

Biological and Ecological Characteristics of Fungi Affecting Seeds of Grain Crops

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The article deals with materials of scientific research on morphological and molecular and genetic identification of fungi affecting the seeds of grain crops. The focus is on the fungi growing in the grain stored. The seeds of grain crops (*Triticum aestivum* L., *Avena sativa* L., *Hordeum vulgare* L., *Zea mays* L., *Oryza sativa* L.) were sampled from granaries of five districts (Talgat, Ili, Karasai, Panfilov and Zhambyl districts) of Almaty Region. The disease agents of fungus etiology of genera of *Ustilago*, *Clodsporium*, *Verticillium*, *Diplodia*, *Macrosporium*, *Alternaria*, *Helminthosporium*, *Fusarium*, *Aspergillus*, *Penicillium*, *Rhizopus*, *Mucor* were identified, which affect safety, quality and safety of grain.

Key words: β -tubulin, fungi, grain crops, identification, ITS, morphology, seed.

The food security of Kazakhstan based on the competitive production is a main goal of the agricultural sector, which is a leading industry of the economy of Kazakhstan. According to statistics, the areas of grain crops are prevailing (up to 13.5 – 14.0 million hectares per annum). The crop yield is growing. The export potential of Kazakhstan is estimated as 5 - 6 million tons per annum, i.e. it is the world's 6th largest grain exporter.

High yields of agricultural crops are based on healthy and good-quality seeds. From harvesting to sowing, the quality of seeds is

determined by many factors, and one of the most important ones is storage conditions (relative humidity, ambient temperature, aeration).

If storage conditions are abnormal, the grain seeds are mainly affected by the fungi of genera of *Aspergillus*, *Penicillium*, *Rhizopus*, *Fusarium*. These fungi decrease the germinating capacity of grain seeds, and change the color of bruchid, cause the death of corcule, and their metabolites are often toxic for plants, animals and human¹.

Species composition of the complex of pathogens on seeds of certain types of crops are not constant, but in certain environmental conditions, it can be maintained.

In Kazakhstan, the defeat of crop seeds microscopic fungi are described in EI Ishpakina (1953), VV Remmeles (1997), Kazenas LD (1956),

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Koishybayev M. (1975), Zhienbayev J. (1974), Sagitov AO (1992), Bostanov AM (2009). However, the biology and taxonomy of these fungi are insufficiently studied. So far, it is difficult to identify the specie composition of small-spore fungi.

When classical methods of identification are used, the identification of fungi by cultural and morphological signs may be unreliable.

Due to development of modern molecular methods, it is now possible to quickly and exactly identify the specie and race of fungi.

MATERIALS AND METHODS

We took the seeds of grain crops from granaries of the five districts (Talgar, Ili, Karasai, Panfilov and Zhambyl districts) of Almaty Region.

The seeds of grain crops were left in a wet chamber. The Petri dishes were kept at the temperature of 21°C. The growth and development were observed every day. On the 7-th day, fungal spores emerged. By the nature of the mycelium growth and sporulation was determined by their species determinants Naumov (1935), Litvinov (1967). Then Petri cup with a nutrient medium Capek passed separately these fungi for further study.

Growth of fungi on agar nutrient storage media at a temperature of 5 °, 15 °, 25 ° and 35 ° evaluated by a colony diameter of each of the form.

As in the cup with Petri Czapek nutrient medium separately subcultured these fungi to determine the nucleotide sequence (sequencing) of beta-tubulin gene. Then, the sediment was dried in test tubes for 30 minutes, and dissolved in 25 microliters of TE-buffer solution (10 mM tris-HCl, pH 7.4; 1 mM EDTA, pH 8.0) or 80 % DMSO.

