DOI: 10.34101/ACTAAGRAR/2/10367

Analysis of the plant physiological effects of late, artificial corn smut infestation using remote sensing methods

László Radócz[1*] – Levente Sápi[2] – Péter Zagyi[1] – Éva Horváth[1] – András Tamás[1]

[1]University of Debrecen, Faculty of Agricultural and Food Sciences and Environmental Management, Institute of Land Use, Engineering and Precision Farming Technology

[2]Hungarian Chamber of Phytosanitary Engineers and Phytomedicines, Hajdú-Bihar County Regional Organisation

*Correspondence: radocz.laszlo@agr.unideb.hu

SUMMARY

In Hungary, corn is also infected by several important pathogens. In this experiment, we analysed the plant physiological effects of artificial late cron smut infestation using remote sensing methods under field conditions We examined the experimental area from which the data comes from with a DJI Phantom 4 Multispectral Drone NDVI and NDRE indices were calculated and analyzed in GIS programs. Individuals treated with a higher dose remained much greener than the untreated control. They also showed significant differences within the indices used.

Keywords: *remote sensing, disease, maize, ustilago maydis, vegetation indices*

INTRODUCTION

Maize is a major crop in continental Europe, including Hungary and it has a wide range of uses (feed, food, fuel, starch production, etc.). In the European Union (EU27), maize production was 70.1 million tons in 2020 (Eurostat, 2021). In Hungary, the area sown to maize was 981 006 hectares and the average yield was 8.5 t ha^{-1} last year (KSH, 2020).

The corn smut pathogen can highly damage the yield, the pathogen can attack every part of the plant, but in an economic view the most important is the crop infections (Frommer et al., 2015).

This is why scientific studies and research on maize are so important, as we can benefit from significantly more information on this key industrial crop. In Hungarian agriculture, an increasing number of farmers are using precision farming tools. In addition, digitalization and IT solutions are increasingly gaining ground in research and development trends. Many examples of these are already working well in practice (e.g. automated steering, RTK systems, phased application technologies, plant health monitoring, spatial zoning, etc.)

Remote sensing is a method that can measure information about the surface of the Earth from the electromagnetic energy emitted or reflected without physical contact (De Jong and Van der Meer, 2007). From the images taken by agricultural UAVs equipped with different cameras, it is possible to produce a variety of vegetation index maps, especially in the case of a multispectral camera that captures images on 6 different channels simultaneously, thus providing a wide range of information. Vegetation indices are based on reflectance by plants. Of all vegetation index maps, NDVI is the most widely used (Bhandari and Kumar, 2012). Normalized Difference Vegatation Index NDVI is defined as the ratio of canopy reflectance measured in the red and near-infrared spectra (Tucker, 1979). Normalized Difference Red Edge Index (NDRE) is also used to measure the amount of chlorophyll and vegetation activity. In the formula Red Edge is used insteaf of Red (Tucker, 1979). Plant pest monitoring can be defined in several ways: detection is the reliable separation of healthy plants from diseased plants. (West et al., 2003) Identification-separation, possibly diagnosis, of specific symptoms (Sankaran et al., 2010). Quantification-the extent, size and severity of the disease (Mahlein et al., 2012).

In this experiment, the corn smut pathogen *Ustilago maydis* DC. CORDA was tested. In the case of the corn smut disease, the major farming damage is caused by the ear infection (Frommer et al., 2015). The smut that develops on maize ears is initially yellowish in color, later becoming whitish and shiny (Szőke et al., 2021a). In this study, late ear infection was carried out and its effects were observed.

MATERIALS AND METHODS

The experimental area was a maize field on the eastern outskirts of Debrecen (*Figure 1*). Coordinates: 47°33'16.9"N 21°41'34.6"E

Soil: humic sand. The field was not irrigateted and there were no use of additional nutrient supply. The agrotechnic work was the same in the whole field, basic traditional corn producing.

The tested feed maize hybrid was: DKC 4596

Three experimental medium plots, each of 400 m^2 in size, have been set up. In the midline of the plots, 5 to 5 rows were artificially infested with 2 ml of inoculum per plant containing the specified amount of sporidium. A veterinary mass inoculation tool was used for the treatments. The rows adjacent to the infected rows were considered as infection-free (control) areas.

