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Effect Of Cow Urine On Fungal Spore Germination

H.S.Ravikumar Patil^{*1}, H.K.Makari¹, H.Gurumurthy¹, S.Mukunda², T.R.Prasheeth Kekuda², D.M.Chetan³, H.S.Anil Kumar³ ¹Department of Biotechnology, GM Institute of Technology, Davanagere-577006, Karnataka, (INDIA) ²Department of Microbiology, S.R.N.M.National college of Applied Sciences, Shimoga, Karnataka, (INDIA) ³Department of Biotechnology, NMAM Institute of Technology, Nitte, Karnataka, (INDIA)

Fax: 08192233344

E-mail: patil_varuni@rediffmail.com Received: 8th October, 2007 ; Accepted: 13th October, 2007

ABSTRACT

This study was aimed to investigate antimicrobial activity of cow urine. The cow urine was effective in controlling spore germination. This study revealed the potency of cow which inhibits fungal spore germination. The urine was tested against pathogens *Fusarium oxysporum* NCIM 1008, *Claviceps purpurea* NCIM 1046, *Rhizopus oligosporus* NCIM 1215, *Aspergillus oryzae* NCIM1212 *Penicillium notatum* NCIM 741 *Trichoderma viridae* NCIM 1051, obtained from NCIM, Pune, *Curvularia* spp, *Alternaria helianthi* and *Cladosporium* spp were collected from the infected plants parts and the activity was tested . There was drastic reduction in the percentage of germination (when compared to control). The length of germ tube and the number of germ tubes formed per spores were also reduced.

INTRODUCTION

The cow is considered to be the "Mother of Indian Culture". The economy of India chiefly depends on the agriculture. The agriculture needs the animals such as cow, ox, buffaloes for manure and the ploughing purposes. So, in olden days there was more encouragement for the domestication of cattle. Next to land, cattle were the precious possessions. Most of their daily routines were dependent on cattle for agriculture operations such as farming, ploughing, threshing, harvesting, transporting. Dung is used as manure and fuel. Urine is used in pest control. But their last intention was milk and its product. Milk, curd, ghee, butter were also having medicinal properties. So, in those days cattle were regarded as God and respected by everyone.

Using cow urine along with *Leucas aspera* extract and asafoetida to control the shoot borer in brinjal has been reported and one of the recent inventions relates to new use of abundantly available cow urine distillate as an enhancer of antibiotic action on the target pathogen. The cow urine distillate helps in enhancing the absorption of antibiotics across the cell membrane in animal cells, gram positive and gram negative bacteria. The cow urine distillate enhances the activity of a drug or active molecule and its availability. Similar activities can also be obtained by using the distillate of the urine of cow at $40-50^{\circ}$ C and from the concentrate, which is

KEYWORDS

Cow urine; Antifungal; Spore germination.

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lyophilized and dissolved for further use. The bioavailability of nutrients and enhancement of antibiotic effect is relevant to human, plant as well as animal health and thus composition and methods of invention are also intended to be used in agriculture and veterinary practice

MATERIALS AND METHODS

The cow urine was collected in the early morning from healthy Indian breed cow (Malnad Gidda) in a stainless steel container(copper, brass, earthen, glass or china clay containers may also be used). After the collection of cow urine was subjected to filtration in two steps. First it was filtered twice using Whatman A 1 filter paper to remove suspended particles in urine. Then the filtrate again filtered through membrane filter (Millipore filters). Final filtrate was used for preparing spore germination inhibition assay and to detect percentage of spore germination with respect to different fungal spores.

RESULTS

In spore germination inhibition assay the percentage of germination of fungal spores in presence of cow urine was observed with reference to distilled water as control. There was a drastic reduction in percentage of germination when compared to control. The length of germ tube and number of germ tubes produced per spore was also reduced in six of the tested organisms. But in the case of *Trichoderma viridae* the percentage of sporulation was enhanced. The results are tabulated in the tables.

DISCUSSION

In spore germination inhibition assay the fungal spores were inhibited to germinate. There was drastic reduction in the percentage of germination (when compared to control). The length of germ tube and the number of germ tubes formed per spores were also reduced.

