Arab Gulf J. Scient. Res., Agric. Biol. Sci., **B7** (1), pp. 113-127 (1989)

Streptomyces - Bacterial Interaction: The Possible Control of Soft Rot Disease of Melon by *Streptomyces* Species

Mary S. Khalil, Mohamed I. Naguib, Youssry E. Saleh

Botany Department, Faculty of Science, Cairo University, Cairo, Egypt

ABSTRACT. Screening for the antimicrobial activity of the culture filtrates of *Streptomyces venezuelae*, *S. rubiginosus* and *S. recifencis* showed that the Gram positive bacterium, *Clostridium acetobutylicum* was completely resistant whereas *Bacillus subtilis*, *B. licheniformis* and *Mycobacterium phlei* while insensitive to the filtrate of either *S. venezuelae* or *S. rubiginosus* were very sensitive to *S. recifencis* filtrate. Coupling the filtrates of *S. venezuelae* with *S. recifencis* reduced or even nullified the latter's potency whereas coupling *S. rubiginosus* with *S. recifencis* lowered the latter's effects. Of the Gram negative bacteria, *Escherichia coli*, *Aerobacter* sp., *Shigella flexneri*, *Pseudomonas putida* and *Erwinia toxica* were totally resistant to the antibiotics produced by the three *Streptomyces* species. *Erwinia carotovora* var. *carotovora*, *E. carotovora* var. *citrullis* and *Serratia marcescens* were sensitive only to the filtrate of *S. recifencis*. *Salmonella typhimurum* was sensitive only to *S. venezuelae* filtrate.

The mixed culture filtrates of *S. rubiginosus* with *S. recifencis* were highly potent against *E. coli* and *Pseudomonas putida*. Growth of *E. toxica* was highly suppressed by the filtrate of the mixed culture of *S. venezuelae* and *S. rubiginosus* but the sensitivity of *E. carotovora* var. citrullis to *S. recifencis* was abolished by mixed cultures with the other two *Streptomyces* species whereas that of *E. carotovora* var. *carotovora* was only attenuated.

Supplementation with nickel noticeably increased whereas supplementation with cadmium or lanthanum slightly decreased the sensitivity of *E. carotovora* var. carotovora to *S. recifencis* filtrate. Nickel-fed cultures of *E. carotovora* var. *citrullis* or *E. toxica* were completely resistant to *S. recifencis* and *S. venezuelae* + *S. rubiginous* filtrates respectively. Nickel-feeding, in combination with lanthanum and/or cadmium had the same effect on *E. toxica* but the high sensitivity of nickel-treated *E. carotovora* var. *carotovora* to *S. recifencis* filtrate was abolished by addition of lanthanum and reduced by replacement or further addition of cadmium.

The toxins produced by *E. toxica* had a broader spectrum than those of *E. carotovora* var. *carotovora* and *E. carotovora* var. *citrullis;* particularly on Gram positive bacteria. Of the tested Gram negative bacteria, only *Serratia marcescens* and *Shigella flexneri* were sensitive as well as *E. carotovora* var. *carotovora* and *E. toxica*.

There has been considerable interest in the possibility that antagonism among microorganisms could be made to play a significant part in controlling plant diseases: *e.g.* antagonism has been found among fungi (Hartley 1921, Machacek 1935, Christensen 1936, Katser 1938, Tervet 1938, Tadashi and Mikami 1969) and among bacteria and actinomycetes (Henry 1931, Mikio and Kaziro 1966, Hans *et al.* 1966, Korotyaev 1966, Omura *et al.* 1982).

Meredith (1946) employed actinomycetes for the control of infection of banana plants by *Fusarium oxysporum cubense.* One method proposed was the addition to the soil of an organism known to produce an antibiotic active against the plant pathogen (Machacek 1935, Tervet 1938). Greig-Smith (1917) showed that actinomycetes could be a factor in the control of the bacterial content of soil, but the "toxin" produced was not elucidated.

Slechta *et al.* (1978) found that the macrolide antibiotic albocycline, when added to a *Streptomyces venezuelae* UC-2560 (WC-3627) fermentation underwent a gradual loss of antimicrobial activity. This inactive product was identified as 2.3-dihydroalbocycline. Ewersmeyer-Wenk *et al.* (1981) isolated a new antibiotic produced by *S. venezuelae*. This new compound could inhibit Gram + ve and to a lesser extent Gram − ve bacteria. Chatterjee and Vining (1981) showed that *S. venezuelae* contains intracellular alpha-glucosidases, induced during growth on certain carbohydrate sources. Such induction was arrested by rifampicin and suppressed by chloramphenicol or streptomycin. Poetsch *et al.* (1985) isolated four new and two known peptide antibiotics containing amicelnomycin (Acm) from a culture of *S. venezuelae*. These di- and tri-peptides exhibited antimicrobial activity against Gram − ve bacteria that could be reversed by the addition of biotin. Also Kern *et al.* (1985) isolated six peptide antibiotics from a culture filtrate of *S. venezuelae* To 2460. All antibiotics inhibited growth by blocking biotin biosynthesis.

In this investigation we have studied the efficacy of culture filtrates of three *Streptomyces* species as antagonists to pathogenic bacteria particularly those causing soft rot disease of melons.

