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(Original Article)

Pesticides Susceptibility and Detoxification Enzyme Activities of *Phthorimaea operculella* (Zeller) (Lepidoptera: Gelechiidae) Under laboratory Conditions

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Abstract

Phthorimaea operculella (Zeller) (Lepidoptera: Gelechiidae), is the most devastating insect pest that attacks potato crops in fields or storage. Pesticides are important to reduce population of this pest. The intent of this study was to investigate the sensitivity levels of *P. operculella* population field comparing with a reference to susceptible strain and biochemical analysis of technique(s) engaged in indoxacarb, sulfoxaflor and emamectin benzoate metabolism to 3 different detoxification enzymes (CPR-DPPH, GST-CDNB, and EST- PNPA). Resistance ratios were 11.9, 1.3 and 3.3 folds for indoxacarb, sulfoxaflor and emamectin benzoate, respectively in *P. operculella* field population. Biochemical analysis displayed that CYP450-DPPH and GST-CDNB activities show no a considerable ($p<0.05$) superfast compared with susceptible strain, furthermore, EST- PNPA activity showed a 2.7 fold increase compared to susceptible population. Bioassay analyses displayed moderate resistance to indoxacarb while a little resistance showed in at field population to emamectin benzoa of *P. operculella*. Esterases have a major role in the increase of resistance to indoxacarb, cytochrome P450 may have an elementary role in resistance against emamectin benzoate, GSTs do not apparently involve in the development of resistance against indoxacarb and emamectin benzoate of *P. operculella*. These results involved important practical application in managing pesticide resistance in *P. operculella* populations.

Keywords: *Phthorimaea operculella, Pesticide resistance, Esterases, GST, P450, Potatoes*

Introduction

Potato, *Solanum tuberosum* L. (Solanaceae) is considered one of the main vegetable crops in the world and Egypt is rated among the world's best potato exporters (Natikar and Balikai, 2018). The potato tuber moth (PTM), *P. operculella* attacks potato crops in the field or storage, the damage starts from the field and then the store with 100% tuber yield loss if no intervention is performed

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(Aryal and Jung, 2019). Larvae of potato tuber moth (PTM) in East Africa attack both leaves and tubers in the field and infested tubers imported in heaps in warm and dry conditions (not refrigerated). (Alvarez *et al*., 2005). Potato tuber moth in orbital regions and other areas has the capability to evolve resistance to pesticides, so it become a mounting agricultural disquiet (Amiri and Bakhsh, 2019).

The neonate larvae of *P. operculella* are more effective under chemical control measures before penetrating the tubers (Valderrama *et al.,* 2007). Potato crop spraying about 10 times during one growing season through unscrupulous of farmers which leads to development of resistance against the applied insecticides (Sharma, 2013). Synthetic insecticides namely organophosphate, carbamates, and pyrethroids, are used for controlling PTM in Egypt. Vegetable farmers applied insecticide one to two per day in order to that, so some resistance was expected would be present (El-kady, 2011). The early detection of development of insecticide resistance which was registered for use against *P. operculella* by determining of basis line toxicity of it is substantial for resistance to insecticide administration. The biochemical mechanisms and physiological for resistance insects of insecticides through four trajectories: metabolism resistance (more rate of detoxification of insecticides), target-site resistance, sequestration of the insecticide, and penetration resistance (Li *et al*., 2007; Ahmad *et al*., 2006; David *et al*., 2013 and Nkya *et al*., 2013). The metabolic detoxification is more important common mechanism of conferring insecticides resistance which mostly involves metabolic enzymes namely GST, carboxy/cholinesterases (CCE), Cyt P450, AChE and Esterases, these enzymes can amplify genes via changes in coding sequence and overexpression to mutate the detoxification capacity and then causing resistance (Li *et al*., 2007; Navarro-Roldan *et al*. 2017and Farouk *et al*., 2021). The insect P450s by participating in the appointed activation of insecticide precursors, is supplying a protection role against xenobiotic, by metabolized the same compounds to avoid toxicity, affecting insecticide selectivity, thus most insect species participate in the output of subaltern metabolites that work as chemical defenses by comparable numbers of the cytochrome P450 in their mitochondrial clans (Jeschke, 2016).

