

1 **ANTIAPPETIZING AND REPELLENT EFFECT OF BIOPESTICIDES ASTOUN 50 EC**
2 **AND NECO 50 EC ON *ELDANA SACCHARINA* Walker (LEPIDOPTERA: PYRALIDAE)**
3 **UNDER INVITRO CONDITIONS**
4
5
6

7 **Abstract**

8 This study is part of the search for means and methods of sustainable control of *Eldana saccharina*,
9 the main pest of sugarcane cultivation in Côte d'Ivoire. Despite efforts to control the pest through
10 the selection of resistant varieties, the cultivation of healthy cuttings and the elimination of
11 alternative host plants, the damage, which is already economically significant, continues to increase
12 in the sugarcane-growing areas of Côte d'Ivoire. This study aims to evaluate the effect of two
13 proven biopesticides, ASTOUN 50 EC and NECO 50 EC, on the palatability of *E. saccharina* larvae
14 and adults. Firstly, different ages of larvae were reared on two nutrient media such as artificial
15 medium and natural medium treated with increasing concentrations of the two biopesticides. 14, 21
16 and 28 days old larvae were incubated and changes in larval palatability were assessed over a 5-day
17 period. The concentrations of biopesticides tested were 0.5, 1, 2, 4, 8 and 10 ml/l. In addition, the
18 repellent effect of the biopesticides on the adults was assessed using the preferential zone
19 method. The results showed that the biopesticide ASTOUN 50 EC is highly antiappetizing for larvae
20 and repellent for adults of *E. saccharina*, unlike NECO 50 EC. It also greatly reduced the rate of
21 oviposition in the treated compartments. This biopesticide can be tested in vivo for use against
22 *Eldana saccharina*.

23 **Keywords:** *Eldana saccharina*, biopesticide, sustainable control, sugar cane, Ivory Coast, Côte
24 d'Ivoire
25

26 **1. Introduction**

27 *Eldana saccharina* is the main constraint to sugarcane cultivation in Côte d'Ivoire [1]. The larva of
28 this pest, which constitutes the harmful phase, digs galleries in the cane stalks and feeds on the cane
29 pulp, which is supposed to accumulate sugar at maturity. The galleries left in the stalks make the
30 plant more vulnerable to disease and other pests such as termites and rodents, as well as to bad
31 weather, which causes more lodging than usual. As a result, *Eldana saccharina* causes huge losses
32 in terms of both quantity and quality in sugar cane cultivation [2, 3]. Statistics show losses of up to
33 0.1% for every 1% of cane attacked, and up to 70% of the sugar content in attacked internodes.
34 Since the 2015-2016 sugar campaigns, when attack rates exceeded the critical threshold of 6% [1],
35 control trials have been carried out in sugarcane production areas in order to get to grips with this
36 pest, whose damage is increasing year on year. These control methods, which are mainly based on

Comment [D1]: Add « s ».

Comment [D2]: Add « s ».

Comment [D3]: Add « Côte d'Ivoire ».

37 eliminating alternative host plants, destroying crop residues and growing resistant varieties, have
 38 shown their limitations. Hence the need to develop other control methods that can be used on their
 39 own or as part of an integrated pest management system. **Its aim is to demonstrate** the efficacy of
 40 biopesticides such as ASTOUN 50 EC and Neco 50 EC, which **are shown** to have insect-repellent
 41 and insecticidal potential against a wide range of pests [4, 5, 6]. It was carried out in vitro on
 42 **Eldana saccharina** larvae and adults.

Comment [D4]: Correct

Comment [D5]: Add « are ».

Comment [D6]: Delete the space.

43 **2. Material and methods**

44 **2.1. Material**

45 The plant material was sugar cane (**Saccharum officinarum**) and the variety was R570 **used for the**
 46 **study**. Cane stalks of this variety with a diameter of between 0.9 and 1.5 cm were cut into pieces 3.5
 47 to 4.3 cm long. These pieces of cane stalk were used to feed the **Eldana saccharina** larvae and
 48 formed the natural nutrient medium. In addition to this nutrient medium, an artificial medium was
 49 prepared following the method used by [7]. The composition of this medium is given in Table 1.

50 The animal material consisted of **Eldana saccharina** larvae of 14, 21 and 28 day old and adults.
 51 Different ages of larvae were used. For adults, only imagos were used.

Comment [D7]: Delete the space in between

52 The technical equipment consisted mainly of rearing dishes for **Eldana saccharina** larvae and
 53 adults, alcohol and sodium hypochlorite.

54 The biological control materials tested consisted of the biopesticides ASTOUN 50 EC and NECO
 55 50 EC. The biopesticide ASTOUN 50 EC comprises neral (39.33%), α -citral (31.89%), sulcatone
 56 (2.49%), oxygenated monoterpenes (73.71%) and hydrocarbon monoterpenes represented by β -
 57 myrcene (26.29%) [8]. As for NECO 50 EC, **its major** components are Thymol, Gamma-Terpinene
 58 and Eugenol [9]. These two biopesticides, obtained **from** Industrial Research Unit of the Université
 59 Félix Houphouët Boigny, are registered in Côte d'Ivoire.

60

61

62 **Table 1:** Composition of the artificial nutrient medium

63

Ingredient	Quantity	Role
Dried and crushed sugarcane	22 g	Nutrients
Chickpea powder	120 g	
Saccharose	10 g	
Casein	12 g	
Baker's yeast	12 g	
Ascorbic acid	4 g	Vitamins
Sorbic acid	2 g	
Agar	10 g	Binding substance

Sterile distilled water	1 000 mL	
Nipagine	1,6 g	
Formaldehyde	1.2 mL	Anti-microbial
Methanol	50 mL	

64
65 Source [10]
66

67 **2.2. Methods**

68 **2.2.1. Multiplication of *E. saccharina* larvae and adults**

69 A basic sample of larvae and pupae was collected from the sugarcane plots. From this sample,
70 rearing was carried out in the laboratory to obtain the number of larvae and adults required for the
71 biopesticide tests. The adults had emerged from the larvae and pupae collected in the field were
72 incubated in rearing boxes under conditions of temperature between 26 to 29°C and relative
73 humidity between 60 and 70%. The eggs were collected every 24 hours and incubated on blotting
74 paper in Petri dishes. Once the eggs hatched, the larvae were incubated to produce 14-, 21- and 28-
75 day-old larvae and adults. The larvae were reared either on pieces of sugarcane stalk or on an
76 artificial nutrient medium. A total of 520 larvae of each age and 96 adults were used in this study.

77 **2.2.2. Preparation of biopesticide concentrations and treatment of nutrient media**

78 The two biopesticides were tested at concentrations of 0.5 ml/l, 1 ml/l, 2 ml/l, 4 ml/l, 8 ml/l and 10
79 ml/l. These concentrations were obtained by diluting distilled water with Tween 20. Pieces of cane
80 stalks were treated by soaking in the various concentrations of ASTOUN 50 EC biopesticide or
81 NECO 50 EC biopesticide for 5 minutes, then exposed to the open air for 5 minutes. As for the
82 artificial medium, the biopesticides were dissolved directly in the nutrient medium. These treated
83 nutrient media were used to feed the larvae. One consisted of untreated nutrient media and the other
84 of nutrient media treated with Tween 20 served as control.

85 **2.2.3. Evaluation of biopesticides on the palatability of *E. saccharina* larvae**

86 This test was carried out on 14-, 21- and 28-day-old larvae. These larvae were deposited on the
87 treated nutrient media and then feeding habit was observed at regular 24-hour intervals according
88 to the method used by [11]. A larva was considered to be feeding on the medium when droppings
89 were present in the Petri dish. The number of larvae having consumed the nutrient media was
90 recorded at each observation and the rate of larvae having consumed the nutrient medium (TLC)
91 was calculated according to the following formula

$$\text{TLC}(\%) = \frac{\text{Number of larvae that consumed the medium}}{\text{Total number of larvae}} \times 100$$

95 TLC : Rate of larvae having consumed the nutrient medium

Comment [D8]: Add « biopesticide ».