Material and Methods

Three species of *Streptomyces* viz. *S. venezuelae* (S_1); *S. rubiginosus* (S_2) and *S. recifencis* (S_3) were used. These were kindly provided by the Microbiological Research Centre (MIRCEN), Cairo.

The antagonistic potential of the broth of each test species as well as that of the mixed cultures of the three species, were tested against twenty different

bacterial species. Of these 11 species were Gram negative including:

Aerobacter sp. (MIRCEN).

Escherichia coli (Amin 1986); Enteric bacteria.

Erwinia carotovora var. *carotovora* (Saleh *et al.* 1984); causing soft rot of melon plants in Egypt.

E. carotovora var. citrullis (Saleh *et al.* 1985); causing wilt and rotting of melon plants in Egypt.

E. toxica (Saleh 1977); toxigenic bacterium infecting the vascular system of melon and water melon plants in USSR.

Proteus vulgaris (MIRCEN); found in fecal matter in many animals, sewage and soil.

Pseudomonas putida (MIRCEN); found in soil and water.

Salmonella typhimurum (Amin 1986); causing food poisoning to man. A natural pathogen for all warm-blooded animals.

Serratia marcescens (MIRCEN) found in water, soil and food.

Shigella flexneri 1 (Amin 1986); the most common cause of dysentry epidemics and sometimes of infantile gastro-enteritis.

Yersinia entrocolitica (Amin 1986); found in the feces and lymph nodes of both sick and healthy animals and man.

The remaining nine species were Gram positive bacteria:

Bacillus licheniformis (MIRCEN); found in soil and water.

B. subtilis (MIRCEN); found in soil and water.

Clostridium acetobutylicum (Sugar and Distillation Company Egypt); a fermenting bacterium.

Micrococcus luteus (MIRCEN); common in soil, dust, water and skin of man and other animals.

M. roseus (MIRCEN); found in dust, water and salt-containing foods.

Mycobacterium phlei (MIRCEN); widely distributed in nature.

Sarcina sp. (MIRCEN); isolated from soil, mud, diseased human stomach and the surface of cereal grains.

Staphylococcus aureus (MIRCEN); found on skin and mucous membranes.

Staphylococcus sp. (MIRCEN).

The *Streptomyces* species were cultured by inoculating 1 ml of spore suspension in 50 ml starch - nitrate medium (starch, 20 g; KNO_3, 2 g; K_2HPO_4, 1 g; $MgSO_4 \cdot 7H_2O$, 0.5 g; NaCl, 0.5 g; $CaCO_3$, 3 g; all constituents were dissolved in one litre distilled water and pH adjusted to 7.0) and incubated for 7 days at

28°C. The mixed cultures were prepared by inoculating 1 ml of a mixed spore suspension (1:1 or 1:1:1 ratio) from $S_1 + S_2$; $S_1 + S_3$; $S_2 + S_3$; $S_1 + S_2 + S_3$.

The antibiotic potential of the filtered broth (by centrifugation at 5000 rpm for 15 minutes) was biologically assasyed by the diffusion plate method according to the following procedure: 0.5 ml aliquots of the filtrate were transferred to wells (10 mm diameter) bored in 10 cm - diameter plates, freshly inoculated with 24-h cultures of the test bacterial suspension containing 5×10^8 cells/ml. The diameter of the inhibition zone, if any, was measured after 24 h at 28°C. Five replicate plates were measured for each treatment.

Since it has been established, in this laboratory, that the virulence and/or potency of *E. carotovora* var. *carotovora* depends, among other factors, on the presence of lanthanum, nickel and/or cadmium, in the culture medium (Khalil and Saleh 1987; Saleh and Khalil 1987), it was thought to elucidate the possible effect of such treatment using 10^{-6} M of the elements and their mixtures, on the resistance of the experimental *Erwinias* to the *Streptomyces* antibiotics. The resistance of the test bacteria and *Streptomyces* species to the toxins produced by the *Erwinias,* if any, was also investigated.

Results and Discussions

Fig. 1 shows that the antibiotics produced by each of the three *Streptomyces* species alone or in mixed cultures were basically potent against the Gram positive *Staphylococcus, Micrococcus* and *Sarcina* species. $S_2 + S_3$ mixed culture differed by showing a very high potency only against *M. roseus.* Of the remaining Gram positive organisms, *Clostridium acetobutylicum* was totally resistant to the *Streptomyces* species in all their combinations whereas *B. subtilis, B. licheniformis* and *Mycobacterium phlei* were totally insenitive to S_1 or S_2 but very sensitive to S_3 antibiotic(s). The broth of the mixed culture of $S_1 + S_2$ inhibited the growth of these bacilli and *Mycobacterium* to a lesser extent than S_3 alone; a phenomenon that was slightly, if at all, affected (Bacilli) or very highly intensified *(Mycobacterium)* by the addition of S_3 to the mixture. Coupling S_1 with S_3 reduced the potency of the latter's antibiotic(s) to almost the same level as that produced by $S_1 + S_2$ (Bacilli) or even totally nullified the inhibitory effects on *Mycobacterium;* a phenomenon that was completely reversed when S_2 was coupled with S_3 where the bacilli were totally insensitive but the growth of *Mycobacterium* was severely suppressed to the same extent as in the mixed culture of $S_1 + S_3$.

