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Effect of foliar application of urea on leaf surface mycoflora of mustard and barley

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The number of fungi/cm² leaf increased insignificantly in the first sampling on treated mustard leaves and in the first and second samplings on barley leaves. A significant decrease in the number of fungi was noted in the rest of the samplings. A little variation in the number of species was recorded between the control and treated leaf samples. *Acrophialophora fusispora, Aureobasidium pullulans, Epicoccum nigrum, Fusarium chlamydosporum, Penicillium citrinum* and *P. rubrum* exhibited favourable effect of urea and thus their percentage distribution increased on the treated leaves.

INTRODUCTION

The effect of foliar spray of urea on the microbial population of leaves has been studied by several workers (Crosse et al. 1968; Ross, Burchill 1968; Burchill, Cook 1971; Hudson 1971). However, no attempt has been made to correlate the population dynamics with the recommended dose of urea which is beneficial for the plants. This lacuna warranted the authors to make a detailed investigation of the problem. The study was intensified for longer duration following two different isolation techniques.

MATERIALS AND METHODS

Mustard (*Brassica campestris* L.) var. YS-42 and barley (*Hordeum vulgare* L.) var. 'Amber' were sown in earthenware pots (18×25 cm) separately (2 plants/pot) and experiments were performed after 30 days of the establishment of the plants. 5 ml/plant of urea (2% for mustard

and 3% for barley) was sprayed on 10 plants. Leaves sprayed with sterilized distilled water served as control. Dates of spraying and sampling were as follows:

Date		Sample No.	Spray No.
December (1976)	2	—	1
	8	1	—
	18	2	—
	22	—	2
	28	3	—
January (1977)	7	4	—
	11	—	3
	17	5	—

On each spraying date leaf surface mycoflora were isolated by dilution plate and washed disks techniques (Dickinson 1971). After the observation the fungi/cm² and their percentage frequencies were calculated.

RESULTS

An insignificant increase in the number of fungi/cm² leaf was observed in the first sampling of treated leaves of mustard and in the first and second samplings of that of barley. A significant decrease in the number was recorded in rest of the samplings in both the cases (Table 1, 2). Statistically significant ($P=0.01$) variation in fungi/cm² leaf in relation to samplings was recorded on mustard leaf but insignificant variation was found on barley leaf. A smaller number of species was recorded from the treated leaf samples in comparison to control in each sampling. The total number of species recorded from treated leaf samples of mustard was smaller than that of control whereas it was the same for treated and control barley leaves; 43 (3 *Phycomycetes* and 40 *Deuteromycetes*) and 40 species (2 *Phycomycetes* and 38 *Deuteromycetes*) were isolated from the control and treated leaf samples of mustard respectively. Total 38 species (3 *Phycomycetes* and 35 *Deuteromycetes*) were isolated from the control as well as treated leaf samples of barley.

Alternaria brassicae, Aspergillus luchuensis, Bipolaris tetramera, Cephalosporium roseo-griseum, Papulaspora sp., Rhizoctonia solani, Rhizopus nigricans and red sterile mycelium were not recorded from the treated leaf samples of mustard whereas *Aspergillus candidus, A. sydowi, A. terreus, Beltrania sp., Memnoniella echinata, Pithomyces maydicus* and brown and white sterile mycelia from the treated leaf samples of barley. Some fungi which were not recorded from the control appeared on the treated leaf samples e.g. *Bispora catenula, Curvularia se-*

negalensis, *M. echinata*, *Myrothecium roridum* and *Pestalotia* sp. on mustard leaf and *Aspergillus luchuensis*, *A. nidulans*, *B. catenula*, *Fusariella indica*, *Humicola grisea*, *Phoma* sp. and red and yellow sterile mycelia on barley leaf. The number of some individual fungi/cm² leaf increased on the treated leaf samples of both the plants e.g. *Aureobasidium pullulans*, *Epicoccum nigrum*, *Fusarium chlamydosporum*. The number of *Acrophialophora fusispora*, *Penicillium citrinum* and *P. rubrum* increased only on treated barley leaf.

DISCUSSION

Increase in the number of fungi/cm² on leaf samples treated with urea in the first few samplings may be attributed fo sufficient supply of nitrogen which favours the growth and sporulation of a large number of leaf surface fungi. Excessive amount of any fertilizer including a nitrogenous one may not be as suitable to the leaf surface mycoflora as its limited amount. This might be the reason for the decrease in number of fungi/cm² in later samplings.

