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MOSQUITOES OF RAIN-WATER DRAINAGE BASINS OF ABHA CITY, KINGDOM OF SAUDI ARABIA AND ITS CONTROL USING TWO FORMULATIONS OF BACILLUS THURINGIENSIS ISRAELENSIS (BTI).

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Key words : . *Culex pipiens*. *Bacillus thuringiensis* var. *israelensis (Bti)* . Culinex. Bacticide .

ABSTRACT

Rain-water drainage basins are small, numerous, widely distributed in Abha City, Kingdom of Saudi Arabia. A preliminary larval survey revealed that, 21 out of 37 surveyed basins support mosquito breeding. *Culex pipiens*, was the most abundant mosquito species in basins. Basins supported mosquito breeding all the year round. Two formulations of *Bacillus thuringiensis* var. *israelensis* (Bti) viz., Culinex tablets, and Bacticide powder were evaluated for their efficacy against mosquitoes breeding in basins. Each basin was treated with the recommended dose of each formulation (One Culinex tablet or 0.5 gm of Bacticide powder) during April and July. 2002. Both formulations induced high larval mortalities for 48 hours post-treatments, but no residual effect was observed one weak post-treatment. In both treatments, Bacticide powder was more effective than Culinex tablets . The possibility of using both formulations by individuals or authorities for mosquito control in this particular breeding place has been discussed.

INTRODUCTION

Abha city lies in the southwestern part of the Arabian Peninsula. The region abounds in mountains, valleys and fertile plains. It has a generally moderate climate throughout the year, heavy rainfall, green pasture and agricultural plateaus. All these factors have contributed in developing the region and have transformed it into a tourist attraction. Mosquito control is routinely made in the area, but no control program is directed to basins made for the

drainage of rain water. These basins are small, numerous, widely distributed in the area, and many of them are suitable for the breeding of mosquitoes.

Bacillus thuringiensis israelensis (B.t.i) has been used since the 80's in several programs for controlling black fly and mosquito species worldwide (De Melo-Santos, *et al.*, 2001). It is considered highly toxic to the target organisms and environmentally safe (de Melo-Santos, 2001, Mittal, 2003, Toma *et al.*, 2003 and Gunasekaran, 2004). Furthermore, no resistance has been detected on populations submitted to long-term exposure to this pathogen (Mittal, 2003 and Liu *et al.*, 2004). *Bti* formulations have been continuously improved to optimize their potency, stability, ease of field application and residual activity (de Melo-Santos, 2001; Blanco *et al.*, 2002; Gunasekaran *et al.*, 2002; Becker, 2003, and Fillinger *et al.*, 2003). Nonetheless, most commercially available *Bti* formulations is short lasting, except in habitats lacking organic substrate; thus it needs to be used frequently, resulting in high costs of management, so a true long lasting formulations still needs to developed (Toma *et al.*, 2003).

The present work aims to through lights on the role of rain water drainage basins, of Abha City, Kingdom of Saudi Arabia, in mosquito breeding and to evaluate the efficiency of a new, slow release tablet formulation of *Bti* (Culinex Tab Plus®), designed for use in containers and small ponds as compared to the widely used wettable powder formulation of *Bti* (Bacticide), against immature mosquitoes breeding in rain-water drainage basins of the Teachers College of Abha, Kingdom of Saudi Arabia, where many complains from mosquito bites were received.

MATERIALS AND METHODS

Monitoring mosquitoes

A preliminary larval survey was carried out for rain-water drainage basins supporting mosquito breeding in Abha city. Three basins, in the Fish market (middle of the City) and another three basins in the Teachers College of Abha (outside the City) were selected and surveyed on weekly basis throughout the period from October 2001 to September, 2002. A net (25 cm in diameter and 10 cm deep) with a long metal handle was used for larval sampling. Collected samples were transported to the

laboratory for identification and counting. All meteorological conditions of the area were kindly provided by the Meteorology and Environmental Protection Administration in Saudi Arabia..

Bti formulations

(Culinex Tab Plus®) tablets: (CULINEX®) is a slow release tablet formulation, developed by Culinex GmbH, Germany, and designed for biological control of mosquito larvae in containers and small ponds. It contains 1,300,000 ITU/tablet (= 3,400 ITU/mg) . The tablets are packed in blistered stripes (10 tablets each).

(Bacticide-DWP): Is a commercial wettable powder formulation produced in India. It contains 50% bacterial spores and 50% delta indotoxin. Its potency is 5400 IU/mg, strain 146 (Batch no: Bio-Bact-13).

