

# EVOLVING EFFECTIVE PRESERVATION TECHNIQUE FOR DISTILLER'S GRAIN

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*Three experiments were conducted to optimize the physical and chemical or combinations thereof, suitable for preservation of barley distiller's grains. In experiment I barley distiller's grain was subjected to two methods of physical preservation (sun drying and shade drying) and in experiment II barley distiller's grain was subjected to chemical method (propionic acid spray). In experiment III certain natural feed processing byproducts with hydrophobic properties like rice bran, cassava thippi and soya bean hull were mixed with DG in varying proportions (50, 60 and 70 per cent) and then was subjected to sun drying or oven drying (55<sup>E</sup>%C) or propionic acid treatment (2% w/v). The method that would reduce moisture content to a target level (<12%) in shortest duration and also arrest further growth of yeast and mould would be considered appropriate for preservation of DG. Results indicate that sun drying was superior to shade drying and 1½ 3 spread thickness was practically suitable for rapid (12-18 h) drying. Results (experiment III) showed that sun drying of distiller's grain and cassava thippi combination in 30:70 ratio for 12 hours was consistently superior in reduction of pH, lowering of yeast growth and total suppression of mould growth. Based on the results of the three experiments it was concluded that DG can be preserved safely for longer periods singly or in combination with cassava thippi (30:70 ratio) followed by sun drying (33<sup>E</sup>% C, 18-24 hrs, spread thickness 1 – 1 ½ 3 ). For short duration storage (7days) 2 % propionic acid spray appears effective as it retards or / and controls growth of moulds and fungi. Shade drying (29<sup>E</sup>% C) and hot air (oven) drying (55<sup>E</sup>% C) are inferior to sun drying.*

## INTRODUCTION

Distiller's grains (DG) is a valuable by-product of liquor industry, especially when cereal grains are used to produce ethanol. Distiller's grains have a moderate content of protein (%) and high level of crude fiber (%) which make it an attractive ingredient to be used in ruminants feeding (Rasco *et al.*, 1989). In spite of abundant availability, the important factor, that limits its utility as animal feed is its short shelf life (3-7 days) and transportation difficulty (Rendell, 2004). Enhancing the storage life of distiller's grains would increase its use as a feed ingredient, avoid its wastage and make its transpor-

tation easier and cheaper. Therefore this study was undertaken with an objective to test effectiveness of certain physical and chemical methods in preservation of DG and enhance its shelf life.

## MATERIALS AND METHODS

### Experimental material and their procurement

Barley distiller's grain are only locally available and was used in the study; (hitherto it will be referred to as distiller's grains). Distiller's grain was collected from Mohan Breweries and

Distilleries Limited. Natural adsorbents (rice bran, cassava thippi and soya bean hull) were collected from different places of Tamil Nadu.

### Experiment I:

Distiller's grains samples were subjected

for sun drying adopting the following. Plastic trays used to spread one kg of distiller's grain at 0.53 (1.3 cm), 13 (2.5 cm), and 1½ 3 (3.8 cm) thicknes(thickness of spread was measured using

**Table 1**

**Effects of spread thickness duration of drying (sun Vs Shade) on moisture contents in samples of distillers grains (Mean ± SE, experiment I)**

