

الداء والدواء في جناحي الذباب

الأستاذ الدكتور / مصطفى إبراهيم حمس

أستاذ الحشرات الطبية ومدير مركز أبحاث ودراسات الحشرات الناقلة للأمراض
كلية العلوم (بنين) - جامعة الأزهر - القاهرة - مصر

المقدمة

Phoresy

Alcanos Greenberg (1973) Taylor (1935)

. Mcoay et al (1982) and Frishman (1980)

Fouda Breznak (1982)

Ghanem et al . Hassan et al (1996, 1998a, 1980b, 2000) (1984)

(1986)

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الطرق والوسائل المستخدمة

١ - جمع الذباب :

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٢ - تشريح الذباب :

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٣ - عزل الكائنات الدقيقة :

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- 1- Nutrient agar emended with 1% yeast extract.
- 2- Nutrient agar emended with 5% sheep blood
- 3- MaConkey's agar
- 4- Starch nitrate agar
- 5- Tryptose blood agar
- 6- Staphylococcus media

(CFU)

Holt et al., (1944)

. Honda et al (2004)

٤ – التحليل الحصري للنشاط ضد الميكروبي :

٥ – عملية التخمير :

٦ – استخلاص وتنقية المركب الأيضي :

Bioautographic technique

Thin layer and column

pH

. chromatography

(UV)

Spectroscopy

(IR)

Spectrophotometer

Hp mudel MS 5988

Mass spectral Data

٧ – تقييم اقل تركيز مثبط للبكتريا (MIC) :

Agar Diffusion Method

النتائج والمناقشة

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Hassan, et al (1998a)

% *Bacillus*

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. *Pseudomonas Erwina Salmonella*

. MacConkey

Ahmed et al (1995)

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Empusa muscae

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Lactobacillus gasseri Salmonella Erwina

L. animalis B. circulans :

. *S. aureus P. aeruginosa B. subtilis*

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B. Circulans

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B. Circulans

Thin layer chromatography

Mass spectra

. () $C_{30}H_{37}N_4SO_9$

Bioautography ()

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. **IR**

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. **UV**

1H -NMR

. (Zhang et al 1999)

Minimum Inhibitory

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. **Concentration (MIC)**

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5 μ g/ml

% ,

S. aureus *B. subtilis* :

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الاستنتاج (وجه الإعجاز العلمي)

B. circulans

B. circulans

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5 . 5 µg/ml

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Table (1): The viable plate count of bacterial flora (CFU/ml) isolated from wings of *P. papatasi*, *M. stabulans*, *M. domestica* and *C. pipiens*.

Medium	<i>P. papatasi</i>		<i>M. stabulans</i>		<i>M. domestica</i>		<i>C. pipiens</i>	
	Right wing	Left wing	Right wing	Left wing	Right wing	Left wing	Right wing	Left wing
Nutrient agar with y. extract	5×10^2	2×10^2	2.9×10^2	3.4×10^2	5.1×10^3	5.1×10^3	Nil	Nil
Nutrient blood	6×10^2	1×10^2	6.7×10^3	5.9×10^3	Nil	4.3×10^3	3×10^2	Nil
MacConkey	Nil	Nil	3.9×10^3	3.9×10^3	Nil	Nil	Nil	Nil
Starch nitrate	1.7×10^2	Nil	5×10^2	4.8×10^2	Nil	Nil	Nil	Nil
Tryptose blood	1×10^2	Nil	3.1×10^3	2.7×10^3	3.3×10^3	3.5×10^3	1×10^2	1.4×10^2
Staphylococcus	Nil	Nil	Nil	Nil	Nil	Nil	Nil	Nil

Table (2): Organisms isolated from wings of the sandfly, the false stable fly, the house fly and the mosquito.

Symbole	Organism
175b	<i>Salmonella arizona</i>
157y	<i>Erwina herbicola</i>
68S	Yeast
165y	<i>Bacillus subtilis</i>
181y	Yeast
191T	Actinomycete
88T	<i>Bacillus circulans</i>
132T	<i>Staphylococcus aureus</i>
127T	<i>Lactobacillus animalis</i>
98y	<i>Bacillus mycoides</i>
113M	<i>Pseudomonas aeruginosa</i>
201T	<i>Lactobacillus gasseri</i>

Table (3): Antagonistic action of bacterial species between each other grown on nutrient broth amended with yeast extract.

