

### Growth Responses of *Fusarium oxysporum* to Metabolites of Some Rhizospheric Microflora of Egyptian Cotton Varieties

DURING studies on the rhizospheric microflora of different cotton varieties grown in Egypt, *Fusarium* wilt-resistant Ashmouni cotton variety rhizosphere proved to be inhabited by the highest percentage of strains of *Bacillus subtilis*, while susceptible Giza 26 cotton variety rhizosphere contained the highest percentage of a strain of *B. megatherium*<sup>1</sup>.

Preliminary screening tests in which each of the bacterial strains was streaked against *Fusarium oxysporum* on either Dox's or soil-extract agar media have proved that *B. subtilis* strains were highly antagonistic to the wilt-inducing pathogen; the former spread rapidly and overgrew *Fusarium* mycelium and encircled it completely. Hyphae of *Fusarium* were lysed and malformed. On the other hand, *B. megatherium* was stimulatory to *Fusarium* mycelial growth. The latter overgrew *B. megatherium* and produced comparatively very abundant conidia with abnormally large size and high septation.

Growth responses of *Fusarium oxysporum*, which proved to be a virulent pathogen causing wilt of either Karnak or Giza 26 cotton variety seedlings, to metabolites of either *B. subtilis* or *B. megatherium* on Dox's liquid medium have been tested by the following two methods: (1) germinative potentialities of *Fusarium* microconidia in hanging-drop cultures of either *B. subtilis* or *B. megatherium* metabolites; (2) growth of *Fusarium* mycelium on differently treated metabolites of either *B. subtilis* or *B. megatherium*. Filtrates of the test organisms were obtained from cultures of two ages, that is, 5- and 10-days old and were either cold-sterilized<sup>2</sup> or autoclaved. Control solutions using either Dox's liquid or water were also used.

The results (Tables 1 and 2) show that considerable suppressed germination of *Fusarium* microconidia and mycelial growth took place in response to either 5- or 10-days old culture filtrates of *B. subtilis* whether unheated or autoclaved. Ageing of culture or application of heat to its filtrate did not appreciably affect the latent period and final percentage of germination or the mycelial dry-weight of *Fusarium*. On the other hand, on using filtrates of *B. megatherium* from either 5- or 10-days old cultures, the final percentage germination of *Fusarium* microconidia rose to 100 per cent. At the same time, *Fusarium* mycelial growth was significantly higher as compared with that on control Dox's liquid media. A stimulatory factor for *Fusarium* microconidial germination and mycelial growth seems to be present in filtrates of *B. megatherium* the potency of which is not affected

Table 1. GROWTH RESPONSES OF *Fusarium* MICROCONIDIA TO METABOLITE-FILTRATES OF *Bacillus subtilis* AND *B. megatherium* AT 30° C.

Experimental medium	Age of culture	Filtrate treatment	Germinative capacities	
			Latent period	Germination (per cent)
<i>B. subtilis</i> filtrate	5 days	unheated	5 hr.	40
		autoclaved	5 hr.	40
	10 days	unheated	5 hr.	48
		autoclaved	5 hr.	50
<i>B. megatherium</i> filtrate	5 days	unheated	5 hr.	100
		autoclaved	5 hr.	100
	10 days	unheated	5 hr.	100
		autoclaved	4 hr.	100
Pure Dox's	—	—	2 hr.	90
Water	—	—	3 hr.	80

Table 2. *Fusarium* MYCELIAL DRY WEIGHT (IN MG.M.) AFTER 10 DAYS AT 30° C. IN RESPONSE TO DIFFERENTLY TREATED METABOLITE-FILTRATES OF *B. subtilis* AND *B. megatherium*. PURE DOX'S LIQUID IS DILUTED WITH WATER AND METABOLITE FILTRATES WITH PURE DOX'S LIQUID\*

Experimental medium	Dilution (per cent)	Filtrate from			
		5-day old cultures		10-day old cultures	
		Un-heated	Auto-claved	Un-heated	Auto-claved
<i>B. subtilis</i> filtrate	100	33	35	29	30
	50	38	40	33	35
	25	45	48	38	41
<i>B. megatherium</i> filtrate	100	40	43	38	42
	50	73	77	80	85
	25	81	85	92	105

\* Mycelial dry weight on 100 per cent pure Dox's liquid, 75 mgm.; on 75 per cent Dox's liquid, 61 mgm.; on 50 per cent Dox's liquid, 40 mgm.

by heating. On the other hand, ageing of culture results in an increased stimulative effect over *Fusarium* mycelial growth.

Further work concerning the nature of the inhibitory anti-fungal metabolites of *B. subtilis* and the stimulatory factor of *B. megatherium* for *Fusarium* growth are still in progress. The former are probably of the nature of an antibiotic<sup>3</sup> while the latter seems to be a growth factor. Difference in the rhizosphere microflora of wilt-resistant and susceptible cotton varieties might explain why *Fusarium oxysporum* fails to invade Ashmouni cotton roots while its pathogenicity is established on susceptible Karnak and Giza 26 cotton varieties.

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<sup>1</sup> Naim, M. S., Mahoud, S. A. Z., and Hussein, A. M., *A'in Shams Sci. Bull.*, No. 1, 173 (1956).

Menon, K. P. V., *Ann. Bot.*, 48 (1934).

<sup>2</sup> Prescott, S. C., and Dunn, C., "Industrial Microbiology", 2nd Edn. (McGraw-Hill Co., 1949).

### A Simple Technique for producing Fruit Bodies of Wood-destroying Basidiomycetes

In attempts to identify some of the many basidiomycete cultures isolated in our investigations of decay, we have tested a number of methods and have found that described here of the greatest value. It produces relatively large, identifiable fruit bodies from cultures representing a wide range of genera and families, and it has given consistent results in the hands of a number of workers, is not laborious, and requires no special equipment or materials.

The method involves establishing the culture to be identified on an enriched sawdust medium in a glass jar and then allowing it to grow through, and fruit on, a block of readily decayed wood incubated in diffuse daylight in a humid but not stagnant atmosphere.

The medium now used is a mixture of air-dry sawdusts (4-24 mesh) of seven timbers in the following proportions: *Pinus radiata* D. Don (sapwood), 25; *Eucalyptus obliqua* L'Herit. (sapwood), 25; *E. diversicolor* F. v. M., 10; *Acacia dealbata* Link., 10; *Nothofagus cunninghamii* Oerst., 10; *Rhizophora* sp. 10; *Ceratopetalum apetalum* D. Don, 10. (This