Duration of drying (hours)	Spread thickness					
	Sun drying			Shade drying		
	½"	1"	1½"	½"	1"	1½"
06	50.6 <sup>d</sup> ± 0.56	62.9 <sup>de</sup> ± 0.45	69.6 <sup>def</sup> ± 0.85	68.1 <sup>def</sup> ± 0.36	74.5 <sup>e</sup> ± 1.02	72.7 <sup>e</sup> ± 0.87
12	8.6 <sup>a</sup> ± 1.25	29.9 <sup>b</sup> ± 1.55	54.2 <sup>de</sup> ± 0.99	59.9 <sup>c</sup> ± 1.06	69.8 <sup>def</sup> ± 0.57	69.7 <sup>def</sup> ± 0.68
18	8.5 <sup>a</sup> ± 0.23	9.7 <sup>a</sup> ± 1.56	23.6 <sup>b</sup> ± 2.20	43.0 <sup>d</sup> ± 0.98	51.7 <sup>de</sup> ± 0.66	66.8 ± 1.56
24	8.6 <sup>a</sup> ± 0.47	9.1 <sup>a</sup> ± 0.13	9.5 <sup>a</sup> ± 0.85	17.2 ± 2.56	46.2 ± 1.96	60.9 ± 2.01
30	8.5 <sup>a</sup> ± 0.36	9.2 <sup>a</sup> ± 0.15	9.5 <sup>a</sup> ± 0.96	12.7 <sup>a</sup> ± 1.45	34.6 <sup>c</sup> ± 2.14	55.7 ± 0.98
36	8.5 <sup>a</sup> ± 0.25	9.3 <sup>a</sup> ± 0.45	9.6 <sup>a</sup> ± 0.33	10.6 <sup>a</sup> ± 0.48	12.7 <sup>a</sup> ± 0.55	47.7 <sup>d</sup> ± 2.11
42	8.6 <sup>a</sup> ± 0.34	9.3 <sup>a</sup> ± 0.45	9.4 <sup>a</sup> ± 0.55	10.5 <sup>a</sup> ± 0.22	11.6 <sup>a</sup> ± 0.38	22.1 <sup>b</sup> ± 0.74
48	8.8 <sup>a</sup> ± 0.18	9.3 <sup>a</sup> ± 0.25	9.5 <sup>a</sup> ± 0.51	10.5 <sup>a</sup> ± 0.22	11.6 <sup>a</sup> ± 0.21	13.5 <sup>a</sup> ± 0.52
56	8.5 <sup>a</sup> ± 0.17	9.3 <sup>a</sup> ± 0.16	9.5 <sup>a</sup> ± 0.25	10.4 <sup>a</sup> ± 0.51	11.0 <sup>a</sup> ± 0.23	12.9 <sup>a</sup> ± 0.65

Initial moisture 78.22 ± 2.35

Mean of three observations. Mean bearing different superscripts differ significantly ( $p < 0.05$ ).

measuring scale) in duplicate. The trays were exposed to dry under direct sunlight (6.30 am to 6.30 pm) and samples were drawn out in 6 hourly intervals for moisture estimation. Drying was continued till the target moisture content (<12 per cent) was achieved and time needed for drying to this target level of moisture was recorded. Similarly,

distiller's grain samples were also subjected to shade drying – indoor at room temperature (29°C) in plastic trays (½", 1", and 1½" thickness spread) in duplicate, sampling procedure was similar to sun drying.

### Experiment II:

This experiment was planned to standardize the level of propionic acid (99 % pure) needed to preserve and extend the shelf life of distiller's grains. Eighteen circular piles (radius 15 cm, height 10 cm and exposed surface area 850 cm square) each containing two kilograms of distiller's grain were heaped up on polythene sheets and propionic acid was sprayed in concentrations of 0, 0.5, 1.0, 1.5, 2.0 and 2.5 % (w/v) with the help of sprayer. Three piles were allotted to control and each of the five concentration of propionic acid

**Table 2**

**Effect of propionic acid spray at graded levels on pH, yeast and mould count in acid treated distillers grain samples collected at two days intervals (Mean  $\pm$  SE, experiment 2).**

