Stability of resistance to Fusarium head blight and *Fusarium* toxin accumulation in winter wheat lines over different environments

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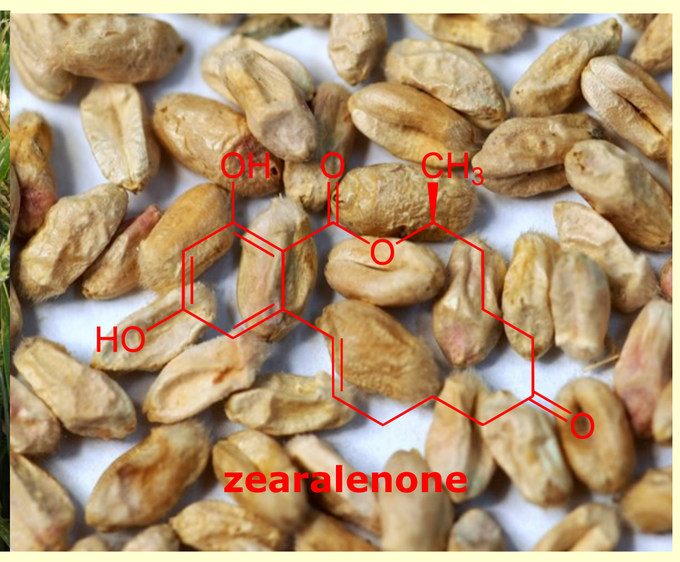

Fusarium head blight (FHB) is a disease of cereals caused by *Fusarium* fungi (mainly *F. culmorum* and *F. graminearum*). These fungi produce toxic metabolites - mycotoxins, with phyto-and zootoxic effects. In the grain of wheat the most common are toxins belonging to trichothecenes B: deoxynivalenol (DON) and nivalenol (NIV), as well as zearalenone (ZEN). There are several types of resistance to FHB: type I – resistance to infection, type II — resistance to spread of *Fusarium* within the head, type III – resistance to *Fusarium* kernel damage. Other types of resistance are: tolerance to FHB or to mycotoxins (DON) (type IV) and type V - resistance to the accumulation of toxins in the grain through their chemical modification (class 1) or blocking synthesis (class 2).
The aim of the research was to assess the variability of winter wheat genotypes in response to infection with *F. culmorum* by examination of the above-mentioned types of resistance and finding genotypes combining resistance of all types and showing stability of reaction over different environments.

Materials and method

To select the forms of winter wheat combining different types of resistance 72 lines were evaluated in the field experiments in two locations (Poznań, Radzików) in years 2014-2016 . They included four resistant checks (R): '20828' [*Fhb1*-], 'A40-19-1-2', 'Arina', 'UNG 136.6.1.1' [*Fhb1+*]; three susceptible checks (S); three lines with a high accumulation of trichothecenes (TOX). Wheat heads were inoculated at the full flowering stage with the spore suspension of aggressive isolates of *F. culmorum* producing DON and ZEN. Head infection severity as well as disease incidence were assessed. From these values Fusarium head blight index (FHBi) was calculated reflecting the percentage of infected spiklest in all of the spikes on a plot. The proportion *Fusarium* damaged kernels was determined visually by splitting the sample into healthy looking kernels and kernels showing different symptoms of damage by *Fusarium*. The value of the FDK (= *Fusarium* damaged kernels) was calculated as a percentage of damaged kernels in whole sample (based on kernel number = FDK). Using the techniques liquid and of gas chromatography as well as immunoenzymatic tests the contents of ergosterol, DON, acetyl derivatives of DON, NIV and ZEN in the grain was analysed.