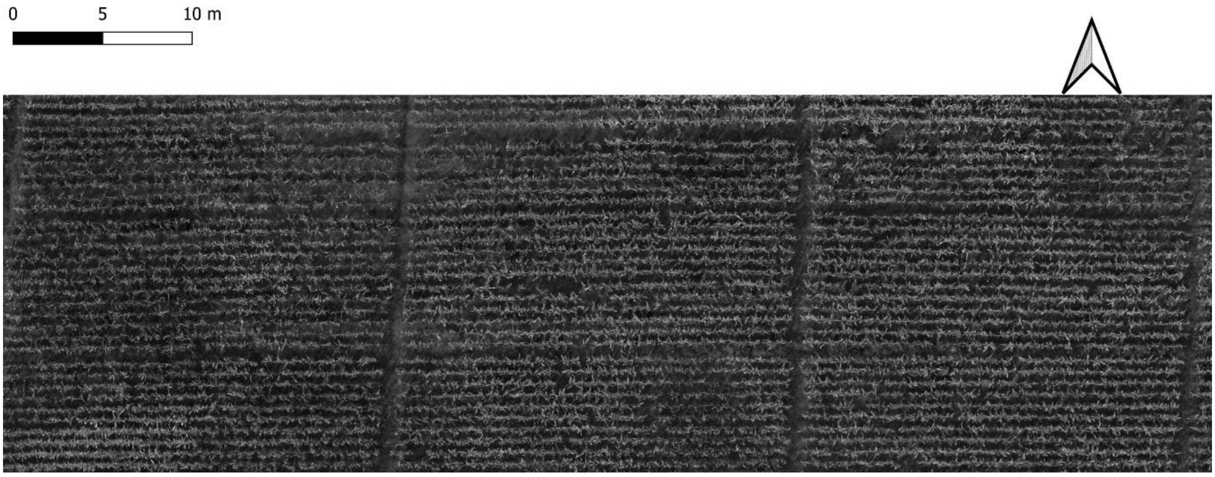
Three different spore concentrations were used in four replications for the ear infestation.

Dates of treatment: The phenological stages of the plants were R1

- 22.07.2021 Low inoculum dose (5.000 sporidia ml^{-1})
- 23.07.2021 Medium inoculum dose (7.500 sporidia ml^{-1})
- 26.07.2021 High inoculum dose (10.000 sporidia ml^{-1})

DOI: 10.34101/ACTAAGRAR/2/10367

Figure 1: **The image of the experimental site**



Aerial photos were taken with a DJI Phantom 4 Multispectral camera. The date of the aerial photography was 16.09.2021 (from 35.5 m height 1.9 cm PX^{-1} resolution Front Overlap : 80% Side Overlap : 80%)

The Multispectral camera captures images in 6 different channels. Therefore, we have a total of 6 orthophotos available to create and analyse vegetation index maps.

- Red Edge (RE): 730 nm ± 16 nm
- Near-Infrared (NIR): 840 nm ± 26 nm
- Green (G): 560 nm ± 16 nm
- Visible Light (RGB)
- Red (R): 650 nm ± 16 nm
- Blue (B): 450 nm ± 16 nm

The date of the aerial photography was 16.09.2021 at the R6 phenological stage of corn (from 35.5 m height 1.9 cm PX^{-1} resolution Front Overlap: 80% Side Overlap: 80%)

The weather was clear and no clouds on the sky.

WebODM software was used to properly put together the images. The orthomosaics were HR (High Resolution) 2 cm $pixel^{-1}$ quality.

Preparing vegetation maps:

The images were processed and evaluated using the Quantim GIS 3.20 geospatial software.