For amplification of a fragment of β -tubulin gene, a pair of primers T1/T2 were used², for ITS-areas (area including a fragment of gene 18S ribosomal riziiform, and internal transcribing spacer 1 [ITS 1] for gene 5,8S ribosomal riziiform, and internal transcribing spacer 2 [ITS 2] and a fragment of gene 28S ribosomal riziiform), ITS1/ITS4³. The amplification was performed in thermal cycler C-1000 (Bio-Rad). 25 mL of reaction mixture contained 1 unit of Taq-polymerase (Helicon, Russia), 0.5 mK of every primer, 200 mK of desoxynucleotid triphosphates, buffer solution (75 mM of Tris-HCl (pH 8.8); 20 mM of $(\text{NH}_4)_2\text{SO}_4$;

0.01% (v/v) Tween 20; 2 mM of MgCl_2). The amplification products were separated by electrophoresis in 1 % agarose gel colored by ethidium bromide. The findings of electrophoresis were visualized in trans-illuminator, and then, the amplicons of required length were extracted by silicon oxide powder⁴.

The sequencing PCR was performed according to traditional chain termination method⁵ by using BigDye Terminator v3.1 Cycle Sequencing Kit (ABI, USA). The base sequences were determined by genetic analyzer ABI PRISM 3500 (ABI-Hitachi, Japan). The base sequences identified were edited in software program VectorNTI.

The base sequences identified were compared to those available in GenBank database, through BLASTn online service. According to the findings of the comparison, the species of the strains were identified.

All the sequences identified were deposited in GenBank database (Table 2). The topicality of all the fungal specie names specified as a result of identification and correctness of names of authors of these specie names were checked through Mycobank nomenclature database.

RESULTS

Collected from cereal seeds were found microscopic fungi of the genus *Ustilago*, *Cladosporium*, *Verticillium*, *Diplodia*, *Macrosporium*, *Alternaria*, *Helminthosporium*, *Fusarium*, *Aspergillus*, *Penicillium*, *Rhizopus* and *Mucor* (Table 1).

Next, we consider the types of fungi that Bole more frequent on the seed crops.

Aspergillus flavus is an opportunistic agent of diseases of grain crops. It is important because it produces aflatoxin as a secondary metabolite in the seeds of a number of crops both before and after harvesting⁶. Aflatoxin is a strong carcinogen, which is strictly regulated in most countries. Under favorable conditions, fungi will grow and produce aflatoxin in any seeds harvested. During storage, the aflatoxin content can be controlled by keeping the humidity below the rate, which is favorable for the growth of *Aspergillus flavus*⁷.

The aerial mycelium of colony is dark green, and the reverse side is yellow. Along its edge, the aerial mycelium is cobweb-like. The conidial holders are 300-1000E5-10 µm, and are colorless, uneven, grooved, and rough, and go from hyphas of submerged substrate mycelium; tip bubble is pear-shaped and sphere-shaped, and its diameter is 20-50 µm, and it is colorless, and carries mainly twin-row sterigmas located throughout all the surface. The primary sterigmas are sized 7-10E3-4 µm, and secondary ones are 7-10E2.5-3 µm. The conidia of phialides are unicellular, sphere-shaped, ovum-shaped, or rarely, pear-shaped, with diameter of 3-5 µm, and are almost smooth, colorless, or light yellow-greenish, and are the chains collected in a column, and heads are radial, and vary in sizes. The sclerotias are gnarly, hard, initially white,

gradually becoming brown, plentiful, and sometimes, rare (Figure 1).

The sequencing of 2-tubulin gene and ITS-5,8S of ribosomal DNA of strains of specie of *Aspergillus* showed that the base sequence with overlapping 100% (identity) and 100% similarity (homology) belongs to genus of *Aspergillus*, species of *Aspergillus flavus* Link.

The sequencing of β-tubulin gene was deposited in GenBank under No. KJ938407.

The genus of *Rhizopus* is classified as a family of Mucoraceae in the procedure of mucorales of phylum of Zygomycota. *Rhizopus oryzae* are the most important and representative agents of zygomycosis.

The conidium holders are linear, often winding, and simple, and sometimes, they have

Table 1. Types of fungi isolated from cereal seeds with granaries Almaty region.

