Biological characteristics of *Beauveria bassiana* (Clavicipitaceae: Hypocreales), collected from overwintering sites of Sunn Pest, *Eurygaster integriceps* (Scutelleridae: Heteroptera), in Iran

Farzaneh Kazemi Yazdi, Jørgen Eilenberg

Dep. of Agriculture and Ecology; Plant and Soil Science, Faculty of Life Sciences, University of Copenhagen, Thorvaldsensvej 40; DK-1871 Frederiksberg C, Denmark. (jei@life.ku.dk)

Introduction

Sunn pest, *Eurygaster integriceps* Puton (Scutelleidae), is a key pest of wheat and barley in Europe, North Africa, the Middle East and Iran (Brown, 1962 and Rassipour *et al*, 1996). Chemical control is the only method used to prevent Sunn pest damage (Javahery, 1995). Due to adverse side effects of insecticides on environment, natural enemies and humans, alternative environmentally-friendly control methods such as biological control can be considered. The potential pathogenicity of entomopathogenic fungi, *Beauveria bassiana*, was studied in the field (Edington *et al*., 2007 and Skinner *et al*, 2007). Our goals were: 1) to find fungal-infested Sunn pests from overwintering sites in Iran, and 2) to isolate and evaluate their biological characterization under laboratory conditions.

Material & Methods

Out of 100 isolates collected from over-wintering sites in some parts of Iran, 9 isolates were chosen. In addition, one highly virulent isolates, which was found in Turkey by other researchers, was included in this research. The insects were exposed to each isolate by placing them in a Petri dish of a well grown fungus (Fig.1) for 45 min (at room temperature), after which, the insects were placed in 15-cm Petri dishes which included a wet cotton ball and 20 wheat grains (at 27° C). The mortality was recorded for 14 days (Fig.2). Data on mortality of *E. integriceps* were analyzed using the proportional odds model (*R* software). The effect of isolate on the mortality of insects was tested using the likelihood ratio test of ordinal regression model (Chi-sq). The duration of experiment (14 days) was divided to 3 groups (1-6, 7-11 and 12-15 days), and the mortality of insects within these groups were tested for different isolates. Pair-wise t-test was used to calculate pair-wise comparisons between isolates with corrections for multiple testing. Based on the above test, two isolates with the highest mortality was chosen for further study. The concentrations for 'IR-K-40' were 23×10^7 ,23×10^6 ,23×10^5 and 23×10^4 conidia/ml, while for isolate 'SPT 22', concentrations were 8×10^7 , 8×10^6 , 8×10^5 and 8×10^4 conidia/ml. For each concentration, the topical method was used where 0.5 µl of each concentration were applied to each insect with a sampler. Dose-response_test for the Isolates 'SPT 22' and 'IR-K-40' were analyzed using the probit model in SAS. The data was corrected with the control mortality. Data were log-transformed where needed.

Results and Discussion

All of the selected isolates showed pathogenicity against *E. integriceps*. Five Iranian isolates 'IR-K- 40', 'IR-K-58', 'IR-K-10', 'IR-K-53' and 'IR-K-43', and one Turkish isolate 'SPT 22' were highly virulent isolates (mortality: 88.7–94.4 %) (Table 1). Isolates 'IR-K-40' and 'SPT 22' were selected for dose response study. The LC50 was 2.9×10^7 conidia/cm2 for both isolates (P<0.0001). The results of this study showed that *B. bassiana* can be isolated from Sunn pest overwintering sites in Kermanshah province of Iran. In addition, the selected isolates were pathogenic to *E. integriceps* in the laboratory. These results will help in selection and development of these fungi for Sunn pest management in future.

Table.1. Mean percentage (range) mortality of *E. integriceps* treated with 10 *B. bassiana* strains and percent mortality of non-treated control

