

MYCOTOXINS INCIDENCE IN WHEAT CULTURE

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ABSTRACT. *Cereals are the most important food sources in the world, directly and indirectly for human consumption and as a factor in livestock production. Wheat is the most important culture, both in surface terms and as a production. Mycotoxins are secondary metabolites of fungi that grow as a result of infection with *Fusarium*, *Alternaria*, *Aspergillus* and *Penicillium*. The qualitative damages caused by presence of mycotoxins exceed the quantitative economic losses. DON's determination from wheat samples was performed by ELISA Method, rapid and sensitive method that provides simultaneous analysis of a large number of samples. DON's concentration varied depending on the variety grown, climatic factors (temperature and humidity) and cultivation technology.*

Key words *wheat, mycotoxin, deoxynivalenol(DON), Elisa method.*

INTRODUCTION

Mycotoxins are the major problem in food safety, they are harmful to both animals and humans (GILBERT AND TEKAUZ 2000; NGOKO ET AL. 2008). There are toxic metabolites produced by fungi and are of considerable concern as they not only cause plant diseases but are also damaging to human and animal organs when contaminated food is ingested (MCMULLEN AND AL., FOROUD AND AL.). Deoxynivalenol (DON) is a common mycotoxin produced by some *Fusarium* species such as *Fusarium graminearum* and *F. culmorum* and is often found in small grains that have been infected with these *Fusarium* species. The most common small grain disease is called *Fusarium* head blight (FHB), which often develops following moist environmental conditions when the head is in flower. The importance of determination of DON level to food safety is due to the severe effect that this mycotoxin has on animal systems. Protein and DNA synthesis in the cell are seriously affected by DON presence. Symptoms exhibited by animals afflicted with trichothecene toxicoses include vomiting, feed refusal, diarrhea, and haemorrhaging of intestines and muscles (UENO Y. AND AL.). DON has also been shown to be neurotoxic and immuno-suppressive (Rotter and al.). Cell signaling pathways are activated by 1 mg DON/kg body weight, through gene induction and activation of several nitrogen-activated protein kinases. DON (≥ 100 ng/mL) activates hematopoietic cell kinase and double-stranded RNA-activated protein kinase, which leads to apoptosis (Pestka and al.). The U.S. Food and Drug Administration (FDA) has set advisory DON levels for wheat-based foods and feeds of no more than 1 $\mu\text{g/g}$ in finished human foods, 10 $\mu\text{g/g}$ in poultry and ruminant feed, and 5 $\mu\text{g/g}$ in other animal feeds. Precise determination of the presence of DON in low amounts is important in monitoring of food and feed for food security. The work aimed to detect DON in wheat samples provided by farmers in Timis and Arad crop years 2014 and 2015. The results showed the importance of the type of cultivated variety, climate and cultivation technology.

MATERIAL AND METHODS

Wheat samples were taken from three farmers in Timis county and three farmers in Arad county. Analysis of each sample was carried out three times in order to avoid the error. Each producer has cultivated one single variety of wheat on seven different plots during 2014 and 2015 years. Deoxynivalenol detection was performed in each plot on the farm by Eliza Method.

A deoxynivalenol specific antibody is coated to a polystyrene microwell. Toxins are extracted from a ground sample with distilled or deionized water. The extracted sample and HRP-conjugated DON are mixed and added to the antibody-coated microwell. DON from the extracted sample and HRP-conjugated DON compete to bind with the antibody coated to the microwell. Microwell contents are decanted and non-specific reactants are removed by washing. An enzyme substrate (TMB) is added and color (blue) develops. The intensity of the color is directly proportional to the amount of bound conjugate and inversely proportional to the concentration of DON in the sample or standard. An acidic stop solution is added which changes the chromagen color from blue to yellow. The microwells are measured optically by a microplate reader with an absorbance filter of 450nm (OD450). The optical densities of the samples are compared to the OD's of the kit standards and an interpretative result is determined. The extraction and detection of DON in a grain sample should be as easy as possible. Solvents containing water can easily extract the water-soluble trichothecene DON. The solvent containing 10% methanol and 90% water gave the best recovery of toxin.

RESULTS AND DISCUSSION

Results obtained are represented in figure 1-6.