Seed	Species of fungi
Triticum aestivum	Helminthosporium sativum Pam., Fusarium proliferatum (Matsush.) Nirenberg ex Gerlach & Nirenberg, Fusarium graminearum Schwabe, Fusarium culmorum (W. G. Sm.) Sacc, Fusarium avenaceum (Fr.) Sacc., Penicillium rugulosum Thom., Penicillium expansum Link, Rhizopus nigricans Ehrenb., Rhizopusoryzae Went & Prins. Geerl., Rhizopus maydis Brydrel, Mucormucedo Fres., Aspergillus niger Tiegh, Aspergillus tubingensis Mosseray, A. glaucus Raper et Fennell, A. flavus Link, Alternaria alternata (Fr.) Keisse, Alternaria tenuis Nees, Macrosporium commune Wall.
Hordeum vulgare	Helminthosporium sativum Pam., Fusarium proliferatum (Matsush.) Nirenberg ex Gerlach & Nirenberg, Fusarium graminearum Schwabe, Penicillium expansum Link, Fusarium culmorum (W. G. Sm.) Sacc, Rhizopus nigricans Ehrenb., Aspergillus niger Tiegh, A. glaucus Raper et Fennell, A. flavus Link, Verticillium sp., Alternaria alternata (Fr.) Keisse, Alternaria tenuis Nees, Macrosporium commune Wall., Ustilagonigra Tapke., Ustilagonuda (Jens.) Kell. St Swing.
Avena sativa	Helminthosporium sativum Pam., Fusarium proliferatum (Matsush.) Nirenberg ex Gerlach & Nirenberg, Fusarium graminearum Schwabe, Fusarium culmorum (W. G. Sm.) Sacc, Fusarium avenaceum (Fr.) Sacc., Penicillium expansum Link, Rhizopus nigricans Ehrenb., Rhizopusoryzae Went & Prins. Geerl., Aspergillus tubingensis Mosseray, A. glaucus Raper et Fennell, A. flavus Link, Alternaria tenuis Nees, Macrosporium commune Wall., Ustilago avenae (Pers.) Jens.
Zea mays	Helminthosporium turcicum Pass., Fusarium graminearum Schwabe, Fusarium oxysporum Schlecht., Rhizopus nigricans Ehrenb., Rhizopusoryzae Went & Prins. Geerl., Aspergillus tubingensis Mosseray, A. glaucus Raper et Fennell, A. flavus Link, Ustilago zeae (Beckm.) Ung., Ustilago maydis (DC.) Cda., Diplodia zeae (Schw.) Lev., Cladosporium zeae Lob.
Oryza sativa	Fusarium graminearum Schwabe, Rhizopus nigricans Ehrenb., Rhizopusoryzae Went & Prins. Geerl., A. glaucus Raper et Fennell, A. flavus Link, Diplodia oryzae Miyake.
Panicum miliaceum	Fusarium graminearum Schwabe, Fusarium avenaceum (Fr.) Sacc., Fusarium oxysporum Schlecht., Rhizopus nigricans Ehrenb., Mucormucedo Fres, A. glaucus Raper et Fennell A. flavus Link
Sorghum vulgare	Penicillium glaucum Fr., Rhizopus nigricans Ehrenb., Aspergillus niger Tiegh, A. fumigatus Fres. A. restrictus G. Sm., A. glaucus Raper et Fennell, A. flavus Link, A. candidus Link, Macrosporium commune Wall.

