# EFFECT OF HEAT TREATMENT ON QUALITY FACTORS AND MYCOFLORA OF SORGHUM

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## ABSTRACT

The potential of using high temperature as a method of preserving sorghum grain quality was investigated. Sorghum grain of different water contents (12 to 18%) were treated at 60, 70, 80 and 90°C for periods of 4, 8 and 12 minutes. Both the germination, free fatty acid content and fungal contamination of the grain was influenced by heat treatment. In general, at 60 and 70°C germination was unaffected or slightly stimulated while at higher temperatures and water contents, significantly reduced, when compared to untreated control grain. The percentage fungal contamination of grain was reduced from 90 to about 5% by heat treatment. However, some fungal species, particularly Eurotium spp., Aspergillus niger and Penicillium spp. could still be isolated from sorghum grain treated at 80-90°C for up to 12 minutes. Subsequently, the dry matter loss, free fatty acid value and percentage germination of both untreated and heat-treated sorghum grain were compared during storage for up to 28 days at the same initial water contents at 25°C. At 17 and 18% w.c. the rate of deterioration in quality was less than that of untreated control grain.

## INTRODUCTION

Sorghum bicolor is an important cereal crop in India and under the prevailing environmental conditions particularly prone to both insect and fungal deterioration. This results in decreased quality particularly of free fatty acids, germination and dry matter loss and can result in the presence of mycotoxins produced by fungal infection, particularly of Aspergillus flavus, Fusarium and Penicillium spp.

The dominant fungi isolated from harvested sorghum grain includes Alternaria alternata, Aspergillus flavus, A.niger, Eurotium spp., Fusarium and Curvularia species (Christensen, 1971; Williams and Rao, 1981; Fahim et al., 1982) Such grain moulds have been shown to result in between 10 and 25% yield losses (Utikar and Shinde, 1985). However, very little information is available on the effect of post-harvest fungal spoilage on quality loss of stored sorghum grain. Futhermore, methods to enable prevention of such spoilage and preserve grain quality for extended time periods would be advantageous, particularly under tropical temperature and humidity conditions.

This study was carried out to determine (1) the effect of heat treatment on the germination, free fatty acid content and fungal contamination of sorghum grain and (2) the potential for heat-treatment to reduce dry matter losses and conserve quality parameters during subsequent storage.

## MATERIALS AND METHODS

### Heat-treatment

Three replicate small (10g) and large (250g) subsamples of sorghum grain (cv. M35-1) were modified to between 12 and 18% water content (w.c., wet weight basis) and sealed in aluminium envelopes. The grain was heat-treated at 60, 70, 80 and 90°C for 4, 8 and 12 minutes.

### Germination tests

Three replicate samples of 25 seeds per heat-treatment were placed on moist absorbent paper, rolled up and placed in upright plastic cylinders on a water tray as recommended by the International Seed Testing Association (ISTA) (ISTA, 1976). The seeds were incubated in a controlled environment chamber at 25°C for 8 days when the percentage germination was assessed.

### Fungal assessment

Fungi present on heat-treated sorghum grain was determined by plating a total of 100 grains per replicate and treatment onto 2% malt extract agar (50 grains) and 2% malt 10% salt agar (50 grains) plates representing water activities of about 0.995 and 0.95 respectively. Grains were also surface-sterilized in 5% sodium hypochlorite prior to plating onto the same agars. The sorghum grain was incubated at 25°C for 7-8 days before determining the genera and where possible species present. For comparison, untreated control sorghum grain was similarly plated out. The mean percentage grain colonised by different fungi in each treatment was determined and compared with untreated control grain.

### Free fat acidity values

The free fatty acid (FFA) values of heat-treated sorghum grain was determined using the American Association of Cereal Chemists (AACC) rapid method (AACC, 1983) and the results are given as Fat Acidity Values (FFA) calculated from the volumes of KOH required to neutralize the fatty acids in the sorghum samples.