There was noteworthy progress in the identification of P450 genes in resistant insects and related systematic mechanisms with it due to expansion molecular and bioinformatics technologies. (Ye *et al*., 2022). Esterases stimulate the diversion of esters for acid and alcohol, so it plays an important role in the toxicity reduction of spinosad and abamectin insecticide in Brazilian populations of *T. absoluta* (Barata *et al*., 2004; Huang and Ottea, 2004; Siqueira *et al*., 2011; Reyes *et al*., 2012 and Hatfield *et al*., 2016). What is produced during insecticides metabolism from lipid peroxidation products, GSTs work on lowering oxidizer stress by unloading types of reaction and detoxification from it (Dauterman, 1985; Grant and Matsumura, 1989; Reidy *et al*., 1990 and Vontas *et al*., 2002). Cytochrome P450-monooxygenases have an effective role in the metabolism of pesticides from various groups, as a basis participate of cartap and spinosad resistance in *T. absoluta* populations (Feyereisen,2005). Cytochrome P450, EST and PSMO enzymes belong to group enzymes that attach a polar group to toxic Materials e.g.,

insecticides or their schism in the body, while GST works on append amino acid, sugar, sulfate, or phosphate group on Phase I product to increase the polarity to excrete from the insect body (Brown and Brogdon, 1987; Bernard and Philogene, 1993; Despres *et al*., 2007; Hatipoglu *et al*., 2015 and Navarro *et al.*, 2020). Cross-resistance is a term for genetically talented characteristics of pests that bears the effect of pesticides from various class have the same influence a result to treatment by some other related chemicals (Georghiou, 1980; Brattsten *et al*., 1986; Ware, 2000; Wang and Wu, 2007 and Yu, 2007). All strains of *P. operculella* showed various degrees of resistance to the 7insecticides (Imidaclopride, primiphos-methyl, deltamethrin, carbosulfan, Aldicarb, Fenitrothion, Lambda-Cyhalothrin) studied, the lowest Behera population was (LC_{50} = 0.52 ppm), while the highest one was recorded at LC_{50} of 715.7 ppm in Damytta population for fenitrothion, while there was moderate resistance in Behera strain (59 and 49.5 fold) to fenitrothion. Dakahlia, Menofia and Behera population gave similarly resistance results to primiphos-methyl (29.2, 28.7 and 23.6 fold respectively), On the other hand Behera strain exhibited unacceptable resistance to deltamethrin (13.7 fold), but the greater resistance to deltamethrin displayed by Damytta, Dakahlia and Menofia strains (81.2,63.7 and 41.4-fold resistance, respectively (El-kady, 2011).

In resistance, cotton leafworm to abamectin GST, oxidases and esterases play a main role as resistance mechanism as in resistance of *P. xylostella* to abamectin (Clark *et al*., 1995 and Wu *et al*., 2001). In the insect nervous system, emamectin benzoate acts as a lasting tonic of the chloride channel, which muscular activity and in the end insect death. (Ishaaya, 2002).

In the current study, pesticides were chosen from a different mode of action mechanism, several reports are available on these pesticides efficacy of *P. operculella* developmental stages, However, up until now, there are no enough reports available regarding its resistance and mechanisms of this resistance, So, it is important to monitor the pesticide efficacy to provide optimal *P. operculella* control and mitigate the potential resistance development to three different detoxification enzymes (CPR-DPPH, GST-CDNB, and EST- PNPA).

Materials and Methods

Insect colonies

PTM lab colony was brought from a population of the biocides unit which collective rearing environment for >5 years at Plant Protection Research Institute (PPRI), Giza governorate. The field strain originated from infested potato foliage in field experiments before pesticides under study application., Experiments were conducted at Plant Protection Department Laboratory and Farm, Faculty of Agriculture, Assiut University, Assiut, Egypt. The colonies were detained in various ecological rearing places at 26 °C ± 1°C with 24-h scot stage in this study to embargo intermixing, the rearing for two colonies happened by using PTMRU method (Taha and Hassan, 2021). First instar larvae were used for bioassays; for biochemical activity assays and cytosol preparation, complete last instar larvae were used.

Pesticides and chemicals

While commercial formulation of indoxacarb (Easo plus® 30%WG), emamectin benzoate (Egy Chem® 5.7%WG) and sulfoxaflor (Closer® 24%sc) were supplied by Starchem, Hebei xingbai and Dow Agro Scinces Co. Egypt, respectively. The other chemicals such as ethanol (analytical grade) and J. T. BAKER Chem. Co was source of potassium phosphate.1-Chloro, 2-4-dintrobenzene (CDNB), reduced glutathione (GSH), p-nitrophenyl acetate (PNPA), Fast blue RR, acetylthiocholine iodide (ASChI), Sigma Chem. Co. which source to brought 5, 5-dithiobis (2-nitrobenzoic acid) (DTNB) and bovine serum albumin (BSA), P. O. Box 14508 St. Louis Mo 63178 USA.