Results

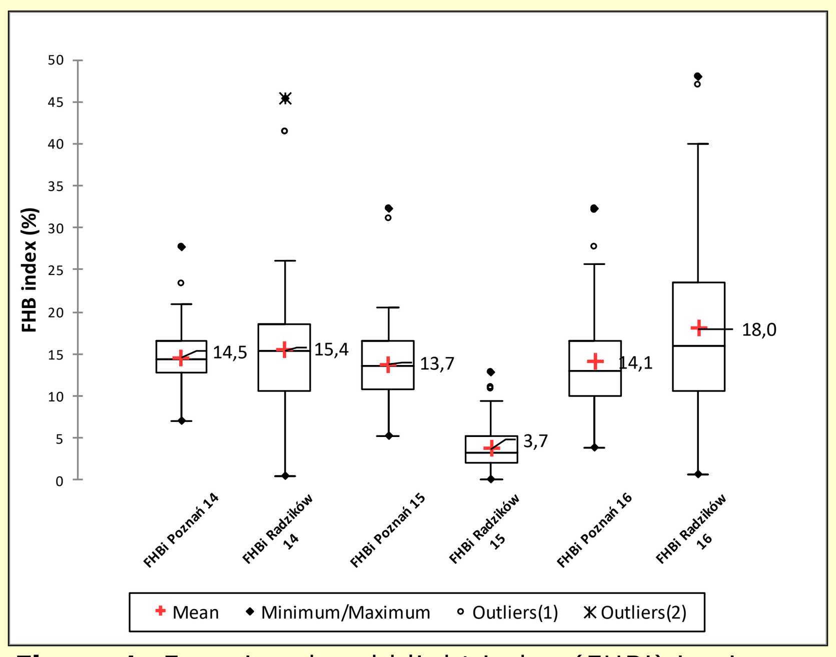

Figure 1. Fusarium head blight index (FHBi) in six environments: three years—2014,2015, 2016; two locations—Poznań, Radzików. Rectangles show the first quartile, the median and the third quartile. Whiskers show the lower and upper limit beyond which the values are outliers.

Average FHBi over six environments was 13.2%; the range of variation from 0 to 48.0% (Fig. 1). Average FHBi in five environments were similar, only in 2015 in Radzików FHBi was very low (3.7%), what was caused by drought during and after wheat flowering. Average FHB indexes for the 72 lines tested in experiments in 2014, 2015 and 2016 correlated significantly (r=0.571 - 0.775). Lines with stable reaction in all six environments were identified - resistant: UNG 136.6.1.1 (VR), A40-19-1-2 (R), POB 679/03, 20828 (R), POB 011, Arina (MR), STH 2041, POB 170/04, STH 9059, SMH 7983, KBP 10 58; susceptible: DD 548/09 (tox), NAD 10079 (S), SMH 8694 (S), SMH 8816 (S) (Fig. 2). Susceptible lines showed higher variability of reaction in different environments that resistant ones. The most stable was reaction of three resistant lines POB 679/03, STH 2041 and SMH 7983.

