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PRESERVATION OF MOIST RAGI GRAINS WITH CERTAIN MILD ACIDS*

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An attempt was made to study the preservative ability of two mild acids Viz., propionic and acetic acids at high moisture contents of the grains of *Eleusine coracana*. Acetic acid proved better than propionic acid for the purpose. A 3% concentration of acetic acid was needed for the prevention and multiplication of all the mycoflora associated with the grains except for *Aspergillus fumigatus* Fres. which needed still a high concentration. The viability of the grains was lost by the treatment (9.14 l lit. per ton) with propionic acid whereas they were found viable with the acetic acid treatment even at 5% concentration.

Healthy and disease free seeds are necessary to raise the healthy crop Grains free of storage fungi are required for the consumption of human beings and livestock. The work of several workers (Grewal and Pal, 1965. Mehrotra, 1976, Pandey, 1982) have shown beyond doubt that a substantial amount of mycoflora is associated with stored ragi in India specially in Kumaun hills. A few of the fungal species associated with the stored grains are known producers of mycotoxins chiefly 'aflatoxins' which are hazardous for the seed health as well as for the health of consumers. The most effective method of preventing deterioration of stored grains by micro-organisms is undoubtedly the drying of grains to the level of 10 to 11% moisture content (Christensen and Kaufmann, 1969) and subsequent storage below 20°C. But this is not often easily possible.

The efficacy of propionic acid to prevent mold and bacterial activity on damp grains is now well established in Europiean countries (Hyde and Burrell, 1973) It exerts a biostatic action in dilute aqueous solution and is biocidal at higher concentration (Huitson, 1968). Feeding experiments with animals have also indicated that the acid is harmless at low concentration (Brousch, 1970, Jones *et al.*, 1970, Young *et al.*, 1970, Sauer 1978) The use of propionic and acetic acids have been recommended by Hall *et al.* (1974) for preservation of corn with high moisture content. Even possibility of preservation of moist hay with propionic acid has been worked out by Nash and Easson, (1977) and the results have been quite encouraging. No work has so far been done on this aspect taking ragi grains into consideration. The present study was, therefore, undertaken to determine the efficacy of propionic and acetic acids in preventing the growth of the fungi more commonly associated with stored ragi grains with high moisture content.

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Table 1 : Effect of propionic acid on spore germination on some storage fungi associated with ragi grains (Number per petri-dish)

Fungal Isolates	Control	ppm 500	800	1000	1200	1500	1800	2000
Aspergillus clavatus	40	32	31	25	14	08	00	00
A. chevalieri	38	42	45	28	12	00	00	00
A. fumigatus	46	47	43	50	40	13	08	00
A. ochraceous	30	19	09	06	00	00	00	00
A. niger	48	38	18	04	00	00	00	00
A. candidus	37	22	26	22	08	00	00	00
A. flavipes	48	39	37	20	25	05	00	00
A. flavus	46	28	22	20	20	06	00	00
Penicillium citrinum	42	37	30	27	10	08	00	00
P. oxalicum	28	28	30	26	30	12	00	00
P. cyclopium	30	36	25	19	02	00	00	00
P. islandicum	42	32	33	27	27	15	00	00
Alternaria alternata	27	29	22	12	08	00	00	00
Curvularia lunata	40	32	29	20	06	00	00	00
Drechslera rostrata	38	48	29	12	07	00	00	00
Cladosporium cladosporioides	32	30	26	18	05	00	00	00
Epicoccum purpurascens	40	29	20	17	07	00	00	00
Fusarium semtectum	34	22	18	07	04	00	00	00
Nigrospora oryzae	28	32	22	12	08	00	00	00
Torula graminis	32	35	20	17	04	00	00	00

Table 2 : Effect of acetic acid on spore germination of some storage fungi associated with ragi grains (Number per petri-dish)