<i>Organism</i>	<i>S. arizona</i> 175b	<i>E. herbicola</i> 157y	<i>B. subtilis</i> 165y	<i>B. circulans</i> 88T	<i>S. aureus</i> 132T	<i>L. animalis</i> 127T	<i>B. mycoides</i> 98y	<i>P. aeruginosa</i> 113M	<i>L. gasseri</i> 201T
<i>S. arizona</i> 175b	x	-ve	+ve	+ve	+ve	2+ve	-ve	+ve	+ve
<i>E. herbicola</i> 157y	-ve	x	+ve	-ve	-ve	-ve	-ve	-ve	-ve
<i>B. subtilis</i> 165y	-ve	+ve	x	+ve	2+ve	3+ve	+ve	-ve	+ve
<i>B. circulans</i> 88T	-ve	-ve	-ve	x	+ve	2+ve	-ve	+ve	-ve
<i>S. aureus</i> 132T	-ve	-ve	+ve	+ve	x	3+ve	-ve	+ve	-ve
<i>L. animalis</i> 127T	-ve	-ve	-ve	+ve	-ve	x	-ve	-ve	-ve
<i>B. mycoides</i> 98y	-ve	-ve	+ve	-ve	-ve	-ve	x	+ve	-ve
<i>P. aeruginosa</i> 113M	-ve	-ve	-ve	-ve	-ve	-ve	-ve	x	-ve
<i>L. gasseri</i> 201T	+ve	-ve	+ve	+ve	+ve	+ve	+ve	-ve	x

-ve = no inhibition zone, +ve = weak inhibition zone, 2+ve = moderate inhibition zone, 3+ve = good inhibition zone.

Table (4): Antagonistic action of most potent bacterial species grown on peptone water during log phase.

Organism	<i>S. aureus</i> 132T	<i>P. aeruginosa</i> 113M	<i>B. circulans</i> 88T	<i>L. animalis</i> 127T	<i>B. subtilis</i> 165y
<i>S. aureus</i> 132T	x	+ve	4+ve	+ve	4+ve
<i>P. aeruginosa</i> 113M	-ve	x	-ve	-ve	-ve
<i>B. circulans</i> 88T	±ve	+ve	x	3+ve	+ve
<i>L. animalis</i> 127T	-ve	+ve	2+ve	x	2+ve
<i>B. subtilis</i> 165y	+ve	+ve	4+ve	2+ve	x

-ve = no inhibition zone, ±ve = doubtful inhibition zone, +ve = weak inhibition zone, 2+ve = moderate inhibition zone, 3+ve = good inhibition zone, 4+ve = very good inhibition zone.

Table (5): Bioautography and migration (R_f) of the active metabolite 88T with various developing solvents.

Developing solvent system	R_f value
Petroleum ether	0.00
Benzene (saturated with water)	0.00
Chloroform (saturated with water)	1.00
Carbon tetrachloride (saturated with water)	0.75
Methanol	0.85
N-Butanol (saturated with water)	0.80
Acetone	0.45
Diethyl ether	0.55
Ethyl acetate	0.50
Amyl acetate	0.00
3% ammonium chloride	0.10
N-Butanol : pyridine : water (1 : 0.6 : 1)	0.00
N-Butanol : Acetic acid : water (2 : 1 : 1)	0.00
Distilled water	0.20
Methylene chloride (1 : 1)	0.00

Table (6): The MIC of active metabolite 88T.

Test organism	MIC ($\mu\text{g/ml}$)
Reference strains:	
<i>Bacillus subtilis</i> NCTC 8236	<5
<i>Bacillus pumilus</i> NCTC 8241	<5
<i>Micrococcus luteus</i> ATCC 9341	12
<i>Staphylococcus aureus</i> NCTC 7447	12
<i>E. coli</i> BPP01	16
<i>Pseudomonas aeruginosa</i> ATCC 10145	83
<i>Klebsiella pneumonia</i> NCIB 9111	18
<i>Candida albicans</i> IMRU 3669	94
<i>Saccharomyces cerevisiae</i> CBS 1171	94
<i>Aspergillus niger</i> LTU 131	>100
Local isolates:	
<i>Bacillus subtilis</i> 165y	<5
<i>Bacillus mycoides</i> 98y	<5
<i>Staphylococcus aureus</i> 132T	<5
<i>Lactobacillus animalis</i> 127T	32
<i>Lactobacillus gasseri</i> 201T	40
<i>Salmonella arizona</i> 175b	<5
<i>Erwina herbicola</i> 157y	>100
<i>Pseudomonas aeruginosa</i> 113M	>100
<i>Yeast</i> 181y	>100
<i>Yeast</i> 68y	>100