These observations indicate that *Streptomyces venezuelae* partially, if not completely, repressed the antagonistic properties of *Streptomyces recifencis* against the Gram positive bacteria whereas *Streptomyces rubiginosus* was totally

repressive (except for *Micrococcus roseus* and *Mycobacterium phlei*). It appears that mixing *S. venezuelae* with *S. rubiginosus* would have initiated the formation of new antibiotic(s) capable of retarding or inhibiting the growth of the bacilli and *Mycobacterium*. Synergism between the antibiotic(s) produced by $S_1 + S_2$ might be the possible explanation for the observed results.

According to Smith (1952) the decomposition products of chloramphenicol, produced by the action of certain bacteria have either a growth-stimulatory effect on bacteria or interfere with the growth inhibitory action of the antibiotic. Kitano and Tomasz (1979) and Leduc *et al.* (1982) have shown that chloramphenicol inhibited the lysis of growing bacterial cultures. Patel (1985) showed that lysis triggered by cell wall active antibiotics is inhibited by chloramphenicol at a concentration of 20 μg/ml.

Fig. 1 further shows that of the Gram negative bacteria *Aerobacter* sp., *Shigella flexneri 1, Pseudomonas putida* and *E. coli* were totally resistant to the antibiotics produced by each of the *Streptomyces* species. Both varieties of *E. carotovora* and *Serratia marcescens* were sensitive to S_3 antibiotic(s) whereas *S. typhimurum* was sensitive only to S_1 antibiotic(s). *Proteus vulgaris* and *Yersinia entrocolitica* were sensitive to S_2 antibiotic(s); the former was less sensitive to S_1 and insensitive to S_3 antibiotics whereas the latter was remarkably sensitive to S_3 and totally resistant to S_1 antibiotic(s).

Mixed culture filtrates of the *Streptomyces* species did not affect the resistance of *Aerobacter* or *Shigella* but the mixed culture of $S_2 + S_3$ was highly potent against *E. coli;* more prominently in the absence of added S_1. This adds further support to the previous suggestion regarding the effect of these mixtures on *Micrococcus* and *Mycobacterium*. Similar observations were noted for the mixed cultures against *Pseudomonas* where the insensitivity of the organism was severely broken by the $S_2 + S_3$ mixture and to a lesser extent by the other mixtures but the three organisms in a mixture seemed ineffective.

The presence of S_3 in any mixture seemed to restore the resistance of *E. toxica* to the antibiotics produced by the other two species but $S_1 + S_2$ mixture was highly inhibitory. On the other hand, the sensitivity of *E. carotovora* var. *citrullis* to S_3 was totally abolished by any of the mixtures whereas that of *Serratia marcescens* was slightly attenuated. Coupling S_1 with S_3 abolished the latter's potency gainst *E. carotovora* var. *carotovora* whereas coupling with S_2 was less effective. Coupling S_1 with either S_2 or S_3 did not affect the potency of the former against *S. typhimurum* but the remaining mixtures abolished its potency. Similarly, the potency of S_3 against *Yersinia entrocolitica* disappeared when mixed with the other organisms whereas that of S_2 was considerably raised when coupled with S_1. Again, S_3 abolished the potency of S_1 against *Proteus,* a phenomenon that was restored by the presence of S_1 that suppressed the potency of S_2 when mixed with it.