The usage of cow urine must be encouraged in the fields. If our farmers are convinced to use cow urine, no doubt there emerges a new way of domestication of Indian breed cows. It is a 100% eco-friendly method. There are no side effects like decrease in soil fertility,

TABLE	1:	Spore	germina	ation	inhibition	assay-A	lternaria
helianth	ii						

E D	n Distilled v	water	Cow urine				
Spore no.	No. of germ tubes	Germ tube length in microns	Spore no.	No. of germ tubes	Germ tube length in microns		
1	4	7,10,8,11	1	1	30		
2	3	20,10,15	2	1	10		
3	5	12,21,7,8,3	3	1	5		
4	4	12,8,15,6	4	1	7		
5	5	8,10,4,5,2	5	1	20		
6	3	2,10,4	6	1	6		
7	3	10,10,2	7	3	10,3,20		
8	2	40,5	8	1	15		
9	3	25,10,3	9	1	6		
10	4	2,4,10,2	10	2	5,7		
11	1	10	11	1	8		
12	3	2,8,6	12	2	12,5		
13	3	5,8,2	13	1	6		
14	3	8,6,30	14	2	7,10		
15	3	8,4,6	15	3	19,1,5		
16	4	15.8.5.7	16	2	10.5		
17	4	20.12.5.3	17	2	6.10		
18	3	9.3.12	18	1	15		
19	4	12.15.3.4	19	2	3.1		
20	4	11.15.7.4	20	3	6.4.1		
21	3	12.9.21	21	1	10		
22	3	6.27.14	22	1	8		
23	2	31.9	23	1	6		
24	5	7 19 6 3 5	24	3	981		
25	4	3 23 17 9	25	1	20		
26	5	12, 18, 9, 6, 3	26	1	8		
27	3	5 11 16	27	1	4		
28	2	21 19	28	1	12		
29	3	7 13 29	29	1	19		
30	3	5 15 10	30	1	15		
31	5	217533	31	2	68		
32	1	17 10 15 6	32	1	9		
32		10754	33	1	9		
34	3	10,7,5,4 10745	34	3	538		
35	2	10,7,45	35	1	8		
36	4	25 15 15 6	36	1	15		
37	4	10 5 3	30	1	13		
38	1	25 10 15 6	38	1	6		
30	4	25,10,15,0	30	1	0		
39 40	2 4	40,15	40	1	4		
40	4	50,0,20,8 8 10 2	40	1	10.9		
41	5	0,10,2 10	41	ے 1	10,8		
42 42	1	15 20	42	1	0		
43	ے 1	10,572	45	1	/		
44	4	10,3,7,3	44 15	1	ð		
45	3	8,10,5	45	1	0		
46	5	2,9,10	40	1	/		
4/	2	6,8 21.7.7	47	1	8		
48	5	21,7,5	48	1	11		
49 50	1	40	49	1	4		
20	ń	15.7.3	50		1		

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Cow urine

No. of

germ

tubes

Germ

tube

length in

microns

3.2

3,5

2,1

1,3

2.3

3,4

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 TABLE 2 : Spore germination inhibition assay- Aspergillus flavus

 TABLE 3 : Spore germination inhibition assay-Aspergillus oryzae

Spore

no.

