New Methods For Bee Venom Production in Honeybee colonies without cheemical used

By

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ABSTRACT

In this study, different types of bee venom electrical devices were used for apitoxine extraction by workers in honeybee colonies (*Apis mellifera L.*) The result indicated that, the amounto of bee venom collected was ranged from 0.255 to 0.439g per colony in on one run (30 min.) / day. Not differences in bee venom produces from F_1 and F_2 of Carniolan, ltalian and Manzala races bees during this experiment.

INTRODUCTION

Collection of Bee – Venom since (1897) by Langer , who reported for the first time was dropped the poison sac togethes with the sting and galnds into 96% alcohol, the residue insoluble in alcohol, was dried at 40 ground to fine powder and extract with water. The other methods were reported as, the rotation and pressure encouraged the bees to sting an absorbent paper on the surface of the cylinder; the venom was separated from the absorbent paper [15] The bee venom could be separated precipitation with trichloro – acetic acid in two fractions. [1] The bee venom could be prepared by mixing fresh sting with quartz and extracted with the finely ground and diluted by hydrochloric acid [35].

The electric shock method to stimulate the bee sting, were used since (1954) by Markoiv and Molnar, it consisted of wooden collector frame with wires that were wound seven millimeters apart. Underneath the wires there was thick rubber sheet to hold the stingers left by the bees after receiveing the shock and an absorbent paper to collect. [33] . The bees sting through a sheet of nylon when shocked, the venom remains on the underside of the sheet when the current is turned off, and sting are with drawn[46] The stinger of the worker bees penetrates through the protective material and the vemnom is deposited dirctly onto the glass sheet, it dries quickly on the glass and is lates scraped off on crystallized form. The collector devices used was electrical current at 4-12.5 V. [7, 34, 26, 31, 2]

In this work some different types of elctrical bee venom collection devices are examind, the present new devices that are being proposed are meant , to improve the capacity of venom collection and provide safety for the operators and minimize bees

loss in hives desinated to this task. This research and facilities were used to evaluate these devices by the National project for Honey – bee Pests Control (N. P. H. P.) at Faculty of Agriculture, Zagazing Univ., Egypt

MATERIALS AND METHODS

For Bee venom collection from honeybee colonies, the experiments were carried out at the apiaries of National project for honeybee pests and Diseases Control, Faculty of Agriculture, Moshtohor, Zagazig University, during the nectar flow seasons in 1997, 1998 and 1999. Also the same experimental work was carried out at the branch of the project in Shobra Kubala, Menofia.

These experiments were conducted on the two generations F1 and F2 of the three races of honeybee; (Manzala, Carniolan and Italian Bees). Eighteen colonies were used in these experiments (3 colonies for each race of honeybee).

Each colony of honeybee was headed with a mated queen in the Langstroth hive that contain 7 combs covered with young bees (3 combs contains brood + 2 combs pollen + 2 combs honey).

In colonies used for venom collection, the erea of brood and honey and pollen were stabilized 228 and 240 inch for brood and honey respectively.

The artificial feeding of coloines was conducted at least 3 months before bee venom collection. The sugar syrup (1: 1w) was used in Moshtohor Feeder plus pollen substitute for estimating the brood rearing activity in the experimental honeybee coloines

The bee venom collection was conducted by Moshtohor device (Khattab, 1997).

The extraction device is powered by continuous electric current of 12 volts (using either a battery or a transformer of 220 volts, alternating the current into 12 volts continuous current.

The above apparatus has been made with different electrical parameters for different research purposes to honeybee venom collection. The electric shock should be kill the bees.

The collection of bee venom was scheduled as follows :

I- Every ten days the electrical devices was attached to each of the three experimental for 30 min. every colony and to provide 3 runs (31/8, 10/9 and 21/9) replication per run was used.

II- Every 3 days for 30 min. a peroid of 22 days strating 27/6/98 and ending 30/8/98.

Five types of devices were utilized these are according to the power used :-

1- Transform from 220 volts to 3 volts at 0.4 A.
2- Transform from 220 volts to 6 volts at 1.0 A.
3- Transform from 220 volts to 12 volts at 1.0 A.
4- Transform from 220 volts to 14 volts at 3.0 A.
5- Wet Battery at 12 volts with 16 A.

Analyses of data :

Data was projected in means together with then respective stander deviation. Also graphocal representations of means are given to the result.

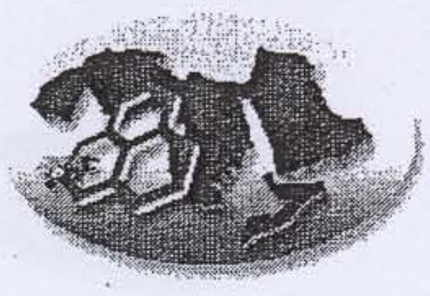
RESULTS & DISCUSSION

Bee venom collection in Honeybees colonies:

Effect of using electrical transformer 220 V. to 1V. at 1A on the mean of bee venom collction in bees races.

