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TABLE OF CONTENTS

СЕЛЬСКОХОЗЯЙСТВЕННЫЕ НАУКИ

СЕЛЬСКОХОЗЯЙСТВЕННЫЕ НАУКИ

PRELIMINARY STUDY ON ANTIFUNGAL ACTIVITY OF A *STREPTOMYCES* SP. STRAIN HU2014 AGAINST PHYTOPATHOGENIC FUNGI

Zhu Hongxia[1, 2],
Wang Xinfa[1, 2]
graduate students
Rozhkova Tetiana[1]
PhD, Associate Professor
[1]Sumy National Agrarian University
Sumy, Ukraine
[2]Henan Institute of Science and Technology
Xinxiang, People's Republic of China

Introduction. Plants are continually under attack from a variety of microbial pathogens that cause diseases. Phytopathogenic fungi are one of the main causes of crop yield reduction, such as wheat scab, pepper phytophthora blight, tomato gray mold and anthracnose disease. At present, the main control methods are chemical control, planting resistant varieties and fumigation, but they also bring adverse effects, such as environmental pollution, pesticide-resistant and crop genetic variability. Therefore, new natural resources are needed to suppress these phytopathogenic fungi. *Streptomyces* is well known for broad-spectrum antifungal and antibacterial activities, and continuously discussed the potential to protect against some of the most damaging cereal crop diseases, particularly those caused by fungal pathogens.

pathogens.

Aim. For this experiment, the goal of our research was to study antifungal activity of a *Streptomyces* sp. strain HU2014. Hopefully it lays a foundation for further exploring the biocontrol mechanism in the future.

Materials and methods. In this study, the tested fungi were *Fusarium graminearum* Schwabe, *Botrytis cinerea* Persoon, *Phytophthora capsici* Leonian and

Colletotrichum gloeosporioides (Penz.) Saccardo. *Streptomyces* sp. strain HU2014 was afforded by Henan Institute of Science and Technology (HIST) in China. The fungi and the strain HU2014 were pre-cultured on potato dextrose agar (PDA) plate at 25°C for about 6 days, respectively. Fungal plugs and the strain HU2014 plugs were co-inoculated on fresh solidified PDA plate by dual culture technique (Royse & Ries, 1978). Inhibition zone were measured and calculation of inhibition rate were executed (S. Z. Li, Lu, Zhang, Gao, & Ma, 2005). Each treatment was considered of 3 replicates and a sole fungal plug as a control. The executed bioassay of the extracellular fermentation of the strain HU2014 was examined by the growth rate method (Fan et al., 2020; Zhang, Li, & Wu, 2009). The strain discs were transferred into ME liquid medium and fermented with shaking incubator for 15 days. Fermentation broth was filtered through a 0.22 μm sterile filter. The extracellular fermentation was made into 5-fold and 10- fold diluted concentration with melted PDA culture broth. Phytopathogenic fungi discs were placed separately on the center of the solidified plates and incubated at 25 °C. Each treatment was considered of 3 replicates and pure PDA medium as a control.

Result and discussion. In the previous stage, we studied the mycelial extract of the strain HU2014 and reached a conclusion that its intracellular products have excellent antifungal activity. For this experiment, we focused on the activity of the strain and its fermentation. Dual culture test showed that the strain HU2014 could produce antifungal substances in the growth process, and the antagonistic rates to all tested pathogenic fungi except *P. capsici* were more than 30 % (Tabl. 1).

Table 1

Antagonistic activity of *Streptomyces* HU2014 against pathogenic fungi

Pathogenic fungus	RCF (mm)	RTF (mm)	Inhibition rate (%)
F. graminearum	40.0±0.0	14.0±0.0	65.00±0.00
P. capsici	39.3±1.2	34.6±0.6	11.86±0.01
C. gloeosporioides	27.7±0.6	18.3±0.6	33.74±0.01
B. cinereal	39.0±1.0	20.0±1.0	48.70±0.03

Each treatment had 3 replicates, and the data showed Mean±SD. Where RCF: Radius of control fungus; RTF: Radius of treated fungus.

The antagonistic effect of *F. graminearum* was the best at 65 % inhibition rate. In the experiment of growth rate method, we measured the inhibitory effect of the extracellular fermentation of this strain with the diluted 5-fold and 10-fold concentration on four kinds of phytopathogenic fungi (Tabl. 2).

Table 2

Antifungal activity of the extracellular fermentation of *Streptomyces* HU2014 against pathogenic fungi

Pathogenic fungus	Inhibition rate (%)			
	5-fold dilution		10-fold dilution	
	48 h	72 h	48 h	72 h
F. graminearum	69.68±4.47^{a*}	57.45±3.93^{a}	47.45±2.19^{b}	43.27±1.09^{c}
P. capsici	7.83±0.70^{d}	2.17±0.81^{e}	9.04±0.40^{d}	-
C. gloeosporioides	40.91±5.21^{c}	33.76±7.96^{b}	12.94±0.56^{d}	15.92±1.91^{d}
B. cinereal	100.00±0.00^{a}	100.00±0.00^{a}	90.86±7.97^{b}	69.16±0.93^{b}

Each treatment had 3 replicates, and the data showed Mean±SD. * mean values followed by different letters in each column are significantly different ($P<0.05$).

The fermentation metabolites of the strain could produce antifungal substances, and the inhibition rate of the extracellular fermentation to *B. cinerea* was 100% with 5-fold dilution and over 69 % with 10-fold dilution at 72 h, and to *F. graminearum* was 69.68% with 5-fold dilution and 47.45% with 10-fold dilution at 48 h. The inhibitory effect against *P. capsici* and *C. gloeosporioides* was poor. The inhibition rate to *C. gloeosporioides* was 33.76% with 5-fold dilution and 12.94% with 10-fold dilution before 72 h, as to *P. capsica*, the inhibition rate was no more than 10% within the time range of all concentration determination. From the above results, we can confirm that those reports in which *Streptomyces* sp. has broad spectrum antibacterial activities on *B. cinerea* (Bi & Yu, 2016; Vijayabharathi et al., 2018) and *F. graminearum* (Colombo et al., 2019; Han et al., 2021). The performance of *B. cinerea* was remarkably different from 5-fold dilution and 10-fold dilution in the bioassay, which may be interpreted as the characteristics of strains for testing, such as growth rate and large genetic variation. At the same time, the sensitivity of different strains to reagents is different (Baraldi et al., 2003). At the same time, we found that

the strain HU2014 scarcely possessed antifungal activity against *P.* capsici, which may be related to the symbiosis of flora. Collectively, the strain HU2014 had good antifungal activity against the above two phytopathogenic fungi.

Conclusions. We know plant diseases cause a significant decline in yield in the world every year. Recently, the biological control of these diseases has been drawing more attention due to its high efficiency and environmental friendliness. The results of this experiment are exciting. The next experiment will further study the isolation and purification of the antifungal substances of this strain HU2014 and the colonization in soil.