96

97 **2.2.4. Evaluation of the repellent effect of biopesticides on *E. saccharina* imagos**

98 The repellent effect of biopesticides was evaluated using the preferential zone method [12]. For this
99 experiment rearing box was divided into two compartments connected by a central opening. On one
100 side were sugarcane leaves treated with biopesticides and on the other, sugarcane leaves treated
101 only with Tween 20. The leaves were soaked for 5 minutes in the different concentrations and then
102 exposed to the air for 5 minutes. Four *E. saccharina* imagos (two females and two males) were then
103 deposited in each compartment. Ten replicates were used for each concentration of biopesticide
104 tested. After 24 hours, the number of adults in each compartment of the rearing box was counted
105 and the percentage repulsion (PR) was calculated using the formula in [12]:

106

$$PR (\%) = \frac{NC - NT}{NC} \times 100$$

107

108 **PR:** Percentage of repellency; **NC:** Number of adults in the compartment treated with biopesticides;

109 **NT:** Number of adults in the compartment treated only with Tween 20.

110 The average repellency percentage for each biopesticide concentration was calculated (PR) and assigned to
111 one of the different repellency classes established by [12], ranging from 0 to V, recorded in Table 2.

112 Similarly, the number of eggs laid in each compartment of the rearing box was counted after 24 hours. The
113 rate of eggs laid in the treated compartment (TOPCT) was then calculated using the following formula:

114

$$TOPCT (\%) = \frac{\text{Number of eggs laid in the treated compartment}}{\text{Total number of eggs laid in the box}} \times 100$$

115

116 **TOPCT :** rate of eggs laid in the treated compartment

117

118

119

120

121

122

Table 2: Biopesticide repellency classification scale

Class	Repulsion interval	Properties
0	PR ≤ 0,1%	Not repellent
I	00,1 < PR ≤ 20%	Very low repellency
II	20,1 < PR ≤ 40%	Weakly repellent
III	40,1 < PR ≤ 60%	Moderately repellent
IV	60,1 < PR ≤ 80%	Repellent
V	80,1 < PR ≤ 100%	Highly repellent

123
124 Source : [12]

125 126 **2.2.5. Data analysis**

127 The data collected were recorded on the computer using Excel 2013 software, which was also used
128 to draw up the graphs, curves and tables.

129 The rates of repulsion and eggs laid in the treated compartment were analysed using Statistica
130 version 7.1 software or an analysis of variance was performed. In the event of a significant
131 difference, the averages obtained were classified into homogeneous groups using the Newmann-
132 Keuls test with a threshold of 5 %.

133

134 **3.RESULTS**

135

136 **3.1.Effect of the biopesticides ASTOUN 50 EC and NECO 50 EC on the palatability of *Edana*** 137 ***saccharinalarvae***

138

139 **3.1.1.Palatability of 14-day-old larvae on treated nutrient media**

140

141 ❖ **In the presence of the biopesticide ASTOUN 50 EC**

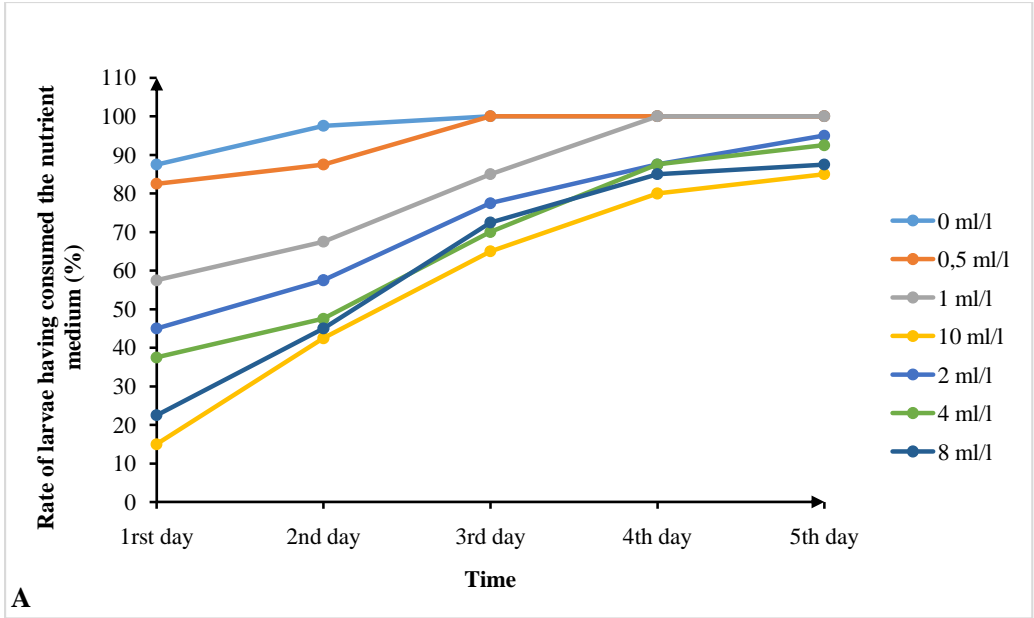
142 One day after culturing on nutrient media treated with the biopesticide ASTOUN 50 EC, all
143 concentrations induced a lower palatability in 14-day-old larvae than in the control, regardless of
144 the nutrient medium. Over 50% of the larvae did not consume the nutrient media treated at
145 concentrations of 2 ml/l and above, whereas 87.5% of the larvae were already feeding on the
146 control. Thereafter, larval palatability increased sharply over time. However, until the end of the
147 observations, the palatability of 14-day-old larvae was significantly reduced by the biopesticide
148 ASTOUN 50 EC from the concentration of 2 ml/l. The lowest rate of larvae consuming the treated
149 nutrient media was obtained with concentrations of 8 ml/l and 10 ml/l (figure 1).

150 ❖ **In the presence of the biopesticide NECO 50EC**

151 The palatability of 14-day-old larvae on nutrient media treated with the biopesticide NECO 50 EC
152 was identical or very close to that of the control up to a concentration of 2 ml/l, throughout the
153 observation period. Already on the first day after incubation, more than 70% of the 14-day-old
154 larvae fed on the nutrient media treated, even at the highest concentrations. Larval palatability also
155 increased sharply over time. At the end of the observations, only 15% of the 14-day-old larvae did
156 not feed on the nutrient media treated with the 8 ml/l and 10 ml/l concentrations. All the other
157 concentrations induced palatability similar to that observed on the control, where 100% of the
158 larvae fed.

159 Over the entire observation period, 14-day-old larvae fed less on nutrient media treated with the
160 biopesticide ASTOUN 50 EC from 2 ml/l and NECO 50 EC from 8 ml/l than the control. After