Table (1) indicated that the means of bee venom collected during 30 min. were 0.31, 0.32 and 0.31 g./ colony with an average of 0.313g. during the first run (31/8/1998). In the second run (10/9/1998) the means of bee venom collected by the same electrical device were 0.20, 0.22 and 0.30g./ colony with an average of 0.24. In case of the 3rd run the means of bee venom collected in 30 min . by the same electrical device were 0.17, 0.20 and 0.27 g./ colony with an average of 0.21g./ colony .Clear from the result that, the highest amount of venom was that obtained In the run in highest with the other two runs. (0.313, 0.24 and 0.21g./ colony .The above results are in agreement with authors who indicated that using the electrical device for bee venom collection is suitable in honey bees colonies (Dotmas & Hider, 1987; Omer et al , 1994 and khattab , 1997) .In addition the amount of venom obtained colony is higher than that obtained by Fakhim – Zadeh, 1998 , of 0.21g. venom colony

Effect of bee venom recreation on the areas of brood reared and the storage of pollen:



Results in table (1) indicated that, the mean area of sealed brood in 1st run was 208" while the areas of sealed brood in the 2nd bees run was 113" , and in the 3rd treatment was 81.6 " . From the above results it is clear that, there was reductions in the brood rearing. This reduction could be due to the consumption of stored pollen and honey when compared with untreated colonies.

In control treatment the area of scaled brood in 1st run was 222", 265, and 279" with an average 255.3", while in the 2nd run was 226", 279, and 275 with an average 256", and in the 3rd run was 229, 259", and 284 with an average 257.3".

In case of pollen stored in the hive during treatments with bee venom devices, the areas in inches of stored pollen are listed in table (1) indicated that , the mean area honey and pollen in 1st run waws 263.33", while the areas of honey and pollen in the 2nd bees run was 220.33" , and in the 3rd treatment was 203".

In control treatment the area of honey and pollen stored in 1st run was 232, 259, and 210" with an average 233.6", while in the 2nd run was 230 , 260 and 210 with an average 233.3", and in the 3rd run was 225, 265, and 215 with an average 235".

From the above results it is clear that, there was reductions in the stored honey and pollen . This reduction could be due to the consumptions of stored pollen and honey when compared with untreated colonies. This reductions could be attributed by that venom is a protein that require feeding on pollen and honey for its production (Omer ,1994) & (Khattab , 1997).

Table (1) Efect of using electrical device; 12V. at 1 A. on venom collection from three beed races pe 30 min.

Date of bee venom collection	No. of combs in hive	Brood combs Inch ^{2}	Brood combs Control Inch ^{2}	Honey and pollen combs inch ^{2}	Honey and pollen Combs control inch ^{2}	No. of dead Bees	Mean of bee venom collection (g/colony)
31/8/98	10	228	222	240	232	5	0.31
	10	226	265	290	259	22	0.32
	8	170	279	260	210	15	0.31
Mean	9.33	208	255.3	263.3	233.6	15	0.313
10/9/98	6	97	226	194	230	31	0.2
	8	108	267	241	260	5	0.22
	8	144	275	226	210	15	0.3
Mean	7.33	113	256	220.3	233.3	17	0.24
21/9/98	6	70	229	150	225	10	0.17
	7	100	259	225	265	5	0.2
	6	75	284	235	215	25	0.27
Mean	7	81.6	257.3	203.3	235	16.6	0.213
$\bar{X}$		135.3	256.2	229	234	14.6	0.25
SE		57.21	22.77	37.22	20.75	9.003	0.054
$\bar{X}$+ SE		192.51	278.97	266.2	254.75	23.603	0.304
$\bar{X}$-SE		78.09	233.43	191.78	213.25	5.597	0.196

* V. Volt , A. = Amber

Table (2) Effect of using electrical transformer 220 V to 6 V. at 1A. on amount of B. V. collection from three F_2 bees races.

Date of bee venom collection	Amount of B. V. collection (g/ colony)		
	Carniolan	Italian	Manzala
8/7/98	0.29	0.27	0.30
	0.28	0.26	0.31
	0.27	0.27	0.29
Mean	0.28	0.26	0.30
11/7/98	0.25	0.28	0.32
	0.29	0.28	0.32
	0.28	0.29	0.34
Mean	0.273	0.283	0.326
14/7/98	0.28	0.28	0.34
	0.25	0.27	0.34
	0.29	0.25	0.29
Mean	0.273	0.266	0.323
$\bar{X}$	0.2753	0.2696	0.3163
SE	0.014	0.011	0.019
$\bar{X}$+ SE	0.2893	0.2806	0.3353
$\bar{X}$-SE	0.2613	0.2586	0.2973

.= Volt, A. = Amber

Table (3) Effects of temperature in honeybee hive and R. H. on the amount of venom collection using electrical transformer 220 V to12 V. at 1A. in three bee races F_1. during 30 min