The growth of some species like *A. fusispora*, *A. pullulans*, *E. nigrum*, *E. chlamydosporum*, *P. citrinum* and *P. rubrum* was stimulated by urea and thus their percentage distribution increased in the treated leaf samples. The disappearance of some species like *Aspergillus brassicae*, *Aspergillus candidus*, *A. luchuensis*, *A. sydowi*, *A. terreus*, *Beltrania* sp., *Bipolaris tetramera*, *C. roseo-griseum*, *M. echinata*, *Papulaspora* sp., *Pithomyces maydicus*, *Rhizopus solani*, *R. nigricans*, brown, red and white sterile mycelia from the treated samples night be attributed to a indirect unfavourable effect of urea. Inter-competition amongst the species might have played a major role in the distribution of the leaf surface microfungi due to which certain fungi susceptible to antagonism could have been suppressed by the tolerant ones. The property of urea acting as an alkali may be one important aspect of suppression of some fungi (Lehman, Hudson 1977).

Chemical and microbial changes in leaves after urea treatment were also reported by Crosse et al. (1968) and Ross and Burchill (1968). Inhibitory effect of urea at 0.1 and 0.2 M solution *in vitro* has also been reported by Agrawal (1975). In the case of *Vinca rosea* L. (=*Lochnera rosea* Reichb.) 5% urea spraying exerted an adverse effect for majority of rhizosphere fungi (Kanaujia 1974). Stimulation of *Cladosporium* spp., *Alternaria* sp. and *Fusarium* spp. on urea treated samples was reported by Burchill and Cook (1971). The also observed that *Aureobasidium pullulans* did not consistently increase by urea treatment.

Table 1

Effect of urea on the number of fungi/cm^2

on mustard and barley leaves recorded by dilution plate technique

Species	Mustard										Barley									
	Sampling dates																			
	8 Dec		18 Dec		28 Dec		7 Jan		17 Jan		8 Dec		18 Dec		28 Dec		7 Jan		17 Jan	
	C	T*	C	T	C	T	C	T	C	T	C	T**	C	T	C	T	C	T	C	T
Choanephora cucurbitarum /Berk. et Rav./ Thaxt.	20	10	16	6	10	5	-	-	-	-	10	4	4	-	4	-	-	-	-	-
Mortierella subtilissima Oud. et Koning	15	20	10	-	10	-	13	-	5	-	20	10	10	8	5	-	-	-	-	-
Rhizophus nigricans Ehr.	-	-	-	-	4	-	-	-	4	-	-	-	4	8	-	-	-	-	-	-
Acrophialophora fusispora /Saksena/ M.B.Ellis	-	18	4	7	8	10	4	-	15	20	12	18	4	4	8	-	40	20	-	-
Alternaria alternata /Fr./ Keissler	20	18	34	9	54	15	50	10	116	101	15	20	26	30	38	20	45	30	98	87
A.humicola Oud.	19	10	20	4	20	5	30	20	70	40	10	4	19	25	14	10	25	8	35	20
Aspergillus candidus Link	-	-	-	-	-	-	-	-	-	-	-	-	4	-	-	-	-	-	-	-
A. flavus Link	10	-	5	-	-	5	10	5	-	-	-	-	-	-	17	-	10	5	-	-
A. fumigatus Fres.	17	19	-	-	-	-	19	-	5	-	9	17	-	-	-	9	5	-	-	-
A. luchuensis Inui	-	-	-	-	-	-	6	-	4	-	-	-	-	-	-	4	-	-	-	-
A. nidulans /Eidam/ Wint.	-	-	-	-	-	-	-	-	-	-	-	4	-	-	-	-	-	-	-	-
A. niger van Tiegh.	-	13	9	-	8	-	6	10	23	20	5	-	-	5	21	13	20	5	10	-
A. sydowi /Bain et Sart./ Thom et Church	-	-	-	-	-	-	-	-	-	-	-	-	-	-	10	-	12	-	-	-
A. terreus Thom	4	-	-	-	-	-	5	10	-	5	-	-	-	-	-	-	-	-	10	-
Aureobasidium pullulans /de Bary/ Arnaud	9	-	10	4	10	15	25	42	20	31	12	4	7	-	8	-	17	-	15	5
Beltrania sp.	-	-	-	-	-	-	-	-	-	-	5	-	-	-	-	-	-	-	-	-
Bipolaris spicifera /Bain./ Subram.	18	25	20	15	20	17	45	31	52	30	20	30	15	20	18	22	10	-	10	-
B.tetramera /McKinney/ Shoemaker	-	-	-	-	8	-	10	-	-	-	-	-	-	-	-	-	-	-	-	-
Bispora catenula /Lév./ Sacc.	-	-	-	-	-	-	-	10	-	-	-	-	-	-	-	-	-	-	-	21