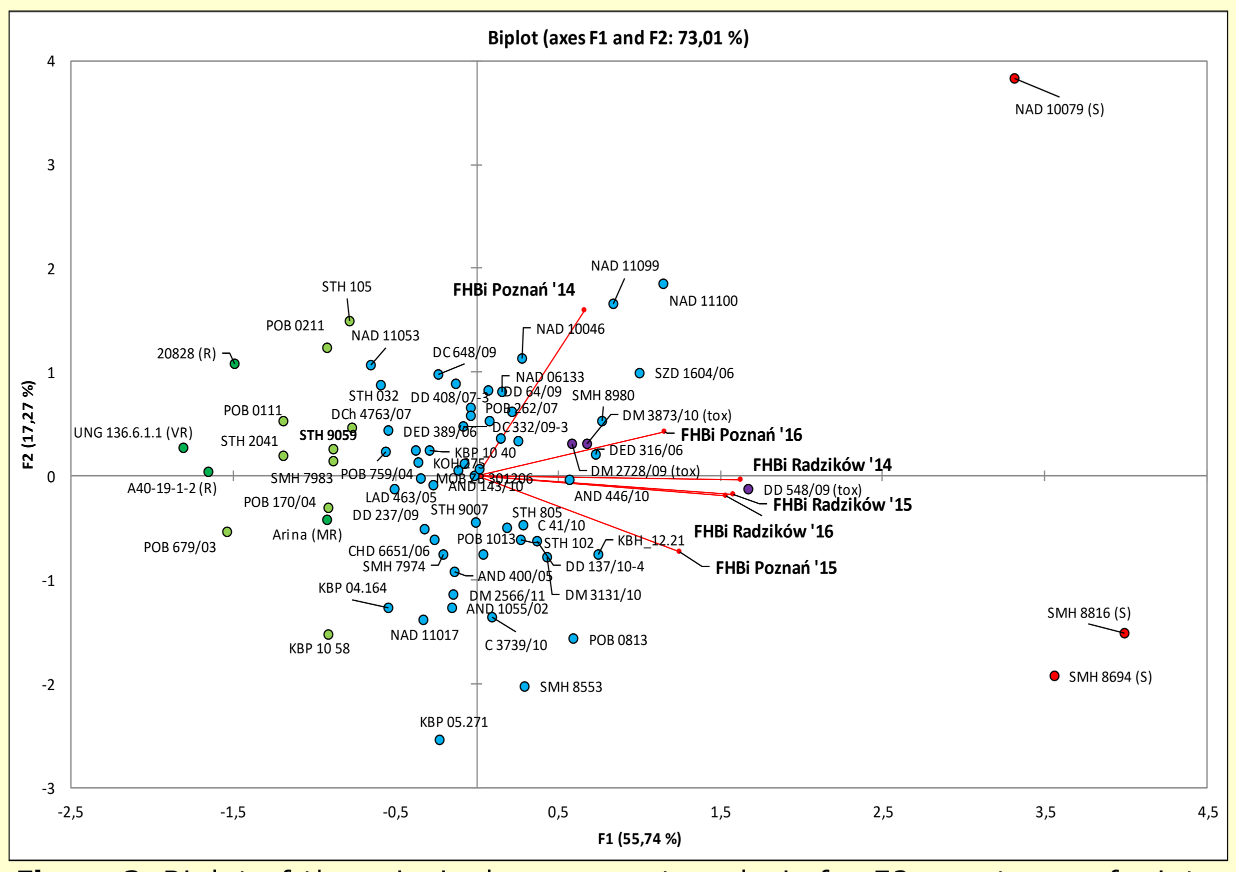

Figure 2. Biplot of the principal component analysis for 72 genotypes of winter wheat. Two first components explained 70.1% of the variability of resistance to Fusarium head blight measured with FHB index (FHBi) in Poznań and Radzików in 2014, 2015 and 2016. Vectors indicate the direction of the increase of the value of the variables. Red circles—susceptible checks, violet circles—toxin checks, green circles—resistant checks, light green circles—the most resistant lines.