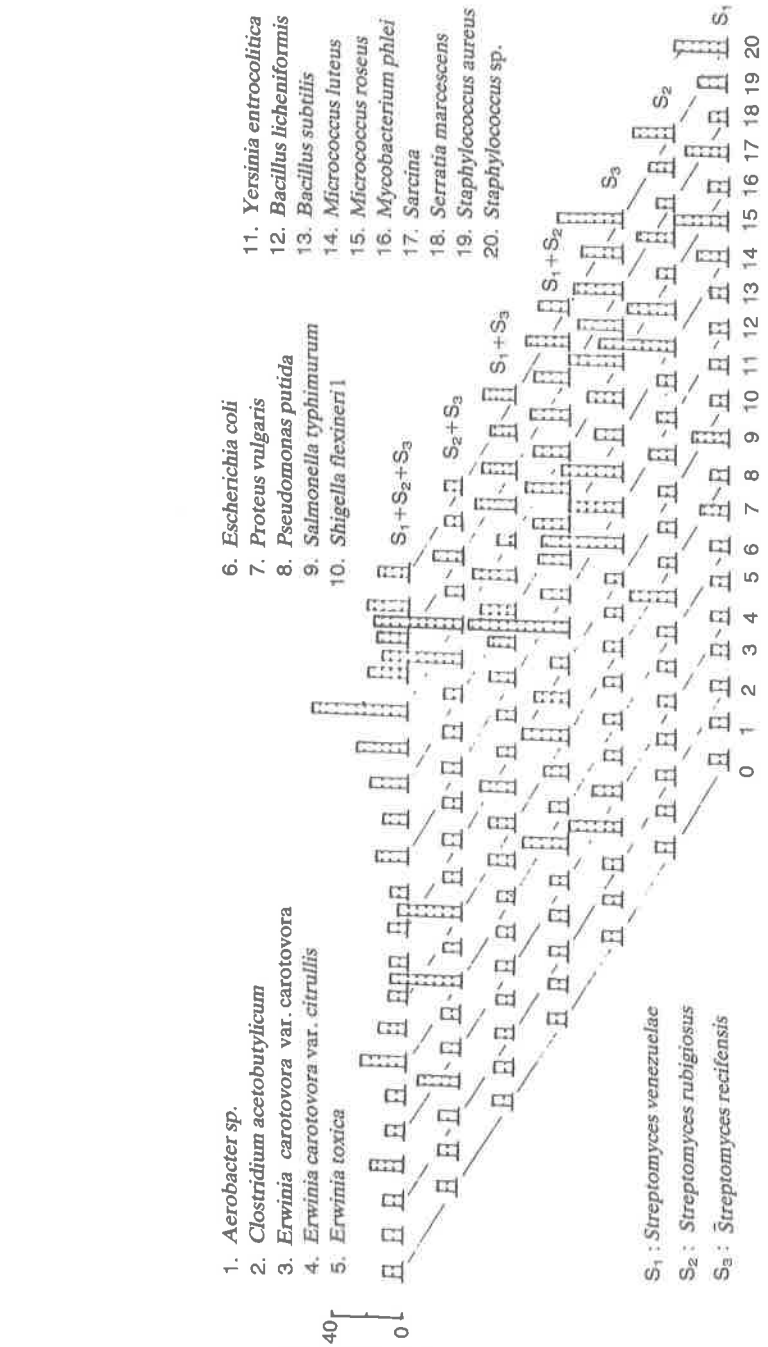

Fig. 1. Antagonistic properties of three *Streptomyces* species against selected members of Gram negative and Gram positive bacteria using 0.5 ml of the broth in 10 mm diameter wells.

These observations indicate that S_3 seemed to be capable of producing antibiotic(s) other than A × −18 reported by Falcao de Morais *et al.* (1957), that were highly potent against several Gram positive and Gram negative bacteria including the plant pathogen *E. carotovora.* A × −18 was reported by Waksman and Lechevalier (1962) as an antifungal antibiotic with slight activity against *Staph. ureus* and *B. anthracis.*

In this connection it may be mentioned that Sabath (1969) found that 15% of *E. coli,* 55% of *Klebsiella,* 47% of *Enterobacter* and 60% of *Serratia* sp. were chloramphenicol resistant. Urban (1972) found that 18% of 94 *Shigella flexneri* and *S. sonnei* strains were also chloramphenicol resistant. Poetsch *et al.* (1985) and Kern *et al.* (1985) were able to isolate four new and two known peptide antibiotics containing amiclenomycin, from a culture of *S. venezuelae.* These were all active against Gram negative bacteria. A synergistic enhancement of the antibiotic activity was found by a polypeptide isolated from the same broth.

Fig. 2 shows that apart from the variations in the rate of growth and/or virulence of the *Erwinias,* nickel noticeably increased whereas cadmium or lanthanum slightly decreased the sensitivity of *E. carotovora* var. *carotovora* to S_3 antibiotics. Nickel - treated *E. carotovora* var. *citrullis* or *E. toxica* were completely resistant to S_3 and $S_1 + S_2$ antibiotics respectivcly. Lanthanum or cadmium hardly affected the sensitivity of the *citrullis* variety but slightly lowered (lanthanum) or even abolished (cadmium) the sensitivity of *E. toxica* to S_3 and $S_1 + S_2$ respectively. The sensitivity of the carotovara variety to $S_2 + S_3$ or $S_1 + S_2 + S_3$ was totally alleviated by adding lanthanum or nickel to its media. Cadmium had similar effects regarding the sensitivity towards $S_2 + S_3$ but exerted no effect toward the sensitivity to $S_1 + S_2 + S_3$. Lanthanum seemed to break down the resistance of the *carotovora* variety of *Erwinia* and the *toxica* species to $S_1 + S_3$ and that of the *citrullis* species to $S_1 + S_2$; a similar phenomenon was also observed for nickel (*citrullis* variety) and cadmium (*toxica* species). Again, nickel or cadmium broke the resistance of the *citrullis* variety to $S_1 + S_3$ and of the *toxica* species to the mixtures containing S_2.

These observations indicate that such elements had disturbed in one way or the other, the physiological activities of the cells that resulted in the unusual sensitivity to the mixed culture broth of the *Streptomyces* species.