Propionic acid (%)	Untreated	0.5	1	1.5	2	2.5
<b>pH</b>						
2 <sup>nd</sup> day	5.03 <sup>gh</sup> $\pm$ 0.003	4.48 <sup>c</sup> $\pm$ 0.05	4.38 <sup>cd</sup> $\pm$ 0.01	3.97 <sup>b</sup> $\pm$ 0.01	3.65 <sup>a</sup> $\pm$ 0.02	3.64 <sup>a</sup> $\pm$ 0.003
4 <sup>th</sup> day	7.49 <sup>f</sup> $\pm$ 0.03	5.32 <sup>g</sup> $\pm$ 0.033	5.26 <sup>i</sup> $\pm$ 0.003	4.87 <sup>f</sup> $\pm$ 0.006	4.32 <sup>c</sup> $\pm$ 0.003	4.30 <sup>d</sup> $\pm$ .003
6 <sup>th</sup> day	7.98 <sup>s</sup> $\pm$ 0.02	5.41 <sup>l</sup> $\pm$ 0.03	5.32 <sup>g</sup> $\pm$ 0.01	5.24 <sup>l</sup> $\pm$ 0.003	4.41 <sup>cd</sup> $\pm$ 0.01	4.36 <sup>def</sup> $\pm$ .003
8 <sup>th</sup> day	8.12 <sup>t</sup> $\pm$ 0.04	6.82 <sup>o</sup> $\pm$ 0.01	6.63 <sup>n</sup> $\pm$ 0.00	6.16 <sup>l</sup> $\pm$ 0.08	5.121 <sup>e</sup> $\pm$ 0.10	5.0 <sup>k</sup> $\pm$ 0.01
10 <sup>th</sup> day	8.49 <sup>u</sup> $\pm$ 0.003	7.16 <sup>d</sup> $\pm$ 0.03	6.96 <sup>p</sup> $\pm$ 0.033	6.46 <sup>m</sup> $\pm$ 0.03	6.00 <sup>k</sup> $\pm$ 0.00	5.96 <sup>s</sup> $\pm$ 0.03
<b>Yeast count (10<sup>6</sup> CFU ml<sup>-1</sup>)</b>						
2 <sup>nd</sup> day	80.0 <sup>j</sup> $\pm$ 2.30	40.0 <sup>f</sup> $\pm$ 0.00	36.0 <sup>def</sup> $\pm$ 1.15	24.7 <sup>b</sup> $\pm$ 0.88	14.3 <sup>a</sup> $\pm$ 0.88	14.3 <sup>a</sup> $\pm$ 0.33
4 <sup>th</sup> day	125 <sup>n</sup> $\pm$ 2.88	52.3 <sup>h</sup> $\pm$ 1.45	47.3 <sup>g</sup> $\pm$ 2.18	34.0 <sup>cde</sup> $\pm$ 1.00	24.0 <sup>b</sup> $\pm$ 1.52	22.3 <sup>b</sup> $\pm$ 1.20
6 <sup>th</sup> day	147.0 <sup>p</sup> $\pm$ 2.64	59.0 <sup>i</sup> $\pm$ 1.00	51.3 <sup>gh</sup> $\pm$ 0.66	38.3 <sup>cd</sup> $\pm$ 0.88	32.7 <sup>cd</sup> $\pm$ 1.20	30.0 <sup>c</sup> $\pm$ 0.00
8 <sup>th</sup> day	160.7 <sup>q</sup> $\pm$ 2.18	140.0 <sup>o</sup> $\pm$ 0.88	126.0 <sup>n</sup> $\pm$ 2.08	120.0 <sup>m</sup> $\pm$ 1.73	93.7 <sup>kl</sup> $\pm$ 2.02	92.0 <sup>k</sup> $\pm$ 3.60
10 <sup>th</sup> day	169.3 <sup>r</sup> $\pm$ 1.33	147.3 <sup>p</sup> $\pm$ 2.84	136.7 <sup>o</sup> $\pm$ 0.88	127.0 <sup>n</sup> $\pm$ 1.52	98.0 <sup>l</sup> $\pm$ 1.53	95.7 <sup>kl</sup> $\pm$ 2.33
<b>Mould count (10<sup>6</sup> CFU ml<sup>-1</sup>)</b>						
2 <sup>nd</sup> day	18.0 <sup>i</sup> $\pm$ 2.30	12.3 <sup>fg</sup> $\pm$ 1.45	11.7 <sup>def</sup> $\pm$ 0.33	9.3 <sup>bc</sup> $\pm$ 0.33	7.3 <sup>ab</sup> $\pm$ 0.33	5.3 <sup>a</sup> $\pm$ 0.33
4 <sup>th</sup> day	26.7 <sup>m</sup> $\pm$ 1.20	20.7 <sup>j</sup> $\pm$ 0.66	16.3 <sup>g</sup> $\pm$ 0.88	12.3 <sup>fg</sup> $\pm$ 0.88	10.0 <sup>cde</sup> $\pm$ 0.57	9.7 <sup>cd</sup> $\pm$ 0.33
6 <sup>th</sup> day	37.7 <sup>p</sup> $\pm$ 0.33	25.3 <sup>lm</sup> $\pm$ 0.33	22.0 <sup>h</sup> $\pm$ 0.57	17.7 <sup>i</sup> $\pm$ 0.88	13.0 <sup>fg</sup> $\pm$ 0.57	12.0 <sup>efg</sup> $\pm$ 0.57
8 <sup>th</sup> day	45.0 <sup>q</sup> $\pm$ 0.57	29.0 <sup>m</sup> $\pm$ 0.57	26.0 <sup>m</sup> $\pm$ 0.57	23.3 <sup>kl</sup> $\pm$ 1.20	16.0 <sup>hi</sup> $\pm$ 0.57	13.0 <sup>fg</sup> $\pm$ 0.00
10 <sup>th</sup> day	49.7 <sup>t</sup> $\pm$ 0.88	33.3 <sup>n</sup> $\pm$ 0.66	29.0 <sup>n</sup> $\pm$ 0.57	24.7 <sup>lm</sup> $\pm$ 0.33	14.0 <sup>eh</sup> $\pm$ 0.57	12.7 <sup>fg</sup> $\pm$ 1.20

