

Biological specifics of some species of fungi on seeds of grain crops

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Abstract. The author pays special attention to fungi developing in stored grain. Seeds of grain crops *Triticum aestivum* L., *Avena sativa* L., *Hordeum vulgare* L., *Zea mays* L., *Oryza sativa* L., *Sorghum vulgare* Pers., *Panicum miliaceum* L.) has been selected in granaries of four regions (Talgar, Ili, Karasaiski, Zhambylski) of Almaty district. Field fungi that are pathogenic fungi of *Helminthosporium*, *Fusarium*, *Alternaria*, *Macrosporium*, *Penicillium*, *Aspergillus*, *Mucor* and *Rhizopus* genera have been found. These fungi affect storing, quality and safety of seeds. In general, species composition and biological specifics of fungi of *Alternaria*, *Helminthosporium*, *Fusarium*, *Macrosporium* genera that affect grain crops in Kazakhstan have been studied insufficiently. [Abdrassulova Z.T., Kuzhantaeva Z.Z., Anuarova L.E. **Biological specifics of some species of fungi on seeds of grain crops.** *Life Sci J* 2014;11(6s):79-82] (ISSN:1097-8135). <http://www.lifesciencesite.com>. 13

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Introduction

Edaphytic (plant pathogenic) microflora consists of microorganisms capable to permeate in inner parts of plants. This group of microorganisms requires special attention because their development in conditions of grain keeping may cause significant damage to crop due to suppression, destruction of plants and decrease of produced grain quality [1]. Some strains of fungi generate 6-7 toxic compounds [2]. Species of *Helminthosporium*, *Fusarium*, *Alternaria* and *Macrosporium* genera belong to this group. Taken together these fungi may cause tremendous economic loss for agricultural production, loss of food for consumption and serious, often mortal diseases of people and animals [3]. Toxicity of metabolites of *Alternaria* species for different organisms including bacterium, birds and mammals has been proved by numerous researches [4;5;6]. Pathogenicity of *Fusarium* strain was proved by pathogenicity tests. The information has been gathered from results of sowing of winter wheat in different climate conditions in Ukraine [7]. Supply of high quality seeds for production sowing in accordance with standards' requirements for seeds irrespective of weather condition is pressing problem now.

Method

Seeds of grain crops: wheat, barley, corn, rice, sorghum, millet has been selected selected in granaries of four regions (Talgar, Ili, Karasaiski, Zhambylski) of Almaty region. Selected seed of grain crops were kept in dampening chamber. Two layers of filter paper were laid on the bottom of Petri dishes and sterilized in exsiccator under the temperature of 130° C for an hour. Pipettes were also sterilized. Sterilized paper in Petri dishes

was moistened for thorough wetting. Seeds were sterilized with 96% spirits for 1 minute. Then seeds were put on wet filter paper in Petri dishes in 1.5 – 2 cm from one another. Petri dishes were kept under the temperature 21° C. Growth and development had been controlled daily. On 7th day spore formation was observed on seeds. Features and the character of growing of mycelium defined the specie of fungi. Then these fungi were subcultured in Petri dishes with nutrient Czapek's medium separately by specie.

Main body

Causative agents of fungoid aetiologically were found in seeds. These are *Helminthosporium sativum*, *Fusarium graminearum*, *Alternaria tenuis*, *Macrosporium commune*, *Mucor mucedo*, *Penicillium* sp, *Aspergillus glaucus*, *Rhizopus nigricans*. Affection of *Macrosporium commune* dominated other sorts of fungi.

Helminthosporium sativum develops on crop seeds (wheat, barley, oats). On nutrient Czapek's medium *Helminthosporium* initially forms rich grey mycelium, than aerial mycelium became dark grey, substrate mycelium became black. Spore formation was weak or there was no spore formation at all. This specie is facultative parasite. In droughty regions of Almaty district seeds as a source of wheat *Helminthosporium* rot, barley, oats has no importance. The main agent of root rot is *Helminthosporium sativum* (see Fig. 1).

Helminthosporium sativum causes grain affection that manifests itself as hidden infection or as black germ of darkening. Black coating of fungi can be easily seen on seeds. Affected seeds lose germinating capacity.