It is interesting to note that the presence of nickel in combination with lanthanum and/or cadmium totally protected the *citrullis* variety against these *Streptomyces* species. Lanthanum mixtures had the same effect on *E. toxica* except against the mixture of $S_2 + S_3$. The high sensitivity of nickel-treated *carotovora* variety to S_3 antibiotics was totally abolished by addition of lanthanum to the former's medium but was only reduced with the replacement or the further

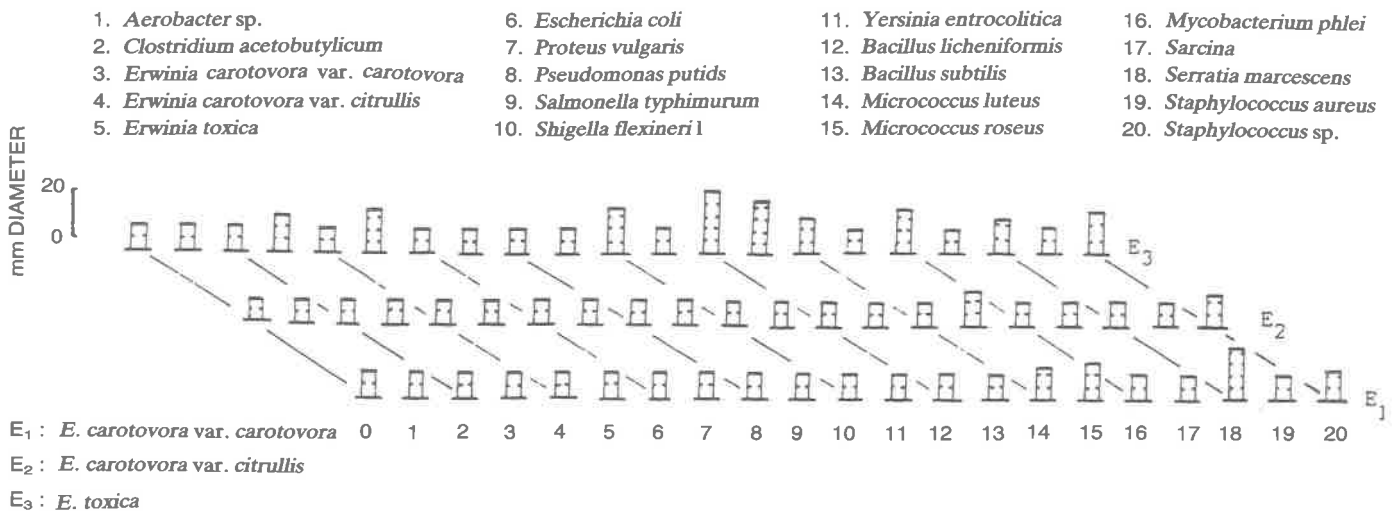

Fig. 2. Antagonistic properties of three *Erwinia* species against selected members of Gram negative and Gram positive bacteria using 0.5 ml of the broth in 10 mm diameter wells.

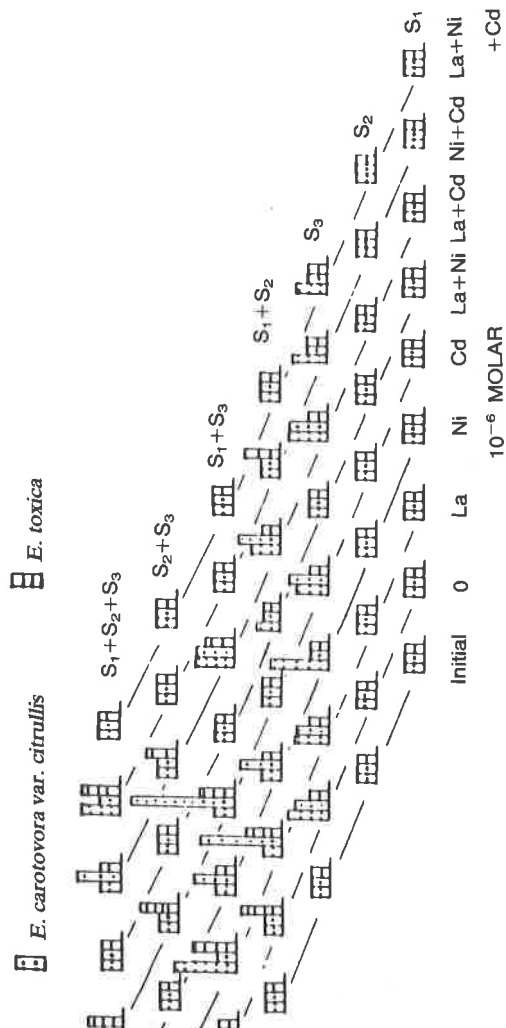

Fig. 3. Effect of various combinations of lanthanum, nickel and cadmium on the resistance of three *Erwinia* species to the antibiotics produced by the three *Streptomyces* species using 0.5 ml of the broth in 10 mm diameter wells.

addition of cadmium to the nutrient medium. Cadmium + lanthanum only slightly altered the sensitivity compared to single metal treatment, but broke the sensitivity of the organism to $S_1 + S_2$ or $S_1 + S_3$ mixed culture broth. A similar phenomenon was also observed for the *citrullis* variety with further sensitivity to the mixture of the three *Streptomyces* species.

Changes in cell wall formation (Tomasz *et al.* 1970) might give a clue to these observations. These observations further show that the mixed elements - with the synergy or antagonism between them (Saleh and Khalil 1987, Khalil and Saleh 1987) - had a more intrinsic effect on the physiology of the test bacteria, leading in most cases to higher resistance and/or tolerance to the antibiotics produced by the *Streptomyces* species.

