INTRODUCTION

Apple orchards occupy 5,163 hectares (out of 630,000 hectares of total area) in the Khorezm, a northwestern region of Uzbekistan. Rosemary, Golden, Reinette Simirenko, semi-dwarf, and dwarf varieties are main varieties grown in the region. The varieties of Reinette Simirenko are prone to be infected with cytosporosis which is caused by a fungus of the genus Cytospora. In 1818, Ehrenberg showed that the Cytospora fungus causes ulcers in the shoots and stems of various plants. So far, studies investigated the nature of Cytospora disease in 85 species of broad-leafed and coniferous trees. The fungus was also isolated from the bark, xylem, and leaves of plants in which no symptoms of the disease were observed. Cytospora mostly affects stonefruit and seed trees. Cytospora can also affect the buds. But it mainly affects stems and shortens tree life, and also lowers the yield. In Italy, the evidence of infection by old walnut trees was reported earlier yet young plants appeared after the infection. In eastern India, chestnut trees (Castanea sativa Mill.) have been infected by natural and anthropogenic factors.

In this case, the appearance of ulcers and the burning of the leaves was observed. This is a very dangerous disease, the results of studies have shown that they belong to the Val- laceae family of the genus Cytospora.

In Uzbekistan, numerous studies addressed the cytosporosis disease, its expansion, and biological properties. Information about the cytosporosis disease and its expansion depending on the groundwater level is rare. This study fills this gap by integrating experimental field-research and statistical analysis of the collected data.

MATERIALS AND METHODS

Study area

The area is a lowland plant located in the North-Western part of Uzbekistan, along the lower reaches of the Amudarya River, between 60°C-61°C longitude and 41°C-42°C latitude, at 113-138m above sea level. The vegetation period of plants is 200–210 days. The climate is extremely continental, with an average annual precipitation of 80-90 mm. Meadow, meadow marshly, marsh-sandy typical alkali soils prevail. The climate of the region is affected by the Kyzylkum and Karakum deserts. The winter here is relatively cold: the average January temperature ranges from –8°C in the north to -2°C in the south. Summer is long and hot here, the average

ABSTRACT

This study analyzed the morphological properties of cytosporosis disease in the Reinette Simirenko variety (Malus) of apple trees and nature of its damage to the causative agents in the Lower Amudarya region (Uzbekistan). The region is characterized with dry climate and shallow groundwater tables which can effect on the spread of the disease. For determining the causes of the disease, the apple trees and the groundwater level were monitored in orchards where the cytosporosis disease is widespread. Based on this monitoring and additional correlation analysis, we found out considerable influence of the groundwater level in the expansion of cytosporosis in apple orchards.

Key Words: Reinette Simirenko, Cytosporosis, Ground-water level, Picnium, Conidium, Irrigation system

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ISSN: 2231-2196 (Print) ISSN: 0975-5241 (Online)
Received: 20.04.2020 Revised: 06.05.2020 Accepted: 18.06.2020 Published: 22.07.2020
July temperature is 28-30°C. The region is in the steppe zone, in the western part of the Khorezm oasis and in the southern part of the Aral Sea, 100 m above sea level. The relief consists of a low plain. It is the old Amudarya delta and consists of river sediments. The western and southwestern parts connecting with Karakum are covered with sand. Of the minerals, there are limestone, sand, clay, and other building materials (Fig 1).

The Amudarya is the sole source of water supply to irrigation sites and residential areas in the region. The region is equipped with a dense set of irrigation and ameliorative systems (Fig 2). In the south of the region, there are salt lakes, wetlands, and saline areas. The soil consists of alluvial deposits of the Amudarya. Meadow, meadow boggy soils are found in the river valley, and sands are in the west.

The roots of fruit trees develop well with soil moisture of 17-18%, i.e. their aboveground parts are normally provided with nutrients and moisture. With an increase in soil moisture of more than 20% or a decrease to 13-15%, the roots do not develop normally, the tree is not provided in sufficient quantities with nutrients and moisture, thin roots slowly decrease. In the Khorezm region, the proportion of irrigated agriculture reaches 100%. Soils have different degrees of salinity; in the autumn and spring months, 5200 m3 of water is required to flush soil salinity. And this leads to a change in the level of groundwater. For irrigated areas, critical groundwater levels are of practical importance, as the raising of groundwater levels to critical is the main cause of salinity.

The critical depth of groundwater is the depth at which groundwater rises through capillary tubes to the root zone of plants and soil salinity begins. To a certain extent, the depth of groundwater at which the processes of salinization and desalination are equal to zero is considered to be a critical depth. Thus, when the groundwater level rises to critical, soil salinity begins. If this depth is below a critical depth, the amount of salt in the soil decreases, and the soils are desalted.