Bioassay method

1. LC_{50} values and Resistance ratios evaluation

Leaf-dip bioassay method (Symington, 2003) was go run on 1st instar larvae of the first laboratory generation of PTM. For 30 seconds cabbage leaf discs were treated with different pesticide concentrations, after drying at room temperature, at which time ten larvae (<24 hours)' were used. Three replicates were done for all treatments. Leaves dipped in water as a control. After placing all larvae, containers were placed in a sitter at 27±1 °C. The results were recorded at 72 hours after moving larvae to leaves. The surviving larvae of the field colony were fed on the treated leaves by the concentration of each LC_{30} value of treatments to complete their development, after that the lives of the last instar larvae were the source of the enzyme extraction procedure.

2. Preparation of cytosols from *P. operculella* larvae for enzyme activities analysis

GSTs, esterase and P450 enzymes activity for larvae of *P. operculella* were measured according to (Hemingway, 1998). In 1 ml of potassium phosphate buffer at 100 mM, 10 larvae in Batches of PTM larvae were homogenized on ice at tissue grinder, pH 7.4 and 1 m M DTT(Dithiothreitol,) 1 mM at 10.000 x g at 4°C for 30 min EDTA (ethylene-diamine-tetraacetic acid), and 1 mM PMSF (Phenyl methyl sulfonyl fluoride) were homogenization and centrifuged. After that, supernatants were collected as enzyme sources. spectrophotometer was used to measure Absorbance levels at particular wavelengths for each enzyme to three replicates per enzyme and blank. Finally, Bradford method (Bradford, 1976) was used to measure protein concentrations, at 595 nm using UV spectrophotometer, CHEM-7.

3. Determination of esterase activity

According to Van Asperen method (Van Asperen, 1962) at volume of 200 µl using (PNPA) as a substrate at 37 °C with 30-s intervals for 10 min, in each cuvette, 0.05 % Triton X-100, 3.8 mM PNPA and 15 µl of supernatant in 100 mM potassium phosphate buffer, pH: 7.0. the reaction was stopped by adding staining reagent 1 mL (50 µl) of BS salt (1%), phosphate buffer (0.04 M). The enzyme

activity was stated as n M /min/mg. protein at 405 nm at spectrophotometric device.

4. Glutathione-*S*-transferase (GST) assay

Method (Habig *et al*., 1974) was used to bioassay. In each cuvette, enzyme source (1 mM) was mixed with 100 mM potassiumphosphate buffer, pH: 7.4, at 25 °C for 5 min., 1 mM of CDNB was added. cuvette contained all reactions except substrate for blank sample. The assay was at triplicate; the absorbance was recorded at 340 nm. appointed activity was calculated as n M/min/mg protein.

5. NADPH(nucleotide adenine diphosphate) -cytochrome P450 activities

The method (Yim *et al*., 2004**)**. potassium phosphate buffer, pH: 7.6 in 1 ml, containing 100 µM of NADPH, ≈100 µg of cytosolic protein and 100 µM of DPPH (as a substrate) The molar suppression coefficient assay at 520 nm was 4.09 mM-1cm-1. The activities of CPR-DPPH were expressed as pmole/min/mg protein.

Statistical analysis

SPSS version 20 software program was used for estimating LC50 values and probit analysis for confidence limits (Abbott, 1925). The ratio between LC50 values of field population to LC50 values of the susceptible population was used for measurement resistance ratios (Torres *et al*., 2002). Similarity or difference in enzyme activities was run to one-way analysis of variance (ANOVA) by using Costat program (Costat, 1998) and significant differences among the means values were determined according to (Duncan, 1955) probability levels of $P = 0.05$. at least significant difference (LSD) SPSS analysis program version 20 was used to appreciate the correlation between the changes in enzyme activities and resistance ratio.

Results and Discussion

Data in Table (1) presented the toxicity of selected pesticides on laboratory. and field strains of *P. operculella.* after 72-h of exposure under laboratory conditions. In general, the selected pesticides demonstrated more toxicity on lab strain rather than field strain. Emamectin benzoate found to be the most potent pesticide among the selected pesticides and the LC_{50} values were 22.96 and 74.86 µg/ml for lab and field strain, respectively. However, indoxacarb and sulfoxaflor were the least toxic pesticides. The LC_{50} values for indoxacarb were 29.65 and 353.65 µg/ml for lab and field strain, respectively. Further, The LC_{50} values for sulfoxaflor were 45.42 and 60.34 µg/ml for lab and field strain, respectively.