Cephalosporium acremonium Corda	4	13	4	8	6	-	-	-	-	-	4	17	7	5	4	-	5	-	25	-
C. roseo-griseum Saksena	4	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Cladosporium cladosporioides /Fres./ de Vries	142	150	213	195	330	300	369	305	428	328	119	125	167	155	213	179	295	191	315	170
C. herbarum /Pers./ Link et Fr.	19	19	30	35	49	71	59	51	78	50	21	4	34	36	17	18	55	35	81	50
Curvularia clavata Jain	13	6	17	6	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
C. lunata /Wakker/ Boedijn	-	10	21	10	4	4	10	4	20	8	-	-	13	18	4	8	7	4	-	-
Drechslera graminea Ito et Kuribayshi	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	5	-
Epicoccum nigrum Link ex Wallr.	-	-	-	-	20	50	16	40	10	45	-	-	-	-	17	20	8	16	15	20
Fusarium chlamydosporum Wollenw. et Reinking	10	-	16	24	20	46	25	62	5	35	4	20	8	25	17	20	10	20	7	10
Memnoniella echinata /Rivolta/ Galloway	-	-	-	14	-	-	-	-	-	-	-	-	-	-	4	-	-	-	8	-
Myrothecium roridum Tode ex Fr.	-	-	-	-	-	-	-	-	-	8	-	-	-	-	-	-	-	-	-	-
Nigrospora oryzae Petch	4	4	8	14	6	8	12	-	-	-	21	26	10	18	12	10	4	8	11	-
N. sphaerica /Sacc./ Mason	-	-	-	-	-	-	-	-	10	-	-	17	-	-	-	-	-	-	-	-
Papularia sphaerosperma /Pers./ v. Höhnel	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	4	-	-	10	-
Penicillium chrysogenum Thom	-	-	-	-	-	-	-	-	21	10	-	-	-	-	-	-	50	21	15	10
P. citrinum Thom	9	-	6	14	10	5	-	127	-	-	-	-	13	16	-	13	18	-	-	-
P. rubrum Stoll	-	-	-	-	-	-	-	25	20	45	-	-	-	-	-	-	-	-	-	-
Phoma sp.	-	-	-	-	-	-	-	-	-	-	-	4	-	-	-	-	-	-	-	-
Trichoderma viride Pers.ex Fr.	-	-	-	-	-	-	167	40	-	-	-	-	-	-	-	-	-	-	12	41
Papulaspora sp.	-	-	-	-	8	-	6	-	5	-	-	-	-	-	18	10	12	4	30	-
Rhizoctonia solani	-	-	-	-	-	-	-	-	12	-	-	-	-	-	-	-	-	-	-	-
Blac sterile mycelium	17	4	17	20	6	25	-	-	30	25	13	-	-	25	-	8	10	25	21	-
Brown sterile mycelium	-	-	25	10	4	-	-	-	-	5	-	-	-	-	-	-	-	-	21	-
Red sterile mycelium	-	-	-	-	-	-	-	-	10	-	-	-	-	-	-	-	-	-	-	10
White sterile mycelium	-	18	4	-	-	-	6	10	5	-	9	-	4	-	-	-	-	-	-	-
Yellow sterile mycelium	-	-	-	14	-	-	10	15	6	-	-	-	-	-	-	-	-	5	-	-
Average no. of fungi/cm^2	354	357	489	409	605	581	903	817	974	806	309	324	349	398	449	368	658	397	754	444
Total no. of species	18	16	20	18	21	15	22	18	24	17	17	16	17	15	19	16	20	15	20	11

C = Control
T = Test

x Treated with 2% urea; xx Treated with 3% urea

Table 2

Effect of urea on frequency of occurrence of the fungal species on mustard and barley leaves recorded by washed disks technique