used. The contents of each pile(s) were mixed well after spraying and kept indoor at room temperature (29<sup>±</sup>°C). Samples were collected on alternate days up to 10 days for estimation of pH (Wilson and Wilkins, 1972) and enumeration of yeast and mould Jenkins (1992)

### Experiment III:

In this experiment distiller's grain was mixed in different proportions (50, 60 and 70 per cent) with each of the three natural adsorbents (rice bran, cassava thippi and soyabean hull). The combinations used were as follows: Distiller's grain + rice bran (DG +RB) – 50:50, 40:60, 30:70; Distiller's grain + cassava thippi (DG + CT) – 50:50, 40:60, 30:70; Distiller's grain + soyabean hull (DG +SH) - 50:50, 40:60, 30:70. The distiller's grains and adsorbent combination lots acid spray (2 % w/v) treatments in the same way as mentioned earlier.

Estimation of pH, yeast and mould count of the samples was carried out as described in experiment II. The design of all the experiments in this study was completely randomized design (CRD). Data were analysed as per procedure of statistical analysis system (SAS/SPPSS, 1999, version 10.0 for windows).

## RESULTS AND DISCUSSION

### Experiment I:

The per cent moisture content in samples of DG collected at 6 hourly intervals during drying is presented in Table 1. In the sun drying process with ½" spread thickness the target moisture content (<12%) was achieved within the first 12 hours of drying, while 18 and 24 h respectively (drying) duration was needed for 1" and 1½" spread thickness to attain the target moisture content (<12

%). In the shade drying process, moisture content below 12 per cent was achieved at 36 and 42 h of drying with spread thickness of ½” and 1” . While target moisture content was not achieved even at 54 h shade drying with 1½” spread thickness. Shade drying has been reported to be a slower process of drying (Bhosale and Arya, 2004). These findings indicate that sun drying was better than shade drying especially with ½” spread thickness.

### Experiment II:

The pH and yeast and mould count in DG samples collected periodically is presented in Table 2. Irrespective of the groups and level of propionic acid an increase in the pH was observed as the days increased. The rate of increase of pH was rapid in control than acid sprayed DG samples. Secondary fermentation has been reported to cause an increase in pH, yeast and mould growth during ensilage of high moisture ear corn after 138 days of ensilage (Sebastian *et al.*, 1996). A significant ( $p < 0.05$ ) reduction in pH was attained at 2 and 2.50 per cent propionic acid treatment at the second day and pH values in these two groups were statistically comparable ( $p < 0.05$ ). Similar trend was noticed with respect to number of yeast and mould as the post treatment days advanced. On the second day the yeast count was significantly ( $p < 0.05$ ) lowest in 2 and 2.50 per cent propionic acid treated groups as compared to control and other treated groups while mould count was significantly ( $p < 0.05$ ) lowest in 2.50 per cent acid treated groups as compared to remaining control and treated groups except 2 per cent acid treated group which has comparable mould count. Propionic acid spray inhibited mould growth and at 2 and 2.5% concentration it was ineffective. Although, Muller and Thaler (1981) reported that propionic acid addition up to 1 per cent level to high moisture corn (19-40 per cent moisture) inhibited inoculated fungi during storage for 6 months at 20°C and these observations are in agreement with the findings of this experiment as number of yeast