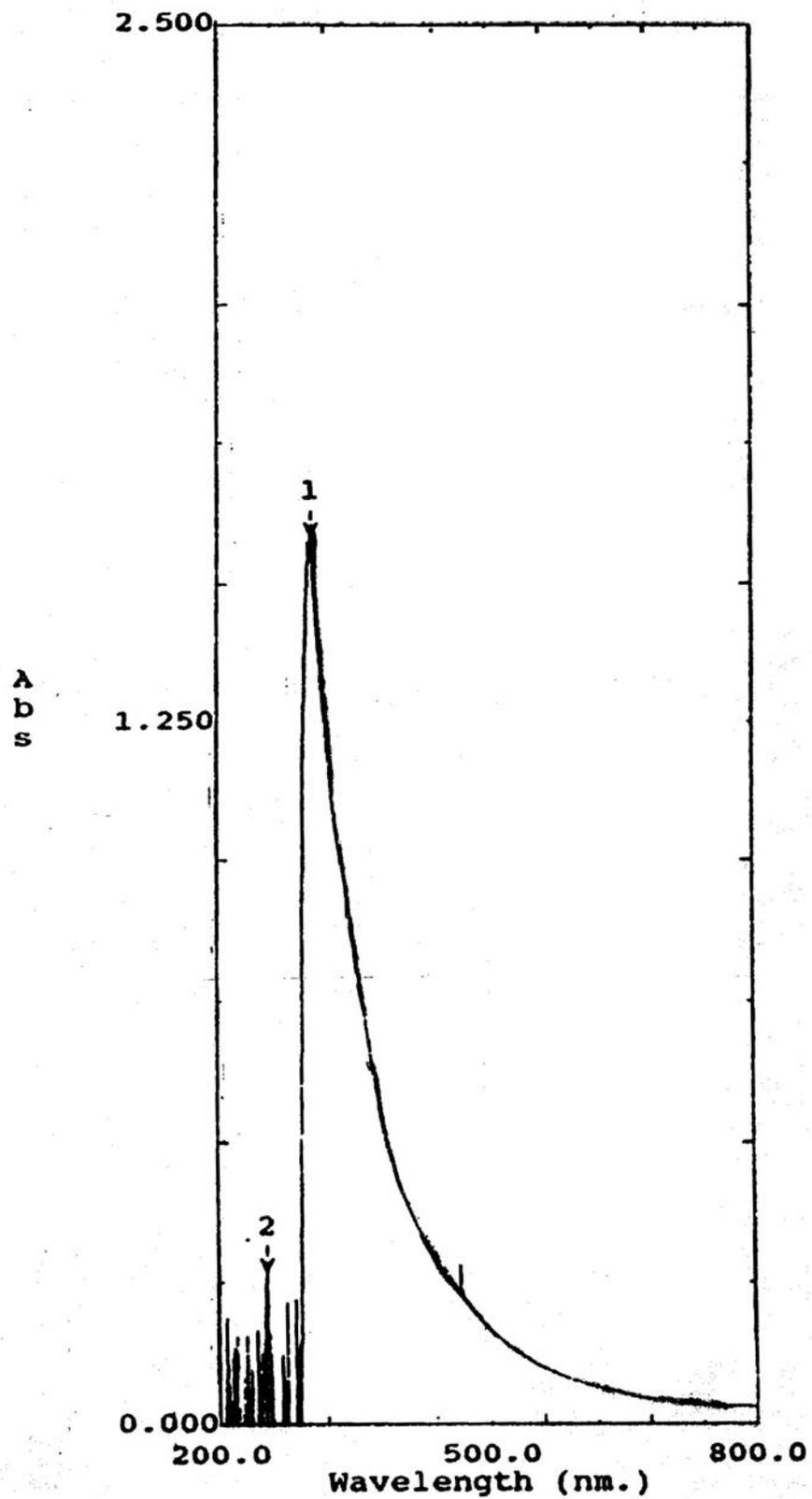


Fig. (1): A simplified scheme for the extraction, isolation and purification of the active metabolite 88T biosynthesized by *Bacillus circulans* 88T.

