Li J., Hao J., Xu Y., Huang K., Tian J., He X., Xu W. Ochratoxin A biocontrol and biodegradation by *Bacillus subtilis* CW 14. *J. Sci. Food Agr.*, 2014, 94(9): 1879-1885 (doi: 10.1002/jsfa.6507).
16. Levitin M.M., Novozhilov K.V., Afanasenko O.S., Mikhailov L.A., Mironenko N.V., Gagkaeva T.Yu., Gannibal F.B. V knige: *Mikologiya segodnya. Tom 2* [In: Mycology today. V. 2]. Moscow, 2011: 261-274 (in Russ.).
17. Gagkaeva T.Yu., Gavrilova O.P. *Materialy Mezhdunarodnoi nauchnoi konferentsii, posvyashchennoi 150-letiyu so dnya rozhdeniya chl.-korr. AN SSSR, prof. A.A. Yachevskogo «Problemy mikologii i fitopatologii v XXI veke»* [Proc. Int. Conf. dedicated to 150 anniversary of Prof. A.A. Yachevskii «Challenges of mycology and phytopathology in the 21st century»]. St. Petersburg, 2013: 400 (in Russ.).
18. Christensen C.M., Kaufmann H.H. *Storage of cereal grains and their products. 2nd ed.* American Association of Cereal Chemists. St. Paul, Minnesota, 1974: 158-192.
19. Laptsev G.Yu., Novikova N.I., Il'ina L.A., Ilydyrym E.A., Soldatova V.V., Nikonov I.N., Filipova V.A., Brazhnik E.A., Sokolova O.N. Dynamics of mycotoxin accumulation in silage during storage. *Sel'skokhozyaistvennaya Biologiya [Agricultural Biology]*, 2014, 6: 123-130 (doi: 10.15389/agrobiology.2014.6.123eng).
20. Tolpysheva T.Yu. *Vestnik Moskovskogo Universiteta. Seriya 16: Biologiya*, 2014, 3: 37-41 (in Russ.).
21. Girlanda M., Isocrono D., Bianco C., Luppi-Mosca A.M. Two foliose lichens as microfungus ecological niches. *Mycologia*, 1997, 89(4): 531-536 (doi: 10.2307/3760987).
22. Burdov L.G. *Monitoring mikotoksinov, profilaktika i lechenie mikotoksikozov v Udmurtskoi Respublike. Kandidatskaya dissertatsiya* [Monitoring of mycotoxins, mycotoxicosis prevention and treatment in the Udmurt Republic. PhD Thesis]. Kazan', 2013 (in Russ.).
23. Burkin A.A., Kononenko G.P. Mycotoxin contamination of meadow grasses in European Rus-

- sia. *Sel'skokhozyaistvennaya Biologiya [Agricultural Biology]*, 2015, 50(4): 503-512 (doi: 10.15389/agrobiology.2015.4.503eng).
24. Laptev G.Yu., Novikova N.I., Dubrovina E.G., Il'ina L.A., Ilydyrym E.A., Filippova V.A., Nikonov I.N., Brazhnik E.A. *Kormoproizvodstvo*, 2014, 10: 35-39 (in Russ.).
 25. Boudra H., Morgavi D.P. Reduction in fusarium toxin levels in corn silage with low dry matter and storage time. *J. Agr. Food Chem.*, 2008, 56(12): 4523-4528 (doi: 10.1021/jf800267k).
 26. Mansfield M.A., Jones A.D., Kuldau G.A. Contamination of fresh and ensiled maize by multiple *Penicillium* mycotoxins. *Phytopathology*, 2008, 98(3): 330-336 (doi: 10.1094/PHYTO-98-3-0330).
 27. Teller R.S., Schmidt R.J., Whitlow L.W., Kung L. Jr. Effect of physical damage to ears of corn before harvest and treatment with various additives on the concentration of mycotoxins, silage fermentation, and aerobic stability of corn silage. *J. Dairy Sci.*, 2012, 95(3): 1428-1436 (doi: 10.3168/jds.2011-4610).
 28. Watanabe I., Kakishima M., Adachi Y., Nakajima H. Potential mycotoxin productivity of *Alternaria alternata* isolated from garden trees. *Mycotoxins*, 2007, 57(1): 3-9 (doi: 10.2520/myco.57.3).