In previous investigations, Cerataneana and Popova (1948), Petrovich and Shustova (1951) and Gnytenko (1966, 1968) reported that *E. carotovora* var. *citrullis* had a toxic effect on warm blooded animals including man. Similarly, *E. toxica* was reported by Korobko (1973) as a toxin producer. Accordingly it was thought to screen the possible effects of such toxins on various bacteria including the producing organisms (the Erwinias).

Fig. 3 shows that the toxins of *E. toxica* had a broader spectrum than that of the test varieties of *E. carotovora,* mostly on Gram positive bacteria including *Micrococcus luteus, Bacillus licheniformis, B. subtilis* and *Mycobacterium phlei.* Of the Gram negative bacteria, only *Serratia marcescens* and *Shigella flexneri* were sensitive to *E. toxica* as well as *E. carotovora* var. *carotovora* and *E. toxica.* The broth of *E. carotovora* var. *carotovora* slightly inhibited the growth of both *Staphylococcus* species and the two *Micrococcus* species but was more effective on *Serratia marcescens.* The three filtrates had no effect on the three *Streptomyces* species or their mixed cultures.

These observations point to the fact that the virulence of these organisms is not correlated with the toxins they produced. *E. carotovora* var. *citrullis* (with the least effective toxins) is known to be a well adapted, well developed, more virulent form species of *E. carotovora* var. *carotovora* (Kryvets 1975). *E. toxica* is not yet reported in Egypt.

References

Amin, A.A. (1986) Biochemical and Physiological Aspects of Some Enterobacteriaceae. *Ph.D. Thesis, Faculty of Science, Cairo University.*

Ceratanena, O.N. and **Popova, A.E.** (1948) K vaprosu o vozmognosti otravlenia nekotorium (Arbuzo). *Trudy Sartovoskova Instituta Sanitariya i gigiena No. 4.*

Chatterjee, S. and Vining, L.C. (1981) Nutrient utilization in Actinomycetes induction of alpha-glucosidase in *Streptomyces venezuelae. Can. J. Microbiol.* **27:** 639-645.

Christensen, J.J. (1936) Antibiotics. A Survey of Penicillin, Streptomycin and other Antimicrobial Substances from Fungi, Actinomycetes, Bacteria and Plants. **Florey, H.W., Chain, E., Heatley, N.G., Jennings, M.A., Sanders, A.G., Abraham, E.P.** and **Florey, M.E.** (eds.) Vol. **1:** p. 68. Oxford University Press London (1949).

Ewersmeyer-Wenk, B., Zaehner, H., Krone, B. and **Zeeck, A.** (1981) Metabolic products of microorganisms. 207-Haloquinone: A new antibiotic active against Halobacteria. 1-Isolation, characterization and biological properties. *J. Antibiot.* **34:** 1531-1537.

Falco de Morais, J.O., De Lima, O.G. and **Maia, M.D.** (1957) No estudo Sobra *Nocardia recifei* Lima et alii e sua desigancao como *Streptomyces recifencis. Ann. Soc. Biol. Penambuco* **15:** 239-253.

Greig-Smith, R. (1917) Contributions to our knowledge of soil fertility: the action of certain microorganisms upon the numbers of bacteria in the soil. *Proc. Linnean Soc. N.S. Wales* **42:** 162-166.

Gnytenko, M.P. (1966) Toxichiskii bakterioza arbuzov na youngavastok evropeiskoi chasti RSFSR; parspektivi barba s nimi. *Fsecioyoznim Symposium po Bakterialnim Rostinii, Kiev.* 65-66.

Gnytenko, M.P. (1968) Toxichiski bakterioza arbuzov na youngavastol evropeiskoi chasti RSFSR; parspektivi borba s nimi. *Izd. Nauka Dumko, Kiev.* 334-337.

Hans, B., Ammann, J. and **Muller, W.** (1966) 7-chlor and 7-brom Actinomycine from *Streptomyces* against *B. subtilis. Tetrahedron Lett.* **30:** 3595-3597.

Hartley, C. (1921) Antibiotics. A Survey of Penicillin, Streptomycin and other Antimicrobial Substances from Fungi, Actinomycetes, Bacteria and Plants. **Florey, H.W., Chain, E., Heatley, N.G., Jennings, M.A., Sanders, A.G., Abraham, E.P.** and **Florey, M.E.** (eds.) Vol. **1:** p. 67. Oxford University Press London (1949).

Henry, A.W. (1931) *ibid,* p. 67.

Katser, A. (1938) *ibid.*, p. 68.

Kern, A., Kabatek, U., Jung, G., Werner, R.G., Poetsch, M. and **Zaehner, H.** (1985) Amiclenomycin peptides; isolation and structure elucidation of new biotin antimetabolites. *Liebigs Ann. Chem.* **0**(5): 877-892.

Khalil, M.S. and **Saleh, Y.E.** (1987) Effect of some heavy metals on the pathogenicity of *E. carotovora* var. *carotovora. Proc. 5th Congr. Egypt. Phytopathol. Soc.* 455-464.

Kitano, K. and **Tomasz, A.** (1979) Triggering of autolytic cell wall degradation in *E. coli* by B-lactam antibiotics. *Antimicrob. Agents Chemother.* **16:** 838-848.