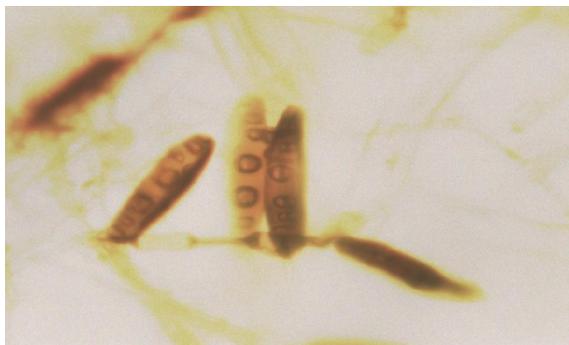


Fig. 1. Helminthosporium sativum

Fungi of Fusarium specie are undemanding for environmental conditions and extremely plastic. In foothills of Almaty district we extracted pure culture Fusarium graminearum from millet seeds. On 3rd day after plating on Czapek's medium white, whitish spiderweb like mycelium develops and pink, orange and violet pads develop on seeds. These pads consist of microconidium spore formation of fungi. Microconidiums have developed in aerial mycelium on simple conidium bearers as accumulations between mycelium floccuses. Microconidiums are unicellular, oval, egg-shaped, ellipsoid. Microconidiums' spore formation are seldom developed, macroconidiums are elongated, multicellular with 3-5 interstices inwardly curved from both sides. Mycelium of these fungi is mainly white, pink-white, pink-violet.

Development of Fusarium graminearum in laboratory conditions has been going on in storing seeds for six years and repeated infections occurred in wet conditions (15%).

Alternaria tenuis (*A.alternata*) was extracted from wheat, barley, oats. *A.tenuis* is most intensively propagating on cereals. Research of *A.tenuis* specie has shown that minimum number of conidium cells is 3, maximum is 14. In most cases total sum of cells in conidium was 16 that refer to maximum number of cell capable of forming in it in case all the interstices have developed. The most persistent features of *A.tenuis*: form, conidium dimentions, number of interstices in conidium, length, width and form of conidium rostell are developed on natural substrate. Secondary features that should be also taken into consideration are color of conidium, conidium's outer shell structure, size, form and the number of interstices (see Fig. 2).

In the form of mycelium *A.tenuis* may remain preserved in grain stored in dry conditions and in low temperature for many years. In normal conditions of bread-grain and fodder grain storage *A.tenuis* was destroyed on 8th year of storage and in laboratory conditions viable mycelium remains preserved longer than 8 years [8].



Fig. 2. Conidium Alternaria tenuis on wheat seeds

Cultivating *A.tenuis* in nutrient Czapek's medium produced cotton-like, intensively growing dark-grey colony. For conidium development on conidium bearers low temperature and darkness are required. So weather conditions impact may accelerate or slow *A.tenuis* shift from one development stage to another, accelerate or slow life cycle of pathogen and thus influence pathogenic mechanism caused by *A.tenuis*.

A.tenuis is a saprophyte but may show parasitical features if a plant is hardly weakened.

Results of artificial infection of germinants of grain crops with conidium *A.tenuis* extracted from *Triticum aestivum* seeds under the temperature +20°C are shown in Table 1.

As Table 1 shows *A.tenuis* development on vegetating leaves goes on weakly because they have limited amount of nutrients that exist on leaves' surface and development is suppressed by specific substances produced by plants. Weakening of a plant on separate leaves gives *A.tenuis* a possibility to develop more intensively.

In nature infection may spread from leaves to other organs of a plant and may reach ripening seeds. Fungi in the form of spores are moved with seeds on new places of cultivating and in favorable conditions of moisture and temperature may affect a plant in germinating stage.

M.V. Gorlenko, E.A. Chinnova and L.M. Levkina (1957) studied forms of *A.tenuis* extracted from different substrates and showed that specie *A.tenuis* is physiologically non-uniform. Forms insulated from germinating seeds that affect young crops are closer to parasites. On the contrary, forms extracted from dead substrate fully manifest their saprophyte nature. M.V. Gorlenko considered *A.tenuis* to be species evolving to parasitism [9].