### Storage studies

Sorghum grain (250 g subsamples) previously heat-treated at 60, 70 and 80°C for 12 min was readjusted as necessary to 13, 15, 17 and 18% w.c. by the addition of small amounts of water and the water content subsequently checked. Ten g subsamples of each treatment were then placed in large plastic sandwich boxes together with salt solutions of the same equilibrium relative humidity (ERH, 65, 75, 80 and 90%) and enclosed in plastic bags. All treatments were stored at 25°C for a total of 28 days. Every seven days 7 replicate subsamples per treatment were removed. Three replicates were dried at 130°C for 18 hrs and weighed on a four decimal point balance to determine dry matter and the other samples used for assessment of germination capacity and FFA levels. The dry matter loss was determined by comparison with the initial dry matter in subsamples prior to start of the experiment. Experiments were similarly carried out with untreated sorghum grain at the same water contents to compare the changes in quality between heat-treated and untreated stored sorghum grain.

## RESULTS AND DISCUSSION

### Effects of heat treatment on germination, FFA and fungal contamination

The effect of the different treatments on germinative capacity is shown in Table 1. The quantity of grain treated, the water content, temperature and time of exposure affected germination of the sorghum grain. In general, high temperature treatment stimulated germination when compared to the untreated control grain. The maximum germination (93%) was obtained with grain of 14% w.c. exposed to 70oC for 4 minutes. At 80°C and high water contents (14-18% w.c.) germination was significantly affected, regardless of exposure time. These results show that slightly different results may be obtained depending in the grain bulk and the time of exposure. For good heat penetration larger bulks of grain would require longer periods of exposure. Previous work with wheat has suggested that grain with 12% w.c. could be treated at up to 75°C for 15 min with no effect on germination (Ghaly and van der Touw, 1982). However, germination was significantly affected when 14% w.c. wheat was treated at 70°C for up to 15 min. Hard and soft varieties of wheat have also been found to differ in susceptibility to heat treatment (Ghaly and Taylor, 1981). However, these studies did not consider effects of heat treatment on grain fungi.

The most common fungal genera isolated from the sorghum grain were Aspergillus, Penicillium, Eurotium, Cladosporium and Alternaria species. Other fungi found included species of Rhizopus, Epicoccum, and Acremonium. The fungi isolated in this study are similar to that found previously on sorghum (Niles, 1976; Fahim et al, 1982). The isolation of fungi from surface-sterilized sorghum was markedly reduced suggesting that most fungi were present on the grain surface as contaminants. Isolation of Aspergillus niger, Eurotium spp. (A.glaucus group) and Penicillium spp. were all affected by the temperature of treatment and the time of exposure (Table 2). A.niger was completely eliminated by 80 C and a 4 min exposure while a small precentage of sorghum grain ( < 10%) still carried Eurotium spp. and Penicillium spp. at 90°C and all three exposure times. Othe fungi such as A.candidus, A.flavus and Cladosporium spp. were eliminated by 70 C and a 12 minute exposure period. Conidia of A.niger and Eurotium rubrum have been found to be sensitive to about 62 and 57°C respectively (Baggerman and Samson, 1988). However, this may be influenced by water availability and the time of exposure. The isolation of both Eurotium spp. and Penicillium spp. from small quantities of heat-treated sorghum at 80 and 90°C suggests that heat penetration of grain may require longer periods of exposure to eliminate fungi. However, this needs to balanced against effects on germination and other quality criteria. These results show that a nucleus of grain contaminated with fungi will remain after heat treatment at these temperatures with the potential to initiate spoilage under suitable water availability and temperature conditions.

The FFA values for untreated sorghum grain was about 13-15. Regardless of water content or heat-treatment the FFA values of sorghum remained between 11 and 15 suggesting little effect on this quality criteria.

Table I. The effect of different heat treatments on germination of 10 g and 250 g sorghum cv.M35-1 at 12, 14 and 16% and 13, 15, 17 and 18% water content respectively. Data are means of three replicates of 25 seeds

10 g heat-treated samples

| Temperature (oC) | 60 | | | 70 | | | 80 | | |
|---|---|---|---|---|---|---|---|---|---|
| Time of exposure (min) | 4 | 8 | 12 | 4 | 8 | 12 | 4 | 8 | 12 |
| Water content (%) | | | | | | | | | |
| 12 | 92 | 89 | 84 | 93 | 87 | 87 | 88 | 52 | 44 |
| 14 | 90 | 84 | 77 | 65 | 73 | 60 | 37 | 0 | 0 |
| 16 | 67 | 65 | 60 | 68 | 67 | 69 | 12 | 0 | 0 |

LSD = 14.6 ($P < 0.05$)