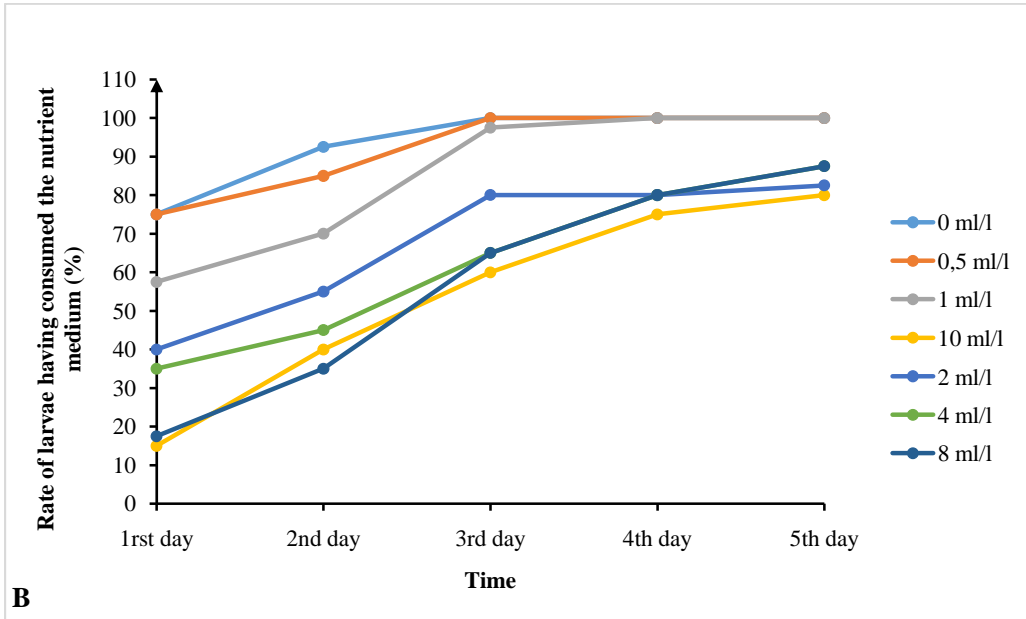
161 culture, the 14-day-old larvae fed less quickly on the nutrient media treated than the control. Larval
162 palatability dropped considerably as the concentration of biopesticides in the nutrient medium
163 increased (figure 2).



164

UNDER PEER

165



166

167

168

169

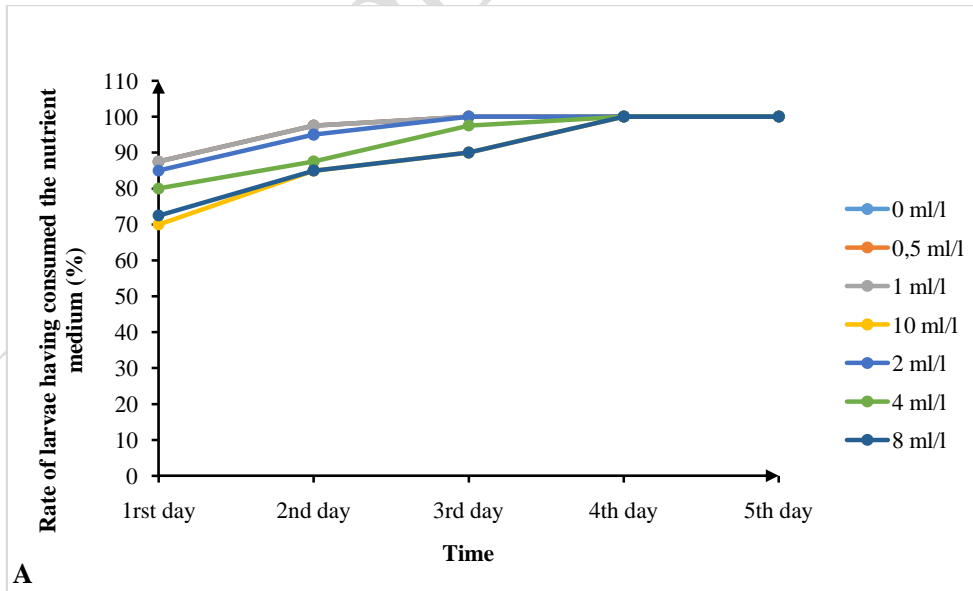
170

171

A : On pieces of cane stalk

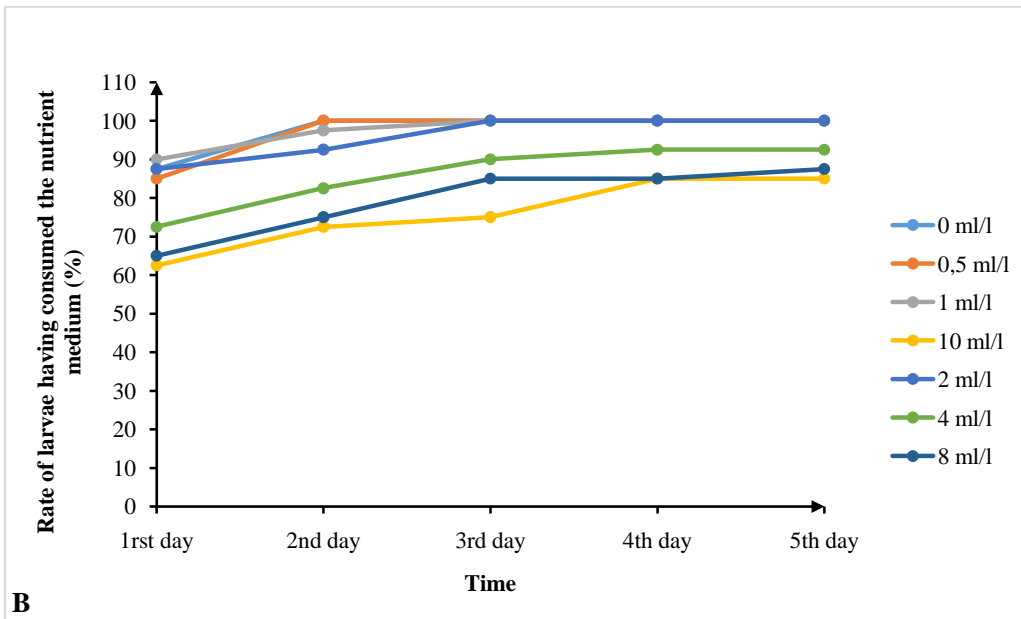
B : On artificial nutrient medium

Figure 1: Changes in the rate of 14-day-old larvae that consumed nutrient media treated with the biopesticide ASTOUN 50 EC



172

173



174
175
176

A : On pieces of cane stalk

B : On artificial nutrient medium

177 **Figure 2:** Changes in the rate of 14-day-old larvae that consumed nutrient media treated with the
178 biopesticide NECO 50 EC

179 3.1.2. Palatability of 21-day-old larvae on treated nutrient media

180
181

❖ In the presence of the biopesticide ASTOUN 50 EC

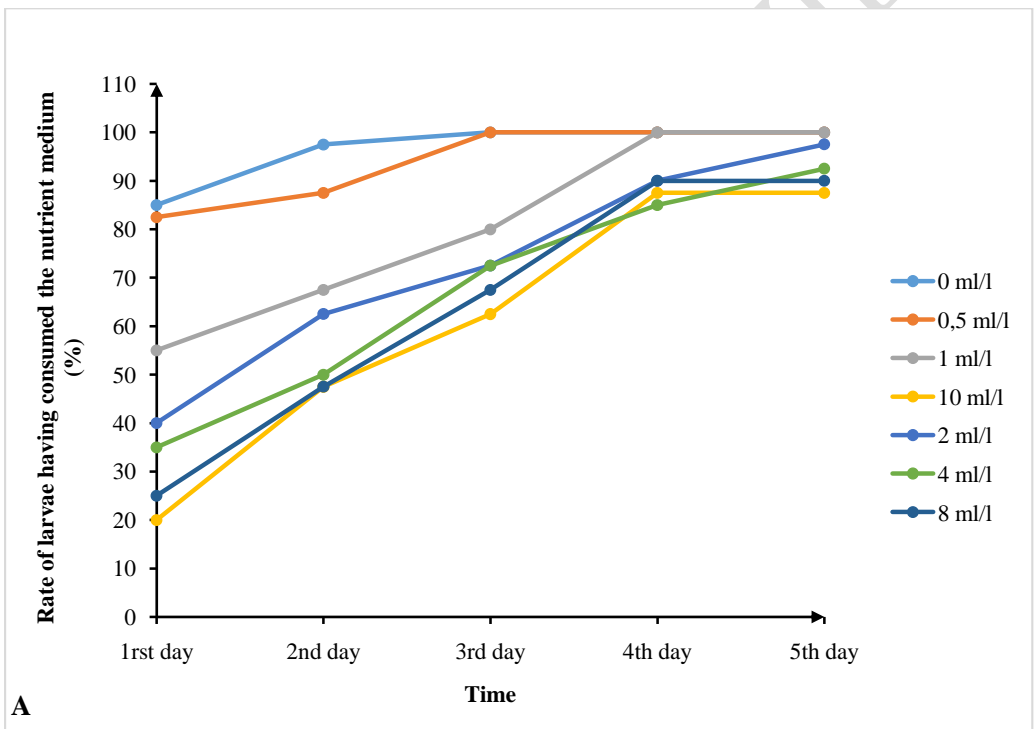
182 A concentration effect on palatability of 21-day-old larvae was observed. On the first day after
183 incubation, more than 50% of the larvae had not fed on the nutrient media treated with a
184 concentration of 2 ml/l, whereas 85% had fed on the control. The feeding rate then increased
185 sharply with time. At the end of the observations, the highest rate of larvae having consumed the
186 nutrient media was observed on the control and the concentrations of 0.5 and 1 ml/l with values of
187 100%. The lowest rates were recorded with concentrations of 8 ml/l and 10 ml/l (figure 3).