Varieties cultivated in Timis County

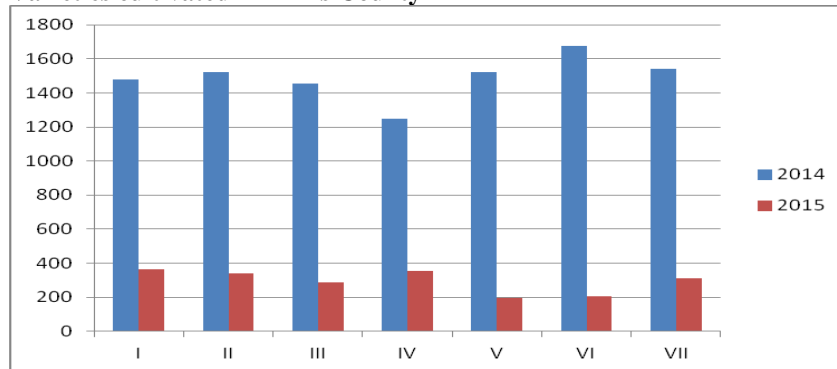


Figure 1. DON content (ppb) in samples of Apache variety

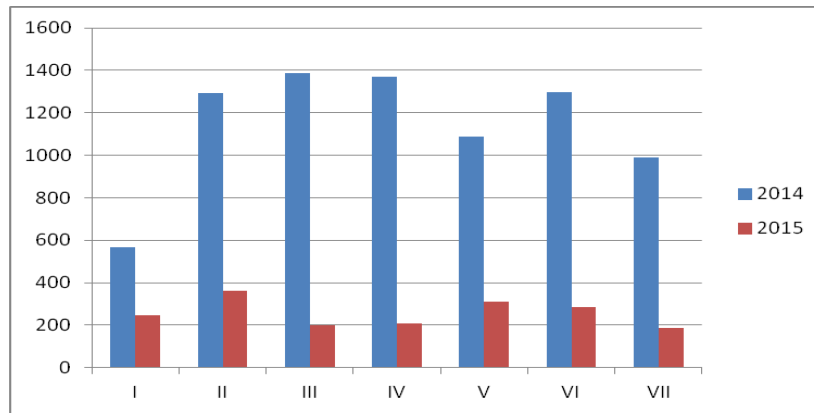


Figure 2. DON content (ppb) in samples of Alex variety

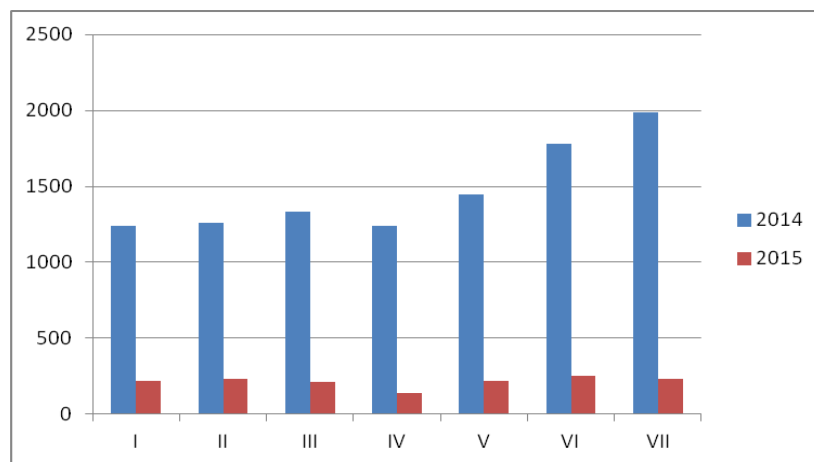


Figure 3. DON content (ppb) in samples of Sofru variety

Varieties cultivated in Arad County

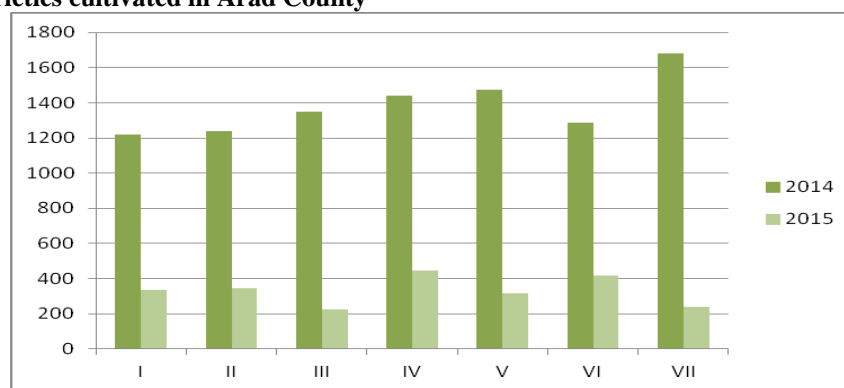


Figure 4. DON content (ppb) in samples of Glossa variety

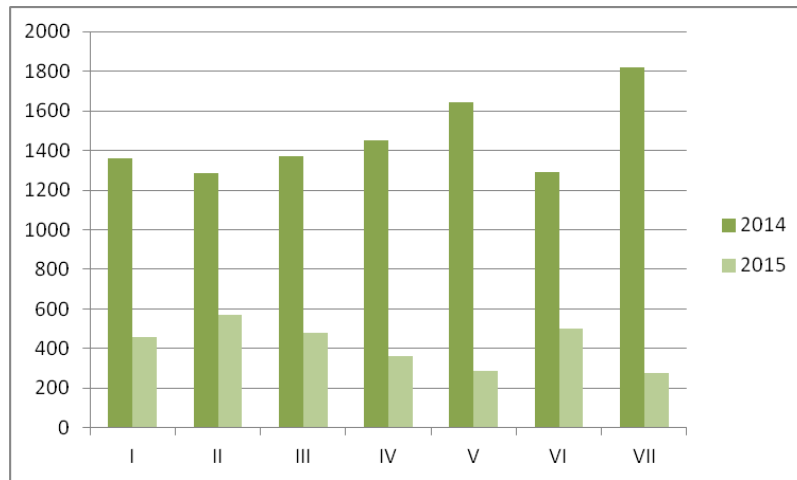


Figure 5. DON content (ppb) in samples of Exotic variety

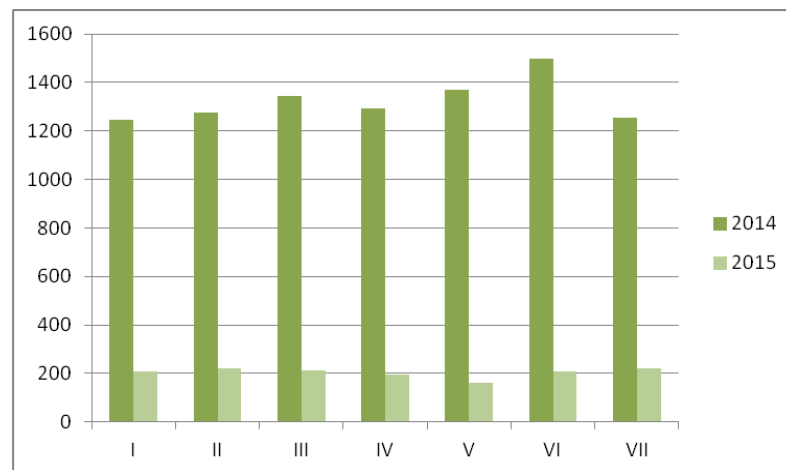


Figure 6. DON content (ppb) in samples of Ingenio variety

CONCLUSIONS

- Ingenio variety cultivated in Arad county showed a good resistance to *Fusarium* infection, only 3 of the 7 plots had more than the allowed DON limit content 1250 ppb in 2014 and in 2015 presented lowest content of DON (223 ppb max)
- 2014 was considered a year with abundant precipitations and an average of 1100 mm in the western countries. 2015, characterized as a drought year in June, July, August and September with an average of 350 mm in the western countries. Thus rainfall are considered as the main climatic factor that influenced the development of *Fusarium* species in wheat and deoxynivalenol infestation of wheat grains.
- The temperature and humidity are key factors favoring contamination of grain with mycotoxins. Among these factors are closely connected, to which it is possible to develop

microorganisms; thus the higher the water content of the grain is increased with the developmental both.

- Prevention of post-harvest contamination is achieved by compliance with optimal condition grain storage temperature, humidity, ventilation to prevent toxin production.

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