Fungal Isolates	Control	ppm 1000	1500	2000	2500	2800	3000	3500
Aspergillus clavatus	40	38	28	22	26	17	00	00
A. chevalieri	38	28	20	32	16	08	00	00
A. fumigatus	37	30	24	27	18	10	06	00
A. ochraceous	30	24	26	22	16	04	00	00
A. niger	48	40	31	32	16	05	00	00
A. candidus	37	34	28	32	21	06	00	00
A. flavipes	48	30	28	20	14	05	00	00
A. flavus	46	29	30	22	22	08	00	00
Penicillium citrinum	42	36	28	25	12	04	00	00
P. oxalicum	28	29	22	27	10	04	00	00
P. cyclopium	30	29	26	17	11	00	00	00
P. islandicum	42	37	36	21	18	11	00	00
Alternaria alternata	27	28	27	24	07	02	00	00
Curvularia lunata	40	22	20	05	00	00	00	00
Cladosporium cladosporioides	32	31	22	16	06	00	00	00
Epicoccum purpurascens	40	38	18	06	00	00	00	00
Fusarium semitectum	34	37	19	12	04	00	00	00
Drechslera restrata	38	37	22	15	10	02	00	00
Nigrospora oryzae	28	21	22	10	06	03	00	00
Torula graminis	32	28	18	12	07	00	00	00

MATERIALS AND METHODS

To assess the effect of propionic and acetic acids on the germination of spores and myceliaparts of storage fungi, a series of experiments were set up following Dwivedi (1978) and Hyde and Burrell (1973). One ml of spore suspension (nearly 30 to 50 spores per ml) of commonly occuring storage fungi was pored in sterilized petri dishes and different concentrations of propionic and acetic acids were added separately to triplicate sets of petri dishes To each of the petridishes 2 ml of cooled, melted Czapek's solution agar medium was added and the dishes were gently swirled to disperse the spores uniformly. Similar experiments without the propionic and acetic acids served as control. All the dishes were incubated at 25±1°C for 5 days and the number of fungal colonies developing in each petri dish was counted.

To see the effect of these acids on the fungal infestation of ragi seeds and percentage of seed germination, different concentrations of propionic and acetic acids were sprayed at the rate of 9.14 lit. per ton (v/w) on the ragi grains following Haitson (1968) and the grains thus treated were stored in polythene bags (0.1 mm thick) at 75% relative humidity (maintained with the help of a saturated solution of sodium chloride in a closed chamber). The moisture content of the grains ranged between 15 and 20 per cent. The grains were sampled out at monthly intervals following James *et al*, (1946) and fungal infestation of each of the samples was determined by duplicate plates method. The germination percentage of the grains at monthly intervals was also recorded.

RESULTS AND DISCUSSION

The rasults of spore germination studies in the presence of propionic and acetic acids have been presented in tables 1 and 2. A perusal of rhe data indicates that propionic acid could inhibit the germination of all the fungi in concentration range of 700-1500ppm except a few. For *Aspergillus clavatus, A. flavus, A flavipes* and *A. fumigatus,* a higer concentration of propionic acid was needed Acetic acid on the other hand was equally effective but needed still a higher concentration. A concentration range of 1200 to 1500 ppm could inhibit all the test fungi except the Aspergilli. However, a concentration range of 3300 to 3500 ppm was the most effective and killed all the fungi including the Aspergilli.

The treatment of stored grains with different concentrations of propionic and acetic acids to test the preservative ability rendered encouraging resuelts (Table 3&4). The mould infestation was prevented when the grains were treated with 0.8% propionic acid except *Aspergillus fumigatus.* This mould. However, could not be controlled even at higher concentration (1.0%) of propionic acid Acetic acid was need in still higher concentration (3.0 to 5.0%) for similar resnlts. It is interesting to note that both the acids completely inhibited the growth of those Aspergilli (*Aspergilluc flavus* and *A. paraciticus*) which are known aflatoxin producers.

Acetic acid has been proved to be better than propionic acid as regards

Table 3 : Effect of propionic acid on the incidence of seed mycoflora and seed germination of radi after different days of storage (RH 75%)