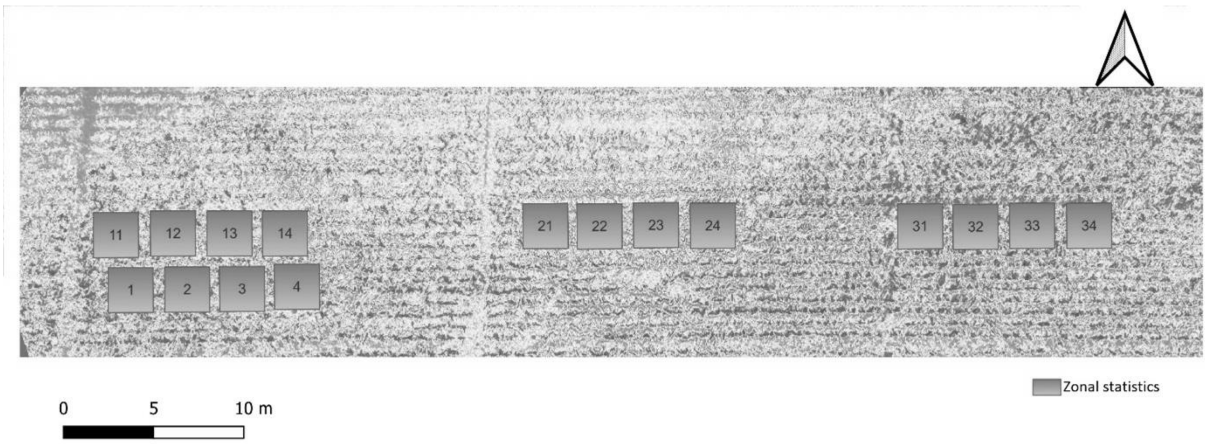
The following two vegetation index maps were created using raster analysis: NDVI and NDRE (*Table 1*)

Table 1: **Calculation formulas for vegetation index maps**

Vegetation index	Formula	References
NDVI	$(R_{NIR}-R_{Red})/(R_{NIR}+R_{Red})$	Tucker (1979)
NDRE	$(R_{NIR}-R_{RedEdge})/(R_{NIR}+R_{RedEdge})$	Barnes (2000)

Once the maps were completed, 2.5x2.5 m "boxes" (as a new shape file layer) were placed in the experimental plots (*Figure 2*) that were important for sampling. The NDVI and NDRE values are taken from these "sampling boxes".

Figure 2: **NDVI map and location of "NDVI" sampling points in the experimental area**



DOI: 10.34101/ACTAAGRAR/2/10367

The process was followed by the main belt statistics, from which we obtained the averages of the various zone statistics.

Zonal statistics:

It calculates some statistical values based on the values of the input raster pixels within certain zones, defined as polygon shape.

The values calculated for the selected zones were:

- minimum
- maximum
- amount
- median
- average
- dispersion
- domain
- minority
- majority

(NDVI mean, NDRE mean). As a next step, the attribute table was exported to MS Excel.

Once the database is in the right format, the analysis can begin. Statistical analyses were performed in Rstudio using random block design (RCBD) and LSD post hoc test.

RESULTS AND DISCUSSION

During the field observation, it was found that the characteristic galls on the artificially infested ears were not able to form in any of the treatments (probably due to the late infestation and the very dry August weather), but in almost all cases ear moulds were formed at the site of infestation (*Figures 3–5*).

Figures 3–5: **Maize ears infected with low (5,000), medium (7,500) and high (10,000) sporidium suspensions (1.5 months post-infection)**

Statistical tests

The NDVI values show (*Figure 6*) that the values of the plots infected with the high (10,000 sporidium) suspension are significantly different from both the control and the low dose (5,000 sporidia ml^{-1}) (c) and the group infected with the medium inoculum dose (7,500 sporidia ml^{-1}) (b). The values for the low inoculum and control plots (c) were similar in that no significant difference was detected between them.

The NDRE map values (*Figure 7*) also show that the values of the high inoculum (10,000 sporidia ml^{-1}) infected plots (a) are significantly different from both the control (c) and the low sporidium (5,000 sporidium ml^{-1}) plots (b). However, the values of the medium (7,500) group (ab) were not significantly different from those of the control group (b). The values for the low concentration (5,000 sporidia ml^{-1}) (b) and untreated control plots (c) were significantly different.

In this experiment, late artificial ear infestation (probably in the very dry August weather) did not cause any more gall formation, but oxidative stress recruitment was likely to have been initiated in the hybrid with good gall resistance. The resulting reactive oxygen species (ROS) and the defence plant physiological reactions to neutralise them enhance the production of various enzymes (Sharma et al., 2012). The components of this defence system are various antioxidant enzymes such as superoxide dismutase (SOD), catalase (CAT), glutathione peroxidase (GPX) and ascorbate peroxidase (APX). These catalyze the neutralization of reactive oxygen species in the plant. However, as the number of inoculum sporidia increases, their production also shows higher levels in the crop. In their artificial inoculation studies with corn smut in the 4–5 leaf (V5 phenological phase) state of maize, Szőke et al. (2021b) have already detected the greening effect of APX. It is likely that as a result of one or even a combination of these enzymes, the infected plants remained in a green state compared to untreated maize.