and mould were significantly lower in acid treated than in untreated DG samples. Earlier reports also indicate that in animal feed stuffs the yeast and mould count ranges between  $10^1$  to  $10^4$  CFU per gram of feed (Wojdat, 2005). In this study the treated distiller’s grain were spread in trays and were kept uncovered state at room temperature (29°C) which might have contributed to yeast and mould growth with advancing days as compared to second day count. Moreover distiller’s grain by itself is a source of (residual) yeast cells as it is a byproduct of ethanol extraction (Mustafa *et al.*, 2000) process.

### Experiment III:

The moisture content in mixture of distiller’s grain and adsorbents is presented in Table 3. The addition of adsorbents decreased the initial moisture content of mixtures as compared to DG alone (control) and the reduction in moisture content was continuous as the levels of adsorbents increased. All combinations of distiller’s grains with adsorbents were capable of reducing the moisture contents below target level (<12%) within 12 hours of sun drying except DG: thippi (40:60 and 30:70); whereas longer duration is needed to achieve target moisture level (<12%) in hot air (oven) drying method (table 3). Rice bran is reported to act as an adsorbent when used in preparation of nutrient blocks (Sausoucy *et al.*, 1988). Bagavathi (1994) also reported a reduction in moisture of mango peel, when cassava thippi was added to it. The pH in mixture of distiller’s grain and adsorbents is presented in Table 4. In this study it was observed that wherever cassava thippi was mixed with DG as adsorbent and dried either in sun or hot air (oven) or treated with 2 per cent propionic acid the pH value was between 4 to 5 and was significantly lower than mixtures of DG and adsorbents tested in this study. Distiller’s grain and cassava thippi mixtures in all proportions subjected to propionic acid treatment had the significantly ( $p < 0.05$ ) lower pH ( $3.91 \pm 0.02$  to  $4.05 \pm 0.02$ ) as compared to other mixtures (DG + RB or DG +SH). The differences between 50;50

**Table 3**

**Effect of propionic acid spray and subsequent drying methods (sun or hot air oven, 55 °C) on moisture content in mixtures of distillers grain and natural adsorbents at six hourly intervals (Mean ± SE, experiment 3)**