188
189

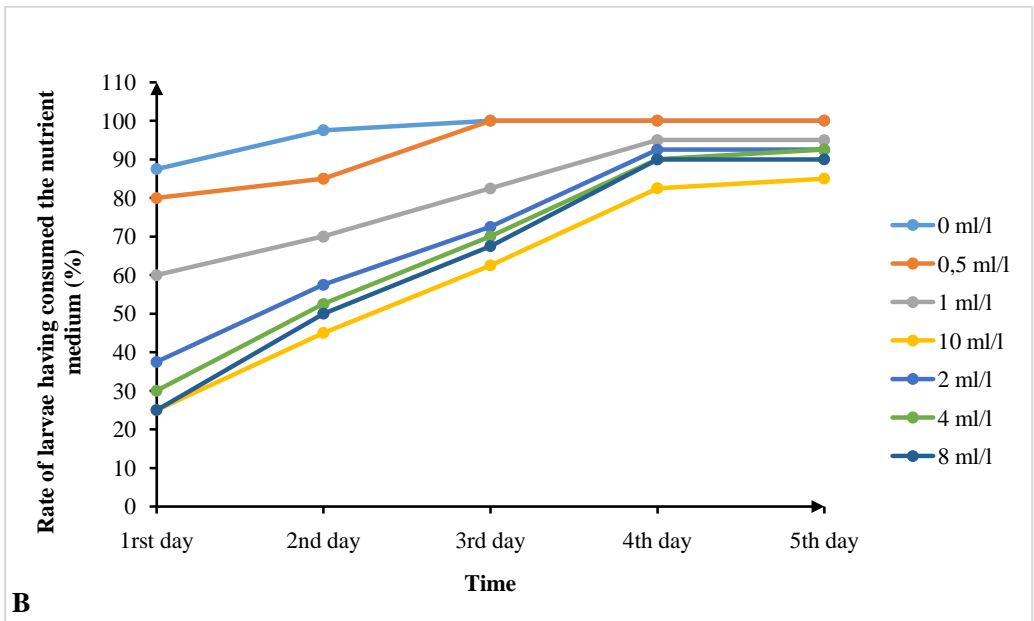
❖ In the presence of the biopesticide NECO 50 EC

190 The results show a concentration effect of NECO 50 EC on the palatability of 21-day-old larvae.
191 Also, the rate of larvae having consumed the treated nutrient media increased significantly with
192 time. On the first day after incubation, more than 65% of the larvae had already fed on the media,
193 whatever the concentration. The 8 ml/l and 10 ml/l concentrations stood out with the lowest rates of
194 larvae having consumed the media, i.e. 62.5 and 72.5% respectively on pieces of cane stem and on

195 artificial media. At the end of the observations, all the concentrations recorded a rate of larvae
 196 having consumed the treated media of over 97%. The control and all concentrations except 10 ml/l
 197 resulted in a 100% rate of larvae having fed. Only 2.5% of the larvae did not feed on the nutrient
 198 media treated with the 10 ml/l concentration (figure 4).
 199 Thus, after incubation, the 21-day-old larvae fed less rapidly on the nutrient media treated than on
 200 the control. Over the entire observation period, 21-day-old larvae fed less on nutrient media treated
 201 with the biopesticide ASTOUN 50 EC than on the control from 8 ml/l onwards. Larval palatability
 202 dropped considerably when the concentration of ASTOUN 50 EC biopesticide or NECO 50 EC
 203 increased in the nutrient medium. The feeding rate of 21-day-old larvae was greater than 80% in the
 204 presence of ASTOUN 50 EC biopesticide and NECO 50 EC.
 205



