

**Brief information about the project**

Name of the project	AP14972820 «Study of the biodiversity of entomopathogenic fungi common in the agrocenoses of southern Kazakhstan»
Relevance	The main idea of the project is to search for insect corpses with signs of mycosis and to select soil samples to isolate new isolates of entomopathogenic fungi. The study of the biodiversity of entomopathogenic fungi common in the agrocenoses of southern Kazakhstan allows us to identify several new isolates adapted to arid conditions. The creation of an extensive collection of entomopathogenic fungi will significantly advance issues related to the development of new biological insecticides.
Purpose	The purpose of this project is to study the biodiversity of entomopathogenic fungi common in the agrocenoses of southern Kazakhstan for use as a biological resource for controlling the number of pests of agricultural plants.
Objectives	<ol style="list-style-type: none"><li>1. Literature review.</li><li>2. Isolation of new isolates of entomopathogenic fungi from soils of agrocenoses of southern Kazakhstan.</li><li>3. Genetic identification of new isolates of entomopathogenic fungi. A PCR analysis of the TEF locus will be performed.</li><li>4. Assessment of morphological and cultural properties, thermal tolerance, and productivity of new isolates. During laboratory experiments, experiments will be laid to evaluate the radial growth of selected isolates in a wide range of temperatures, humidity and to assess their productivity.</li></ol>
Expected and achieved results	New isolates of entomopathogenic fungi from soils of agrocenoses of southern Kazakhstan will be isolated. The morphological and cultural properties, thermal tolerance and productivity of the new isolates will be evaluated. Genetic identification of new isolates of entomopathogenic fungi will be carried out. A PCR analysis of the TEF locus will be performed. It is planned to publish 2 (two) articles in journals from the first three quartiles by impact factor in the Web of Science database or having a CiteScore percentile in the Scopus database of at least 50. The results obtained within the framework of the ongoing project can be used by research institutions and universities of biological and agricultural profiles. The obtained new strains can be used in the future in the creation of a biological product against harmful insects.
Research team members with their identifiers (Scopus Author ID, Researcher ID, ORCID, if available) and links to relevant profiles	Abdukerim Rauza, PhD, <a href="https://orcid.org/0000-0002-4745-1437">https://orcid.org/0000-0002-4745-1437</a> , <u>Scopus Author ID: 57201500323</u>
List of publications with links to them	
Patents	-

<p><b>Section of the calendar plan: № 1.</b> <b>Literature review.</b></p>	<p>January 2023</p>	<p>According to the expected result of the contract calendar plan: The work was carried out with literature in the library and in available Internet resources, using the keywords "Beauveria" and "Metarhizium". The search was performed using the following resources: Web of Science (<a href="http://www.webofknowledge.com">http://www.webofknowledge.com</a>), Scopus (<a href="https://www.scopus.com">https://www.scopus.com</a>), CAB Abstracts (<a href="https://www.cabdirect.org">https://www.cabdirect.org</a>) and Google Scholar (<a href="https://scholar.google.com">https://scholar.google.com</a>), CyberLeninka, eLibrary Library, PubMed, SciFinder, AGRIS - International System for Agricultural Science and Technology, Biodiversity Heritage Library, Bookshelf (<a href="http://www.ncbi.nlm.nih.gov">www.ncbi.nlm.nih.gov</a>), Elsevier Publishing House (on the ScienceDirect platform), MDPI (Multidisciplinary Digital Publishing Institute), Nature Journals, Taylor &amp; Francis Publishing House, Wiley Publishing House (Wiley Online Library), Library Hub Discover, Department of the State Scientific Research Institute of the Siberian Branch of the Russian Academy of Sciences, Library of Congress, National Electronic Library (NEB), The Russian State Library (RSL), Library of the Russian Academy of Sciences (BAN). More than a hundred scientific articles on the isolation of entomopathogenic fungi have been studied in detail. After reviewing them, we concluded that recently, the issue of isolating new isolates from different types of soils has been actively studied all over the world. It is planned to use this data for further work.</p>
<p><b>Section of the calendar plan: № 2.</b> <b>Literature review.</b></p>	<p>February 2023</p>	<p>According to the expected result of the contract calendar plan: Work has been carried out with literature in the library. Scientific articles on the study of morphological features of entomopathogenic fungi are analyzed.</p>
<p><b>Section of the calendar plan: № 3.</b> <b>Literature review.</b></p>	<p>March 2023</p>	<p>According to the expected result of the contract calendar plan: Work has been carried out with literature in the library. Studies on the influence of abiotic factors on the spread of entomopathogenic fungi are considered.</p>
<p><b>Section of the calendar plan: № 4.</b> <b>Literature review.</b></p>	<p>April 2023</p>	<p>According to the expected result of the contract calendar plan: Work has been carried out with literature in the library. Scientific articles on the study of the pathogenic of entomopathogenic fungi on different groups of harmful insects are analyzed.</p>
<p><b>Section of the calendar plan: № 5.</b> Search and isolation of new isolates of entomopathogenic fungi from soils of</p>	<p>May 2023</p>	<p>According to the expected result of the contract calendar plan: To conduct route surveys of agroecosystems in the southern region, a list of farms engaged in crop production in the Turkestan region has been compiled. The first route trip was to the farms of the Sauran district: Kartex Agro – 350 hectares</p>