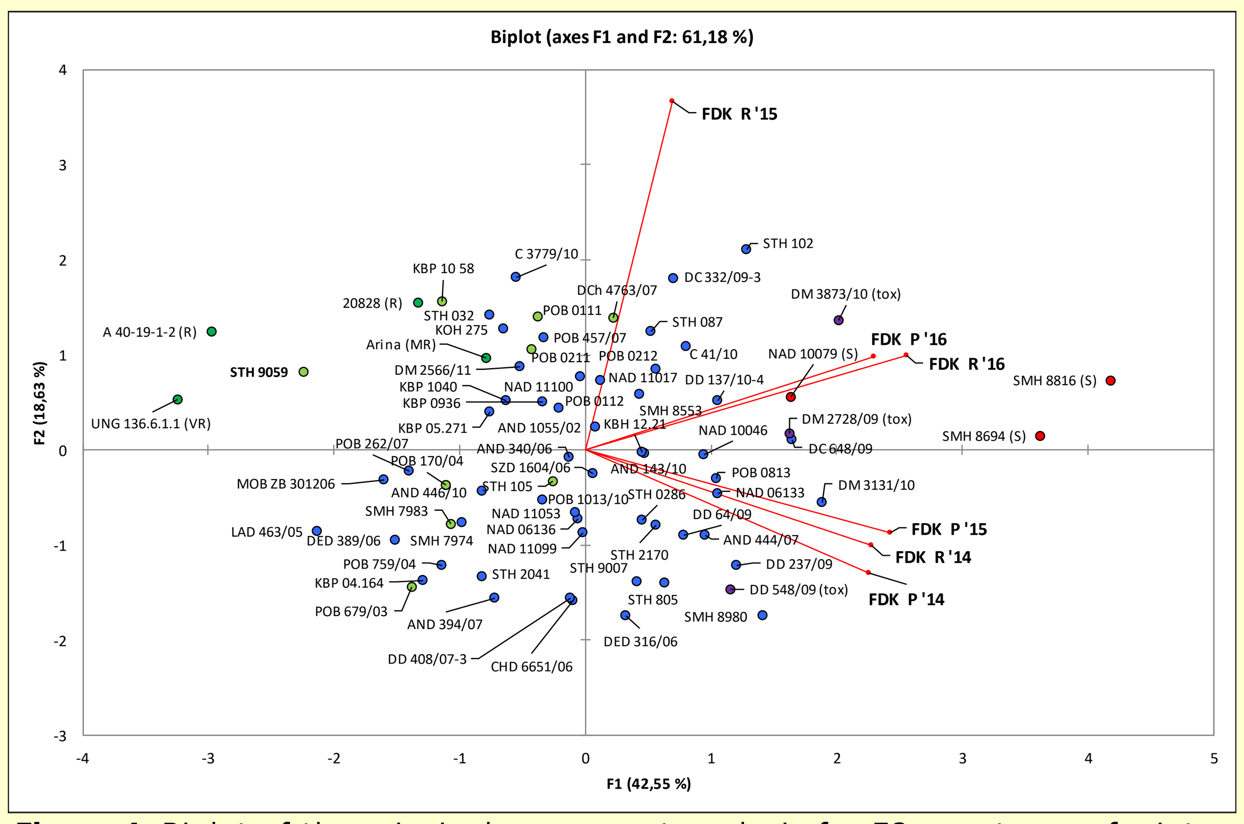

Figure 4. Biplot of the principal component analysis for 72 genotypes of winter wheat. Two first components explained 70.1% of the variability of resistance to Fusarium head blight measured with proportion of Fusarium damaged kernels (FDK) in Poznań (P) and Radzików (R) in 2014, 2015 and 2016. Vectors indicate the direction of the increase of the value of the variables. Red circles—susceptible checks, violet circles—toxin checks, green circles—resistant checks, light green circles—lines the most resistant to head infection.

Average FDK over six environments was 24.6%; the range of variation from 1.8 to 91.5% (Fig. 3). Average FDK in Poznan and Radzików in 2015 and 2016 were similar. In 2014 in Poznań FDK was very high (61.8%), what was caused by rainy weather before harvest. In Radzików FDK in 2015 was also higher than in other years. Average FDK values for the 72 lines tested in experiments in 2014, 2015 and 2016 correlated significantly (r=0.263 - 0.488), however coefficients were lower than for FHB indexes. Lines with stable reaction in all six environments were identified - resistant: UNG 136.6.1.1 (VR), A40-19-1-2 (R), STH 9059, LD 463/05, MOB ZB 301206; susceptible: SMH 8694 (S), SMH 8816 (S) (Fig. 4). Susceptible lines showed higher variability of reaction in different environments that resistant ones. The most stable was reaction of highly resistant checks UNG 136.6.1.1 (VR) and 20828 (R), and resistant lines C 3779/10, POB 0813 and STH 9059.

Average FHB indexes and FDK values for the 72 lines tested in experiments in 2014, 2015 and 2016 correlated significantly (r=0.400 - 0.698).

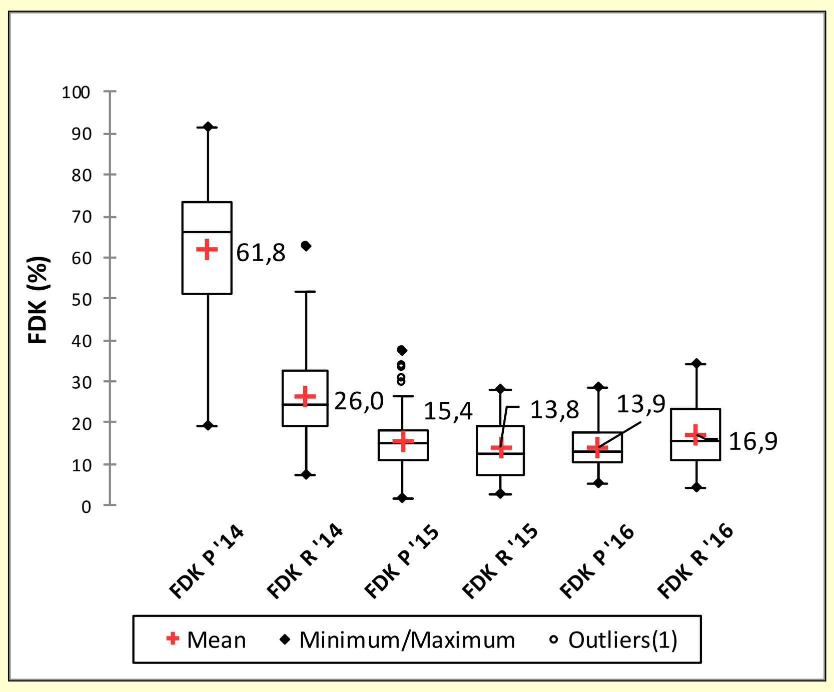

Figure 3. Fusarium damaged kernels (FDK) in six environments: three years—2014, 2015, 2016; two locations — Poznań, Radzików. Rectangles show the first quartile, the median and the third quartile. Whiskers show the lower and upper limit beyond which the values are outliers.