206
 207

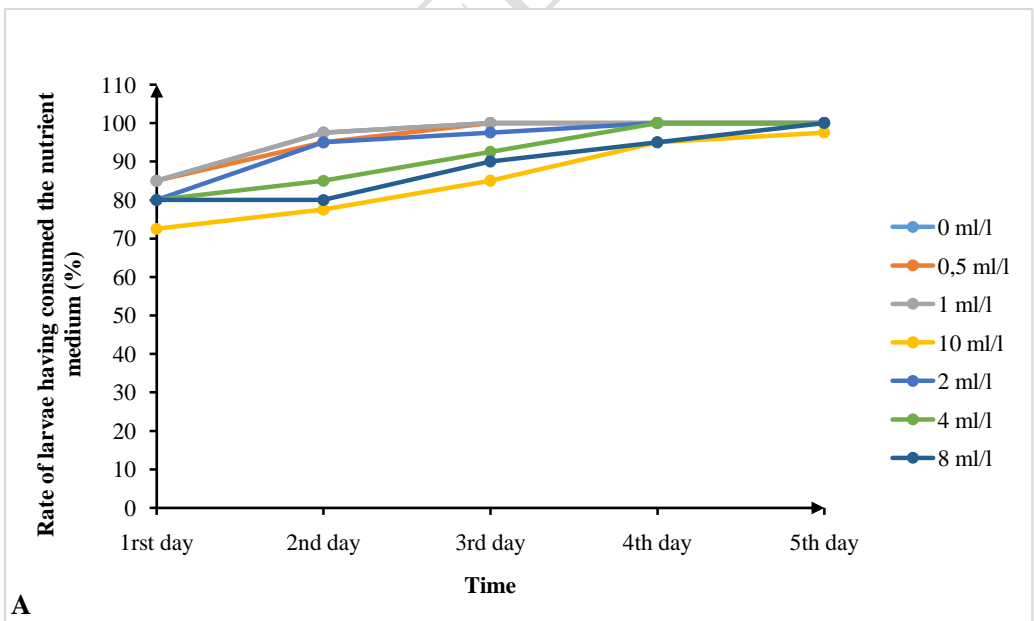


208
209
210
211
212

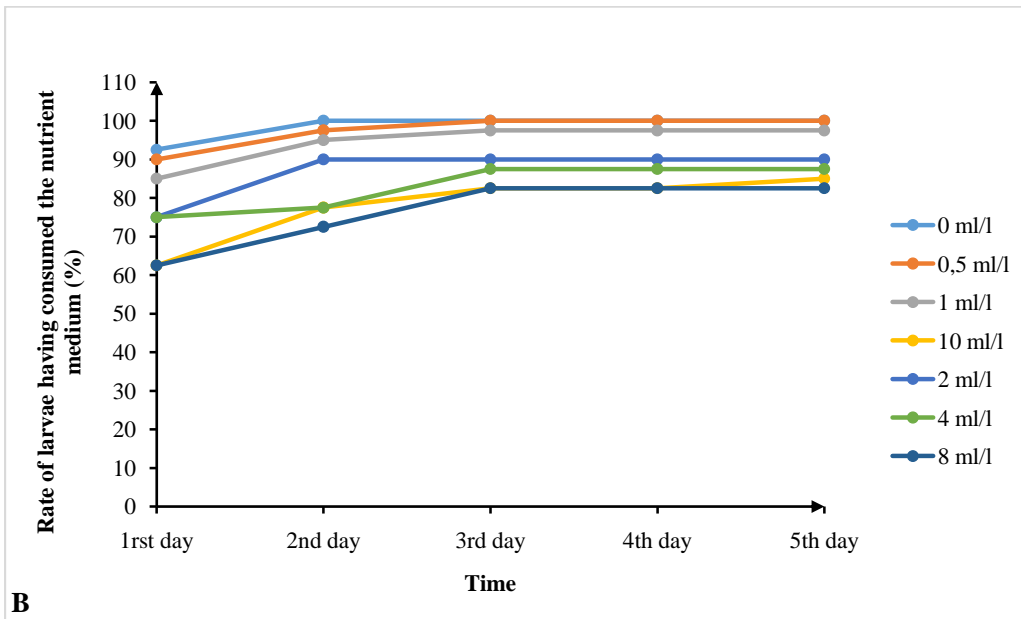
A : On pieces of cane stalk

B : On artificial nutrient medium

Figure 3: Changes in the rate of 21-day-old larvae that consumed nutrient media treated with the biopesticide ASTOUN 50 EC



213
214



215

216

217

218

219

220

221

222

223

224

225

226

227

228

229

230

231

232

233

234

235

236

A : On pieces of cane stalk

B : On artificial nutrient medium

Figure 4: Changes in the rate of 21-day-old larvae that consumed nutrient media treated with the biopesticide NECO 50 EC

3.1.3. Palatability of 28-day-old larvae on treated nutrient media

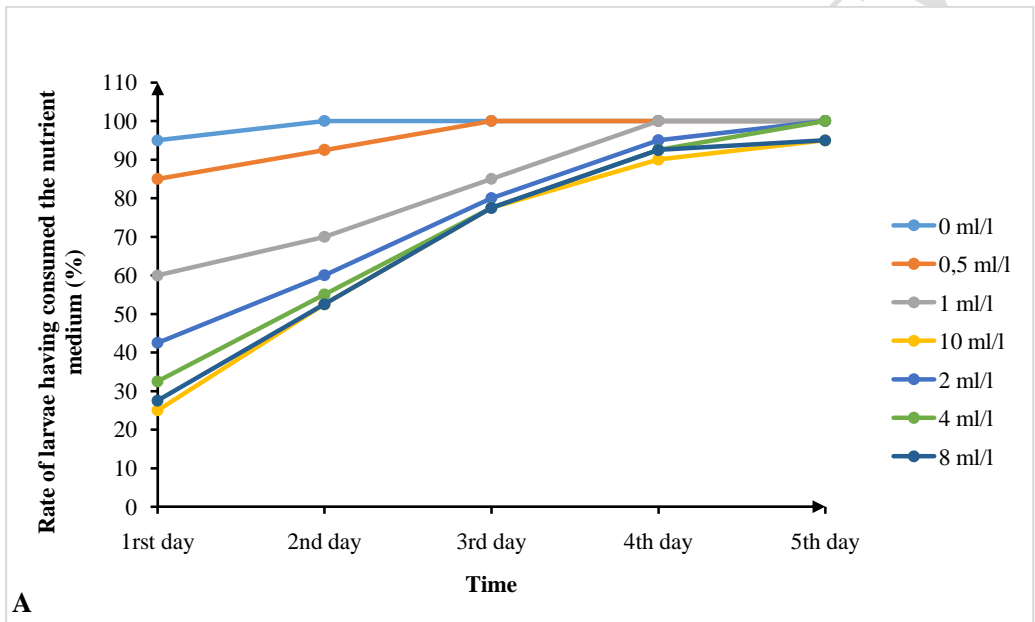
❖ In the presence of the biopesticide ASTOUN 50 EC

The results show that the rate of larvae having consumed the treated media increased considerably over time and decreased with increasing biopesticide concentration. On the first day after incubation, more than 50% of the 28-day-old larvae had not yet fed on the nutrient media treated at a concentration of 4 ml/l, whereas the control recorded a rate of more than 80%. At the end of the observations, 100% of the larvae had fed on the two artificial media up to a concentration of 4 ml/l. On the nutrient media treated with concentrations of 8 ml/l and 10 ml/l, only 5% of the larvae did not feed (figure 5).

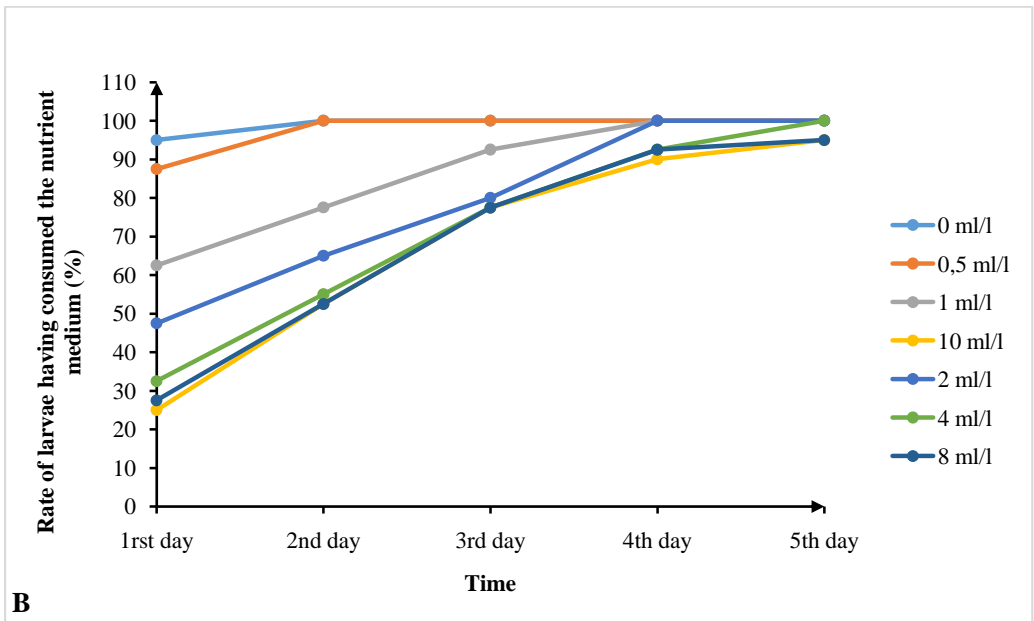
❖ In the presence of the biopesticide NECO 50 EC

The results show that the rate of larvae that consumed the treated nutrient media increased sharply over time for all the concentrations tested. A concentration effect was observed during the first few days of incubation. On the first day after incubation, more than 75% of the larvae fed on the treated nutrient media, whatever the biopesticide concentration. By the third day after culture, only 5% of the larvae were not feeding on the highest concentrations. At the end of the observations, all the

237 larvae fed on the pieces of cane stalk whatever the concentration and only 5% did not feed on the
238 artificial medium for the concentrations of 8 and 10 ml/l (figure 6).
239 In short, over the entire observation period, 28-day-old larvae fed less on nutrient media treated
240 with the biopesticides ASTOUN 50 EC and NECO 50 EC than on the control at 8 ml/l or more.
241 After culture, the 28-day-old larvae fed less rapidly on the nutrient media treated than on the
242 control. Larval palatability dropped considerably when the concentration of biopesticides in the
243 nutrient medium increased. The rate of 28-day-old larvae consuming treated nutrient media was
244 greater than 90% in the presence of the biopesticide ASTOUN 50 EC and NECO 50 EC.
245