Table 2. Nucleotide sequence of the genes of fungi storage

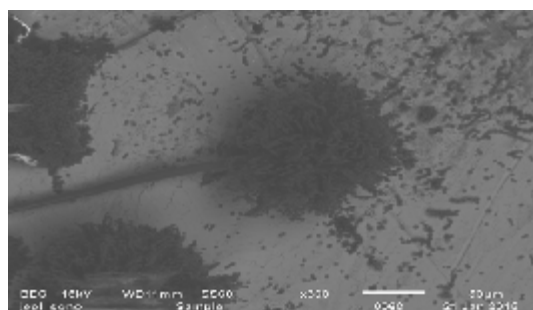
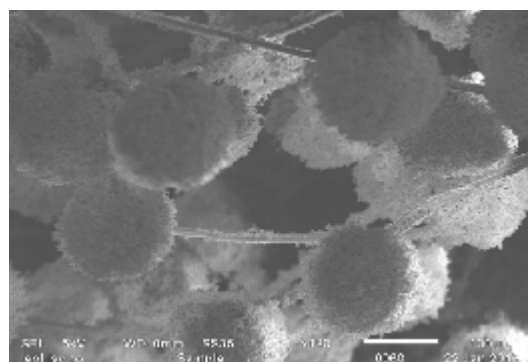
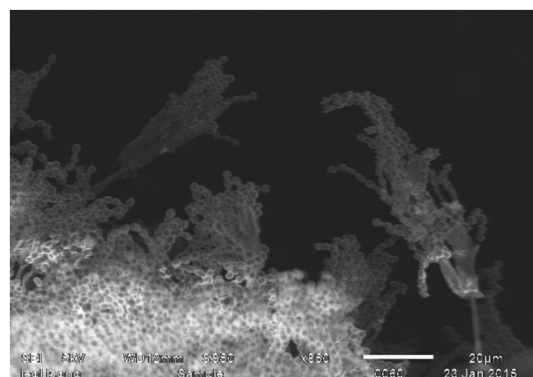
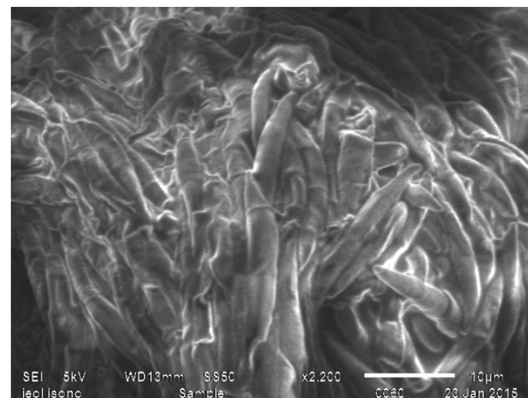
Name of fungi	The obtained nucleotide sequences and account number in GenBank
<i>Aspergillus flavus</i> Link.	Sequencing of the gene beta-tubulin (deposited in GenBank under N° KJ938407): TGGTAACCAAAATCGGTGCTGCTTTCTGGTATGTCTCAATGCCTTCGAGTTAGTATGCTTTGGACCAAGGA ACTCCTCAAAAGCATGATCTCGGATGTGTCTGTATATCTGCCACATGTTTGCTAACAACCTTGCAGGC AAACCATCTCTGGCGAGCACGGCCTTGACGGCTCCGGTGTGTAAGTACAGCCTGTATACACCTCGAACG AACGACGACCATATGGCATTAGAAGTTGGAATGGATCTGACGGCAAGGATAGTTACAATGGCTCCTCCG ATCTCCAGCTGGAGCGTATGAACGCTACTTCAACGAGGTGCGTACCTCAAAATTCAGCATCTATGAA AACGCTTTGGCAACTCTGACCGCTTCTCCAGGCCAGCGGAAACAAGTATGTCCCTCGTGCCGCTCTCGTT GATCTTGAGCCTGGTACCATGGACGCCGTCGGTGCCGGTCCCTTCGGTCAGCTCTTCCGTCCCGACAAC TCGTTTTCCGGCCAGTCCGGTGTGGTAAACAACCTGGGCCAAGGGTCACTACACTGAGGGTGC
<i>Aspergillustubingensis</i> Mosseray.	Sequencing of the gene beta-tubulin (KJ938412): CTTGTGCTAACTGCATGTCTTCGTCGCTTCAATAGGTTACCTCCAAACCGGCCAGTGTGTAAGTGCCAA TATGTTCTTCCAATGATTGCCCTCCCGGGTCTTGATTGGTGTTCGGTGGACTAAACAACAAATGATGGT GGTTAGGGTAACCAAAATGGTGTGCTTTCTGGTACGTATTCAGTCCACTGGATTGGGGATGGAACAT CATCTCTCAAGCTATCTCAGCTTGAGTTAGATGTTATCCATCGGGGATATAGCTATCGGGTTAAGAACA CGTCTAACAACTCAACAGGCAGACCATCTCTGGCGAGCACGGCCTTGACGGCTCCGGTGTGTAAGTACA ACTTTTTCACACCTCTCAATTTGGTCAATGTGGAAGGATTGGGTTTTCCTGACGCGCAGGATAGTTAC AATGGCACCTCCGACCTCCAGCTGAGCGCATGAACGTCTACTTCAACGAGGTATAGATCACACCGCTCCC TGAGTTTTTTCACGACAATATCATCAATGTCTGACCACTTCAGCAGGTAGCGGTAACAAGTATGTCCC CCGTGCCGTCTCGTCAATCTCGAGCCCGGTACCATGGACGCCGTCCGTGCCGGTCCCTTCGGCCAGCT