<p>agrocenoses in southern Kazakhstan.</p>		<p>grow cotton; Azim Ata – 352 hectares grow cotton; Turan – 413 hectares grow cotton; IS Zhangir – 235 hectares grow cotton, wheat, corn; YSD-agro – 430 hectares grow cotton, wheat, corn; Nabihan – 954 ha cotton, wheat, corn are grown; Mehman – 435 ha cotton, wheat, corn are grown; IS-Zhangir – 250 hectares grow wheat; Koskargan – 290 hectares grow cotton; Asha – 484 hectares grow cotton, wheat, corn; Husan-ata – 400 hectares grow wheat; Irisbai-ata – 230 hectares grow cotton; Yntymak agro – 480 hectares grow alfalfa; Saidmurat – 210 hectares grow cotton, corn; Yugnak – 1,433 hectares cotton is grown; Dinmuhamed – 1,632 hectares cotton, wheat, corn, alfalfa, garden are grown; Turkestan- agro – 1,075 hectares wheat, corn, alfalfa are grown; Agro zhetik – 1,342 hectares wheat, corn, alfalfa are grown. Soil samples were taken from different points of each site at 10 m from each other using a sampler to a depth of 20 cm in alcohol-sterilized ziplock plastic bags. The soil is ground mechanically, sifted using a sieve with holes with a diameter of 2 mm to clean it from foreign materials, then sent to storage at a temperature of 4 °C for further work.</p>
<p><b>Section of the calendar plan: № 6.</b> Search and isolation of new isolates of entomopathogenic fungi from soils of agrocenoses in southern Kazakhstan.</p>	<p>June 2023</p>	<p>According to the expected result of the contract calendar plan: According to the list, the following visits were to the farms of the Otyrar district: Asia agro – 1000 hectares grow wheat; Ontutik-Altayr – 250 hectares grow corn; Kazyna 2030: 500 hectares grow alfalfa; Otyrar sut-et – 1050 hectares grow corn, vegetables, and gardens; and Akkum: 5141 hectares are engaged in gardening. Further route surveys were carried out in the farms of the Zhetisai district: Nuraly Zhol Zhh – 497 hectares grow cotton, garden; Arsenal garden – 592 hectares grow cotton, garden; Bakhytzhan – 520 hectares grow cotton; Ketebai – 225.5 hectares grow cotton; Ali – 650 hectares grow cotton; Maktaly Zher – 388 ha cotton is grown; Nurasyl – 250 ha cotton is grown; The next trip was to the farms of the Ordabasy district: ConSaMe – 456 ha wheat is grown; Migrazh-B – 400 ha wheat is grown; 1-mamyr – 497 ha wheat is grown; Mator – 200 ha wheat is grown; Baisal – 200 ha wheat is grown wheat; Abduali – 238 ha wheat is grown; Shubar – 300 ha wheat is grown; Nuraly – 271 ha wheat is grown; Ramadan Agro – 723 ha cotton is grown; Zhandos – 335 ha wheat is grown; Bimukhanov SH – 200 ha wheat is grown; Torebek-Nur – 200 hectares grow wheat; Yntymak – 600 hectares grow wheat; Kolbai – 750 hectares grow wheat; Tokbolat – 600 hectares grow wheat; Ykylas temir – 500 hectares grow wheat; Ak Nur – 200 hectares grow wheat; Kalash – 224 hectares grow wheat; Batsu Agro – 400 hectares grow</p>