250 g heat-treated samples

| Temperature (oC) | 60 | 70 | 80 |
|---|---|---|---|
| Time of exposure (min) | 12 | 12 | 12 |
| Water content (%) | | | |
| 13 | 70 | 76 | 38 |
| 15 | 64 | 60 | 15 |
| 17 | 84 | 78 | 14 |
| 18 | 80 | 65 | 12 |

LSD = 12.6
Control germination = 78%

Table II. The effect of different heat treatments on dominant fungal contaminants of sorghum grain cv. M35-1 at 12% water content.

| | Mean percentage (%) colonisation of grain | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Temperature ($^{o}C$) | 60 | | | 70 | | | 80 | | | 90 | | |
| Time of exposure (min) | 4 | 8 | 12 | 4 | 8 | 12 | 4 | 8 | 12 | 4 | 8 | 12 |
| Eurotium spp.* | 98 | 94 | 82 | 96 | 40 | 80 | 64 | 46 | 30 | 4 | 8 | 6 |
| Aspergillus candidus | 12 | 4 | 2 | 0 | 2 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| A.flavus* | 0 | 2 | 2 | 4 | 2 | 4 | 2 | 0 | 0 | 0 | 0 | 0 |
| A.niger | 24 | 44 | 8 | 38 | 34 | 36 | 26 | 0 | 0 | 0 | 0 | 0 |
| Penicillium spp. | 12 | 16 | 16 | 14 | 20 | 16 | 24 | 10 | 0 | 4 | 4 | 4 |

*, assessed on 2% malt 10% salt agar

Storage studies of heat-treated and untreated sorghum grain

Germination of untreated sorghum grain stored for a period of 28 days differed from heat-treated grain (Table III). Untreated grain stored at 17 and 18% w.c. began moulding after two weeks storage accompanied by a reduction in germination, particularly after 28 days storage. Little change was observed in germination of control grain stored at 13 and 15% w.c. when compared with that of heat-treated grain at 60 or 70°C. However at 18% w.c., germination of sorghum grain heat-treated at both 70 and 80°C was significantly reduced over the 28 day period. This suggests that direct heat damage of the seed as well as fungal spoilage may have contributed the loss in germination obtained when compared to untreated sorghum grain at the same w.c.

Table III. Germination of untreated and heat-treated sorghum grain cv. M35-1 of various water contents during storage for up to 28 days.

| | | Percentage germination | | | |
|---|---|---|---|---|---|
| Storage time (days) | | 7 | 14 | 21 | 28 |
| Water content (%) | Treatment* (°C) | | | | |
| 13 | Control | 84 | 83 | 79 | 87 |
| | 60 | 70 | 77 | 79 | 79 |
| | 70 | 74 | 67 | 68 | 72 |
| | 80 | 39 | 30 | 37 | 32 |
| 15 | Control | 67 | 71 | 72 | 64 |
| | 60 | 74 | 68 | 79 | 77 |
| | 70 | 63 | 72 | 83 | 58 |
| | 80 | 16 | 42 | 32 | 16 |
| 17 | Control | 77 | 72 | 70 | 64 |
| | 60 | 84 | 70 | 81 | 75 |
| | 70 | 79 | 60 | 65 | 54 |
| | 80 | 16 | 16 | 18 | 11 |
| 18 | Control | 89 | 84 | 67 | 27 |
| | 60 | 79 | 60 | 28 | 12 |
| | 70 | 68 | 42 | 40 | 11 |
| | 80 | 14 | 18 | 11 | 2 |

LSD = 10.4

*, heat treatment time was 12 min

Free fatty acid (FFA) values of the control and heat-treated grain are shown in Table IV. The FFA value of untreated grain was unchanged with 13 and 15% w.c. while that of the 17 and 18% treatments increased with time of storage, indicative of fungal spoilage. However, there was a delay in the increase in FFA values of heat-treated sorghum grain at both 17 and 18% w.c. particularly during the first two weeks of storage. This was further reflected in the level of dry matter lost in untreated and heat-treated stored sorghum grain (Table V). During the 28 day storage period at 25°C the dry matter loss was greater in untreated than the heat-treated grain at both 17 and 18% w.c. although there was no dry

Table IV. Changes of free fatty acid (FFA) values of untreated and heat-treated sorghum cv. M35-1 of different water contents during storage for up to 28 days at 25°C.