9,3

Distilled water Cow urine							Distilled water			
Spore no.	No.of germ	Germ tube length in	Spore no.	No. f germ	Germ tube length in	Sj	Spore	No. of germ	Germ tube	
1	2		1	1 1		1	10.	tubes	length in	
2	1	5	2	1	3				microns	
3	1	10	3	1	2		1	1	8	
4	2	3.7	4	1	2		2	1	6	
5	3	7,4,5	5	1	5		3	2	10,2	
6	1	15	6	1	3		4	2	5,5	
7	1	20	7	1	4		5	2	5,3	
8	2	15,18	8	1	3		6	1	10	
9	1	20	9	1	3		7	2	25,6	
10	2	10,12	10	1	3		8	1	18	
11	1	9	11	1	2		9	1	15	
12	2	7,5	12	1	4		10	1	12	
13	1	12	13	1	2		11	1	3	
14	1	15	14	1	3		11	1	11	
15	1	10	15	1	3		12	1	11	
16	1	7	16	1	3		15	2 1	4,21	
17	1	5	17	1	4		14	1	12	
18	1	8	18	1	2		15	I	3	
19	1	9	19	1	2		16	1	9	
20	3	/,5,8	20	1	4		17	1	8	
21	1	10	21	1	4		18	3	5,3,2	
22	1	11	22	1	2		19	2	3,2	
23	1	13	23	1	2		20	2	10,7	
24	1	22	24	1	2		21	1	1	
25	1	20	25	1	23		22	1	1	
20	1	9	20	1	3		23	1	30	
28	1	12	28	1	4		22 24	1	11	
29	1	17	29	1	2		24 25	1	12	
30	2	21.10	30	1	4		25 26	2	12	
31	1	5	31	1	3		20	2	4,2	
32	1	13	32	1	3		21	2 1	12,5	
33	1	17	33	1	5		28	1	14	
34	1	11	34	1	3		29	1	16	
35	1	8	35	1	2		30	2	12,5	
36	1	16	36	1	4		31	3	1,8,2	
37	1	19	37	1	3		32	2	5,8	
38	2	7,9	38	1	2		33	1	3	
39	1	11	39	1	2		34	1	3	
40	1	21	40	1	3		35	1	6	
41	1	18	41	1	4		36	1	7	
42	1	14	42	1	5		37	1	18	
43	1	23	43	1	3		38	1	8	
44	1	12	44	1	3		39	1	11	
45	1	52	45	1	4		40	1	9	
40 17	1	4	40 17	1	3		70 //1	2	1 4	
4/ 19	1	24 10	4/ /Q	1	2 2		+1 17	∠ 1	1,4	
40 40	1	10	40 40	1	∠ 1		42 12	1	Э 10	
49 50	1 2	11 / 0	49 50	1	4 2		43	1	19	
	L	4,9	50	1	<u>∠</u>		44	2	3,4	
bioaccum	ulation	, biomagni	fications e	etc. Cov	v urine also		45	1	14	
acts as a 1	nitroge	n source f	or plants	It also	consists of		46	1	3	
acto uo u I			- Prunto.		- 51151515 01		47	1	11	

bioaccumulation, biomagnifications etc. Cow urine also acts as a nitrogen source for plants. It also consists of sulphur, copper, phosphate, sodium, potassium, manganese, carbolic acid, calcium, sodium chloride, vitamins which provide favorable microenvironment for the

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oxysporum					viriaae							
Distilled water				Cow urine			istilled w	ater	Cow urine			
Spore no.	No. of germ tubes	Germ tube length in microns	Spore no.	No. of germ tubes	Germ tube length in microns	Spore no.	No. of germ tubes	Germ tube length in microns	Spore no.	No. of germ tubes	Germ tube length in microns	
1	1	11	1	1	04	1	2	3,2	1	1	1	
2	1	15	2	1	02	2	2	5,4	2	1	1	
3	1	21	3	1	03	3	1	4	3	1	1	
4	1	08	4	1	06	4	1	3	4	1	3	
5	1	06	5	1	02	5	2	1,2	5	1	4	
6	1	17	6	1	07	6	3	1,2,4	6	1	1	
7	1	19	7	1	04	7	1	8	7	1	1	
8	l	15	8	l	02	8	1	6	8	1	2	
9	l	09	9	l	10	9	1	10	9	1	3	
10	1	06	10		05	10	1	6	10	1	1	
11	1	26	11	2	06,03	11	1		11	1	1	
12	1	11	12	1	03	12	2	2,6	12	1	1	
15	1	12	13	1	04	13	2	10,11	13	1	1	
14	1	19	14	1	03	14	2	1,2	14	1		
15	1	28	15	1	02	15	2 1	8,5 20	15	1	2	
10	1	28	10	1	01	10	1	20	10	1	5	
17	1	10	17	1	01	17	1	12	17	ے 1	1	
10	1	10	10	1	05	10	1	12	10	1	2 1	
20	1	07	20	1	05	19 20	1	2,3,7,3,2	20	1	1	
20	1	04	20	1	01	20	1	0	20	1	1	
21	1	08	$\frac{21}{22}$	1	03	21	1	9	$\frac{21}{22}$	1	1	
22	1	11	22	1	05	22	2	63	22	1	1	
23	1	13	23	1	07	23	1	13	23	2	23	
25	1	14	25	1	09	25	2	35	25	1	2,5	
26	1	07	26	1	09	25	1	3,5	26	1	2	
27	1	09	27	2	02.03	20	1	7	20	1	3	
28	1	06	28	1	06	28	1	8	28	1	4	
29	1	05	29	1	05	29	1	12	29	1	1	
30	1	04	30	1	07	30	1	10	30	2	1.1	
31	1	12	31	1	06	31	1	13	31	1	1	
32	1	15	32	1	07	32	1	8	32	1	1	
33	1	18	33	1	08	33	3	9,4,9	33	1	1	
34	1	12	34	1	02	34	2	19,11	34	1	1	
35	1	07	35	1	03	35	1	5	35	1	1	
36	1	03	36	1	05	36	1	3	36	1	1	
37	1	02	37	2	03,05	37	1	4	37	1	1	
38	1	06	38	1	09	38	1	5	38	1	1	
39	1	07	39	1	10	39	1	6	39	1	1	
40	1	15	40	1	03	40	2	7,4	40	1	1	
41	2	08,09	41	1	03	41	2	4,5	41	1	1	
42	3	02,05,01	42	1	07	42	2	7,6	42	1	1	
43	2	03,02	43	1	06	43	3	7,4,9	43	1	2	
44	3	02,05,04	44	1	04	44	1	11	44	1	1	
45	2	02,01	45	1	10	45	1	15	45	1	1	
46	2	02,01	46	1	03	46	2	13,2	46	1	3	
47	2	01,02	47	1	03	47	1	6	47	1	1	
48	2	03,04	48	1	03	48	1	3	48	1	1	
49	2	02,03	49	1	02	49	2	5,3	49	1	2	
50	2	04,01	50	1	05	50	1	12	50	1	1	