		wheat; Sabyr – 336 hectares grow wheat; Zhanuzak – 250 hectares grow wheat; Badam – 1898 hectares grow wheat. Soil samples were taken from different points of each site at 10 m from each other using a sampler to a depth of 20 cm in alcohol-sterilized ziplock plastic bags. The soil is ground mechanically, sifted using a sieve with holes with a diameter of 2 mm to remove foreign materials, and sent to storage at a temperature of 4 °C for further work.
<p><b>Section of the calendar plan: № 7.</b> Search and isolation of new isolates of entomopathogenic fungi from soils of agroecosystems of southern Kazakhstan.</p>	July 2023	<p>According to the expected result of the contract calendar plan:</p> <p>According to the list, the following visits were to the farms of the Sairam district: Zher Ana Birlik – 217.4 hectares grow wheat; Dikhan and K – 655 hectares grow wheat, safflower, potatoes; Karasai Batyr-2 – 200 hectares grow wheat, alfalfa; Otemis ata – 455 hectares grow wheat, safflower; Poshan ata – 440 hectares grow wheat; Samat SHT – 500 hectares grow wheat; Akbura – 220 hectares grow wheat; Akbulak and K – 367 hectares grow wheat; Isamitdin A – 250 hectares grow barley; Omir-Darkhan – 565 hectares grow wheat; Yrysdaulet – 236 hectares grow wheat; MTC COMPANY – 212 hectares grow wheat, safflower; Taskeshu – 417 hectares grow wheat, safflower; Zholyimbek – 207 hectares grow safflower; Mir Alisher Navoi – 380 hectares grow wheat, barley; Navruz XXI – 572 hectares grow wheat, alfalfa; Mehnat XXI asr – 551 hectares grow wheat, alfalfa; Mehnatobod – 347 hectares grow wheat, barley, garden; Agroservice Karamurt – 270 hectares grow wheat; Enbekshi 21 – 283 hectares grow wheat; Ibrahim Karasu BI – 364 hectares grow wheat, barley, alfalfa; Altyn tobe – 342 hectares grow wheat, alfalfa; Akarys and SO – 355 hectares grow wheat; Nur-17 – 150 hectares grow wheat; Madi-7 – 97 hectares grow wheat; Kyzyl Kyshlak – 232 hectares grow wheat, alfalfa; Mankent -1 – 951 hectares grow wheat, alfalfa; O.Kurbanov – 919 hectares grow wheat, alfalfa, grapes; Kazybek – 200 hectares grow wheat, safflower; Nyshanbek – 2,186 hectares of wheat are grown; Nur-Kol – 3,910 hectares of wheat are grown. Soil samples were taken from different points of each site at 10 m from each other using a sampler to a depth of 20 cm in alcohol-sterilized ziplock plastic bags. The soil is ground mechanically and sifted using a sieve with holes with a diameter of 2 mm to clean them from foreign materials and sent to storage at a temperature of 4 °C for further work.</p>
<p><b>Section of the calendar plan: № 8.</b> Search and isolation of new isolates of</p>	August 2023	<p>The final route trips were to the farms of Saryagash district: Myrzakul-Products – 16.3 hectares grow wheat; Temirshi-TEK – 500 hectares grow wheat; Kazygurt-Baiterek – 500 hectares grow safflower;</p>