Date of bee venom collection	No. of combs in hive	No. of dead Bees	Amount of bee venom Temp in hive C°	Temp in hive C°		R. H. in hive	
				Bofore	After	Before	After
31/8/98	10	5	0.31	34	37	65	67
	10	22	0.32	35.5	38.5	70	82
	8	15	0.31	35.7	38.4	55	70
Mean	9.33	15	0.313	35.06	37.96	63.33	73
10/9/98	6	31	0.2	33.9	37.1	51.9	65
	8	5	.22	35.52	36.4	70.3	72.4
	8	15	0.3	36.3	40.3	63.7	64.4
Mean	7.33	17	0.24	35.24	37.9	61.96	67.2
21/9/98	6	10	0.17	32.9	35.5	60	75
	7	5	0.2	35.9	36.5	76.2	79
	6	25	0.27	36.8	37.9	76.7	79
Mean	7	16.6	0.213	35.7	36.63	70.96	77.6
$\bar{X}$			0.255	33.18	37.5	65.4	72.6
SE			0.054	0.76	1.35	8.24	6.13
$\bar{X}$+ SE			0.309	33.94	38.85	73.64	78.73
$\bar{X}$- SE			0.201	32.42	36.15	57.16	66.47

Comparison selective between types of electrical bee venom collector device:

The final results nedicated that, the battery device at 12 V. was the best one (average 0.478g.) while the other devices were 0.446g . in case the device (14 V. at 3A.) 0.439 g. in case (12 V. at 1A.) ; 0.426g in case of (3V. at 0.4A.) and the electrical transformer 220 V. to 6 V. at 1A . came the last one which produced 0.420g./ 30 min / treatment during the experiment.

From table (8) it is obvious that the amount of venom was different among the devices used . The highest amount of venom (0.478) was obtained when the wetbattery (12 volt 16 A) was used in comparison with other devices and the least amount (0.42) was obtained when (6 volt and 1A) device was used . so it is better to use the wetbattery (12v volt 16 A) provided that it well no do any harm to the colony or increase the mortality rate due to its application.

This result agreed with the recommendation by Alexandru (1983) who in a similar study used a device of 12 volt battery . and also agreed with Anne and gard (1996).

Table (8a) Amount of bee venom secreted by pure of honeybee colony (in gm.)

Generation / Transformer	Italian	Carnionlan	Manzala
3 V. at 0.4A.	0.33	0.315	0.36
6V. at 1 A.	0.345	0.325	0.375
12V. at 1A.	0.35	0.335	0.385
14 V. at 3A.	0.36	0.36	0.41
Wetbattery 12 V. with 16A.	0.38	0.365	0.435
X	0.353	0.340	0.393
SE	0.0166	0.0167	0.0265

Table (8b) amount of bee venom affected by bees races and generation (F_1 and F_2) (in gm.)

Generation / Transformer	Italian		Carniolan		Manzala		Total	Mean
	F_1	F_2	F_1	F_2	F_1	F_2		
3V. at 0.4A.	0.313	0.35	0.3	0.33	0.35	0.37	2.013	0.426
6V.at 1A.	0.34	0.35	0.32	0.33	0.32	0.35	2.10	0.42
12V. at 1A.	0.34	0.36	0.32	0.35	0.36	0.39	2.17	0.439
14V. at 3A.	0.35	0.37	0.35	0.37	0.4	0.42	2.23	0.446
Wetbattery 12V. with 16A.	0.37	0.39	0.35	0.38	0.42	0.45	2.39	0.478
Total	1.713	1.82	1.64	1.76	1.9	2.01	10.963	0.438
Mean	0.342	0.364	0.328	0.352	0.38	0.402	10.903	0.436

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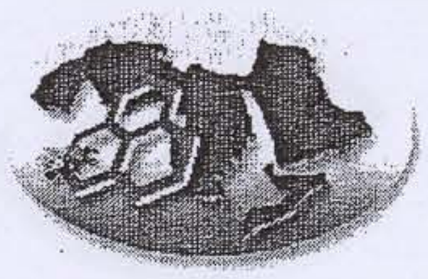

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٦- ســاعة / طائفة كل ١٠ أيام علي الترتيب وأعطت السلالة الهجين الثاني المنزلة أعلي متوسط لإنتاج السم مع هذا الجهاز.

٧- اســتخدمت جهاز آخر يعمل ببطارية سائلة ١٢ فولت قوة ١٦ أمبير كبديل لاستخدام التيار الكهربي في المناطق التــي لا يصــل إلــيها كهــرباء وأوضحت النتائج علي سلالة الهجين الثاني المنزلة والإيطالي والكرنيولي حيث أعطت بالترتيب ٠,٤١ ، ٠,٣٧، ٠,٣٤٦ جرام / طائفة عند تشغيل الجهاز لمدة نصف ساعة.

ثانياً: دراسة تأثير المواسم المختلفة علي تجميع سم النحل:

أجــريت مقارنــة بــين متوســط إنتاج السم خلال ثلاثة أشهر وهي شهر يونيو ، وشهر أكتوبر ، وشهر ديسمبر ، وأعطي شهر يونيو أكبر كمية من السم حيث أعطي متوسط إنتاج ٠,٣٣ جرام بينما أعطي شهر أكتوبر متوسط ٠,٣١٣ جرام أما شهر ديسمبر فقد أعطي متوسط قدره ٠,٢٤٣ جرام سم/ طائفة.