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Life history of *Plodia interpunctella* Hübner on sunflower seeds: Effects of seed qualitative traits and the initial seed damage

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ABSTRACT

Sunflower seeds are regularly infested by *Plodia interpunctella* during storage. Although this pest prefers damaged seeds, in practice it can infest undamaged seeds as well. This research assessed the influence of the sunflower seed type (oil, protein for human consumption and bird-feed) and the initial seed damage during post-harvest processing (dehulled kernels, 10, 20, 30% of damaged seeds and undamaged seeds) on development of *P. interpunctella* (larval mortality, larval development, mean developmental duration, adult emergence and fecundity). Biochemical analysis of seeds, kernels and hulls detected the highest content of phenols in the seed and hull and tocopherols in the kernel of the oil type hybrid. The antioxidative activity was the highest in the seed, kernel and hull of the protein type for bird feed. The shortest development (39.5 days) and the highest fecundity (91.3) were on the oil type seeds, while the longest development (42.1 days) and the lowest fecundity (68.1) were on the seeds of the protein type for bird feed. The highest mortality of larvae was on the undamaged seeds of the protein type for bird feed and human consumption (21.3% and 14.0%, respectively). The type of sunflower and the level of initial damage affected larval mortality, developmental duration and fecundity. The mean developmental duration and the number of emerged adults were dependent only on the initial seed damage. Principal component analysis detected strong positive correlation between mortality and development with the tocopherol content on the undamaged seeds while fecundity was associated with the state of kernel and the amount of tannins, proteins and oil content in the seed. The undamaged seeds of the protein type for the bird feed were the least suitable for the development of this pest, while the oil type kernels were the most suitable.

1. Introduction

The sunflower (*Helianthus annuus* L.) is the most important oil crop in the Republic of Serbia and one of the four major oil crops in the world (Anandhan et al., 2010; Balalić et al., 2012). According to FAO estimates, it is cultivated on over 26.2 million hectares in more than 70 countries (FAOSTAT, 2016). The EU sunflower seed production in 2016 was 34.0 million tonnes, a decrease of −14.8% compared to 2014, followed by an increase of 10.7% between 2015 and 2016 (FAOSTAT, 2016). Two basic types of sunflower seeds are cultivated: the oil type for the production of vegetable oils, and the non-oil type (protein, confectionery or nibbling), intended for the human consumption and bird-feed (Jocić et al., 2015).

The quality of sunflower seeds is influenced by the genetic traits inherited from their parent lines, germination and vigour, but also by the post-harvest processing and storage conditions (Zambolim, 2005; Prole et al., 2010). One of the main concerns about the long-term seed storage is the physical damage of the seeds, which

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is not always visible. The sunflower seeds have an easily cracking shell, which makes them very susceptible for the development of pests in storages (Beratlief and Iliescu, 1997). Seed damage could occur in production, harvesting, drying or storing process, but could be also caused or enhanced by storage pests, and usually results in the reduction of the seed germination (Stejskal et al., 2014; dos Santos et al., 2016). Stored product pests can sense certain volatiles which are released by the damaged seeds (Trematerra et al., 2000, 2007, 2013, 2015, 2016; Athanassiou et al., 2006). Under these conditions, sunflower grains are regularly infested, mainly by secondary pests such as *Plodia interpunctella* (Hübner, 1813), *Oryzaephilus surinamensis* (L., 1758) *Cryptolestes ferrugienus* (Steph, 1893), *Tribolium castaneum* (Herbst, 1797) and *T. confusum* (Du Val, 1863), which prefer the damaged seeds, because they feed primarily on the germ or seed/grain dust (Storey, 1987; Golob et al., 2002; Silhacek and Murphy, 2005). Out of all mentioned species, *P. interpunctella* is one of the most important pests frequently found in sunflower storages, which often causes deterioration of the seeds (Atanasov, 1974; Beratlief and Iliescu, 1990; Gvozdenac et al., 2018).

The development of *P. interpunctella* is highly dependent on the nutritive quality of the available food (Locatelli and Limonta, 1998; Vukajlović and Pešić, 2012; Predojević et al., 2017; Vukajlović et al., 2017). Proteins, polyunsaturated fatty acids, vitamins and steroids in a diet are very important for the fast and successful development of this moth (Vukajlović and Pešić, 2012; Vukajlović et al., 2017; Predojević et al., 2017). Oilseeds, like the sunflower are rich in proteins and fats (Beratlief and Iliescu, 1997; Sarwar et al., 2013), which is why they are very suitable for a strong colonization of *P. interpunctella*. A protein-rich diet, such as a germ in seeds or an oilseed itself is very suitable for *P. interpunctella*, as it leads to short developmental duration and high fecundity (Almaši, 1984; Silhacek and Murphy, 2005, 2006; Onaolapo et al., 2017).