246
247

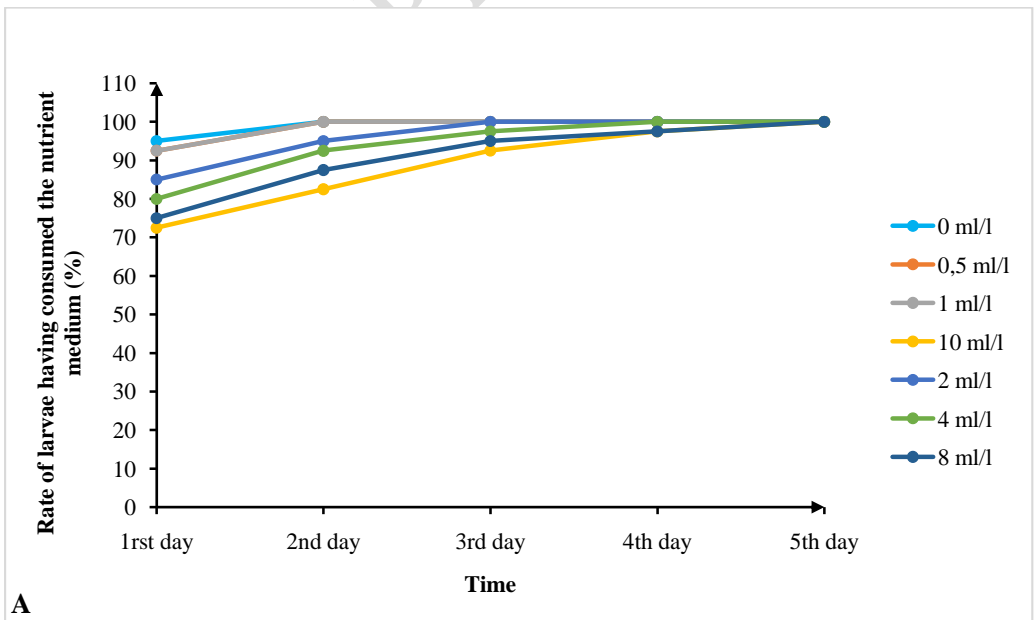


248
249
250
251
252
253
254

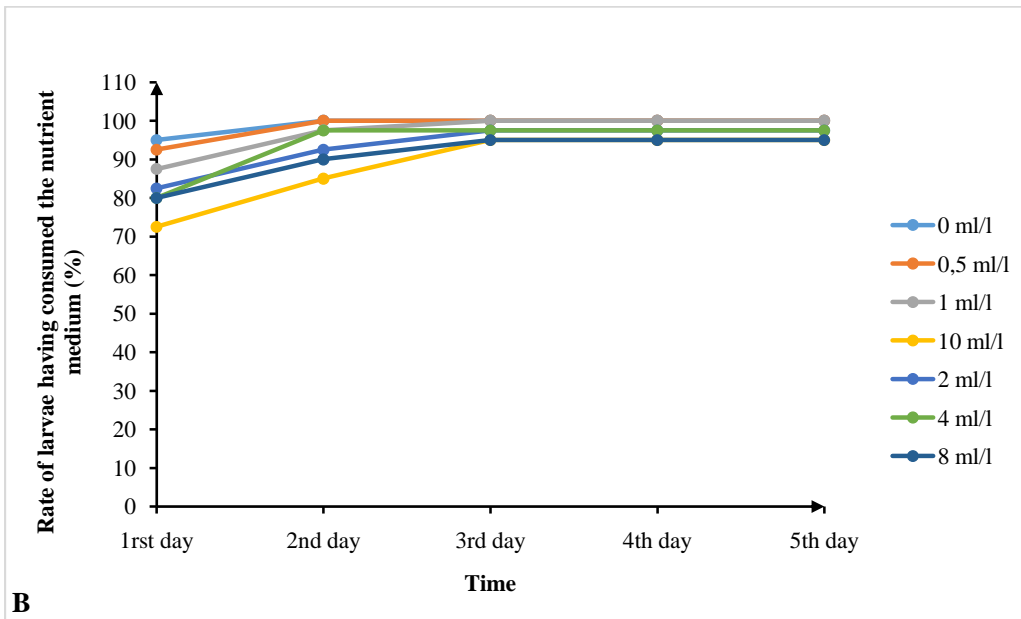
A : On pieces of cane stalk

B : On artificial nutrient medium

Figure 5: Changes in the rate of 28-day-old larvae that consumed nutrient media treated with the biopesticide ASTOUN 50 EC



255
256



257
258
259

260

261

262

263

264

265

266

267

268

269

270

271

272

273

274

275

276

277

278

A : On pieces of cane stalk

B : On artificial nutrient medium

Figure 6 : Changes in the rate of 28-day-old larvae that consumed nutrient media treated with the biopesticide NECO 50 EC

3.1.4. Larval palatability as a function of age, nutrient medium and biopesticide

The rate of *Eldana saccharina* larvae consuming the treated nutrient media varied significantly with age. The highest rate was recorded for 28-day-old larvae (98.75%). Larvae aged 14 and 21 days fed less on the treated nutrient media, with rates of 95.10 and 95.18% respectively. Thus, the older the larvae, the more indifferent they were to the treatments (table 3).

A biopesticide effect was observed on the palatability of *E. saccharina* larvae. This effect was significant only for 14-day-old larvae and for all ages combined (Table 4). The palatability of the larvae was higher on nitric media treated with the biopesticide NECO 50 EC than on those treated with ASTOUN 50 EC with 97.38 and 95.30 %.

A significant effect of the nutrient medium was recorded on the palatability of the larvae except at the age of 28 days. Larval palatability was higher on pieces of cane stem, with rates of 97.14 and 97.68% at 14 and 21 days of age respectively, compared with 93.04 and 92.68% on artificial medium. For all ages combined, the rate of larvae having consumed the treated media was higher on pieces of cane stem than on artificial medium, with 98.04 and 94.64% respectively (table 5).

279 **Table 3:** Rate of larvae consuming treated nutrient media as a function of age

Age of larva	Rate of larvae feeding (%)
14 days	95,10 ^b
21 days	95,18 ^b
28 days	98,75 ^a
Average	96,33929
C.V (%)	6,76000
Probability	0,000003

280 ^a values followed by different letters are significantly different at the 5% threshold according to the Newman-Keuls test.

281 C.V. = Coefficient of variation

282

283 **Table 4:** Rate of larvae consumed the treated nutrient media according to the biopesticides tested

284

Biopesticides	Rate of larvae feeding (%)			Average
	14-day-old larvae	21-day-old larvae	28-day-old larvae	
ASTOUN 50 EC	92,68 ^b	94,64 ^a	98,57 ^a	95,30 ^b
NECO 50 EC	97,50 ^a	95,71 ^a	98,93 ^a	97,38 ^a
Average	95,10	95,18	98,75	3,66
C.V (%)	7,98	7,46	3,36	6,76
Probability	0,000004	0,000004	0,000004	0,015212

285

286 ^a values in the same column followed by different letters are significantly different at the 5% threshold according to the
287 Newman-Keuls test. C.V. = Coefficient of variation

288 **Table 5:** Rate of larvae having consumed the treated nutritive media as a function of the nutritive
289 medium

Milieux nutritifs	Rate of larvae feeding (%)			Average
	14-day-old larvae	21-day-old larvae	28-day-old larvae	
Artificial nutrient medium	93,04 ^b	92,68 ^b	98,21 ^a	94,64 ^b
Piece of cane stem	97,14 ^a	97,68 ^a	99,29 ^a	98,04 ^a
Average	97,14	95,18	98,75	96,34
C.V (%)	7,98	7,46	3,36	6,76
Probability	0,000003	0,000003	0,000003	0,009898

290 ^a values in the same column followed by different letters are significantly different at the 5%
291 threshold according to the Newman-Keuls test. C.V. = Coefficient of variation

292

293 **3.1.5. Percentage of repulsion of imagos by biopesticides**

294 The repellent effect of the biopesticides ASTOUN 50 EC and NECO 50 EC on *E. saccharina* adults
 295 increased with the dose of the product. For ASTOUN 50 EC, it increased from 30 to 90 % and for
 296 NECO 50 EC from -5.41 % to 17.5 % when the concentration was increased from 0.5 to 10 ml/l
 297 (Tables 6). Also, for all doses tested, the biopesticide ASTOUN 50 EC had a more pronounced
 298 repellent effect than NECO 50 EC on *E. saccharina* adults (Tables 7).