According to the slope values of the toxicity selected pesticides of *P. operculella* demonstrated relative high homogeneity response to sulfoxaflor pesticide and slope value was 1.08). In contrast, *P. operculella* exhibited heterogeneity response to the rest of selected pesticides for both strains (laboratory and field strains).

Based on the resistance ratio (RR) values, *P. operculella* (field strain) stated most resistance to indoxacarb and the RR value was 11.93-fold followed by emamectin benzoate (RR value was 3.26-fold) and sufloxaflor (RR vale was 1.33-fold).

Table 1. Effectiveness of some pesticides against 1st instar larvae of laboratory and field strains of *P. operculella* in dipping bioassay after 72-h of exposure

Pesticides	Strain	LC_{50} (µg/ml) [a] (95% Cl)	Slope ± SE [b]	Resistance ratio (RR) [c]
Indoxacarb	Lab	29.65 (12.12-51.89)	0.94 ± (0.11)	-
	Field	353.65(168.97-1303.51)	0.59 ± (0.15)	11.93
Sulfoxaflor	Lab	45.42 (24.16-71.77)	1.08 ± (0.14)	-
	Field	60.34(25.10 – 109.69)	0.75 ± (0.15)	1.33
Emamectin benzoate	Lab	22.96 (8.97-76.14)	0.58± (0.06)	-
	Field	74.86(27.36 – 265.98)	0.67 ± (0.10)	3.26

[a] LC: lethal concentration; CI: confidence interval; [b] SE: standard error of mean.
[c] Resistance ratio (RR)=LC_{50} of filed strain / LC_{50} of lab strain. The resistance ratios were considered to be significant if their value was greater than 1: as described by Torres-Vila et al., (2002) based on the followed scale: Susceptibility (RR=1), low impedance (RR=2-10) or moderate (RR=11-30), high resistance (RR=31-100) and very high resistance (RR>100).

Data in Table (2) demonstrated the results of the effects of indoxacarb pesticide on three different detoxification enzymes on *P. operculella* under laboratory conditions. The high level of enzyme activity was GST for lab. and field strain and the values were 317.2 and 345.4, respectively. However, CPR was the second active enzyme, and the values were 74.6 and 92.4, respectively. Further, the least active enzyme among the tested enzymes was EST and the values were 32.6 and 86.5, respectively. According to the fold increase in activity for the three enzymes for both strains, EST was the highest among the tested enzymes by 2.7-fold followed by CPR with value of 1.2-fold and the least fold increase in activity was GST and value was 1.1-fold.

Table 2. Effect of indoxacarb on detoxification enzyme activities, GST-CDNB, CPR-DPPH and EST-PNPA, on lab and field strains of *P. operculella*

Enzyme	Lab Population		Field population		
	(N)*	Activity ±SE[1]	(N)*	Activity ±SE[1]	Fold increase in Activity [2]
CPR-DPPH	22	74.6±1.8 a	20	92.4 ± 5.03 b	1.2
GST-CDNB	22	317.2±1.4 a	22	345.4 ± 3.2 a	1.1
EST- PNPA	26	32.6±2.3 a	18	86.5 ± 3.7 b	2.7

[1] pmole min^{-1} mg $protein^{-1}$ for enzyme activities ±Standard Error of Mean
[2] Fold increase =Enzyme activity in field strain / Enzyme activity in lab strain
* Means with the different letter within the same row are insignificantly different ($P \leq 0.05$) according to Duncan's Multiple Range Test
* Sample size indicate number of pools which contains ≈10 larvae of *P. operculella*

The effects of the emamectin benzoate on three specific detoxifying enzymes on *P. operculella* were shown in Table (3) under laboratory conditions. The lab. and field strains both had high levels of the enzyme GST, with values of 289.5 and 351.7, respectively. The values for CPR, on the other hand, were 83.7 and 141.3, respectively, making it the second active enzyme. Furthermore, EST had values of

34.3 and 36.9, being the least active enzyme among those examined. According to the fold increases in activity for the three enzymes in both strains, CPR had the highest value (1.7-fold) followed by GST, which had a value of 1.2-fold, and EST, which had the lowest value (1.1-fold).