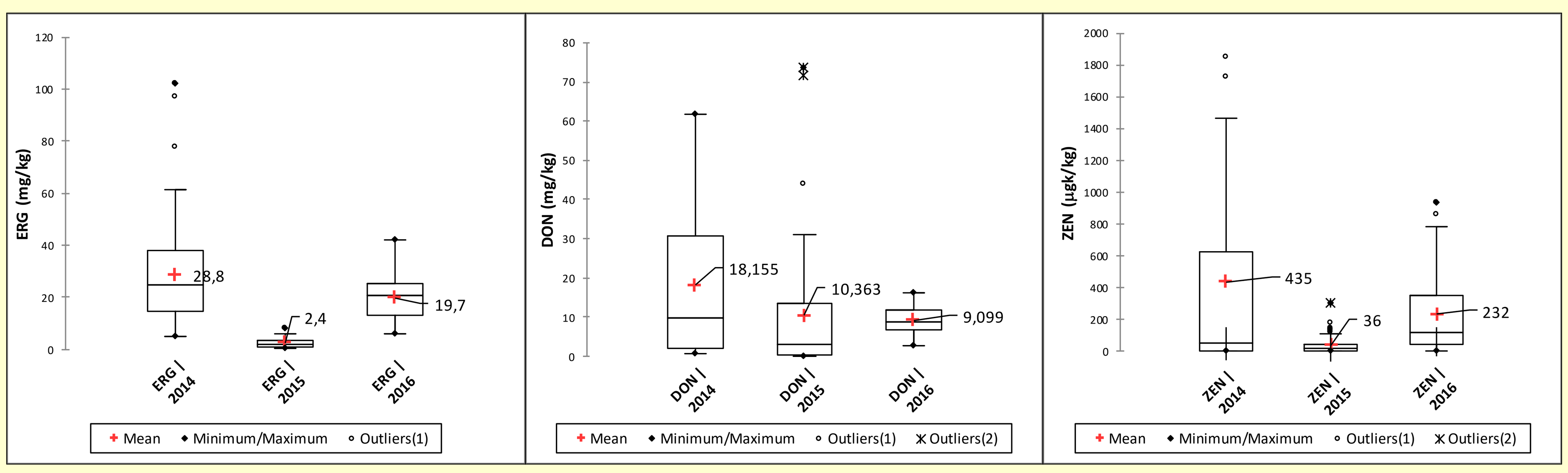

Figure 5. Content of ergosterol (ERG), deoxynivalenol (DON) and zearalenone (ZEN) in the grain of 28 wheat lines in years 2014, 2015, and 2016 (averages from two locations — Poznań, Radzików). Rectangles show the first quartile, the median and the third quartile. Whiskers show the lower and upper limit beyond which the values are outliers. In 2014 the highest concentration of ZEN was 4522 µg/kg.