299 3.1.6. Laying rate in the treated compartment

300 The laying rate of *E. saccharina* females in the presence of NECO 50 EC changed from 48.73 to
 301 46.73% with the increase in concentration from 0.5 to 10 ml/l. However, this decrease was not
 302 significant at the 5% level. In the presence of ASTOUN 50 EC, the laying rate of *E.*
 303 *saccharina* females in the treated compartment changed from 41.29 to 4.09% with the increase in
 304 concentration from 0.5 to 10 ml/l. The analysis of variance revealed a significant effect of the
 305 concentration on the spawning rate in the treated compartment. Thus, as the concentration of
 306 ASTOUN increased, the females moved further away and laid eggs in untreated areas (table 8). In
 307 comparison, the oviposition rate of *E. saccharina* females in a treated area was statistically lower in
 308 the presence of the biopesticide ASTOUN 50 EC than in the presence of NECO 50 EC from the
 309 dose of 2 ml/l (table 9).

310 **Table 6:** Percentage of repulsion of *E. saccharina* imagos by the biopesticide ASTOUN 50 EC and
 311 NECO 50 EC

Concentration	Percentage of repulsion	
	ASTOUN 50 EC	NECO 50 EC
0,5 ml/l	30,00 ^a	-5,00 ^a
1 ml/l	45,00 ^b	7,50 ^a
2 ml/l	60,00 ^c	10,00 ^a
4 ml/l	67,50 ^c	12,50 ^a
8 ml/l	87,50 ^d	12,50 ^a
10 ml/l	90,00 ^d	17,50 ^a
Average	63,33	9,17
C.V (%)	0,39	3,33
Probability	0,000000	0,026070

323 ^aaverages in the same column followed by a different letter are statistically different at the 5% threshold according to
 324 the Newman-Keuls test.

325
 326
 327

328 **Table 7:** Percentage of repulsion of *E. saccharina* imagos by the biopesticide ASTOUN 50 EC and
 329 NECO 50 EC

Product	Percentage of repulsion					
	0,5 ml/l	1 ml/l	2 ml/l	4 ml/l	8 ml/l	10 ml/l
ASTOUN 50 EC	30,00 ^a	45,00 ^a	60,00 ^a	67,50 ^a	87,50 ^a	90,00 ^a
NECO 50 EC	5,00 ^b	7,50 ^b	10,00 ^b	12,50 ^b	12,50 ^b	17,50 ^b
Average	12,5	26,25	35	40,00	50,00	53,75
C.V (%)	2,47	1,09	0,97	0,92	0,93	0,8
Probability	0,007183	0,001187	0,000108	0,000071	0,000007	0,000001

330

331 ^aaverages in the same column followed by a different letter are statistically different at the 5% threshold according to
 332 the Newman-Keuls test.

333

334

335 **Table 8:** Rate of eggs laid in the compartment of the rearing box treated with the biopesticides
 336 ASTOUN 50 EC and NECO 50 EC

Concentration	Rate of eggs laid in the treated compartment	
	ASTOUN 50 EC	NECO 50 EC
0,5 ml/l	41,29 ^a	48,73 ^a
1 ml/l	37,15 ^b	48,18 ^a
2 ml/l	16,95 ^b	45,17 ^a
4 ml/l	9,76 ^b	45,49 ^a
8 ml/l	4,69 ^b	49,25 ^a
10 ml/l	4,09 ^b	46,73 ^a
Average	18,99	47,26
C.V (%)	78	57
Probability	0,000028	0,999180

337

338 ^aaverages in the same column followed by a different letter are statistically different at the 5% threshold according to
 339 the Newman-Keuls test.

340

341 **Table 9:** Comparative effect of the biopesticide ASTOUN 50 EC and NECO 50 EC on the
 342 oviposition rate of *E. saccharina* females

Egg-laying rate in the treated compartment	
--	--

biopesticides	0,5 ml/l	1 ml/l	2 ml/l	4 ml/l	8 ml/l	10 ml/l
ASTOUN 50 EC	48,73 ^a	48,18 ^a	45,17 ^a	45,49 ^a	49,25 ^a	46,73 ^a
NECO 50 EC	41,29 ^a	37,15 ^a	16,95 ^b	9,76 ^b	4,69 ^b	4,09 ^b
Average	45,01	42,67	31,06	27,63	26,97	25,41
C.V (%)	71	57	75	120	102	114
Probability	0,614703	0,327538	0,003342	0,011174	0,000007	0,000125

343 *averages in the same column followed by a different letter are statistically different at the 5%
344 threshold according to the Newman-Keuls test.

345 4. DISCUSSION

346 The palatability of *E. saccharina* larvae decreased significantly with increasing concentration of the
347 biopesticide ASTOUN 50 EC. Similarly, this biopesticide had a very marked repellent effect on *E.*
348 *saccharina* adults and considerably reduced the oviposition rate of females in the treated
349 compartments. These results indicate that the biopesticide ASTOUN 50 EC has a repellent effect on
350 *E. saccharina* larvae and adults. This justifies the fact that *E. saccharina* larvae feed faster on
351 control nutrient media than treated nutrient media as well as the effects observed on adults. Our
352 results are similar to those of [13] who showed that the essential oils of *Cymbopogon citratus* and
353 *Ocimum canum* have an antiappetizing effect on *Cylas puncticollis* Boheman resulting in
354 reduced consumption of sweet potato roots compared to the control. After 5 days of observation, a
355 rapid increase in the rate of larvae having consumed the treated nutrient media was observed. This
356 progressive feeding of larvae on treated nutrient media due to either to the larvae's accommodation
357 to the odour emitted by the biopesticides or to the loss of the latter's repellent effect over time. The
358 latter would be attributable to the high volatility of the active ingredients in the biopesticide
359 ASTOUN 50 EC. As indicated by the work of several authors [14, 15, 16], the essential oil of
360 *Cymbopogon citratus*, the main component of the biopesticide ASTOUN 50 EC, is highly volatile,
361 which severely limits its repellent activity over time. In the presence of NECO 50 EC, the
362 palatability of the larvae was lower than that of the control only on the first day after culturing and
363 for the highest concentrations. After that, the rate of larvae feeding on the treated nutrient media
364 increased very rapidly to equal that of the control in most cases. Thus, biopesticide ASTOUN 50
365 EC and NECO 50 EC has a repellent effect against *E. saccharina* larvae. However, its action is very
366 limited in time. These results are in line with those of [17], who demonstrated that *Ocimum*
367 *gratissimum* L, whose essential oil is the main component of NECO [4], is a repellent plant against
368 the lepidopterans *Hellula undalis*, *Spodoptera littoralis* and *Plutella xylostella*. In addition, several
369 laboratory and field studies have demonstrated the insect-repellent and/or insecticidal effect of the

370 NECO biopesticide against a wide range of stock predators, fungal and bacterial agents and borers
371 [4, 5, 18, 19, 20].

372 Comparative analysis of these two biopesticides in terms of their effect on the palatability of *E.*
373 *saccharina* larvae and repellency of adults revealed significant differences. The biopesticide
374 ASTOUN 50 EC had a more marked repellent effect on larvae, imagos and greatly reduced the
375 oviposition rate of *E. saccharina* females than the biopesticide NECO. Thus, the biopesticide
376 ASTOUN 50 EC was more insect repellent than NECO 50 EC on *E. saccharina* larvae and adults.
377 This difference in efficacy could be explained by the difference in composition of these two
378 biopesticides. The biopesticide ASTOUN 50 EC includes neral, α -citral, sulcatone, oxygenated
379 monoterpenes and β -myrcene [8] whereas NECO 50 EC has Thymol, Gamma-Terpinene and
380 Eugenol as active ingredients [9]. This result is consistent with those of [6] who demonstrated a
381 difference in the larvicidal effect of topical application of the essential oils of *Zingiber officinale*,
382 *Cymbopogon citratus* and *Ocimum gratissimum*.