Table 3. Effect of emamectin benzoate on detoxification enzyme activities, GST-CDNB, CPR-DPPH and EST-PNPA, on lab and field strains of *P. operculella*

Enzyme	Lab Population		Field population		
	N	Activity ±SE	N	Activity ±SE	Fold increase in activity
CPR-DPPH	20	83.7±1.4 a	22	141.3±4.5 b	1.7
GST-CDNB	22	289.5 ±0.8 a	22	351.7± 2.7 b	1.2
EST- PNPA	26	34.3±0.7a	24	36.9±2.08 a	1.1

In Table (4), the effects of sulfoxaflor on *P. operculella* on three distinct detoxification enzymes are displayed under laboratory conditions. With values of 308.2 and 353.6, respectively, the laboratory strain and field strain both had significant amounts of the enzyme GST. Comparatively, CPR had values of 73.1 and 79.2, making it the second active enzyme. EST was the least active enzyme among those tested, having values of 41.8 and 75.7, respectively. According to the fold increases in activity for the three enzymes in both strains, EST was the most active (1.8-fold), followed by GST (1.1-fold), and EST (1.1-fold).

Table 4. Effect of sufloxaflor on detoxification enzyme activities, GST-CDNB, CPR-DPPH and EST-PNPA, on lab and field strains of *P. operculella*

Enzyme	Lab Population		Field population		
	N	Activity ±SE	N	Activity ±SE	Fold increase in activity
CPR-DPPH	22	73.1±2.06 a	22	79.2±3.2 a	1.1
GST-CDNB	22	308.2±1.2 a	18	353.6± 3.3 a	1.1
EST- PNPA	24	41.8±0.7 a	22	75.7±2.7 b	1.8

In general, there are many of resistance mechanisms whether behavioral or physiological, these physiological of them includes any barrier or processes as sequestration, transport, metabolism and excretion that impede arrival of pesticide concentration capable of affecting, this biochemical assay can supply statement about the presence of resistance mechanisms specific in *P. operculella* populations. The main objective of this part of the present study was to further elucidate the role of esterase's, monooxygenases and Glutathione-S-transferases as more important common mechanism of conferring pesticides resistance in larval resistance of a field strain of *P. operculella* to some candidate pesticides.

In order to analyze the GST enzyme systems in *P.* operculella samples, CDNB, the general substrate for GSTs, was used to determine the biochemical activity in field population under varying pesticides. GST-CDNB activities showed a statistically significant ($p<0.05$) 1.2-fold increase only with emamectin benzoate pesticide Table (3). The activities of EST- PNPA showed a statistically significant 1.8- and 2.7-fold increase in exhibition field population to sufloxaflor and indoxacarb, respectively (Table 4 and 2). However, field population did not show a-significant ($p<0.05$) increase in EST-PNPA activities under emamectin benzoate exposure.

NADPH-cytochrome P450 reductase (CPR) is a key enzyme that transfers electrons from NADPH to cytochrome P450-monooxygenases (CYP450), detoxifying xenobiotics such as pesticides. CPR enzyme activates CYP450s to metabolize pesticides. Any increase in CPR activity would consequently lead to an increase in CYP450s activities. CPR-DPPH activity of field population showed a statistically significant ($p<0.05$) 1.7-fold increase compared to the susceptible population under exposure to emamectin benzoate pesticide Table (3), while did not show a significant ($p<0.05$) increase with indoxacarb and sufloxaflor pesticide. This increase is parallel to the resistance ratios for emamectin benzoate, 3.3 fold.

Esterases and P-450 mono-oxygenases involvement in the opposition development of emamectin benzoate activity in *P. solenopsis*, As in involvement of tebufenozide in *S. exigua*, acetamiprid in Plutella *xylostella* Linnaeus and imidacloprid in *Bemisia tabaci* (Sayyed and Crickmore, 2007; Jia *et al*., 2009 and Wang *et al*., 2009). Cytochrome P450 enzymes have a major role in abamectin resistance development in field populations of *T. absoluta*, and GSTs might have a minor role in this resistance development. esterase activity did not have an important function in abamectin resistance mechanisms (Konus, 2014).

Activity of PMSO and GST play a role of emamectin benzoate resistance in *B. tabaci* while the reduction of emamectin benzoate efficacy on *C. pomonella* and *L. botrana* is related to increased EST activity (Kang *et al*. 2006; Reyes *et al*. 2007 andYu 2008).