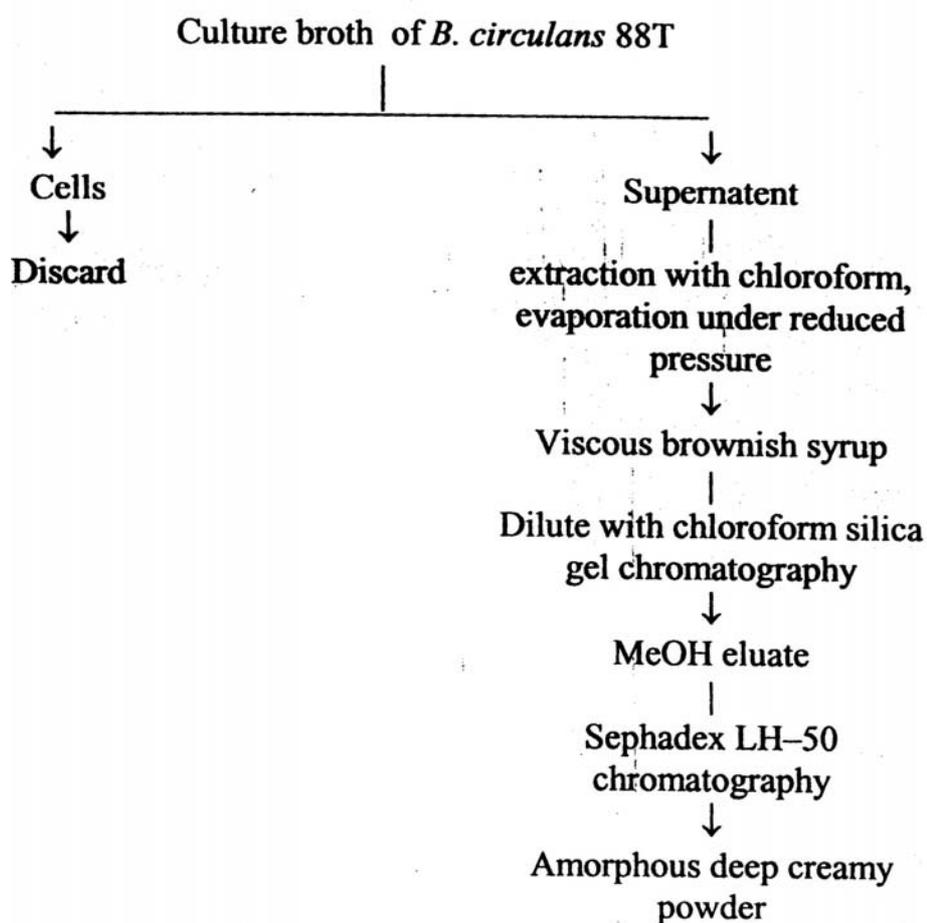


Fig. (2): A simplified scheme for the extraction, isolation and purification of the active metabolite 88T biosynthesized by *Bacillus circulans* 88T