Seed Mycoflora	After 30 days % concentration*					After 60 days % concentration*					After 90 days % concentration*				
	C	a	b	c	d	C	a	b	c	d	C	a	b	c	b
Mucor hiemlis	2	—	—	—	—	—	—	—	—	—	1	—	—	—	—
Rhizopus nigricans	—	—	—	—	—	1	—	—	—	—	2	—	—	—	—
Absidia ramosa	1	6	—	—	—	2	1	—	—	—	—	—	—	—	—
Syncephalastrum racemosum	—	—	—	—	—	1	—	—	—	—	1	—	—	—	—
Aspergillus clavatus	2	—	2	—	—	1	—	—	—	—	2	3	1	—	—
A. chevalieri	—	2	—	1	—	2	—	—	1	—	2	3	1	—	1
A. fumigatus	4	4	3	2	1	3	2	4	4	1	4	3	2	1	2
A. ochraceous	2	—	1	2	2	1	2	—	—	—	2	1	—	—	—
A. niger	2	2	2	1	—	2	1	1	1	—	3	2	1	1	—
A. candidus	1	—	—	2	2	2	—	—	—	1	1	—	—	—	—
A. flavus	2	—	2	1	1	2	3	2	1	1	2	1	1	—	—
A. nidulans	—	1	—	—	—	—	—	—	—	—	2	—	—	1	1
A. flavipes	1	—	1	—	—	1	—	1	—	—	—	—	—	—	—
A. terreus	—	2	—	1	—	—	—	1	—	—	2	—	—	1	—
Penicillium lapidosum	2	—	1	2	1	1	1	—	—	—	—	—	—	—	—
P. implicatum	—	2	—	1	—	1	—	—	—	—	2	1	1	—	—
P. chrysogenum	—	1	—	1	—	2	—	—	—	—	—	—	—	—	—
P. oxalicum	—	1	1	—	—	—	—	—	1	—	1	—	1	—	—
P. cyclopium	2	—	1	1	—	1	1	—	—	—	2	—	1	—	—
Chaetomium globosum	2	1	—	—	—	2	—	—	—	—	1	—	—	—	—
Pithomyces maydicus	1	—	1	—	—	1	—	1	—	—	1	—	—	—	—
Epicoccum purpurascens	2	1	—	—	—	1	1	—	—	—	1	—	—	—	—
Nigrospora oryzae	1	—	1	—	—	1	—	1	—	—	1	—	—	—	—
Cladosporium Cladosporioides	1	—	1	—	—	1	1	—	—	—	1	—	—	—	—
Fusarium semitectum	—	1	—	—	—	1	1	—	—	—	1	—	—	—	—
Torula graminis	2	1	—	—	—	—	—	—	—	—	—	1	—	—	—
Trichoderma viride	1	1	—	—	—	—	1	—	—	—	—	—	—	—	—
Alternaria alternata	4	2	—	3	—	3	1	1	—	1	3	1	—	—	—
Drechslera australiensis	2	1	—	1	—	2	—	—	—	—	1	—	—	—	—
D. rostrata	4	1	—	2	—	4	1	2	—	—	2	—	—	—	—
Black sterile mycelium	1	—	—	—	1	2	—	1	—	—	1	—	—	—	—
White sterile mycelium	1	1	—	—	—	1	—	—	—	—	—	—	—	—	—
Total No. of colonies	43	25	17	21	08	42	16	15	08	04	42	16	09	04	04
Seed germination %	86	94	87	92	88	86	32	32	22	12	82	00	00	00	00

*C = control, a = 0.4%, b = 0.6%, c = 0.8%, d = 0.0%

Table 4 : Effect of acetic acid on the incidence of seed mycoflora and seed germination of ragi after different days of storage (RH 75%)