orvsnorum

 TABLE 4 : Spore germination inhibition assay- Fusarium
 TABLE 5 : Spore germination inhibition assay-Trichoderma

 viridae

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Di	stilled w	ater		Cow uri	ne	
Spore no.	No. of germ tubes	Germ tube length in microns	Spore no.	No. of germ tubes	Germ tube length in microns	
1	1	4	1	2	3,2	
2	2	3,5	2	1	4	
3	1	12	3	1	4	
4	1	3	4	1	3	
5	1	4	5	1	6	
6	1	14	6	1	7	
7	1	8	7	3	4,2,2	
8	2	7,9	8	2	3,2	
9	2	4,6	9	3	2,3,2	
10	1	35	10	2	9,7	
11	1	10	11	1	10	
12	l	12	12	l	4	
13	l	15	13	l	0.5	
14	1	12	14	l	8	
15	1	3	15	l	2	
16	1	15	16	1	2	
1/ 10	1	6 20	1/	1	3 11.0	
18	1	20	18	2	11,9 876	
19	1	5	19	3 1	8,7,0	
20	1	0	20	1	2	
21	1	4	21	1	2	
22	2	3,5	22	1	3	
23	2	4,7	23	1	J 4	
24 25	2	31	24 25	1		
25	1	12	25	1		
20	1	7	20	2	62	
28	2	67	28	2	2,1	
29	1	4	29	$\frac{1}{2}$	1.2	
30	1	14	30	2	3.2	
31	2	20.36	31	1	2	
32	1	25	32	1	5	
33	1	9	33	1	3	
34	1	27	34	2	6,2	
35	1	9	35	1	2	
36	2	2,2	36	1	1	
37	2	3,3	37	1	1	
38	2	10,4	38	1	1	
39	2	12,3	39	1	2	
40	2	5,6	40	1	3	
41	2	18,10	41	1	1	
42	4	15,6,3,4	42	1	3	
43	1	10	43	1	1	
44	1	9	44	1	4	
45	2	3,9	45	1	3	
46	1	6	46	1	1	
47	1	20	47	1	2	
48	1	4	48	1	2	
49	1	5	49	1	3	

TABLE 6: Spore germination inhibition assay-Cladosporium

plants in the soil.

Cow urine may be used in integrated pest management (IPM) along with several plant extracts which enhances the antifungal activity and prevents insects. Cow urine can be used along with the agnihothra ash(which may be regarded as the Homa farming technique). Since both are proved to be antifungal agents the pathogens are successfully controlled.

CONCLUSION

Based on the results obtained, it may be concluded that the cow urine possess activity against both vegetative phase and sporulation in fungi. Thus cow urine is found to be acting on different phases of life cycle of target fungi. Thus the cow urine may be used as an inducer for the production of secondary metabolites in industries and thus the products can be commercialized.

Cow urine reduces colony diameter with remarkable reduction in extent of sporulation. This result suggests that cow urine will be the cheapest and natural fungicide.

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