383 5. Conclusion

384 This study is part of the search for means and methods to control *Eldana saccharina*, the main
385 sugarcane stalk borer in Côte d'Ivoire. The effect of two biopesticides, ASTOUN 50 EC and NECO
386 50 EC, was tested on the palatability of the pest's larvae and adults. The results showed that these
387 two biopesticides had an antiappetizing effect on larvae and a repellent effect on *Eldana saccharina*
388 adults. However, the biopesticide ASTOUN 50 EC was the most effective. It is highly repellent to
389 adults, considerably reduces the oviposition rate of females in treated areas and is highly
390 antiappetizing. This biopesticide could be tested in the field for effective use against *Eldana*
391 *saccharina* in sugar cane.

392

393 References

- 394 1. Kouamé DK, Péné CB and Zouzou M. Assessment of varietal resistance of sugarcane to the
395 African tropical stem borer (*Eldana saccharina* Walker) in Côte d'Ivoire. *Journal of Applied*
396 *Biosciences* 2010 ; 26: 1614-1622.
- 397 2. Goebel FR and Way MJ. Investigation of the impact of *Elana saccharina* (Lepidoptera:
398 Pyralidae) on sugarcane yield in field trials in Zululand. *Pocceeding of South Africa Sugarcane*
399 *Technologists Association*, 2003 ; 77: 256-265.
- 400 3. Assefa Y, Conlong DE and Mitchell A. Phylogeography of *Eldana saccharina* Walker
401 (Lepidoptera: Pyralidae). *Annals of the Entomological Society of France*, 2006 ; 42 (3-4): 331-
402 338.
- 403 4. Kassi MF, Badou JO, Tonzibo FZ, Salah Z, Bolou ABB, Camara B *et al.* Antifungal
404 potential of the essential oil of *Ocimum gratissimum* in the biological control of banana black

405 streak disease caused by *Mycosphaerella fijiensis* morelet (mycosphaerellacea). *African*
406 *Agronomy*, 2014 ; 26 (2): 1 - 11.

407 5. Kobenan KC, Tia VE, Ochou GEC, Kouakou M, Bini KKN, Dagnogo M, Dick AE and
408 Ochou OG. Comparison of the insecticidal potential of the essential oils of *Ocimum*
409 *gratissimum* L. and *Ocimum canum* Sims on *Pectinophora gossypiella* Saunders (Lepidoptera:
410 Gelechiidae), insect pest of cotton in Ivory Coast. *European Scientific Journal edition*,2018 ; 14
411 (21): 1857 – 7881.

412 6. Ouattara D, Kouamé KD, Johnson F, Kassi KFJM, Yao KJ-E, Yao K *et al.* In-vitro
413 Biocontrol of Sugarcane Stem Borer (*Eldana saccharina* Walker) with Essential Oils from
414 Aromatic Plants. *Asian Research Journal of Agriculture*, 2022 ; 15 (4): 161-169.

415 7. Kouamé KD, Kassi KFJM, Kouamé KG, Kouassi KV, Dove JH, Ng Cheong LR, Zouzou M.
416 Soil moisture management and mulch impact on sugarcane yields under irrigated and rainfed
417 conditions in Côte d'Ivoire. *Journal of Biodiversity and Environmental Sciences*, 2018 ; 12 (5):
418 381-390.

419 8. Kobenan KC, Kouakou BJ, Bini KKN Kouakou M, Dick AE and Ochou OG. Effects of the
420 Essential Oils of *Ocimum gratissimum* L. and *C. citratus* Stapf on the Growth and Production
421 Parameters of Cotton in Côte d'Ivoire. *European Journal of Scientific Research*,2019 ; 154 (1),
422 21-35.

423 9. Minader (Ministry of Agriculture and Rural Development). List of pesticides approved and
424 authorized in Côte d'Ivoire as of March 15, 2018. *Directorate of Plant Protection, Control and*
425 *Quality*, 2018 ; 100 p.

426 10. Kla YF, Identification of insects (parasitoids and others) resulting from the breeding of
427 caterpillars and pupae of *Eldana saccharina* Walker at the Zuénoula Integrated Agricultural
428 Unit. End of studies dissertation in Engineering at the Ecole Supérieure d'Agronomie de
429 Yamoussoukro (INPHB),2012 ; 69 p.

430 11. Sotondji FA, Djihinto CA, Kobi DKO, Dannon E, Zodome G, Adjou E *et al.* Laboratory
431 evaluation of the larvicidal effect of cashew balm and three vegetable oils based plants of
432 *Tephrosia Purpurea*, *Ricinus Communis* and *Thevetia Neriifolia* to control populations of
433 *Plutella Xylostella* L. 1758 (Lepidoptera, Plutellidae). *European Scientific Journal*, 2020 ; 16
434 (24), 29-59.

435 12. McDonald LL, Guy RH and RD, Speirs. Preliminary evaluation of new candidate
436 materials as toxicants, repellents and attractants against stored product insects. *Marketing.*
437 *Res. Rep. No. 882.* Washington: Agric. Res. Service, US. Department of Agriculture, 1970 ;
438 183 p.

- 439 13. Tia VE, Cisse M, Douan GB and Kone A. Comparative study of the insecticidal effect of
440 the essential oils of *Cymbopogon citratus* DC and *Ocimum canum* Sims on *Cylas puncticollis*
441 Boheman, a sweet potato weevil. *International Journal of Biological and Chemical*
442 *Sciences*,2019 ; 13(3): 1789-1799.
- 443 14. Fradin MS, Day JF. Comparative efficacy of insect repellents against mosquito bites. *New*
444 *England Journal Medicine*, 2002 ; 347: 13-8.
- 445 15. Thorsell W, Mikiver A, Tunon H. Repelling properties of some plant materials on tick
446 *Ixodes ricinus* L. *Phytomedicine*, 2006 ; 13: 132-4.
- 447 16. Katz TM, Miller JH, Hebert AA. Insect repellents: historical perspectives and new
448 developments. *Journal of American Academy of Dermatology*, 2018 ; 58: 865-71.
- 449 17. Yarou BB, Silvie P, Komlan FA, Mensah A, Alabi T, Verheggen F, Francis F. Pesticidal
450 plants and protection of market gardening in West Africa (bibliographic summary).
451 *Biotechnology, Agronomy, Society and Environment*, 2017 ; 21 (4), 288-304.
- 452 18. Camara B, Koné D, Kanko C, Anno A and Aké S. Antifungal activity of the essential oils
453 of *Ocimum gratissimum* L., *Monodora myristica* (Gaaertn) Dunal and two synthetic products
454 (Impulse and Folicur), on the mycelial growth and in vitro spore production of *Deightoniella*
455 *torulasa* (SYD.) ELLIS. *Revue Ivoirienne des Sciences et technologies*,2007 ; (9): 187 - 201
- 456 19. Tano DKC., Tra BCS., Kouassi KA, Ossey CL and Soro S. Impacts of *Podagrica*
457 *decolorata* Duvivier 1892 attacks on okra cultivation and control of these adults using the
458 biopesticide NECO 50 EC (Daloa, Ivory Coast). *Journal of Applied Biosciences*,2019 ; 143:
459 14692 – 14700.
- 460 20. Kouassi AM, Ouali N’Goran S-WM, Akesse EN, Coulibaly A. Evaluation of the
461 performance of an anti-insect net in an integrated control system against the main pests of
462 cabbage in Korhogo, northern Ivory Coast. *Journal of Applied Biosciences*,2022 ; 177: 18424 –
463 18433.