Isolate	species	Mortality		
		Day 6	Day 11	Day 15
control	*B. bassina*	0.1(0-0.3) %	2.5(0.8-7.8) %	5.6(1.8-16.2) %
IR-K-40	*B. bassina*	22.5(7.1-52.3) %	88.0(65.9-96.5) %	94.4(81.7-98.4) % * abce
IR-K-58	*B. bassina*	17.6(5.3-45.1) %	84.3(58.3-95.4) %	92.5(76.7-97.9) % * abce
SPT 22	*B. bassina*	16.0(4.8-41.7) %	82.8(56.1-94.7) %	91.7(74.7-97.6) % * abce
IR-K-10	*B. bassina*	14.6(4.3-39.5) %	81.2(53.1-94.3) %	90.9(72.3-97.4) % * abcde
IR-K-53	*B. bassina*	13.1(3.8-35.6) %	79.1(50.0-93.5) %	89.7(69.8-97.1) % * abcde
IR-K-43	*B. bassina*	11.9(3.5-33.7) %	77.3(47.5-92.7) %	88.7(67.6-95.9) % * abcdef
IR-K-31	*B. bassina*	7.3(2.1-22.6) %	66.5(34.9-88.0) %	82.1(55.3-94.4) % * bcdefg
IR-K-37	*B. bassina*	5.0(1.4-16.2) %	56.9(26.4-82.6) %	75.3(75.3-91.8) %* defghij
IR-K-36	*B. bassina*	2.6(0.7-8.9) %	40.3(15.7-71.0) %	61.0(30.1-85.0) %* ij

A * indicates a significant difference of the isolates from the control isolates followed by the same letter were not significantly different for each column.
Yellow color: high virulent
Blue color: medium virulent
Red color: low virulent

Table.2. Percent corrected cumulative mortality of *E. integriceps* treated with four concentrations of isolates IR-K-40 and SPT 22 and percent mortality of non-treated control

Isolate	species	Mortality		
		Day 6	Day 11	Day 15
control	*B. bassina*	2.7(1.5-4.3) %	7.1(4.6-10.8) %	9.7(6.5-14.7) %
SPT 22($8x10^{4a}$)	*B. bassina*	2.3(0.8-6.1) %	6.1(2.2-15.4) %	8.3(3.1-20.4) %
SPT 22($8x10^5$)	*B. bassina*	4.6(2-10.1) %	11.8(5.4-24.0) %	15.9(7.5-30.7) %
SPT 22($8x10^6$)	*B. bassina*	2.1(6.3-21.9) %	27.7(15.8-43.9) %	35.0(18.9-49.4) % *
SPT 22($8x10^7$)	*B. bassina*	35.8(22.9-51.2) %	60.9(45.3-74.5) %	68.7(50.8-78.5) % *
IR-K-40($23x10^4$)	*B. bassina*	4.4(1.9-9.7) %	11.4(5.2-23.1) %	15.32(7.2-29.7) %
IR-K-40($23x10^5$)	*B. bassina*	2.2(0.9-6.0) %	6.0(2.4-15.2) %	8.2(3.0-20.2) %
IR-K-40($23x10^6$)	*B. bassina*	12.8(7.0-22.2) %	29.1(17.4-44.3) %	36.6(20.9-49.9) %*
IR-K-40($23x10^7$)	*B. bassina*	51.7(35.9-67.2) %	75.0(61.0-85.1) %	80.8(66.2-87.7) %*

A * indicates a significant difference of the isolates from the control. If the confidence interval contains zero then the isolates are not significantly different from control.
[a] The conidia concentration (conidia/ml)

Fig. 1:Petri dish containing a well-grown fungi

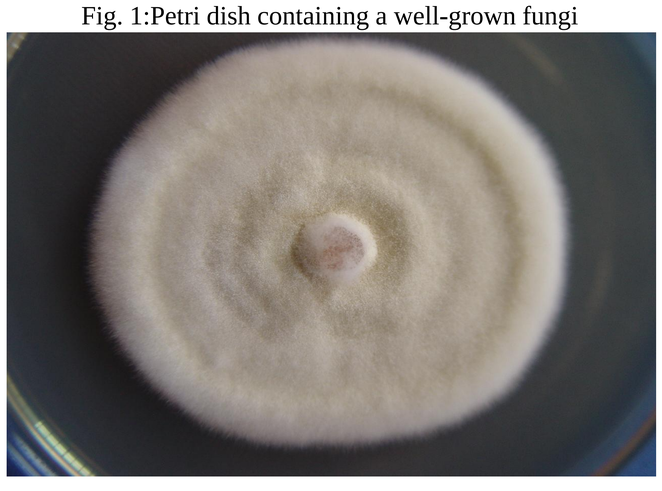

Fig. 2: A dead Sunn pest infected by *B. bassiana*

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