<i>Aspergillustubingensis</i> Mosseray.	Sequencing of the ITS-regions (KJ938413): ACCTCCATCCGTGTCTATTATACCTGTTGCTTCGGCGGGCCCGCCGCTTGTCCGCCGCCGGGGGGGGC CCTTTGCCCCCCGGGGCCCGTGCCCGCCGGAGACCCCAACAGAACACTGTCTGAAAGCGTGCAGTCTGA GTTGATTGAATGCAATCAGTTAAAACCTTCAACAATGGATCTCTGGTTCCGGCATCGATGAAGAACGC AGCGAAATGCGATAACTAATGTGAATTGCAGAATTCAGTGAATCATCGAGTCTTTGAACGCACATTGCG CCCCCTGGTATTCCGGGGGGCATGCTGTCCGAGCGTCATTGCTGCCCTCAAGCCCGGCTGTGTGTGG GTCCGCTCCCGCTCTCCGGGGGGACGGGCCGAAAGGCAGCGCGGCCACCGCTCCGATCCTCGAGC GTATGGGGCTTTGTACATGCTCTGTAGGATTGGCCGGCGCTGCCGACGTTTTCCAACCATTTTTTCCA GGTTGACCTCGGATCAGGTAGGGATACCCGCTGAACCTAAGCATATCAATAAGCGGAGGA
<i>Penicillium expansum</i> Link.	Sequencing of the gene beta-tubulin (KJ938414): AACGGCCCTGAGCCATGACCCCACTCCCAACAGATCTTTTGCTAATATGATCTAGGTTACCTCCAAA CCGGCCAGTGTGTAAGTTGACATGGAACATTCTTGGAACATTCTTGGATTCTGTTGGACTAAATTTGGA ATTGGGTTATAGGGTAACCAAAATGGTGCCGCTTTCTGGTAAGTGCCGAGCTTTTTTTCCGGTTGGGTA TCAATTGACAAATTAATACTGGATTGCAAGCAACCATCTCTGGCGAGCACGGTCTCGATGGTGATGG ACAGTAAGTTCAACGGTGATGGGTTTCTAGTAGATCACACGTCTGATATCTTGCTAGGTACAATGGTAC CTCCGACCTCCAGCTCGAGCGTATGAACGTCTACTTCAACCATGTGAGTACACCGACTAGTTACCGAAT AATCGTGCAATCATCTGATCGGATCTTTTCTTGATAATCTAGGCCAGCGGTGACAAAGTACGTTCCCGT GCCGTTCTCGTCAATTTGGAGCCCGGTACCATGGACGCTGTCCGTCCGGTCCCTTCGGCAAGCTTTTTCC GCCCGACAACCTTCGTTCTCGGTTCAGTCCGGTGCTGGTAACAACCTGGGCCAAGGGTCACTA
<i>Penicillium expansum</i> Link.	Sequencing of the ITS-regions (KJ938415): TGGGTCCAACCTCCACCGTGTTATTACCTCGTTGCTTCGGCGGGCCCGCCTTAAGTGGCCGCCGGG GGGCTCACGCCCCCGGGCCCGCCGCGCCGAGACACCCCGAACTCTGCCTGAAGATTGCTGCTGAG TGAAAAATATAAATTATTTAAACCTTCAACAACGGATCTCTGGTTCCGGCATCGATGAAGAAGCAGC