In some insects such as *Phenacoccus solenopsis* and *Spodoptera litura* the detoxify indoxacarb were related with PMSO and EST enzymes activity while in *C. rosaceana* and *L. botrana* larvae, the PMSO, EST, and GST enzymes play an important role (Sayyed and Wright 2006; Sayyed *et al.* 2008; Afzal and Shad 2015 and Hafez *et al*. 2020).

For *Cydia pomonella* and *Choristoneura rosaceana* strains, resistance ratio in the field population for indoxacarb 72-fold higher compared to control population, but under rearing 10 generations, indoxacarb record low or moderate (<10-fold) resistance coefficient (Ahmad *et al*., 2002; Dunley *et al*., 2006; Mota-Sanchez *et al*., 2008 and Civolani *et al*., 2014). In the time that EST was excelled of the resistance situations than GST enzyme which recorded 63% vs 36%. Indoxacarb detoxification for *C. rosaceana* larvae is related with EST, Cytochrome P450 and GST enzymes, on the other GST and PMSO. Involved on the emamectin benzoate resistance in *B. tabaci* (Kang *et al*., 2006; Reyes *et al*., 2007; Yu, 2008 and Hafez *et al*., 2020)

Emamectin benzoate was a 1–3-fold higher insecticidal vigor than other avermectins, it has broad-spectrum, progress thermal stability and ultra-high efficiency, so, it considered as pronounced as more environment safely to replace other neurotoxic insecticides e.g. (Zhou *et al*., 2016 and Mermer *et al*., 2023). Combinations and exchange between vehicles are strategies may delay the increase resist of insecticide instead of use of emamectin benzoate continuously (Consortium, 2013).

This study may help growers in integrated pest management programs for *P. operculella* and strategize their pesticides use in which relieve of pesticide resistance development via create better pesticide application methods that focus on prioritization and understanding of the potential risks of pesticide resistance, this could also give researchers the necessary time to discover and register new pesticides for efficient pest control. In addition, there is a need for standardization in bioassays and enzymatic analyses for *P. operculella* in order to provide comparable results between different experiments from various locations. Lastly, more research is needed to monitor pesticide toxicity and measuring detoxification enzyme levels for effective *P. operculella* control in potato production areas.

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أنشطة الإنزيمات وتحليل مستويات الحساسية في فراشة درنات البطاطس لبعض مبيدات الآفات المستخدمة

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الملخص

تعد فراشة درنات البطاطس (Zeller) *Phthorimaea operculella* من الآفات الهامة التي تهاجم المحصول في الحقل والمخزن وتعد السيطرة على هذه الافه من خلال استخدام المبيدات الحشرية طريقة فعالة وغير مكلفة وبالتالي شائعة الاستخدام بين المزارعون. بسبب الاستخدام الجائر لهذه المركبات كانت هناك ضرورة لدراسة مستويات حساسية اثنين من السلالات (الحقلية والمعملية) وتتبع ظهور صفة المقاومة ومستوياتها والتحليل الكيميائي الحيوي لبعض النظم الأنزيمية المسئولة عن هذا السلوك لمركبات الإندوكساكارب والسلفوكسافلور والإيمامكتين بنزوات وهي الاستريز، السيتوكروم والجلوتاثيون ترانزفيريز.

وكانت نسب المقاومة 11.9، 1.3 و3.3 مرة لكل من الإندوكساكارب والسلفوكسافلور والإمامكتين بنزوات على التوالي في السلالة الحقلية.

أظهر التحليل الكيميائي الحيوي أن أنشطة CYP450-DPPH GST-CDNB لا تظهر سرعة كبيرة ($p < 0.05$) مقارنة بالسلالة الحساسة علاوة على ذلك، أظهر نشاط EST-PNPA زيادة بمقدار 2.7 ضعفًا مقارنة بالحساسة.

أظهرت تحاليل التقييم الحيوي مقاومة متوسطة للإندوكساكارب بينما أظهرت مقاومة قليلة في السلالة الحقلية للإمامكتين بنزوا. تلعب الإستريزات دورًا رئيسيًا في زيادة مقاومة الإندوكساكارب، وقد يكون للسيتوكروم P450 دور أساسي في المقاومة ضد بنزوات الإيمامكتين، ولا يبدو أن GSTs تشارك في تطور المقاومة ضد الإندوكساكارب وإيمامكتين بنزوات في فراشة درنات البطاطس. تضمنت هذه النتائج تطبيقًا عمليًا مهمًا في إدارة مقاومة مبيدات الآفات في *P. operculella*.