Field trials

Two field trials were carried out in April, and July, 2002 to evaluate the efficiency of 2 *Bti* based formulations against mosquitoes breeding in rain water drainage basins located in the Teachers College of Abha. The basins (60 cm length x 60 cm width and different depths ranged from 0.8 to 1.2 meters) are lined with Cement. Each basin has an outlet tube of a height of about 50 cm above the bottom of the basin. Basins are covered with perforated metal covers to allow the passage of rain water into it. Basins are usually filled with rain water to a depth of (30 – 50 cm). A complete chemical analysis of water was made before the application of larvicides. Two basins were treated with Culinex Tab Plus (1 tablet/each), and another two basins were treated with Bacticide powder (0.5 gm/each), and two untreated basins were left as controls. The powder was stirred in one liter of breeding water and poured on water surface. Basins were observed 24 and 48 hours post-treatment then on a weekly basis. The impact of both formulations was determined by calculating the number of preimaginal stages/10 dips.

RESULTS AND DISCUSSION

Monitoring mosquitoes

Rain-water drainage system is very important in Abha City, where rain fall is common throughout most of the year (Table.1). Basins used to collect rain water are small, numerous, widely distributed, and many of them are suitable for the breeding of mosquitoes. The preliminary larval survey revealed that, 21 out of 37 surveyed basins were positive for mosquitoes (56.75%). Mosquito control is routinely made in Abha City using fogging machines, but no control program is directed to these basins, especially outside the City center, where many complains from mosquito bites were received. Basins supported mosquito breeding all the year round. *Culex pipiens*, was the most abundant mosquito species in basins. The prevalence of this species in drainage water collections of Abha city was also observed by Abdullah and Merdan (1995). The authors stated that fresh drinking water reservoirs is one of the best breeding habitat in many surveyed areas of Abha City.

The highest larval abundance was observed from March to August, and from April to November in site A and site B, respectively (Table 1). These results are in accordance with what have been found by Abdullah and Merdan (1995) who reported that the density of *Cx.* pipiens was relatively high during raining seasons as will as during the period (March – November). It seems that low abundance of larvae during January, February and December is due to low temperature. The sudden decrease in abundance during September can be correlated to the heavy and/or successive rains during August (12 rainy days). Heavy rains fill the basins and either kill or strongly wash larvae outside the basins. In February and April, heavy rains did not affect larval abundance because rains fall only for 3 and 2 days, respectively so that new eggs can hatch and the population can be rebuild again.

Efficiency of *Bti* formulations

The results obtained (Fig. 1) showed that basins treated with Bacticide powder (0.5 gm/basin) .in April and July, showed 100% reduction in larval density, 24 and 48 hours post-treatment. In April trial this reduction in density was reduced to 82.77%, one weak post-treatment: whereas in July, a dramatic increase in larval density was observed, so that the density

was almost equal to the pre-treatment density. In April, basins treated with Culinex tablets (1 tablet/basin), showed 89 and 95.4% reduction in larval density 24 and 48 hours post-treatment, respectively; but one week post-treatment, this reduction in density was reduced to 45.8%. In July the mean initial mortality achieved was 100% for 24 and 48 hours, then larval density was greatly increased one week post-treatment.

During the bioassay period, 5.6 and 4.9 mm of rain were recorded in April and July, respectively. The average water temperature were 17.4 and 21.7 °C in April and July, respectively (Table. 1). Results of chemical analysis of water in the treated basins (Table. 2) indicate that water is chemically suitable for drinking i.e. it has no adverse effect on the used formulations .

Most of the studies in the literature agree upon the effectiveness of *Bti* formulations against many mosquito species under different environmental conditions, but a greet contradiction about the [redacted] of these formulations was found. Our results indicate that the larvicidal activity of Culinex tablet formulation (3,400 ITU/mg) only lasted for about 48 hours against *Culex pipiens* mosquito in rain-water drainge basins of Abha City. [redacted] results were found by Toma *et al.*, [redacted] in Italy. The authors stated that, in most cases, the larvicidal activity of the Vectobac DT tablets formulation (3400 ITU/mg) only lasted [redacted] . Other studies revealed that *Bti* tablet formulations provided a residual control for a relatively longer period. [redacted] reported that Vectobac *Bti* tablets (1900 ITU/mg) provided a residual control of A. aegypti larvae in desert coolers and tire dumps for 2 and 3 weeks, respectively. Also [redacted] de Melo-[redacted] *et al.*, (2001), in Brazil, reported that (C4p1) *Bti* tablet formulation (1146 ± 52 ITU/mg) was effective against *Aedes aegypti* under simulated field conditions for [redacted] days in shade. Whereas [redacted] *et al.* (2004) reported a very long persistence of tablets formulation. The authors stated that VectoBac tablets at the dosage of 1 tablet or 0.37 g per 50 liters of water provided excellent control for about [redacted] to 112 days [redacted].