Table 1. Specifics of infection of germinants with conidium *Alternaria tenuis*, extracted from *Triticum aestivum* seeds

Germinants of grain crops	Characteristics of germinants of grain crops	Extent of injury
1	2	3
Triticum aestivum	Vegetating above-ground organs of germinants	No infection
	Leaf detached from stem	Conidium has abundantly developed on big yellowed spots on leaves
Hordeum vulgare	Vegetating above-ground organs of germinants	No infection
	Leaf detached from stem	Conidium has abundantly developed on big yellowed spots on leaves
Avena sativa	Vegetating above-ground organs of germinants	No infection
	Leaf detached from stem	Conidium has abundantly developed on big yellowed spots on leaves
Sorghum vulgare	Vegetating above-ground organs of germinants	No infection
	Leaf detached from stem	Conidium has developed only around inoculums

D.N. Terevnikova-Babayana and L.S. Zakiyan (1969) showed that observed in nature strains of parasite and saprophyte species *Alternaria* (*Alternaria tenuis* Nees, *Alternaria chartarum* and others) are significantly variable in parasite features and adapt rapidly to strong affection of plants where they have developed on and extracted from. For example *A.tenuis* strain extracted from tomatoes affects tomatoes in highest degree. Degree of affection of pepper and potatoes is lower and the lowest degree of affection is for aubergines. Being processed through potatoes fungi becomes capable to affect potatoes in highest degree and lower – tomatoes and pepper [10].

In case of grain material storing under required moisture lower than 14% population of microorganisms reduces and ratio of different species of microorganisms changes. But infection by field fungi *Alternaria*, *Helminthosporium*, *Fusarium*, *Macrosporium*, *Rhizopus* is retained in seed mass for several years [8].

Macrosporium is close to *Alternaria* by life pattern and morphology (conidium are also many-cell, dark colored). But conidium does not form chains and have no rosette or is elongated apical cell. *Macrosporium commune* was observed on seeds of wheat, barley and oats. Typical feature of this species is structure and conidium bearers ability to proliferate. The cell of conidium bearer that carries conidium is inflated near its top. After forming of first conidium conidium bearer keeps on growing through inflated top moving first conidium aside. After a certain period of growth the top of conidium bearer became inflated again, next conidium forms that moves aside conidium bearer by new emergence. Three or more proliferations usually take place. Young conidium *Macrosporium commune* have round, egg or reverse egg form (matured conidium

retain this form or became wide elliptical) or slightly elongated (see Fig).

**Fig. 3. *Macrosporium commune* conidium**

Macrosporium commune is cosmopolite plant, it may be found on plant residues, on seeds of wheat, barley and oats. The fungi may remain alive for several years. In dampening chamber seeds of wheat, barley and oats affected by *M.commune* are being covered with dark grey mycelium and conidium of fungi became black and does not germinate. It is mostly *saprophyte and facultative parasite*. It is the cause of wheat, barley and oats seeds death.

We have carried out experiments on artificial infecting of vegetative organs of germinants of grain crops with conidium *M.commune* in laboratory conditions (see Table 2).

M.commune is representatives of filamentous fungi that may be found in soil. It also abundantly populates cultivated soils.

Table 2. Specifics of infection of germinants with conidium *M.commune*, extracted from *Triticum aestivum* seeds

Germinants of grain crops	Characteristics of germinants of grain crops	Extent of injury
1	2	3
Triticum aestivum	Vegetating organs of germinants	Conidium has abundantly developed on infected roots
	Leaf detached from stem	No infection
Hordeum vulgare	Vegetating organs of germinants	Conidium has abundantly developed on root
	Leaf detached from stem	Conidium has abundantly developed on yellowed leaf
Avena sativa	Vegetating organs of germinants	Conidium has abundantly developed on roots
	Leaf detached from stem	Conidium has abundantly developed on yellowed spots
Sorghum vulgare	Vegetating organs of germinants	No infection
	Leaf detached from stem	Conidium has developed on yellowed leaf around inoculums
Panicum miliaceum	Vegetating organs of germinants	No infection
	Leaf detached from stem	Conidium has abundantly developed on yellowed leaf
Zea mays	Vegetating organs of germinants	No infection
	Leaf detached from stem	Conidium has abundantly developed on yellowed leaf

So in experiments on vegetating organs of grain crops germinants infection with *M.commune* fungi affected roots and leaves.