The Khorezm region differs from other regions of the republic by its dry climate, soil irrigation methods, soil salinity, surface groundwater in some places. In the Khorezm region, there are more than 2,000 wells for measuring the level of groundwater and the degree of salinity, of which samples are taken every 10-15 days.

**METHODS**

According to the methods of Abdullaev et al. (2002) the study area comprised the province of Khorezm (Republic of Uzbekistan), an ancient irrigated oasis along the Amu Darya River and the surrounding desert. The studies were conducted in 2013-2018 in the apple orchards of the Khanka, Urgench and Khiva districts of the Khorezm region. Apple orchards were established in 2005 and diseases were observed in 2013-2018. The coordinates of the studied areas were determined using GPS and entered into the database. In these fields, cytosporosis was studied in 50 sites. In this case, 50 trees were studied diagonally in each field. A total of 2500 trees were studied in 50 areas. Samples of the bark of infected trees were taken and examined in the laboratory. The bark consisting of infected cells was taken at a size of 5 mm. To preserve conidia of bark fungi and to remove other microorganisms, the pieces were placed for several minutes in a 70% alcohol solution. Pieces of bark were removed from alcohol and, to remove residual alcohol, they were placed for an hour into sterile water at a temperature of 25°C. After conidia, the bark was taken using an inoculation loop and seeded in starvation agar. The studied culture was studied under an Optika microscope at 1200 fold magnification.

**STATISTICAL ANALYSIS**

The results were studied by comparison in the Fotohifer 2007 program. The change in groundwater level in 2013-2017 was determined by the formula

\[ E = J_1 - J_2 \]
here:
E - differences between the maximum and minimum depth of groundwater in the period 2013-2017.
J₁ - maximum depth
J₂ - is the minimum depth.

Statistical calculations were carried out by the Excel program. Wells for measuring the depth of groundwater were introduced into the GIS map.

The degree of infection of apple orchards was determined by the formula

\[ D = \frac{A}{B} \]

here:
D is the degree of infection of the orchard;
A is the number of infected trees;
B is the number of trees examined.

RESULTS

In the studies, the signs of cytosporosis in the shoots of an apple tree of the Reinette Simirenko variety (Malus) were manifested in the form of soft spots, the red-brown color of various sizes (Fig. 3).

As the disease progressed, these spots passed into healthy tissues and covered the main stem. Particularly, the spot annularly grasped the lower part of the stem and led to the rapid drying of the upper part. Under favorable conditions for the development of the disease within ten days, the spots reached a size of 10x15 cm and after two months led to the drying of the main stem. Droplets appear on the surface of the new spots, which then dry. The point of contact with healthy tissue remains wet. In some cases, several spots are combined to form a circle. Damaged fabric hardens over time, various cracks appear on the surface. As a result, healthy tissue is separated from the damaged crack. The dead bark turned into red or brown and was easily separated from wood. As a result of the formation of pycnids under the dead tissue, multiple bulges are formed. This leads to a surface roughness of the shoots.

The shoots of the apple tree are also affected by cytosporosis. The causative agent of apple cytosporosis in the shoot bark forms conical pycnids 0.9-1.4 mm in size. Pycnids are multi-chambered, have one entrance. Conidiophores are colorless, extensive, simple, size 13.5 - 28.7 x 2 - 3 microns. The ripened conidia in the pycnidia are attached to each other by a mucus-like liquid; leakage in the form of a belt is observed. Conidia are colorless when flowing out of pycnids, they are initially brilliant yellow, then brown and finally turn dark, waxy, 4.7 - 7.3 x 1 - 2 μm in size (Fig. 4).

As a result of morphological studies, it was found that white colonies grow well in agar water. As a result of comparing the morphological properties and other signs of a fungus that causes harm to apple trees of the Reinette Simirenko variety with the information of Fotouhifar, K.B., Hedjaroude, Gh.A., Ershad, D., Moussavi, S.M., Okhovvat, S.M. and Javan-Nikkhah, M. (2008) for the first time in the Khorezm region, the species of Cytospora sp., close to the species of Cytospora schulzeri, was identified.

Cytosporosis is more common in trees under stressful conditions. They are heat, lack of moisture, and other key factors. In the conditions of the northern regions of Uzbekistan, the relationship of damage from the groundwater level was studied and monitoring of changes in the groundwater level was carried out. The results are shown in the pictures. As can be seen from the data presented for 2013-2017, when studying the level of groundwater and damage to apple trees, tree disease was established.