Although, the formulations used in our study and in the study of Toma *et al.*, (2003) contain more ITU (3400 ITU/mg) than the formulations used by Batra *et al.*, 2000 (1900 ITU/mg), and de Melo-Santos *et al.*, 2001 (1146 ± 52 ITU/mg); the last two formulations provided a longer

persistence in the field. This may indicate that physical and chemical characterists and nature of the breeding habitat as well as the efficiency of formulations are more important than the applied dose of *Bti*. The duration of effectiveness of slow release Bti formulations in containers is strongly influenced by the presence of an organic substrate (Ali *et al*., 1994 and Sulaiman et al., 1997) and by the absence of other biotic and abiotic factors that may occur in natural habitats (Nayar *et al*., 1999). When tablets sank. the active ingredient settled among decayed organic materials that had settled at the bottom of the basins and out of the feeding area of the larvae (Toma *et al*., 2003).

In summary. this trial demonstrated that both formulations tested (Culinex tablets and Bacticide powder) have provided control of Cx. pipiens. breeding in rain-water drainage basins. lasting 48 hours . Despite the low residual activity of both formulations, Tablet formulation seems to be an excellent choice for treatment of fresh water larval habitats. because it does not require safety measures and supervision(Batra *et al*., 2000). it does not change the odor of water and it decrease the turbidity of water(Mulla *et al*., 2004): it can be easily transported, stored and used especially in small water containers as rain-water drainage basins. The tablet as well as powder formulations should be applied in basins within 7 days after rains.

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(Table 1) Metorological conditions and monthly abundance of *mosquitoes** in rain-water drainge basins at two sites of Abha, Saudi Arabia from October, 2001 to September 2002.**

Months	No. of rainy days days*	Mm rain*	Av. water Temp °C	% Relative humidity*	No of larvae/10 dips**	
					Site A	Site B
Jan.	0	0	11.3	63	6	2
Febr.	3	34	14.3	65	12	8
Mar.	0	0	15.6	63	85	13
April	2	5.6	17.4	56	[illegible]	[illegible]
May	3	7.5	19.3	42	175	[illegible]
June	6	11.5	21.5	43	86	[illegible]
July	2	4.9	21.7	40	214	[illegible]
Aug.	12	20.1	19.7	53	[illegible]	[illegible]
Sept.	1	15.5	19.1	34	21	9
Oct.	0	0	16.8	36	34	[illegible]
Nov.	0	0	14.5	37	17	87
Dec.	1	9.8	12.4	68	11	9

* : Data kindly provided by Meterology and Environmental Protection Administration, Saudi Arabia