Proportions		Initial moisture	Sun drying (33° C)				Hot air (oven) drying (55° C)			
			Duration in hours (post mixing)				Duration in hours (post mixing)			
			6	12	18	24	6	12	18	24
Distiller's grain (100%) control		71.71 ± 0.43	60.13 ± 1.30	36.23 ± 2.29	10.61 ± 0.18	10.50 ± 0.04	60.90 ± 0.14	48.26 ± 0.11	36.39 ± 0.10	27.86 ± 0.47
Distiller's grain + Rice bran	50:50	38.56 ± 10.22	19.20 ± 0.66	11.58 ± 0.12	11.50 ± 0.24	10.80 ± 0.14	30.21 ± 0.17	24.12 ± 0.24	18.01 ± 0.03	13.29 ± 0.22
	40:60	30.63 ± 0.35	14.97 ± 0.95	10.75 ± 0.70	10.66 ± 0.16	10.52 ± 0.26	22.52 ± 0.18	16.51 ± 0.10	11.65 ± 0.24	11.03 ± 0.19
	30:70	25.14 ± 0.39	10.77 ± 0.75	10.23 ± 0.60	10.56 ± 0.35	10.43 ± 0.14	18.00 ± 0.10	12.12 ± 0.18	11.29 ± 0.14	10.85 ± 0.37
Distiller's grain + Cassava thippi	50:50	40.24 ± 0.94	20.68 ± 0.90	11.35 ± 0.39	11.00 ± 0.74	10.63 ± 0.17	32.20 ± 0.25	26.06 ± 0.18	19.59 ± 0.05	14.00 ± 0.14
	40:60	33.26 ± 0.83	17.99 ± 0.55	12.23 ± 0.35	11.68 ± 0.08	10.91 ± 0.17	25.17 ± 0.29	19.27 ± 0.13	12.09 ± 0.33	11.01 ± 0.26
	30:70	29.40 ± 1.42	15.00 ± 0.54	12.07 ± 0.50	11.26 ± 0.34	10.37 ± 0.07	21.05 ± 0.30	15.36 ± 0.11	11.23 ± 0.09	10.76 ± 0.25
Distiller's grain + soy bean hulls	50:50	43.49 ± 0.10	20.08 ± 0.83	11.13 ± 0.16	10.95 ± 0.30	10.64 ± 0.24	43.49 ± 0.22	35.31 ± 0.12	29.45 ± 0.21	20.89 ± 0.36
	40:60	34.67 ± 0.48	17.21 ± 1.00	10.59 ± 0.67	10.50 ± 0.37	10.64 ± 0.12	34.67 ± 0.36	26.51 ± 0.32	20.26 ± 0.17	14.73 ± 0.24
	30:70	28.55 ± 0.45	12.49 ± 0.36	11.26 ± 0.17	10.99 ± 0.16	10.52 ± 0.01	28.55 ± 0.21	20.11 ± 0.18	13.69 ± 0.26	11.22 ± 0.73

Mean of three samples  
Spread layer thickness 1½"

and 30:70 (DG:CT) were statistically significant ( $p < 0.05$ ) while non significant between 50:50 and 40:60 ratios. The pH of sun dried Distiller's grain cassava thippi mixture in 30:70 proportion also was significantly ( $p < 0.05$ ) low ( $3.99 \pm 0.003$ ). Whenever, cassava thippi is used as adsorbent with DG the pH value reduced because cassava thippi as such is a low (4-7) pH material. Yeast

count ( $\times 10^6$  CFU ml<sup>-1</sup>) in distiller's grain and adsorbent mixtures, is presented in Table 4. In all combinations of distiller's grain with adsorbents all applied treatments were able to reduce yeast count significantly ( $p < 0.05$ ) as compared to distiller's grain alone. As the level of adsorbents increased in mixtures irrespective of adsorbent used there was a significant ( $p < 0.05$ ) decrease in yeast count in sun dried lots. In oven dried lots the number of yeasts in

CT groups decreased with increasing proportions of CT in mixtures A significantly ( $p < 0.05$ ) low yeast count was recorded in distiller's grain to rice bran (30:70) and distiller's grain to cassava thippi (30:70) lots subjected to sun drying and also in distiller's grain to rice bran (50:50) lots only. However DG plus CT lots treated with acids having higher proportions of CT (60 and 70) recorded numerically higher number of yeasts though they were statistically comparable with one another (Table 4). Moulds count ( $\times 10^6$  CFU ml<sup>-1</sup>) in distiller's grain adsorbent mixtures is presented in Table 4. Similar to yeast count studies, all distiller's grain adsorbent mixtures dried in sun have significantly ( $p < 0.05$ ) lower or comparable mould count as the level of adsorbents increased irrespective of the adsorbents added; No mould growth was detected in sun dried distiller's grain cassava thippi mixtures (40:60 and 30:70) and distiller's grain to rice bran (30:70) mixture. Oven drying of mixtures (DG + adsorbents) has higher or comparable number of yeast cells when compared with sun dried lots having similar adsorbents and levels. Moulds were absent in oven dried lots having RB at higher (70) proportion. In propionic acid treatment no mould growth was detected in distiller's grain to cassava thippi (40:60 and 30:70) mixtures. The sun dried distiller's grain cassava thippi mixture (30:70) was found consistently superior in reduction of pH and yeast growth and evincing no mould growth. Though distiller's grain is a source of residual yeast (Mustafa *et al.*, 2000) their numbers were consistently reduced by drying methods irrespective of adsorbents proportions used in the study. Mould growth was not detected in sun dried distiller's grain, cassava thippi (40:60, 30:70) and in other adsorbents and DG combinations also mould count was well below  $1.33 \times 10^6$  CFU ml<sup>-1</sup> after sun drying. The rapid reduction of moisture to the target level (<12 %), possibly would not have been conducive for the mould growth. The longer duration needed to achieve the target moisture level in oven drying process proved conducive for mould growth. Bhat *et al.* (1998) reported that in sorghum