Korotyaev, A.I. (1966) Effect of chloramphenicol on alpha-glutaric dehydrogenase activity in *E. coli* and **Proteus vulgaris. Mikrobiologiya 35:** 13-19.

Korobko, A.P. (1973) *Erwinia toxica* sp. nova. An agent of cucumber bacteriosis. *Mikrobiol. Zh. An Ukr-SSR* **35:** 699.

Kryvets, I.O. (1975) *Erwinia carotovora* in the lupin rhizosphere. *Mikrobiol. Zh. Kiev* **37:** 161-165.

Leduc, M., Kasra, R. and **Van Heijenoort, J.** (1982) Induction and control of the autolytic system of *E. coli, J. Bacteriol.* **152:** 26-34.

Machacek, J.E. (1935) Antibiotics. A Survey of Penicillin, Streptomycin and other Antimicrobial Substances from Fungi, Actinomycetes, Bacteria and Plants. **Florey, H.W., Chain, E., Heatley,**

N.G., Jennings, M.A., Sanders, A.G., Abraham, E.P. and **Florey, M.E.** (eds.) Vol. **1:** p. 68. Oxford University Press London (1949).

Meredith, C.H. (1946) *ibid*, p. 53.

Mikio, K. and **Kaziro, Y.** (1966) Mechanism of action of chromomycin A_3: Inhibition of nucleic acid metabolism in *B. subtilis* cells. *J. Biochem.* 59-66.

Omura, S., Iwai, Y., Hinotozawa, K., Tanaka, H., Takahashi, Y. and **Nakagawa, A.** (1982) OM-704 A: A new antibiotic active against Gram +ve bacteria, produced by *Streptomyces* sp. *J. Antibiot.* **35:** 1426-1429.

Patel, M.V. (1985) An agar plate method for the screening of antibiotics triggering autolytic enzymes. *J. Antibiot.* **38:** 527-529.

Petrovitch, S.L. and **Shustova, L.N.** (1951) K vaprosu o mikroflora arbuzov. *Gigiena i Sanitariya* **3:** 41-44.

Poetsch, M., Zaehner, H., Werner, R.G., Kern, A. and **Jung, G.** (1985) Metabolic products from microorganisms. 230-amiclenomycin peptides new antimetabolites of biotin taxonomy, fermentation and biological properties. *J. Antibio.* **38:** 312-320.

Sabath, L.D. (1969) Current concepts: Drug resistance of bacteria. *New Eng. J. Med.* **280:** 91.

Saleh, Y.E. (1977) Biological Diseases of Melons and Water-Melons in the Ukrainian - SSR. *Dessert. Candidat. Inst. Microbiol. Virol. Acad. Sci.* Ukrainian SSR.

Saleh, Y.E. and **Khalil, M.S.** (1987) Further studies on the interaction of some heavy metals on the susceptibility of the host and potentiality of the pathogen causing soft rot disease of melons. *Proc. 5th Congr. Egypt. Phytopathol. Soc.* 465-477.

Saleh, Y.E., Naguib, M.I. and **El-Hendawy, H.H.** (1984) Biological studies on bacteria pathogenic to melons in Egypt caused by *Erwinia carotovora* (Jones) Holland var. *carotovora. Egypt. J. Bot.* **27:** 75-80.

Saleh, Y.E., Naguib, M.I. and **El-Hendawy, H.H.** (1985) Biological studies on bacteria pathogenic to melons in Egypt caused by *Erwinia carotovora* (Jones) Holland f. sp. *citrullis* Sizova. *Proc. Egypt. Bot. Soc. (Ismailia Conf.)* **4:** 1130-1148.

Slechta, L., Ciadella, J. and **Hoeksema, H.** (1978) Biomodification of albocycline by *Streptomyces venezuelae. J. Antibiot.* **331:** 319-323.

Smith, G.N. (1952) In The Actinomycetes. Vol. III. Antibiotics of Actinomycetes. **Waksman, A.S.** and **Lechevalier, A.H.** (eds.) p. 257. The Williams and Wilkins Co. Baltimore (1962).

Tadashi, A. and **Mikami, Y.** (1969) Identification keys for antibiotics producing *Streptomyces.* Antifungal antibiotic producers. *Ann. Rep. Inst. Food Microbiol.* Chiba Univ. **22:** 59-79.

Tervet, I.W. (1938) Antibiotics. A Survey of Penicillin, Streptomycin and other Antimicrobial Substances from Fungi, Actinomycetes, Bacteria and Plants. **Florey, H.W., Chain, E., Heatley, N.G., Jennings, M.A., Sanders, A.G., Abraham, E.P.** and **Florey, M.E.** (eds.) Vol. **1:** p. 68. Oxford University Press London (1949).

Tomasz, A., Albino, A. and **Zanati, E.** (1970) Multiple antibiotic resistance in a bacterium with suppressed autolytic system. *Nature* **227:** 138-140.

Urban, T. (1972) Transferable multiple drug resistance of *Shigella* strains isolated in Sweden. *Scand. J. Inf. Dis.* **4:** 221.