	Mustard										Barley									
	Sampling dates																			
	8 Dec		18 Dec		28 Dec		7 Jan		17 Jan		8 Dec		18 Dec		28 Dec		7 Jan		17 Jan	
	C	T	C	T	C	T	C	T	C	T	C	T	C	T	C	T	C	T	C	T
Alternaria alternata /Fr./ Keissler	34	64	56	36	84	40	36	40	56	68	38	34	60	36	64	68	26	28	88	56
A. brassicae /Berk./ Sacc.	-	-	-	-	-	-	-	-	8	-	-	-	-	-	-	-	-	-	-	-
A. humicola Oud.	8	12	10	4	8	12	10	4	20	4	-	-	-	-	-	-	8	4	-	-
Ascochyta sp.	-	-	-	-	-	12	-	-	10	-	-	-	-	-	-	-	-	-	-	-
Aspergillus niger van Tiegh.	-	-	-	-	-	-	-	-	-	-	-	-	-	-	76	-	-	-	4	-
Aureobasidium pullulans /de Bary/ Arnaud	34	16	20	12	-	-	2	8	4	16	-	10	20	16	18	18	4	4	4	28
Bipolaris spicifera /Bain./ Subram.	-	-	10	4	24	12	20	12	4	4	14	4	20	16	20	10	-	-	-	4
B. tetramera /McKinney/ Shoemaker	4	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Cladosporium cladosporioides /Fres./ de Vries	40	80	80	80	96	72	56	76	80	60	40	78	80	64	86	88	68	80	92	88
C. herbarum /Pers./ Link et Fr.	30	52	70	64	20	50	40	70	30	50	10	10	20	18	16	26	26	18	20	10
Curvularia clavata Jain	16	-	-	-	10	-	-	-	-	-	-	-	-	-	12	12	8	-	4	-
C. lunata /Wakker/ Boedijn	-	4	10	4	20	8	10	8	4	8	10	6	8	16	4	8	8	10	-	-
C. pallescens Boedijn	6	4	-	-	-	-	-	-	10	-	-	-	-	-	-	-	-	-	-	-
C. senegalensis /Speg./Subram.	-	-	-	-	-	-	-	-	10	4	-	-	-	-	-	-	-	-	-	-
Drechslera graminea Ito et Kuribayshi	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	4	-	-	4	-
D. Sorokiniana /Sacc./ Subram. et Jain	-	-	-	-	-	-	10	4	8	-	-	-	-	-	-	-	-	-	-	-
Epicoccum nigrum Link.ex Wallr.	-	-	-	-	24	24	8	4	20	-	-	-	-	-	28	20	8	4	12	-
Fusariella indica Roy et B.Rai	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	16	-	-
Fusarium chlamydosporum Wollenw. et Reinking	-	8	-	-	-	28	-	-	-	-	4	-	-	-	-	-	-	-	-	-
Humicola grisea Traaem	-	8	6	-	-	-	-	-	-	-	-	-	-	4	-	-	-	-	-	36
Nigrospora oryzae Petch	-	8	16	4	-	-	-	-	-	-	8	10	20	16	10	12	8	4	4	8
N. sphaerica /Sacc./ Mason	-	-	6	-	-	-	20	20	4	4	10	6	40	26	-	-	18	4	4	12
Penicillium chrysogenum Thom	-	-	-	-	10	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
P. citrinum Thom	-	-	-	-	6	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Periconia minutissima Corda	-	-	-	-	-	-	-	-	4	4	-	-	-	-	-	-	-	8	8	8
Pestalotia sp.	-	-	-	4	-	2	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Pithomyces maydicus /Sacc./ M.B.Ellis	-	-	-	-	-	-	-	-	-	-	-	-	-	-	12	-	-	-	-	-
Stemphylium verruculosum Zimm.	8	-	-	-	-	-	4	4	-	8	-	6	20	-	12	-	-	-	20	-
Trichoderma viride Pers.ex Fr.	-	-	-	-	-	-	16	-	-	-	-	-	-	-	-	-	-	-	-	-
Trichothecium roseum /Pers./ Link ex Fr.	-	-	-	-	44	10	20	4	-	4	-	-	-	-	72	8	8	32	-	-
Black sterile mycelium	14	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	8	-	4	-
Total number of species isolated	10	10	10	9	11	11	13	12	14	12	8	9	9	9	13	11	12	12	13	9

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