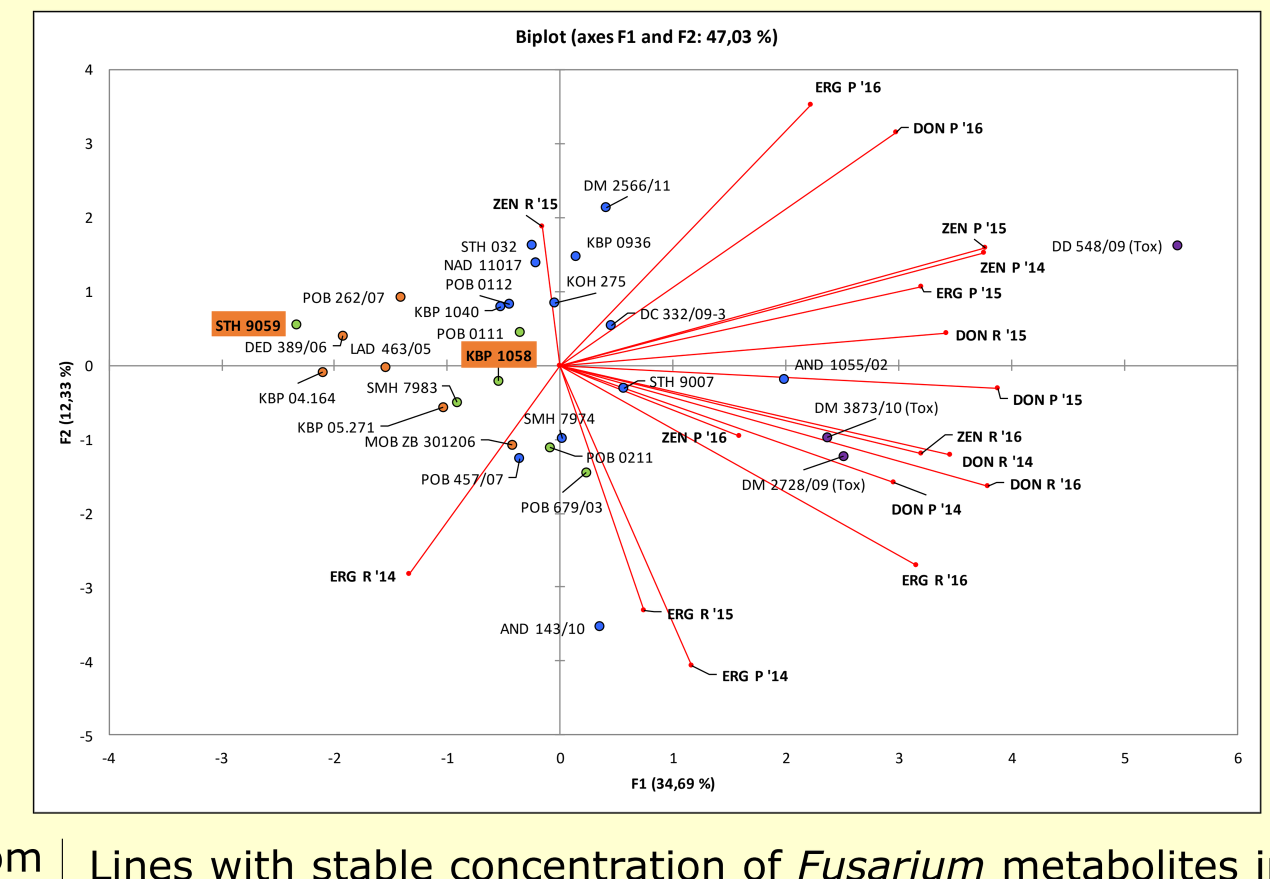

Figure 6. Biplot of the principal component analysis for 28 genotypes of winter wheat. Two first components explained 70.1% of the variability of concentration of ergosterol (ERG), deoxynivalenol (DON) and zearalenone (ZEN) in grain in Poznań (P) and Radzików (R) in 2014, 2015 and 2016. Vectors indicate the direction of the increase of the value of the variables. Violet circles—toxin checks, light green circles—lines the most resistant to head infection, orange circles—lines the most resistant to kernel damage, orange labels—lines resistant to both head infection and kernel damage.

Average ERG content in grain over six environments was 16.9 mg/kg; the range of variation from 0.2 to 102.3 mg/kg (Fig. 5). Average ERG concentration in 2014 and 2016 were similar. In 2015 in ERG content was very low (2.4 mg/kg). In Radzików average ERG concentration was two times lower than in Poznań (10.4 and 23.4 mg/kg, respectively). Average DON content in grain over six environments was 12.532 mg/kg; the range of variation from 0 to 73.650 mg/kg (Fig. 5). The highest DON content was detected in grain in 2014, the lowest in 2016. In Radzików average DON concentration was about seven times lower than in Poznań (3.645 and 21.433 mg/kg, respectively). Average ZEN content in grain over six environments was 243 µg/kg; the range of variation from 0 to 4522 µg/kg (Fig. 5). The highest ZEN content was detected in grain in 2014, the lowest in 2015. In Radzików average ZEN concentration was much lower than in Poznań (16 and 451 µg/kg, respectively). Only traces of ZEN were detected in grain samples in Radzików in 2015.

Lines with stable concentration of *Fusarium* metabolites in all six environments were identified - low conc.: STH 9059, DED 389/06, LAD 463/05, KBP 04.164, POB 262/07; high conc.: DD 548/09 (tox), DM 3873/10 (tox), DM 2728/09 (tox) (Fig. 6). Two lines (STH 9059, KBP 1058) combined low head infection, low kernel damage and low toxin accumulation. Six low kernel damage and low toxin concentration and two low head infection and low toxin accumulation.
FHBi and FDK correlated significantly with concentration of *Fusarium* metabolites (r=0.484-0.749), despite FHBi vs. ERG. DON contents in years 2014-2016 correlated significantly (r=0.442, 0.386, 0.649). ERG concentration in 3 years did no correlate significantly and only concentration of ZEN in 2014 and 2015 correlated significantly (r=0.626).

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