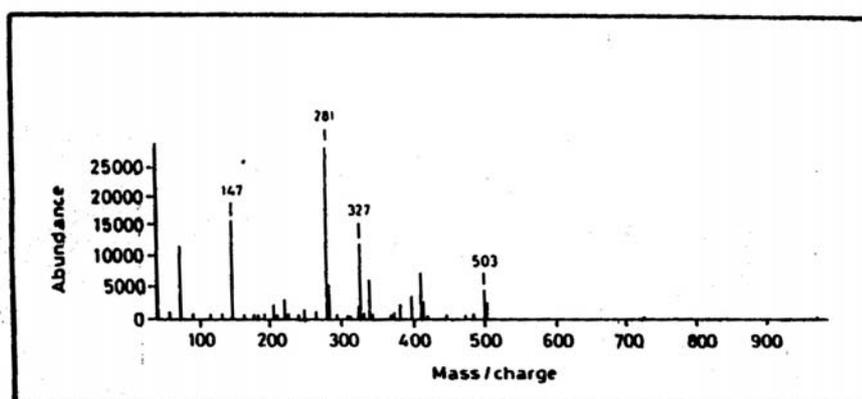


Fig. (3): Mass spectrum of the active metabolite 88T

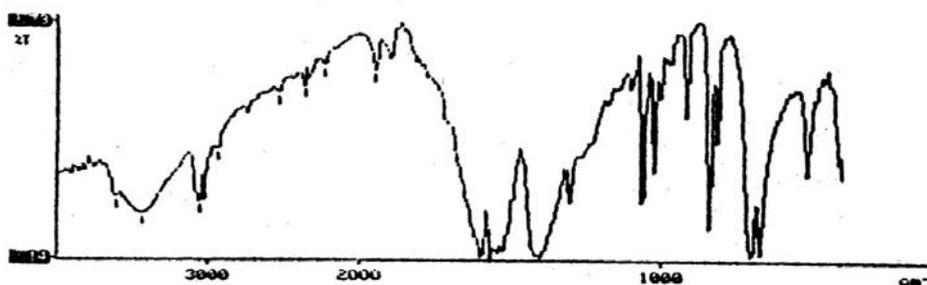


Fig. (4): IR spectrum of the active metabolite 88T.

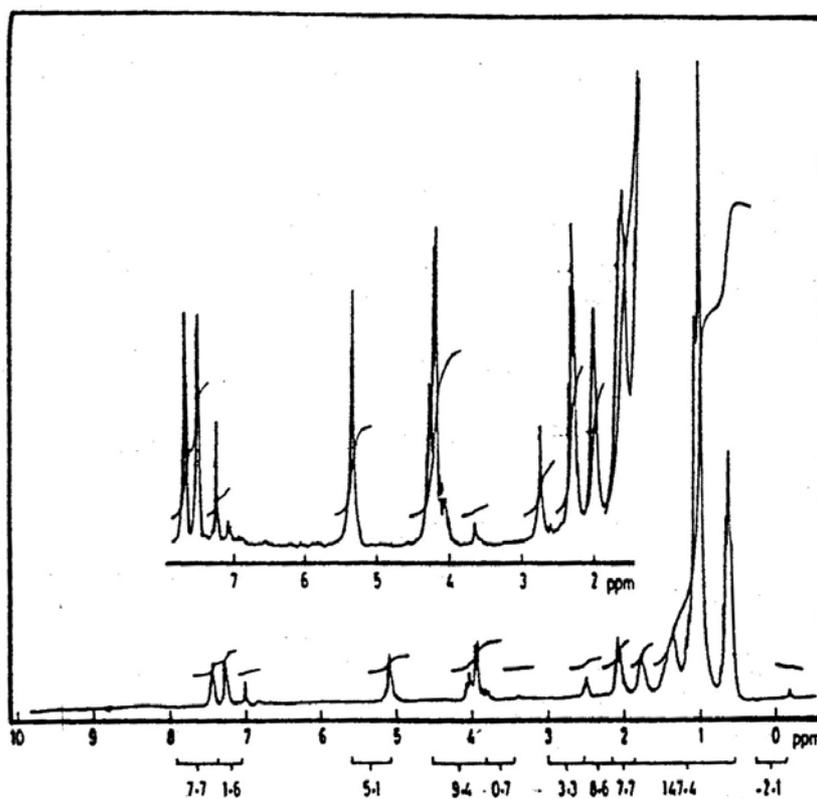


Fig. (5): ^1H -NMR spectrum of the active metabolite 88T.

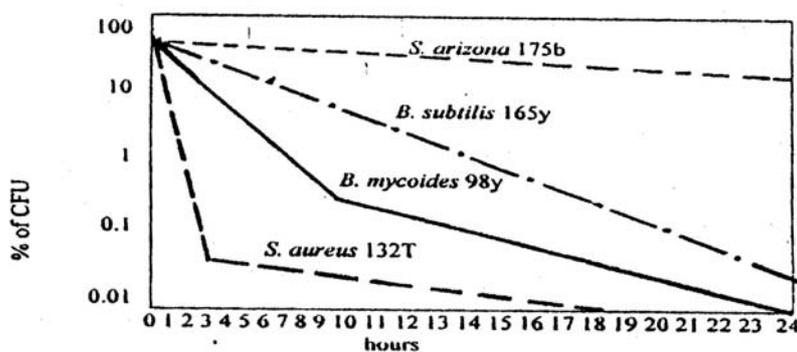


Fig. (6): The time killing curves of the active metabolite 88T using *Bacillus mycooides* 98y, *Bacillus subtilis* 165y, *Staphylococcus aureus* 132T and *Salmonella arizona* 175b.

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المراجع

العربية :

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