** : Three basins were surveyed from each site

*** : Most larve were *Culex pipiens*

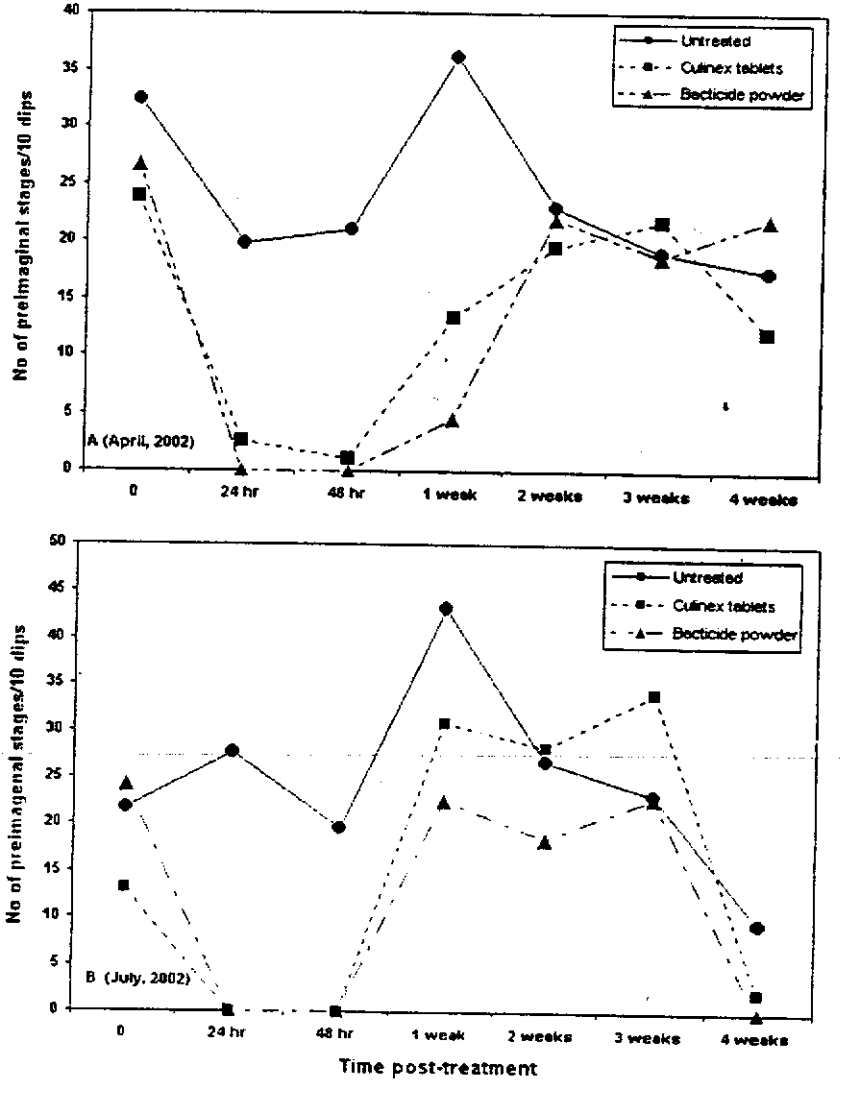
Site A : Teachers College

Site B : Fish market

(Table 2) Chemical characterstics* of water in rain-water drainge basins, Teachers College, Abha, Saudi Arabia, April, 2002.

Chemical characteristics	Mg/liter	Chemical characteristics	Mg/liter
pH value	7.5	Chloride	14
Total alkalinity	70	Sulphate	6.58
Total hardness	64	Calcium	24.05
Total dissolved solids	90	Magnesium	0.9827
Ammonia	Nil	Iron	Traces
Nitrite	Nil	Turbidity (NTU)	2.5
Nitrate	Traces	Odour	Nil

* : Water analysis was kindly made by the Central Lab., Minstry of Water, Asir Directorate



(Fig.1) Efficiency of Culinex tablets and Bacticide powder *(Bti)* against *Culex pipiens* in rain-water drainage basins of Abha, Saudi Arabia during April and July, 2002.

بعوض أحواض صرف مياه المطر بمدينة أبها – المملكة العربية السعودية، ومكافحته باستخدام مستحضرين من بكتريا باسيلس ثيرونجينسس إزرائيلينسس

عبد الوهاب عبد المقصود إبراهيم

كلية المعلمين في أبها – المملكة العربية السعودية

تتناول هذه الدراسة أهمية أحواض صرف ماء المطر، الواسعة الانتشار، بمدينة أبها ، المملكة العربية السعودية كمكان لتوالد البعوض. وقد أبان الحصر المبدئي وجود يرقات البعوض في ٢١ حوضا من مجموع ٣٧ حوضا تم فحصها. وكانت يرقات الكيولكس بيبينز هي أكثر الأنواع تواجدا في هذه الأحواض وعلى مدار العام. كما تم تقويم كفاءة مستحضرين من بكتريا باسيلس ثيرونجينسس إزرائيلينسس (أقراص كيولينكس وبودرة الباكتيسيد) في هذه الأحواض خلال شهري أبريل ويونيو، لعام ٢٠٠٢. وبمعاملة بعض هذه الأحواض بجرعات المستحضرين الموصي بها (قرص كيولينكس أو نصف جرام باكتيسيد لكل حوض) نتجت نسبة عالية من الإماتة بين اليرقات ولمدة ٤٨ ساعة، ولم يظهر لأي من المستحضرين أثر باقي بعد أسبوع من المعاملة. وتبين من كلا المعاملتين أن مستحضر الباكتيسيد أكثر كفاءة من أقراص الكيولينكس. وقد ناقش البحث إمكانية استخدام الأفراد أو الهيئات للمستحضرين في مكافحة يرقات البعوض في أحواض صرف ماء المطر.