Violation of required storage conditions quality and quantity of grain microflora compound drastically changes: it reduces up to full elimination

of the quantity of bacteria (*Pseudomonas* sp., *Bacterium herbicola*) and field fungi, amount of moulds increases that relate mainly to genera *Penicillium*, *Aspergillus*, as well as *Mucor*, *Rhizopus* [11]. Development of these bacteria on nutrient Czapek's medium was not observed.

Conclusion

Studying of grain crops seeds from granaries of four regions of Almaty *Helminthosporium sativum* fungi species were found on seeds of wheat, barley and oats, *Fusarium graminearum* on seeds of millet, barley and oats, *Mucor mucedo*, *Penicillium* sp, *Aspergillus glaucus*, *Rhizopus nigricans* on all seeds. So storing infection on seeds is reliable way to preserve and wintering of agent in nature.

On Czapek's medium *Fusarium graminearum* produces white cotton-like mycelium colonies with pink and violet pads with micromycelium and rarely macroconidium spore bearings. On Czapek's medium *Alternaria tenuis*, *Macrosporium commune* produce mycelium colonies initially grey than dark grey color with specific conidium.

A. tenuis extracted from wheat seeds affected detached leaves of grain crops (wheat, barley, oats) but vegetating leaves were not infected. *Macrosporium commune* extracted from *Triticum aestivum* (temperature 20 °C) affected roots of vegetating getminants of *Triticum aestivum*, *Hordeum vulgare*, *Avena sativa*. There is no data about root infection by species of genus *Macrosporium* in literature.

On Czapek's medium storage fungi *Mucor mucedo*, *Rhizopus nigricans*, *Penicillium* sp, *Aspergillus glaucus* produce mycelium colonies with abundant spore bearing.

Alternaria tenuis affects only weak vegetating organs of grain crops and *Macrosporium commune* affects roots and weak leaves of grain crops.

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References

1. De Lucca A.J., 2007. Review. *Rev Iberoam Micol.* 24(1):3-13.
2. Repedkiene J, L. Levinskaite, A. Paskevicius and V. Raudoniene, 2013. Toxin-producing fungi on feed grains and application of yeasts for their detoxification. *Pol J Vet Sci.* 16(2):391-3.
3. Zukiewicz-Sobczak W.A., G. Cholewa, E. Krasowska, J. Chmielewska-Badora, J. Zwoliński and P. Sobczak, 2013. Rye grains and the soil derived from under the organic and conventional rye crops as a potential source of biological agents causing respiratory diseases in farmers. *Postepy Dermatol Alergol.* 30(6):373-80.
4. Müller M.E., I. Steier, R. Köppen, D. Siegel, M. Proske, U. Korn and M. Koch, 2012. Cocultivation of phytopathogenic *Fusarium* and *Alternaria* strains affects fungal growth and mycotoxin production. *J Appl Microbiol.* 113(4):874-87.
5. Scott P.M., W. Zhao, S. Feng and B.P. Lau, 2012. *Alternaria* toxins alternariol and alternariol monomethyl ether in grain foods in Canada. *Mycotoxin Res.* 2012 Nov;28(4):261-6.
6. Kosiak B., M. Torp, E. Skjerve and B. Andersen, 2004. *Alternaria* and *Fusarium* in Norwegian grains of reduced quality — a matched pair sample study. *Int. J. Food Microbiol.* 93: 51—62
7. Kriuchkova, L.O., 2013. Micromycetes related with wheat affections in different regions of Ukrain. *Microbiology*, 75(4): 59-68.
8. Abdrassulova, Zh.T., 2013. Specifics of preserving vitality of species fungi affecting seeds of grain crops. International Conference of Students, Magistrants and Young Scientists "Lomonosov".
9. Gorlenko, M.V., E.A. Chinnov and L.M. Levkina, 1953. Reports of the Academy of Sciences of the USSR,.
10. Teterevnikova-Babayana, D.N. and L.S. Zakiyan, 1969. Proceedings of the Session of Transcaucasian Council on Coordination of Scientific and Research on Plants Protection.
11. Bostanova, A.M., 2009. Fungi on seeds of grains and legumes crops during storage. *Biological Diversity and Sustained Development of Nature and Science*, pp: 23-26.