GAAATGCGATACGTAATGTGAATTGCAAAATTCAGTGAATCATCGAGTCTTTGAACGCACATTGCGCCCC CTGGTATTCCGGGGGGCATGCTGTCCGAGCGTCATTGCTGCCCTCAAGCCCGGCTGTGTGTGGGGCC CGTCTCCGATTCCGGGGGACGGGCCGAAAGGCAGCGCGGCCACCGCTCCGGTCTCGAGCGTATG GGGCTTTGTACCCGCTCTGTAGGCCCGCGCGGCTTCCGATCAACCCAAATTTTTATCCAGGTTGAC CTCGGATCAGGTAGGGATACCCGCTGAACCTAAGCATATCAATAAGCGGAGGA
<i>Rhizopus oryzae</i> Went & Prins. Geerl.	Sequencing of the ITS-regions (KJ938408): TTACCTTAGGGTTTCTCTGGGGTAAGTGATTGCTTCTACACTGTGAAAAATTTGGCTGAGAGACTCAGAC TGGTCATGGGTAGACCTATCTGGGGTTTGATCGATGCCACTCCTGGTTTCAGGAGTACCTTTCATAATAA ACCTAGAAAATTCAGTATTATAAAGTTTAATAAAAAACAACCTTTTAAACAATGGATCTCTGGTCTCCCAT CGATGAAGAACGTAGCAAAAGTGCAGATAACTAGTGTGAATTGCATATTCAAGTGAATCATCGAGTCTTTGA ACGACGCTTGACATCTATGGTTTTTCTATAGAGTACGCTGCTTCAGTATCATCAAAACCCACACATAA CATTTGTTTATGTGGTATGGGTCGATCGCTGTTTTATTACAGTGAGCACCTAAAATGTGTGTGATTTT CTGTCTGGCTTGCTAGGCAGGAATATTACGCTGGTCTCAGGATCTTTTTTTGGTTCCGCCAGGAAGTA AAGTACAAGAGTATAATCCAGTAACCTTTCAAACTATGATCTGAAGTCAGGTGGGATTACCCGCTGAAC TAAGCATATCAATAAGCGGAGGA
<i>Fusarium proliferatum</i> (Matsush) Nirenberg ex Gerlach & Nirenberg.	Sequencing of the gene beta-tubulin (KJ938410): GTATTGTGCTGTGACGCGTTGAGTTTACCGTGCCCTGATTCTACCCCGCTGGGTGGTGGCAGCTCAA CGACATTGCACGATAGCTAGCAGCTTAACTACCTTCTGTCAAGACGAAGAAGCTAATCAGATCTTTT CTCTACGATAGGTTACCTCCAGACCGGTCAAGTGGTCAAGTGTCTCATCGCTTCTCAACGTCGCATGCGG GGGATGCTCACAACGTTTATCAGGGTAACCAAAATGGTGCTGCTTTCTGGCAAAACCATCTCTGGCGAGC ACGGCTCGACAGCAATGGTGTCTACAACGTTACCTCCGAGTCCAGCTCGAGCGCATGAGTGTCTACT TCAACGAGGTATGCCTTAACAGTCAATGCCAATAATCCACAAGCTCACACAACCTAGGCCTCTGGCAACA AGTATGTTCCCGAGCCGCTCTCGTCAATCTTGAGCCTGGTACCATGGAGTCTTTTTTTGGTTCCGCCAGGA CGTCAAGCTCTTCCGTCCCGACAACCTTCGTTTTTCGGTCAAGTCCGGTGTGGAACAACCTGGGCCAAGG TCACTA