<p>entomopathogenic fungi from soils of agrocenoses of southern Kazakhstan.</p>		<p>Saken firmasy – 709 hectares grow wheat; Emesh-Ata – 200 hectares grow wheat; Nurtas and K – 457.8 hectares grow wheat; T.N.N.-K – 321 ha grow barley; Kuralbek-Zh – 283.7 ha grow wheat; Beybars-B – 100 ha grow wheat; Kural-2030 – 250 ha grow wheat; D-Amir and K – 462 ha grow wheat; Saryagash-2 – 332 ha grow wheat; Kulamkha – 224 hectares grow wheat; Nurbakhyt – 100 hectares grow wheat; Taskotan – 800 hectares grow wheat; Baktybai-Sh – 300 hectares grow wheat; Zainil ata – 500 hectares grow wheat; Bakytzhan-E – 350 hectares grow wheat; Iztai ata – 250 hectares grow wheat; Nur Astan-Kz – 440 hectares grow wheat; Kamshat – 340 hectares grow wheat; Laura – 228 hectares grow wheat; Salihat – 556 hectares grow wheat; Rahat-Nurbolat – 900 hectares grow wheat; Astyk-7 – 500 hectares grow wheat; Algabastau – 135 hectares grow wheat; Kosagash-2 – 100 hectares grow wheat; Kobdi – 263 hectares grow wheat; Aubakir – 200 hectares grow wheat; Mehri – 500 hectares grow wheat; Tanabayev Nurgali – 100 hectares grow wheat; Shugyla – 219 hectares grow wheat; Omar ata – 392 hectares grow wheat; Betburys – 400 hectares grow wheat; Dauren-S.K. – 300 hectares grow wheat; Shaikhan 2030 – 1111 hectares grow wheat; Indira 2005 – 125 hectares grow wheat; Aizhol – 118 hectares grow wheat; Sat – 200 hectares grow wheat; Zainil – 827 hectares grow wheat; Kanatbek – 150 hectares grow wheat; Ordash Nury – 157 hectares grow wheat; Krasnivodopad – 607 hectares grow wheat; That harvest – 400 hectares grow wheat; Akniyet – 248 hectares grow wheat; Aganai – 140 hectares grow wheat. Soil samples were taken from different points of each site at 10 m from each other using a sampler to a depth of 20 cm in alcohol-sterilized ziplock plastic bags. The soil is ground mechanically and sifted using a sieve with holes with a diameter of 2 mm to clean them from foreign materials and sent to storage at a temperature of 4 °C for further work.</p>
<p><b>Section of the calendar plan: № 9.</b> Isolation of new isolates from soil samples.</p>	<p>September 2023</p>	<p>In laboratory conditions, isolation of entomopathogenic fungi was carried out using insect baits. <i>Tenebrio molitor</i> L larvae were used at a late stage. 250 g of the collected soil from agrocenoses was placed in a plastic pot. Five <i>Tenebrio molitor</i> L larvae with nutrition were also placed there. The pots were stored at a temperature of 25 ± 10 C and a relative humidity of ≥ 80% in the dark. The larvae were monitored daily for signs of pathology to make sure that the insects were not infected. After two weeks, dead insects were removed and placed in a humid chamber (relative humidity ≥ 80%) at a temperature of 25 ± 10 C for 7 days to promote the appearance of</p>

mycosis. When mycosis appeared, conidia were collected from the insect's surface and transferred to the Saburo nutrient medium. As a result, 173 soil samples were analyzed in laboratory conditions, in which flour crunch larvae were placed.



		
<p><b>Section of the calendar plan: № 10.</b> Isolation of new isolates from soil samples.</p>	<p>October 2023</p>	<p>Isolation of entomopathogenic fungi into a pure culture was carried out according to a standard procedure. A small piece of mycelial-spore plaque from the insect corpse was transferred with a dissecting needle to a Petri dish on a Saburo nutrient medium and then placed in a thermostat. Replanting was carried out three times to obtain a pure culture of entomopathogenic fungi. As a result, 30 isolates of entomopathogenic fungi were isolated, which were previously identified as <i>Beauveria bassiana</i> sensu lato and 2 isolates of <i>Metarhizium anisopliae</i>.</p>



<p><b>Section of the calendar plan: № 11.</b>  Determination of the effect of air temperature and humidity on the intensity of development of entomopathogenic microorganisms in surface culture under laboratory conditions.</p>	<p>November 2023</p>	<p>To determine the effect of temperature on the intensity of radial growth, entomopathogenic fungi were dispersed by injection into the center of a Petri dish with the same medium layer. The cups were placed in a thermostat with a fixed temperature (+100, +150, +200, +250, +300 and +350 C). For 29 days (starting in October), with an interval of 2 days, the colonies were measured in two mutually perpendicular directions (cross to cross) and the diameter of the colonies was determined. The repetition rate in the experiments was 4 times. The observations have shown that the most favorable temperatures for the growth of colonies of isolates of entomopathogenic fungi on the Saburo nutrient medium are +200C and +250C. There are no exceptions among the isolates of entomopathogenic fungi. With deviations from optimal temperatures, a decrease in colony growth is observed. However, several promising isolates of entomopathogenic fungi have been identified, which are able to tolerate temperature fluctuations.</p>