| Storage time (days) | | Free fatty acid values | | | |
|---|---|---|---|---|---|
| Water content (%) | Treatment* (°C) | 7 | 14 | 21 | 28 |
| 13 | Control | 13 | 13 | 13 | 13 |
| | 60 | 13 | 15 | 15 | 19 |
| | 70 | 10 | 13 | 17 | 17 |
| | 80 | 15 | 15 | 15 | 19 |
| 15 | Control | 13 | 13 | 15 | 17 |
| | 60 | 13 | 17 | 17 | 17 |
| | 70 | 15 | 17 | 17 | 19 |
| | 80 | 15 | 17 | 15 | 19 |
| 17 | Control | 13 | 24 | 24 | 30 |
| | 60 | 13 | 17 | 21 | 19 |
| | 70 | 15 | 15 | 21 | 17 |
| | 80 | 10 | 13 | 17 | 21 |
| 18 | Control | 13 | 33 | 54 | 86 |
| | 60 | 13 | 21 | 45 | 71 |
| | 70 | 13 | 17 | 27 | 65 |
| | 80 | 15 | 15 | 19 | 71 |

LSD = 12.9
*, heat treatment for 12 min

Table V. The percentage dry matter loss of untreated and heat-treated sorghum cv. M35-1 at different water contents during storage at 25°C for up to 28 days.

| Storage time (days) | | Percentage dry matter (%) | | | |
|---|---|---|---|---|---|
| Water content (%) | Treatment* (oC) | 7 | 14 | 21 | 28 |
| 17 | Control | 0.38 | 0.46 | 0.36 | 0.86 |
| | 60 | 0.03 | 0.03 | 0.07 | 0.51 |
| | 70 | 0.01 | 0.11 | 0.12 | 0.12 |
| | 80 | 0.15 | 0.16 | 0.13 | 0.10 |
| 18 | Control | 0.78 | 0.82 | 0.73 | 1.23 |
| | 60 | 0.09 | 0.10 | 0.45 | 0.45 |
| | 70 | 0.06 | 0.07 | 0.69 | 1.03 |
| | 80 | 0.04 | 0.05 | 0.86 | 0.96 |

LSD = 0.11
*, heat treatment for 12 min

matter loss at 13 and 15% w.c. There were some discrepancies in the results obtained at different heat treatment temperatures although the steady state environmental conditions were maintained at all times. Heat-treatment, by reducing the population of fungal contaminants to a lower level, probably delayed the rate of colonisation even under conducive environmental conditions (18% w.c.). This was supported by the observation that visible moulding was delayed by 7 days in the 17 and 18% w.c. treatments.

These results suggest that potential exists for using heat-treatment as a method of conserving sorghum grain quality over short periods of time and delaying the onset of fungal spoilage.

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# EFFETS DU TRAITEMENT THERMIQUE SUR LES FACTEURS DE QUALITE ET LA MYCOFLORE DU SORGHO

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**RESUME**

On a examiné la possibilité d'utiliser les hautes températures comme méthode de préservation de la qualité des grains de sorgho. Des grains de sorgho possédant différents degrés d'humidité (12, 14 et 16 %) ont été traités à 69, 79, 80 et 90 C pendant un temps de 4, 8 et 12 mn. La germination, le contenu en acides gras libres et la contamination fongique ont tous été influencés par le traitement par la chaleur. En général, à 60 et 70 C, la germination du sorgho a été légèrement stimulée tandis qu'elle a été nettement plus réduite à plus haute température, comparée à celle du sorgho non traité. Le pourcentage de contamination fongique du grain était réduit de 90 à environ 5 % au cours du traitement thermique. Cependant, quelques espèces fongiques, particulièrement *Eurotium spp.*, *Aspergillus niger* et *Penicillium spp.* pouvaient encore être isolées après des durées de traitement allant jusqu'à 12 min à 80 - 90 C.

Par conséquent, la perte de matière sèche, le taux d'acides gras libres et le pourcentage de germination, à la fois des grains traités par la chaleur et des grains non traités, ont été comparés lors du stockage allant jusqu'à 28 jours à différents degrés d'humidité et à 25 C. L'utilisation possible de telles techniques pour améliorer la stabilité du stockage du sorgho et empêcher les pertes de qualité est discutée. Une telle information pourrait aider à développer des techniques d'exposition au soleil convenables sous les tropiques.