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A COMPARISON STUDY OF BIOTIC FACTOR'S EFFECT ON PHOTOSYNTHESIS PROCESSES OF SOYBEAN BY USING MULTISPEQ DEVICE ON PHOTOSYNQ ORG PLATFORM

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ABSTRACT

One of the modern devices for monitoring photosynthesis is MultispeQ that is the open-source scientific instruments designed to collect high-quality data for in situ plant studies. Soybean (Glycine max (L.) Merr) is an important oilseed crop in Kazakhstan, but in recent years its production affected by widespread soil-borne diseases especially root rot by Fusarium spp in Western Kazakhstan. Monitoring plant physiological responses to biotic vectors are unique to evaluate the effects of different vectors on host processes involved in plant growth and yield. The aim of the study was comparison study of biotic factors (Fusarium infection and genotype). Soybean physiology was studied with the MultispeQ device on photosyng.org platform to test 12 different genotypes. R software was used to identify the most important physiological parameters in selecting the most adaptive varieties of soybean. Results showed that soil infection of F. equiseti significantly impacted and increased to linear electron flow (LEF) chlorophyl fraction indexes on Samer-1, Samer-2, Toury, Anastasya, Samer-3, Samer- 5, Belor, Swapa and Cheremosh soybean genotypes with respect to non-inoculated genotypes. The general distribution of LEF chlorophyl fraction indexes before inoculation decreased as compared to ones after experiment. Our results suggested that soil infection affected LEF chlorophyll fraction.

Keywords: multispeQ, linear electron flow, photosynthesis, soybean, two-way analysis of variance, R, fungal infection, genotype.

INTRODUCTION

The influence of biotic factors is reflected in the process of plant photosynthesis (Baghbani *et al*., 2019; Macioszek *et al*., 2019; Jimenez-Garcia *et al*., 2018). Kalaji *et al*. (2014) explained different properties of Chl-a fluorescence signal changes in photosynthesis due to using wider range of fluorescence techniques. Meantime Walker *et al.* (2018) indicated that the availability of appropriate instrumentation for the application of plant phenotyping techniques is severely limited. To address these issues, Kuhlgert *et al.* (2016) have developed a low-cost, yet sophisticated open-source scientific instrument, MultispeQ, to collect high-quality field data. But analyzing big data need special statistical approaches in plant pathology.

Soybean also is one of the important agronomic crops drawing greater attention in Kazakhstan, due to its increasing demand as an oilseed crop and animal feed to support food security (Makulbekova *et al*, 2017). However, soybean, as any other crops, is susceptible to fungal diseases with soil infection (Fones *et al*., 2017; Kalaitzandonakes *et al*., 2019; Chang *et al*., 2018; You and Barbetti, 2017). It is reported that the main agent of root rot of soybeans in Western Kazakhstan is *Fusarium equiseti* (Kuldybayev *et al*., 2019).

Resents studies on data analysis in plant pathology were focused on Gramene 2016 platform as a comparative plant genomics and pathway resources (Tello-Ruiz *et al*., 2016), plant Reactome, a resource for plant pathways and comparative analysis (Naithani *et al*., 2017), tools for comparative plant genomics and pathway resources explanation package to manipulate and visualize variant call format data in R (Knaus and Grünwald, 2017), developing optical sensors for accurate and objective detection of plant diseases (Kuska and Mahlein, 2018), application of statistical tools for data analysis and interpretation in rice (Nayak *et al*., 2018)

Our preliminary research showed that soil infection of *F. equiseti* can impact to LEF chlorophyll fractions of soybean; however, more in-depth information is needed to explain how soil infection affected plant physiology. The objective of our study was evaluate soil infection of *F. equiseti* can impact to predicted physiological variables of soybean.

METHODS AND MATERIALS

Experimental Design

The study was conducted in the Kazakh-Japanese Innovation Center of Kazakh National Agrarian University in 2019. A completely randomized design was set up with 12 soybean genotypes and replicated 5-7 times. Twelve soybean cultivars (Maple Ridge, Toury, Isidor, Samer-1, Samer-2, Samer-3, Saer-5, Swapa, Belor, Anastasya, Tanais, and Cheremosh) were conducted on resistance to soil infection with *F. equiseti* conidia suspension isolated from soybean root rot cuts from susceptible genotypes. Each soybean genotype treatment was replicated 4 times. Seeds were planted in 5-6 cm depth in 500 mL plastic pots.

Data Collection and Analysis

The project registered on the Photosynq platform, project ID 6871 [project title is "Impact of F. equiseti for soybean physiology"]. It was taken 715 measurements on soybean seedlings by using two MultispeQ 1.0 devices. The MultispeQ device equipped with relative humidity and temperature sensors and a CO_2 sensor to measure photosynthetic and biochemical parameters of soybean plants at flowering and seed formation stages. The leaf temperature differential, leaf ambient humidity, leaf ambient temperature, leaf angle, fractions of LEF, NPQt, Phi2 и PhiNO and relative chlorophyll were measured (Kuhlgert *et al*., 2016).

The following dependent variables were used: 1: Ambient temperature; 2: Leaf angle; 3: Leaf Temperature Differential - the difference between the leaf temperature and the ambient temperature; 4: LEF chlorophyll fraction; 5: PAR - photosynthetically active reaction, 6: NPQt - chlorophyll fraction; 7: Phi2 chlorophyll fraction; 8: PhiNO chlorophyll fraction; 9: PhiNPQ chlorophyll fraction; 10: relative chlorophyll.