Waksman, S.A. and **Lechevalier, H.A.** (1962) The Actinomycetes, Vol. III, p. 197. The Williams and Wilkins Co. Baltimore.

(Received 10/04/1988;
in revised form 17/10/1988)

تداخل البكتيريا والأستربتوميسيتات
١ ـ دراسة أولية على امكانية مقاومة مرض العفن الطرى للشمام بواسطة أنواع من الأستربتوميسيتات

ماري صبحي خليل و محمد إبراهيم نجيب و يسري السيد صالح

قسم النبات ـ كلية العلوم ـ جامعة القاهرة ـ مصر

في دراسة لتأثير نشاط راشح كل من الأستربتومايسيس فينزويلي و الأستربتومايسيس روبيجينوزس، الأستربتومايسيس راسيفنسيز فقد أظهرت البكتريا الموجبة لصبغ جرام والكلوستريديام اسيتوبيوتيريكم مناعة كاملة في حين كانت بكتيريا باسيلس ساتيلي والباسيلس ليكينوفورميس والميكوباكتيريام فلاي غير حساسة لراشح كل من ستربتومايسيس فينزويلي وستربتومايسيس روبيجينوزس ولكنها أظهرت حساسية واضحة مع راشح الأستربتومايسيس راسيفنسيس فقد وجد أن قدره النوع الأخير قد قلت أو أختفت نهائياً. كذلك عند خلط كلا من نوعى الأستربتومايسيس روبيجينوزس على الأستربتومايسيس راسيفنسيس قد عمل أيضاً على خفض القدرة التأثيرية للنوع الأخير.

ومن البكتريا السالبة لصبغ جرام مثل الأشيريشيا كولاي والأيروباكتر وشيجيلا فلا كسنيرى وبسيدوموناس بيوتيدا واروينيا توكسيكا فقد كانت لها درجة مقاومة كاملة لأنواع المضادات الحيوية التي تفرز بواسطة الثلاثة أنواع من استربتوما يسيتات. أما بكتريا الأروينيا كاراتوفورا صنف كاراتوفورا وبكتريا أروينيا كاراتوفورا صنف سينترواليس وسيراشيامرسيسنس فقد كانت حساسة لراشح الأستربتومايسيس راسيفنسيس فقط.

أما بكتريا السلمونيلا تيفيموريام فقد أظهرت حساسية فقط مع راشح

الأستربتومايسيس فينزويلي.

وفي خليط لمزرعة من الأستربتومايسيس روبيجينوزس وستربتومايسيس راسيفنسيس فقد كانت لها قدرة واضحة ضد الأشيريشيا كولاى والبسيد ومونـاس بيوتيدا. أما نمو بكتريا الأروينياتوكسيسكا فقد أحبط بنسبة كبيرة بواسطة راشح خليط من مزرعة من الأستربتومايسيس فينزويلي وستربتومامسيس روبيجينوزس. أما حساسية بكتريا الأروينيا كاراتوفورا صنف سيترواليس للأستربتومايسيس راسيفنسيس فقد أضمحلت عند خلطه مع النوعين الأخرين من الأستربتوميسيتات. أما مع بكتريا الأروينيا كاراتوفورا صنف كاراتوفورا فقد ضعف هذا التأثير فقط.

كذلك فقد وجد أن استجابة بكتريا الأروينيا كاراتوفورا صنف كاراتوفورا لراشح الأستربتومايسيس راسيفنسيس تتأثر بإضافة بعض العناصر الثقيلة حيث ان اضافة عنصر النيكل يزيد هذه الحساسية بدرجة ملحوظة في حين أن عنصري الكادميوم واللانثانام فهما يقللان إلى حد ما هذه الحساسية. أما عند إضافة النيكل إلى مزرعة من بكتريا الأروينيا كاراتوفورا صنف سيترواليس وبكتريا الأروينيا توكسيكا فقد أبدت هذه البكتريا مناعة واضحة لراشح كل من الأستربتومايسيس راسيفنسيس ومخلوط من الأستربتومايسيس فينزويلي والأستربتومايسيس روبيجيمنوزس على التوالي.

أما إضافة عنصر النيكل إلى اللانثانام مع/ أو الكادميوم فقد أبدت نفس التأثير على الأروينيا توكسيكا.

أما شدة حساسية بكتريا الأروينيا كاراتوفورا صنف كاراتوفورا المعاملة بالنيكل لراشح الأستربتومايسيس راسيفنسيس فقد اختفت هذه الحساسية عند اضافة عنصر اللانثانام وقلت أيضاً عند احلاله بالكادميوم أو بإضافة زيادة منه.

أما السموم التي تتكون بواسطة بكتريا الأروينيا تولسيكا فقد أبدت مجالاً تأثيرياً واسعاً أكثر من الأروينيا كاراتوفورا صنف كاراتوفورا والأروينيا كاراتوفورا

صنف سيترووليس خاصة البكتريا الموجبة لصبغ جرام فقد كانت بكتريا السيراشيا مرسيسنس والشيجيلا فلاكسنيرى والأروينيا كاراتوفورا صنف كاراتوفورا حساسة لهذه السموم .

هذا بالإضافة إلى حساسية بكتريا الأروينيا توكسيكا نفسها لهذه السموم .