DISCUSSION

In 2013, cytosporosis was found in the apple orchards of our route. Moreover, infection in one orchard was 12%, in two orchards was 4%, in 12 orchards was 2%. In the remaining 35 orchards, the disease has not been established. The groundwater level in the studied orchards was at least 110 cm, the maximum depth was 312 cm. The dependence of the spread of cytosporosis on the groundwater level was 0.8% (Fig. 5).
The degree of damage of the orchards studied in 2014: in 1 orchard was 36%, in 2 orchards of 11 orchards was 20%, in 1 orchard was 18%, in 1 orchard was 16%, in 10 orchards was 2%, then in 13 orchards, they were not affected by cytosporosis. The maximum indicator of the depth of groundwater is 300 cm, the minimum is 121 cm. The dependence of the development of cytosporosis on the level of groundwater was 6.62% (Fig. 6).

In 2016, the maximum degree of damage to apple orchards is 76%, and the minimum rate is 4%, while all apple orchards are affected by cytosporosis. The maximum groundwater level is 306 cm, and the minimum rate is 140 cm. The dependence of the development of cytosporosis on the groundwater level was 0.08% (Fig. 8).

The maximum degree of infection of orchards with cytosporosis in 2017 was 88%, the minimum indicator is 8%. In the studied orchards, there are no uninfected trees. The maximum depth of groundwater is 298 cm, the minimum level is 125 cm. The dependence of the development of cytosporosis on the level of groundwater is 7.2% (Fig. 9).

The effect of the difference between the maximum and minimum levels of groundwater on the damage to apple trees in orchards in 2013 was studied. The correlation was 67%. This shows that a wide range of groundwater levels leads to an increase in the tendency of trees to be affected by cytosporosis. Since this changes the soil moisture regime. A decrease in groundwater levels, an arid climate leads to a moisture deficit, an increase in groundwater levels affects the adapted water regime of apple trees. And this leads to difficulty in breathing the root system. In an area with elevated groundwater levels, salinization is intensifying.

These processes cause stress conditions for the development of apple trees. The development of cytosporosis under stress...
conditions has been shown in the works of many scientists\textsuperscript{26,27}, the deterioration of soil aeration and moisture conduction can also lead to an increase in the tendency of trees to cytosporosis\textsuperscript{28}.

\begin{figure}[h]
\centering
\includegraphics[width=\textwidth]{figure10.png}
\caption{The dependence of the average infection of apple trees on the difference between the maximum and minimum levels of groundwater.}
\end{figure}

A dry climate, a high level of groundwater (100 - 300 cm) in the Khorezm region leads to the rise of salts dissolved in groundwater through capillaries to the surface. High evaporation from the soil surface of water leads to salinization of the soil. High soil salinization rates do not allow for the normal development of apple trees.

\section*{CONCLUSIONS}

A groundwater level raise which is common in the Khorezm region negatively affects the growth of the Reinette Simirenko varieties of the apple trees. As a result, favorable conditions appear for the development of the cytosporosis disease. To prevent such cases, it is necessary to regulate groundwater tables and implement agrotechnical measures in time based on the results of the continuously monitoring of groundwater levels.

\section*{Acknowledgments:}
The authors are grateful for the use of materials from the scientific project “Sustainable management of land and water resources in Uzbekistan - the ZEF / UNESCO project in the Aral Sea Region” of the Center for Development Research (ZEF) of Bonn University.

\section*{Conflict of interest:}
None

\section*{Financial support:}
None

\section*{REFERENCES}

1. Ikram Abdullaev, Zafar Matyakubov, Manzura Doschanova, Abdulla Iskandarov (2020). The study of population and colony interrelation of Anacanthrotermes turkestanicus (Isoptera: hodotermitidae) in Khorezm oasis // Journal of Critical Reviews ISSN- 2394-5125 Vol 7, Issue 2, 2020, pp.387-390.

2. Abdullaev I.I., Khamraev A.S.H., Martius Ch., Nurjanov A.A., Eshchanov R.A. (2002) Termites (Isoptera) in Irrigated and Landscapes of Central Asia (Uzbekistan). Sociobiology, 40(3): pp.605-614.

3. Adams G.C., Surve-Iyer R.S., Iezzoni A. (2002) Ribosomal DNA sequence divergence and group I introns within Leucosomata species. L. cinctum, L. persoonii, and L. parapersoonii sp. nov., ascomycetes that cause Cytospora canker of fruit trees. Mycologia 94, pp.947–967.

4. Adams G.C., Wingfield M.J., Common R., Roux J. (2005) Phylogenetic relationships and morphology of Cytospora species and related teleomorphs (Ascomycota, Diaporthales, Valsaceae) from Eucalyptus. Studies in Mycology 52, pp.1–144.