DOI: 10.34101/ACTAAGRAR/2/10367

Figure 6: **Results of NDVI statistical tests: different letters indicate significantly different groups**
(Treatments with the same letter are not significantly different)

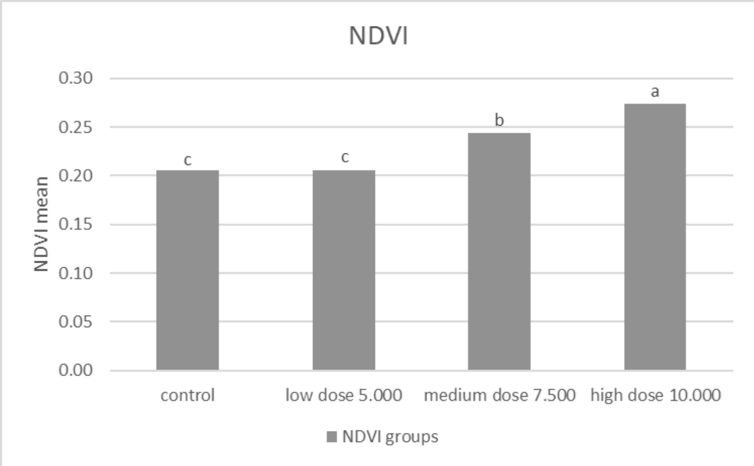
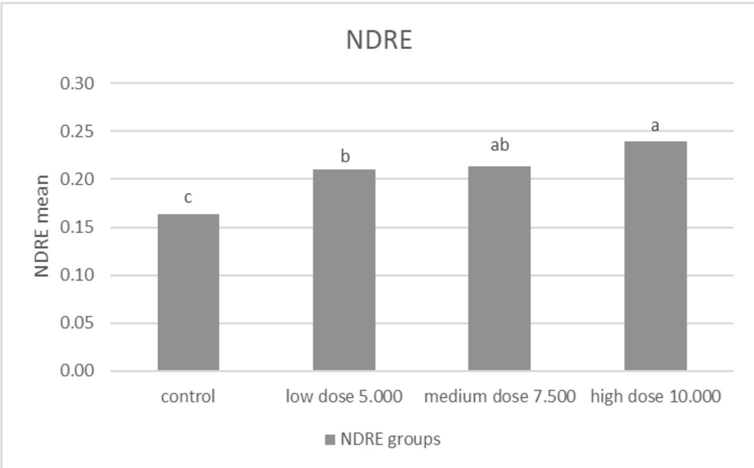


Figure 7: **Results of NDRE statistical tests: different letters indicate significantly different groups**
(Treatments with the same letter are not significantly different)



CONCLUSIONS

The late corn smut infestation no longer formed smut on the ears, but we observed that all treated ears developed ear mould.

We further observed that the treated plants were still much greener than the controls at this time, as evidenced by the fact that NDVI averages were significantly higher in the two plots with higher inoculum concentrations compared to the untreated control and the low concentration treatment. Here the amount of inoculum concentration also resulted in further significant differences for the two higher concentration treatments.

The incubation period for corn smut infection is usually 2 weeks. During this time, the pathogen takes the nutrients it needs from the maize using its own enzymes. Later on, galls develop at the site of infection (Pécsi, 1997). However, Mazeheri-Naeni and colleagues (2015) have also found that the pathogen is able to pass between root cells, but that no galls usually form on the plant when infected in this way.

To elucidate this phenomenon, we plan to continuously analyse the changes in plant antioxidant enzyme levels in response to late corn smut infection (mainly in a forage hybrid with good resistance) under large plot field conditions.

ACKNOWLEDGEMENTS

The study research is the project TKP2020-IKA-04 of the National Research With support from the Development and Innovation Fund, from 1.1 to TKP2020 application program and EFOP-3.6.3-VEKOP-16-2017-00008 was implemented in the financing of a number of projects.