Statistical processing of data was carried out using the R-studio program. A two-way analysis of variance (ANOVA) test was conducted with variable 1: inoculation (with 2 level - inoculated, and with non-inoculation by *F. equiseti* and 2: genotype (12 genotypes of soybean) were used as predictor factors. The two-way ANOVA test was performed before and during experiment. The significance all variables were evaluated with P-value in R Studio software (Aphalo, 2017) with these commands:

```
> M1<-aov( data$variable~genotype+ infection+ genotype:infection,data = data)
> summary (M1).
```

RESULTS

While the first measurement by MultispeQ device (before inoculation) did not significantly affect the infection to all variables, at the end of second measurement (10 days after inoculation) we observed that the infection factors significantly influenced LEF chlorophyll fraction variable (P=0.01). The impact of soil inoculation with *F. equiseti* to LEF chlorophyl fraction were detected in Samer-1, Samer-2, Toury, Anastasya, Samer-3, Samer-5, Belor, Swapa and Cheremosh genotypes (Table-1 and Figure-1). The fungal infection increased chlorophyll fraction LEF indices and root LEF indexes on infected plants than that of the non-infected plants (Table-1).

Table-1. Impact of inoculation with *F. equiseti* to indexes of soybean root rot and the LEF chlorophyll fraction parameters.

Genotype	Root rot indexes (%)		Photosynthetically parameters LEF (nm)			
			Before inoculation		10 days after inoculation	
	inoculated	non-inoculated	inoculated	non-inoculated	inoculated	non-inoculated
Samer-1	2.3	2.0	13.8	5.0	9.2	6.5
Samer-2	3.3	2.0	13.3	41.8	8.5	6.9
Isidor	2.8	1.3	10.3	5.9	6.1	6.0
Toury	1.5	0.3	11.7	10.5	10.4	7.6
Belor	1.0	0.0	23.0	12.1	8.5	8.2
Swapa	3.8	0.0	45.6	14.4	9.2	9.7
Cheremosh	1.0	0.8	29.7	9.7	10.9	9.0
Anastasya	3.5	3.3	45.9	27.9	11.2	8.7
Samer-3	2.8	1.8	19.1	39.0	18.7	13.7
Maple Ridge	1.3	0,0	13.7	18.7	6.6	6.2
Tanais	1.3	0,0	9.8	6.3	9.3	7.0
Samer-5	4.5	3.3	18.8	34.3	20.8	8.1
P-value	infection factor		0.28		0.01	

In contrast, the impact of soil inoculation with *F. equiseti* to LEF chlorophyl fraction were detected in Samer-2, Isidor, Toury, Belor, Swapa and Cheremosh genotypes.

The general distribution of LEF chlorophyl fraction before inoculation were minimal - 1.7 nm, maximal - 117.0, 1st quantile - 6.9, median - 11.4, mean - 20.5, 3rd quantile - 20.5 nm. After 10 days of inoculation with *F. equiseti* were noted the dicreasing of indexes of LEF. The minimal one was 1.2 nm, maximal - 77.3, 1st quantile - 5.5, median - 7.8, mean - 9.5, 3rd quantile - 10.85.

DISCUSSIONS

Our results showed that soil infection of *F. equiseti* have increased the LEF chlorophyl fractions in Samer-1, Samer-2, Toury, Anastasya, Samer-3, Samer 5, Belor, Swapa and Cheremosh soybean genotypes as compared with non-inoculated ones. The early fungal infection of *Fusarium oxysporum* have led to decrease net photosynthetic rate, which mainly resulted from stomatal limitation and to damage to chloroplasts, contributed to the reduction in the photosynthetic capacity of plants in the later stages of infection (Dong *et al*., 2012). Bauriegel and Herppich (2011) have reported the impact of *F. culmorum* on wheat ears to chlorophyll fluorescence variables. The photosynthetic efficiency of infected ears as compared to healthy ears decreased and shown that the disease severity highly correlated with this parameter.

The general distribution of LEF chlorophyl fraction indexes before inoculation decreased with respect to the ones after experiment. As described by Kuhlgert *et al*. (2016), the dependency of LEF on PAR fits reasonably well onto a typical photosynthesis saturation curve with a half saturation point. Huang et al. (2018) examined how selective photosytem (PSII) photoinhibition influenced LEF in the attached leaves of shade-demanding plant Panaxnotoginseng, which displayed a selective PSII photodamage under strong illumination. With these treatments, the LEF was significantly decreased under all light levels but acidification of the thylakoid lumen changed negligibly. The decrease in LEF under low light was also positively correlated with the extent of PSII photoinhibition. Results indicated that the residual PSII activity is an important determinant of LEF in this shade-adapted species, which provide new insight into how strong illumination affects the growth of shade-demanding plants.

CONCLUSIONS

Results showed that soil infection of *F. equiseti* impacted and significantly increased the linear electron flow (LEF) chlorophyl fraction in Samer-1, Samer-2, Toury, nastasya, Samer-3, Samer-5, Belor, Swapa and Cheremosh soybean genotypes than that of the non-inoculated ones. The general distribution of LEF chlorophyl fraction indexes before inoculation decreased with respect to the ones after experiment. Our next research will be focused on impact of fungal infection to stomatal cunductivity and plant physiology.

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