Table 3. Drastic temperature fungi isolated from cereal seeds

The name of the fungi	Temperature, °C Min	Temperature, °C Optimum	Temperature, °C max
<i>Rhizopus oryzae</i> Went & Prins. Geerl.	-5-(+)5	15-20	25-35
<i>Fusarium proliferatum</i> (Matsush)			
Nirenberg ex Gerlach & Nirenberg.	6-8	26-28	32-37
<i>Aspergillus tubingensis</i> Mosseray.	3 - 5	15-25	30-45
<i>Aspergillus flavus</i> Link	3 - 5	15-27	30-47
<i>Penicillium expansum</i> Link.	5	15-20	25-35

double, triple or improper verticillated branches, 300-55E11-22, and are light-brown, and have many oil drops, and are batched (by 2-3 in a batch), and rarely, they go from rhizoids as single ones. The conidia are sphere-shaped, and their diameter is 100-250 μm , and their color is brownish red. The column is semisphere-shaped, and its diameter is 44-77E33-66 μm , and its color is brown. The sporangiospores are deformed sphere-shaped, angular, and are like wide ellipsoid, sized 4.4-8.8E4.4-7.7 μm , and their color is

brownish red, muddy-olive, with strongly lineolated and stroke-like shell. The chlamidiospores are sphere-shaped, and are like wide ellipsoids, ellipsoid cylindrical, and their diameter is 44-55 μm . The zygospores are sphere-shaped, and sometimes, they are squeezed from sides, and their diameter is 132-165 μm , and their color is dark-brown, and have large warts. The copulating spurs have the same size. The fungal colonies growing on Czapek's medium are dense- or loose tomentous, and have well-developed aerial

**Fig. 1.** *Aspergillus flavus***Fig. 2.** *Aspergillus tubingensis***Fig. 3.** *Penicillium expansum***Fig. 4.** *Fusarium proliferatum*

mycelium, and later, they become light-yellow or brownish-gray

The sequencing of β -tubulin with reference primers was impossible, therefore, additionally, ITS-areas of ribosomal operon were sequenced. According to the findings of BLAST-analysis, it is the base sequence with 100% overlapping and 100% similarity, and it belongs to genus of *Rhizopus*, specie of *Rhizopus oryzae* Went & Prins. Geerl.

In 2013-2014, fungi *Aspergillus tubingensis* from berries of various agroclimatic areas of Spain⁸, and Argentina vineyard soils were sampled and studied⁹. The sclerotia formed by *Aspergillus* of section Nigri was taken from a population of one and the same field in North Carolina, USA, and were identified as *A. tubingensis* based on genealogical analysis. It was shown that ascospores from *A. tubingensis* differ from species of section of Flavi by net-like pattern of ascospores and presence of two ridges forming equatorial trench. Sexual reproduction in *A. tubingensis* may be useful for production of ferments and organic acids by using recombination mediated gen engineering for industrial strains¹⁰.

In 2013, it was identified in the seeds of grain corns in Almaty Region of Kazakhstan.

The fungal colonies are pinkish-gray and reddish-brown gradually changing to black. The conidial holders are 200-400E7-10 μ m, and diameter of final bubble is 20-60 μ m. The sterigmas are tristichous: primaryones is 20-30 μ m, secondary ones are 6-10 μ m, and the conidia are round, and their diameter is 2.5-4 μ m (Figure 2).

The sequencing ITS-areas of ribosomal operon and β -tubulin gene of fungi of genus of *Aspergillus* was deposited in GenBank. The findings of BLAST-analysis: overlapping 100%, similarity 99%.

The findings of identification showed that the base sequence belongs to specie of *Aspergillus tubingensis* Mosseray.

Among all the species, which produce blue mold, *Penicillium expansum* is the most widespread and virulent one. Patulin is a micotoxin, and it is produced by various strains of specie *Penicillium expansum*, which are the most widespread. The micotoxin has toxic effect on animals, and some of adverse effects include reproductive toxicity and adverse influence on

endocrine system of the animals.

Morphologically, the species differ by their brushes as spore-forming structure, which produces long green chains of unicellular spores of philiada. They are well known as affecting agents, and are used for production of antibiotics such as penicillin and griseovulfin, and at the same time, produce toxic metabolites (micotoxines) such as ochratoxin A, patulin and penitrem A in foodstuffs and grain, and they play important role in production of Camember and blue cheeses.

The sequencing of β -tubulin gene and ITS-areas was deposited in GenBank. According to the findings of BLAST-analysis: with 100% overlapping and 100% similarity belongs to genus of *Penicillium*, specie of *Penicillium expansum* Link (Figure 3).

Fungi of specie *Fusarium* live as saprophytes or parasites on various plants. From 2007 through 2009, in United States, the isolates of *F. proliferatum* were tested for pathogenicity on soy plants grown in hothouse.