Most studies on the development of the *P. interpunctella* have been carried out on stored cereals or cereal products. However, the information on the development of this species on the sunflower is scarce. Several facts inspired this research. Namely, there is a constant increase in the sunflower production (from 167 000 ha to 231 000 ha) and the seed export (from 15 671 t to 136 580) in Serbia, in the last five to seven years, 2011–2018 (Annonimus, 1, 2017). At the same time, there is a registered increase of *P. interpunctella* population in Serbian storages which indicates the growing importance of this moth as a pest of sunflower seeds. The last published research on *P. interpunctella* as a pest of sunflower in Serbia date back from 1980s (Vukasović et al., 1966; Stojanović and Kosovac, 1974; Almaši, 1984). Therefore, the increase of economic importance and damages caused by this pest imposed a need for a more detailed study of *P. interpunctella* on sunflower. Therefore, the aim of this work was to assess the influence of different types of the sunflower seeds (oily and protein) and the level of the initial seed damage that occurs during the post-harvest processing, on *P. interpunctella* life history parameters.

2. Materials and methods

2.1. Insect culture

The *P. interpunctella* culture used in this research originates from the population reared for ~50 generations in transparent plastic containers for mass rearing (5 L), in a thermostat chamber, at 28±1 °C, r.h. 60 ± 10% and photoperiod 14:10 (L:D), on standard a laboratory diet (Silhacek and Miller, 1972), consisted of ground dog meal (10%), rolled oats (4%), white cornmeal (26%), whole wheat flour (23%), wheat germ (2%), brewers' yeast (5%), glycerol (16%), and honey (14%). 100 pairs of one-day-old adult males and females *in copuli* were isolated with an entomological aspirator from the containers for mass rearing and placed into 1 L glass jars where the females laid eggs. The one-day-old eggs used in the experiment were carefully removed from the ovipositional jars with a brush made from fine hairs. Before the experiment, eggs were observed under a binocular microscope to eliminate those with obvious deformities. Only undamaged, whole one-day-old eggs were used in the experiment.

2.2. Sunflower seeds as a feeding nutrient

The seeds of three sunflower types, namely hybrids, were used in this research. Hybrid Leone is the oily type (OT) with a high oil (46–48%) and low protein content in seeds (20–22%). NS Colonel is a protein type hybrid intended for human consumption (PT) with the lower oil content (35%) and higher protein content (26%). Lactal is a protein hybrid intended for bird-feed (PBF) with a low content of oils (34–38%) in kernels and white hulls. The seeds were obtained from 2016 vegetation season, from The Institute of Field and Vegetable Crops, Novi Sad, Republic of Serbia.

2.3. Qualitative biochemical analysis of sunflower seeds

The biochemical analysis encompassed the determination of the content of total phenols, total tannins, flavonoids, phenolic compounds and tocopherol. Whole seeds (achenia), hulls and kernels (1 g) were grounded to a fine powder in a mill and extracted after 24 h with 50 mL of 70% methanol. The extracts for the spectrophotometric analysis (*Thermo Scientific* Evolution *220 spectrophotometer, Madison WI, USA)* were vacuum-filtered through the sintered glass funnel and kept refrigerated until the analysis. All extractions were performed in triplicates (Bereksi et al., 2018).

The total phenolic content (TP) was determined using a Folin-Ciocalteu colorimetric method (Nagavani and Raghava Rao, 2010) and the results were expressed in milligrams of quercetin equivalents per 1 g of a plant material (mg QE/g). Extract (20 μL) was mixed with 3.4 mL of deionized water, 0.4 mL of 20% sodium carbonate and 0.2 mL of Folin-Ciocalteu reagent, which was previously diluted (1:2) with distilled water. After incubation at room temperature for 30 min, the absorbance of the reaction mixture was measured at 720 nm using an UV/VIS spectrophotometer. The data are reported as means for three replications. The total tannins (TT) content was determined by the Folin-Ciocalteu procedure, after the removal of tannins by their adsorption on an insoluble matrix PVPP (polyvinylpolypyrrolidone). Briefly, 1 mL of extract, in which the total phenolics were determined, was mixed with 100 mg PVPP, vortexed, kept for 15 min at 4 °C and then centrifuged for 10 min at 3000 rpm. In the clear supernatant the non-tannin phenolics were determined the same way as the TP. The calculated values were subtracted from the TP content, and the TT content was expressed in milligrams of quercetin equivalents (QE) per 1 g of the plant material (Nagavani and Raghava Rao, 2010).

DPPH (1,1-Dyphenyl-2-picrylhydrazyl) radical scavenging activity. Scavenging of free radicals was tested in a DPPH (2,2-diphenyl-1-picrylhydrazyl) acetone solution (Lai and Lim, 2011). DPPH is a stable free radical and accepts an electron or hydrogen to become a stable diamagnetic molecule. Plant extracts (200 μL) were added to 2.0 mL of 50 μM acetone DPPH solution. The mixture was left in the dark for 30 min before reading the absorbance at 517 nm with 70% methanol as blank. Radical scavenging activity was expressed as mg trolox equivalents (TE) per gram of plant material (mg TE/g). The ferric-reducing antioxidant power (FRAP) assay was carried out according to the procedure described in the literature (Valentão et al., 2002).

FRAP assay is based on the ability of antioxidants to reduce Fe 3 + into Fe 2 + in the presence of 2,4,6-tri (2-pyridyl)-s-triazine