grain, harvested after rains and heaped in piles, the high moisture content was conducive for mould growth.

Based on the findings of three experiments conducted in this study it can be concluded that the distillers grain can be preserved safely for longer periods singly or in combination with cassava thippi (30:70 ratio) followed by sun drying (33°C for 18 – 24 h, spread thickness 1 to 1½") for short duration storage (7 days) 2% propionic acid spray appears to be effective as it retards or/ and controls growth of moulds and fungi.

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**Table 4**  
**pH, Yeast and Mould count ( $10^6$  CFUml<sup>-1</sup>) in distiller's grain adsorbent mixtures**  
**subjected to sun and oven drying ( $1\frac{1}{2}$ " thickness spread) and 2 per cent propionic**  
**acid treatment (Mean  $\pm$  SE, experiment 3)**

	Treatment	Sun	Oven	Propionic acid
<b>pH</b>	DG+RB 50:50	6.64 <sup>f</sup> $\pm$ 0.04	6.16 <sup>o</sup> $\pm$ 0.03	5.47 <sup>g</sup> $\pm$ 0.01
	DG+RB 40:60	6.26 <sup>ps</sup> $\pm$ 0.02	6.16 <sup>o</sup> $\pm$ 0.01	5.51 <sup>kl</sup> $\pm$ 0.01
	DG+RB 30:70	6.21 <sup>op</sup> $\pm$ 0.01	6.28 <sup>q</sup> $\pm$ 0.02	5.59 <sup>l</sup> $\pm$ 0.01
	DG+CT 50:50	4.84 <sup>g</sup> $\pm$ 0.02	4.24 <sup>ae</sup> $\pm$ 0.06	3.91 <sup>a</sup> $\pm$ 0.02
	DG+CT 40:60	4.38 <sup>f</sup> $\pm$ 0.01	4.22 <sup>ae</sup> $\pm$ 0.04	3.96 <sup>ab</sup> $\pm$ 0.01
	DG+CT 30:70	3.99 <sup>b</sup> $\pm$ 0.00	4.18 <sup>cd</sup> $\pm$ 0.01	4.05 <sup>bc</sup> $\pm$ 0.02
	DG+SH 50:50	6.22 <sup>op</sup> $\pm$ 0.04	5.33 <sup>h</sup> $\pm$ 0.01	5.59 <sup>lm</sup> $\pm$ 0.01
	DG+SH 40:60	6.07 <sup>n</sup> $\pm$ 0.04	5.48 <sup>ik</sup> $\pm$ 0.02	5.62 <sup>m</sup> $\pm$ 0.01
	DG+SH 30:70	5.77 <sup>n</sup> $\pm$ 0.01	5.35 <sup>lm</sup> $\pm$ 0.01	5.64 <sup>m</sup> $\pm$ 0.01
	DG -100	6.19 <sup>op</sup> $\pm$ 0.01	6.16 <sup>o</sup> $\pm$ 0.03	4.31 <sup>del</sup> $\pm$ 0.01
<b>Yeast count (<math>10^6</math> CFU ml<sup>-1</sup>)</b>	DG+RB 50:50	17.0 <sup>l</sup> $\pm$ 0.57	20.0 <sup>k</sup> $\pm$ 0.57	5.0 <sup>ab</sup> $\pm$ 0.57