In a series of tests performed in 2008 and 2009, *F. proliferatum* was identified in the plantations in Malaysia¹¹, and its main evidences are round brown lesion with sporodochias, and formations of white mycelium on the surface of the lesion.

Fusarium proliferatum (Matsushima) Nierenbergis known all over the world as a moderately aggressive agent for a few plants¹². In addition, the agent may live as endophyte without any external evidences of disease of host plant.

Typical are dark-blue, round or egg-shaped perithecia, and their surface is slightly rough, sized 0.2-0.3E0.2-0.4 μ m. The bags are cylindrical, and flask-shaped, sized 81-15E9-18 μ m, with 4-6, or rarely 3, single- or unclearly rowed elongated elliptical ascospores with one wall, sized 10-24E4-9 μ m. In the conidial stage, macroconidia are formed in sporodochias or pionnotes. They are slim, awl-shaped, slightly sickle-shaped or almost linear, narrowed from both sides, and with a leg at the base, with 3-7 walls. The chlamidospores are absent. Microconidia are grouped in chains or false heads. The aerial mycelium is powdery, white, whitish-pink, or yellow (Figure 4).

The sequencing of β -tubulin gene was deposited in GenBank under No. KJ938409.

The findings of BLAST-analysis showed

that the base sequence has 100% overlapping and 100% similarity, and belongs to genus of *Fusarium*, specie of *F. proliferatum* (Matsush) Nirenberg ex Gerlach & Nirenberg.

The most important environmental factor that determines the life activity of fungi is temperature. Development of fungi is possible only at certain temperatures, which are different for different species and groups of organisms. For each type of fungus, there are three temperature ranges (cardinal temperatures) within which they are able to develop: optimal, minimum and maximum.

Thus, when the maximum and minimum temperature limits may still develop mild mushroom (slow growth and reproduction), and beyond it stops completely Temperature limits for certain types of fungi shown in Table 3

The above drastic temperature can be shifted in one direction or another under the influence of the conditions of existence of the fungus.

CONCLUSION

Since recently, much attention has been paid to mold fungi, which both change nutritional value of grain, and produce toxic substances. Such species as *Penicillium expansum* are pathogenic for grain, and decrease its germinating capacity, or cause rootlets to under-develop. In addition, they produce carcinogenic metabolite patulin, which is hazardous neurotoxin for hosts. The content of patulin in foodstuff is a problem of healthcare.

In 2013, *Penicillium expansum* was isolated from wheat grains taken from the granaries of Almaty Region of Kazakhstan. Its morphological features and genetic sequence were studied. According to the findings of genetic analysis, with 100% overlapping and 100% similarity, it belongs to genus of *Penicillium*, specie of *Penicillium expansum* (Link).

Aspergillus flavus, this species can be defined by its ability to produce aflatoxins.

Rhizopus >rCz05 is fungus, which lives in the world of dead organic substance.

The strains of *Rhizopus* including R. >rCz05 are used (mainly in Asia) in food industry, and in general its products are recognized as safe.

The sequencing of α -tubulin gene with reference primers failed, therefore, additionally,

ITS-areas of ribosomal operon were sequenced. The base sequence showed 100% overlapping and 100% similarity, and belongs to genus of *Rhizopus*, specie of *Rhizopus oryzae* (Went & Prins. Geerl).

By its morphological signs, *Aspergillus tubingensis* is similar to *Aspergillus Niger*. Therefore, previously it was difficult to identify the specie by morphological characteristics. To determine specie of *Aspergillus tubingensis*, sequencing was performed. The base sequence of gene belongs to specie *Aspergillus tubingensis* Mosseray.

Fusarium proliferatum is the main source of micotoxins in foodstuff and fodders and a result these organisms pollute agricultural products, in particular, grain crops. The prolonged consumption of the food polluted with thetoxins may be carcinogenic, particularly, causes esophageal cancer. *Fusarium* colonizes a wide range of host plants. In 2013, in Almaty Region of Kazakhstan, it was sampled from the seeds of barley.

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