DOI: 10.34101/ACTAAGRAR/2/10367

REFERENCES

Barnes, E.M.–Clarke, T.R.–Richards, S.E.–Colaizzi, P.D.–Haberland, J.–Kostrzewski, M.–Waller, P.–Choi, C.–Riley, E.–Thompson, T. (2000): "Coincident detection of crop water stress, nitrogen status and canopy density using ground based multispectral data." Proc. 5th Int. Conf. Precis Agric. Bloomington, MN, USA

Bhandari, A.K.–Kumar, A. (2012): “Feature Extraction using Normalized Difference Vegetation Index (NDVI): A Case Study of Jabalpur City”, Proceedings of Communication, Computing & Security. Procedia Technology Volume 6, pp. 612–621. https://doi.org/10.1016/j.protcy.2012.10.074

De Jong, S.M.–Van der Meer, F.D. (Eds.). (2007): Remote sensing image analysis: including the spatial domain (Vol. 5). Springer Science & Business Media.

Frommer, D.–Veres, Sz.–Lévai, L.–Radócz, L.: 2015. Régi-új betegség a kukorica golyvásüszög (*Ustilago maydis*). Journal of Agricultural Sciences. 66: 43–46.

https://ec.europa.eu/eurostat/statistics-explained/index.php?title=Agricultural_production_-_crops#Cereals

https://www.ksh.hu/stadat_files/mez/hu/mez0072.html).

Mahlein, A.K.–Oerke, E.C.–Steiner, U.–Dehne, H.W. (2012): Recent advances in sensing plant diseases for precision crop protection. Eur J Plant Pathol 133:197–209. doi:10.1007/s10658-011-9878-z

Mazaheri-Naeini, M.–Sabbagh, S.K.–Martinez, Y.–Séjalon-Delmas, N.–Roux, C. (2015): Assessment of *Ustilago maydis* as a fungal model for root infection studies. Fungal Biolodgy, 119 (2–3), pp. 145–153. https://doi.org/10.1016/j.funbio.2014.12.002

Nageswara, P.P.R–Shobha, S.V.–Ramesh, K.S.–Somashekhar, R.K. (2005): “Satellite-based assessment of Agricultural drought in Karnataka State, Journal of the Indian society of remote sensing”, 33 (3), pp. 429-434., 2005

Pécsi, S. (1997): A kukorica betegségei. in: Glist, M.–Horváth, J.–Kuroli, G.–Petróczi, I. (szerk.): Növényvédelem. Mezőgazda Kiadó, Budapest. pp. 49–60.

Sankaran, S.–Mishra, A.–Ehsani, R.–Davis, C. (2010): A review of advanced techniques for detecting plant diseases. Comput. Electron. Agr. 72, 1–13. https://doi.org/10.1016/j.compag.2010.02.007

Sharma, P.–Jha, A.B.–Dubey, R.S.–Pessarakli, M. (2012): Reactive oxygen species, oxidative damage, and antioxidative defense mechanism in plants under stressful conditions. J Botany. 2012:1–26. doi:10.1155/2012/217037

Szőke, L.–Kovács, G.–Biró, Gy.–Rácz, D.–Radócz, L.–Takácsné, H.M.–Kovács, B.–Tóth B. (2021a): Hormonkezelések hatása a golyvásüszög fertőzés tüneteinek mérséklésére. Növénytermelés 70. 2 p.1–22.

Szőke, L.–Moloi, M.J.–Kovács, G.–Biró, Gy.–Radócz, L.–Hájos, M.–Kovács, B.–Rácz, D.–Danter, M.–Tóth, B. (2021b): The Application of Phytohormones as Biostimulants in Corn Smut Infected Hungarian Sweet and Fodder Corn Hybrids. Plants. 10(9):1822. https://doi.org/10.3390/plants10091822

Tucker, C.J. (1979): Red and photographic infrared linear combinations for monitoring vegetation. Remote Sens. Environ. 8, 127–150. https://doi.org/10.1016/0034-4257(79)90013-0

West, J.S.–Bravo, C.–Oberti, R.–Lemaire, D.–Moshou, D.–McCartney, H.A. (2003): The potential of optical canopy measurement for targeted control of field crop diseases. Annu. Rev. Phytopathol. 41, 593–614. doi: 10.1146/annurev.phyto.41.121702.103726