5. Adams G.C., Roux J., Wingfield M.J. (2006) Cytospora species (Ascomycota, Diaporthales, Valsaceae): introduced and native pathogens of trees in South Africa. Australasian Plant Pathology 35, pp.521–548.

6. Ariyawansa H.A., Hyde K.D., Jayasiri S.C., Buyck B. et al (2015) Fungal diversity notes taxonomic and phylogenetic contributions to fungal tax. Fungal Diversity 75, pp.27–274.

7. Bertrand P, English H. (1976a) Virulence and seasonal activity of Cytospora leucostoma and Cytospora cineta in French prune trees in California. Plant Disease Reporter 60: pp.106–110.

8. Biggs A.R (1989) Integrated approach to controlling Leucostoma canker of peach in Ontario. Plant Disease 73: pp.869–874.

9. Biggs A.R, Grove G.G.(2005) Leucostoma canker of stone fruits. The Plant Health Instructor on:DOI: 10.1094/PHI-1-2005-1220-1001. Cytospora canker of fruit and nut crops.

10. Conrad C., Schorcht G., Tischbein B., Davletov S., Sultanov M., and Lamers JPA (2012) Agro-Meteorological Trends of Recent Climate Development in Khorezm and Implications for Crop Production.

11. Chang L.S., Iezzoni A.F., Adams G.C., Ewers F.W. (1991) Hydraulic conductance in susceptible versus tolerant peach seedlings infected with Leucostoma persoonii. Journal of the American Society for Horticultural Sciences pp.18-20.

12. Dar M.A., Rai M.K. (2014) Occurrence of Cytospora castanea sp. nov., associated with perennial cankers of Castanea sativa "www.mycosphere.org Article Mycosphere Copyright © 2014 Online Edition Doi 10.5943/mycosphere/5/6/5.

13. Dukhovny V.A., Sokolov V. (2001) Diagnostic Study Rational and Efffective Use of Water Resources in Central Asia/ UN Special Program For economics of Central Asia. Tashkent, Uzbekistan –Bishkek.

14. Istin M.M. (1965) The drying of the branches of the apple tree. Plant protection against pests and diseases.pp 40–42.

15. Isroilov A (1974) Apple tree diseases and measures to combat them in the Tashkent region diss. Abstract. Candidate of Agricultural Sciences. Tashkent. p. 5.

16. Gutner I.S. (1935) Materials for the monograph of the genus CytosporaUSSR Academy of Sciences pp.18-20.

17. Guyon J., Jacobi W.,Meintyre G. (1996) Effects of environmental stress on the development of Cytospora canker of aspen. Plant Dis. 80: pp.1320-1326.

18. Wang Y.L., Lu Q., Decock C., Li Y.X., Zhang X.Y. (2015) Cytospora species from Populus and Salix in China with C. davidiana sp. nov. Fungal Biology 119, pp.420-432.

19. Rahimboeva F.M. (1967) The experience of studying the hydro-geological and reclamation conditions of the Khorezm region. Tashkent, pp.20-26.

20. Ribakov A.A., Ostroukhova S.A. (1981) Fruit growing in Uzbekistan. T, Teacher, p.65.

21. Lu G., Velentini N. (2006) La corilicoltura in Italiya e nel mondo Petriya 16: pp.7-18.
22. Sheraliev A.SH, Rahimov U.H. (2017) Plant Immunity. TASHDAU Publishing House, Tashkent. p.4.
23. Shmid S.M. (1985) Assessment of improving conditions for irrigated land in Uzbekistan. SANIIRI scientific communication Tashkent.
24. Speilman L. (1985) A monograph of Valsa on hardwoods in North America Can. J. Bot63: pp.1355-1387.
25. Scorchini M. (2006) Le Principali aversita dei nocecolo nel Lazio. Petria 16: pp.31-44.
26. Hujaev O.T. (2010) Olma, nok, behing keng tarqalgan kasalliklari va ularga qarshi kuh choralari. Doktorlik diss. Avtoref. Tashkent. p.6.
27. Fan X., Hyde K.D., Liu M., Liang Y., Tian C. (2015) Cytospora species associated with walnut canker disease in China, with description of a new species C. gigalocus. Fungal Biology 119: pp.310–319.
28. Fomenko T.M. (1975) Cytosporosis of the apple tree in the Voronezh region. Abstract of dissertation for the degree of candidate of agricultural sciences Moscow.
29. Fotouhifar K.B., Hedjaroude G.H.A., Ershad D., Moussavi S.M., Okhovvat S.M. and Javan-Nikkhah M. (2008) New records of form-genus Cytospora in Iran (II). Rostaniha, 9(1): pp.49-66.