Seed Mycoflora	After 30 days % concentration*					After 60 days % concentration*					After 90 days % concentration*				
	C	a	b	c	d	C	a	b	c	d	C	a	b	c	b
Mucor hiemlis	2	2	—	—	—	—	—	—	—	—	1	—	—	—	—
Rhizopus nigricans	—	—	—	—	—	2	—	—	—	—	—	—	—	—	—
Absidie ramosa	1	—	—	—	—	1	—	—	—	—	—	—	—	—	—
Syncephalastrum racemosum	—	—	—	—	—	1	—	—	—	—	1	—	—	—	—
Aspergillus clavatus	2	1	—	—	—	1	2	—	—	—	2	2	2	1	—
A. chevalieri	—	1	—	1	—	2	—	1	—	—	2	—	—	—	—
A. fumigatus	4	4	3	2	1	3	1	3	2	1	4	3	2	1	1
A. ochraceous	2	1	—	—	1	1	—	—	—	—	2	—	—	—	—
A. niger	2	2	2	1	—	2	—	1	1	—	3	2	1	—	—
A. candidus	1	—	—	1	1	2	—	—	—	—	1	—	—	—	—
A. flavus	2	2	1	—	—	2	—	1	—	—	2	1	—	—	—
A. nidulans	—	1	2	1	—	—	2	1	—	—	2	1	—	—	—
A. flavipes	1	—	1	—	—	1	—	—	1	—	—	—	—	—	—
A. terreus	—	2	—	—	—	—	1	—	—	—	2	—	—	—	—
Penicillium lapidosum	2	—	2	—	—	1	—	—	—	—	—	—	—	—	—
P. implicatum	—	2	—	—	—	1	2	—	—	—	2	2	—	—	—
P. chrysogenum	—	1	—	—	—	2	1	—	—	—	—	—	—	—	—
P. oxalicum	—	1	1	—	—	—	2	—	—	—	1	—	—	—	—
P. cyclopium	2	1	—	—	—	1	1	—	—	—	2	—	1	—	—
Chaetomium globosum	2	—	1	—	—	2	—	—	—	—	1	—	—	—	—
Pithomyces maydicus	1	—	—	—	—	1	—	—	1	—	1	—	—	—	—
Epicoccum purpurascens	2	1	—	—	—	1	—	—	—	—	1	—	—	—	—
Nigrospora oryzae	1	1	—	—	—	1	—	2	—	—	1	—	—	—	—
Cladosporium Cladosporiodes	1	1	—	—	—	1	—	—	—	—	1	—	—	—	—
Torula graminis	2	1	—	—	—	—	—	—	—	—	—	1	—	—	—
Trichoderma viride	1	—	1	—	—	1	—	—	—	—	—	—	—	—	—
Alternaria alternata	2	1	—	—	—	3	—	—	—	—	3	1	—	—	—
Drechslera australiensis	2	—	—	—	—	2	1	—	—	—	1	—	—	—	—
D. rostrata	4	2	—	2	—	4	1	1	—	—	2	—	—	—	—
Black sterile mycelium	1	—	—	—	—	2	—	—	1	—	1	—	—	—	—
White sterile mycelium	2	—	—	—	—	1	—	—	—	—	—	—	—	—	—
Total No. of colonies	42	28	14	06	03	42	14	10	06	01	39	13	08	02	01
Seed germination %	84	82	87	92	94	86	87	79	92	90	82	87	88	90	93

*C = control, a = 1.0%, b = 2.0%, c = 3.0%, d = 5.0%

the preservation of grains is concerned though it is of course required in higher quantity. Propionic acid has a pungent odour and taste and thus causes discomfort if treatment is given in a closed room and it is slightly corrosive also. Acetic acid on the other hand do not possess these characters. Further the grains were seen to loose their viability and did not germinate when treated with propionic acid and stored for more than two months (Tables 3, 4). The grains treated with acetic acid were found viable and maintained even at higher concentration (5%). Hyds and Burrell (1973) have recommended the use of formic and acetic acids separately or in combination with propionic acid for the preservation of moist grains in storage. Schroeper (1964) has advocated infrared drying and sodium propionate treatment of the grains prior to storage to prevent the field fungal infestation in rough rice. The results reported herein indicate that acetic acid can be successfully used to prevent the deterioration of giains by mould infestation. Similar observation have been made by Owivedi (1978) while studying the fungi associated with the cereal grains.

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RESEARCH NOTES :

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INDUCED HERMOPHRODITISM IN CASTOR

Physical mutagens are found to induce male sterility in castor (Sivaraman, 1973) permitting the production of hybrid seeds simple and easy. Konwar (1960) and Kulkarni *et al.* (1967) reported the appearance of bisexual flowers in a short castor mutant treated with fast neutron at 2.5 x 10^{13} n/cm^{2}.

The present report describes an induced true breeding hermophrodite castor mutant is lated from an M_2 population of gamma irradiated strain of castor, SA 2.

With the object of inducing variability and to isolate mutants with early flowering, high yield and oil content, a project was initiated at the Agricultural College and Research Institute, Coimbatore. Dried seeds of the castor strain SA 2 with 5 0% moisture were subjected to gamma irradiation at 10, 20, 30, 40, 60, 70 and 80 Krad using 100 dry seeds in each dose. The M_1 plants were raised in isolation and observed for plant morphology. In the M_2 generation, an array of mutants with fasciated stem, drooping spike, chlorophyll mutants of different grades, leaf shape, capsule with warty surface, complete pistillate flowers, others with male and female organs combined in the same flower, seed with modified size, shape and seed coat colours were recorded. At 50 Krad, one M_2 plant progeny se-