	DG+RB 40:60	8.3 <sup>el</sup> $\pm$ 0.33	11.0 <sup>g</sup> $\pm$ 0.57	7.0 <sup>del</sup> $\pm$ 0.57
	DG+RB 30:70	5.0 <sup>ab</sup> $\pm$ 0.57	7.7 <sup>del</sup> $\pm$ 0.33	14.7 <sup>i</sup> $\pm$ 0.33
	DG+CT 50:50	11.0 <sup>g</sup> $\pm$ 0.57	13.7 <sup>h</sup> $\pm$ 0.33	4.7 <sup>a</sup> $\pm$ 0.33
	DG+CT 40:60	6.3 <sup>bcd</sup> $\pm$ 0.33	8.7 <sup>el</sup> $\pm$ 0.66	5.7 <sup>abc</sup> $\pm$ 0.33
	DG+CT 30:70	4.7 <sup>a</sup> $\pm$ 0.33	7.7 <sup>del</sup> $\pm$ 0.33	6.0 <sup>abc</sup> $\pm$ 1.00
	DG+SH 50:50	19.3 <sup>k</sup> $\pm$ 0.33	23.3 <sup>i</sup> $\pm$ 0.66	14.0 <sup>hi</sup> $\pm$ 0.57
	DG+SH 40:60	10.3 <sup>g</sup> $\pm$ 0.33	12.7 <sup>h</sup> $\pm$ 0.33	16.7 <sup>j</sup> $\pm$ 0.66
	DG+SH 30:70	6.33 <sup>bcd</sup> $\pm$ 0.33	10.00 <sup>g</sup> $\pm$ 0.00	18.66 <sup>k</sup> $\pm$ 0.33
	DG -100	23.66 <sup>l</sup> $\pm$ 0.33	37.66 <sup>m</sup> $\pm$ 1.45	24.33 <sup>l</sup> $\pm$ 0.33
<b>Mould count (<math>10^6</math> CFUml<sup>-1</sup>)</b>	DG+RB 50:50	0.33 <sup>ab</sup> $\pm$ 0.33	0.60 <sup>abc</sup> $\pm$ 0.00	0.66 <sup>bc</sup> $\pm$ 0.00
	DG+RB 40:60	0.33 <sup>ab</sup> $\pm$ 0.33	0.33 <sup>ab</sup> $\pm$ 0.33	0.33 <sup>ab</sup> $\pm$ 0.33
	DG+RB 30:70	0.00 <sup>a</sup> $\pm$ 0.00	0.00 <sup>a</sup> $\pm$ 0.00	0.33 <sup>ab</sup> $\pm$ 0.33
	DG+CT 50:50	0.33 <sup>ab</sup> $\pm$ 0.33	0.66 <sup>abc</sup> $\pm$ 0.66	0.33 <sup>ab</sup> $\pm$ 0.33
	DG+CT 40:60	0.00 <sup>a</sup> $\pm$ 0.00	0.66 <sup>abc</sup> $\pm$ 0.33	0.00 <sup>a</sup> $\pm$ 0.00
	DG+CT 30:70	0.00 <sup>a</sup> $\pm$ 0.00	1.33 <sup>c</sup> $\pm$ 0.33	0.00 <sup>a</sup> $\pm$ 0.00
	DG+SH 50:50	1.33 <sup>c</sup> $\pm$ 0.00	1.33 <sup>c</sup> $\pm$ 0.33	0.66 <sup>abc</sup> $\pm$ 0.33
	DG+SH 40:60	1.33 <sup>c</sup> $\pm$ 0.33	2.33 <sup>d</sup> $\pm$ 0.33	1.00 <sup>bc</sup> $\pm$ 0.00
	DG+SH 30:70	1.00 <sup>bc</sup> $\pm$ 0.00	3.00 <sup>de</sup> $\pm$ 0.57	1.00 <sup>bc</sup> $\pm$ 0.00
	DG -100	3.66 <sup>e</sup> $\pm$ 0.33	5.00 <sup>f</sup> $\pm$ 0.57	3.00 <sup>de</sup> $\pm$ 0.57

Mean of three observations. Mean bearing different superscripts differ significantly ( $p < 0.05$ ).

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