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Title:

**High-temperature drying of organically grown bread rye**

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Summary:

Mycotoxin producing fungi are naturally occurring components in cereals. When produced, some of the mycotoxins are toxic to humans and animals. Heat treatment is a method to reduce fungal abundance on cereals. The objectives of this work have been to provide more knowledge on this topic, and specifically to optimise the drum drying technique with reference to reducing the dissemination and proliferation of fungi on organically grown rye without reducing the baking quality of the grain.

The results show that drum drying can be a very efficient method in reducing fungi colonising on grain. For rye it is possible to achieve high effects on fungi without reducing the baking abilities. When drying at a constant maximum grain temperature of 64°C, the treatment resulted in less than 1% of the yeast propagules and less than 2% of the filamentous fungi, compared to what was found in the reference grain.

As drying temperatures and time for the grain to be treated are essential, the drum drier must be equipped with an effective control unit. The experiments show that a control system using fixed constant maximum grain temperature is most suitable.

## 1. Introduction

Mycotoxin producing fungi are naturally occurring components in cereals. When produced, some mycotoxins are toxic to humans and animals. Several circumstances, pre-harvest as well as post-harvest, may be of importance for the extent of mycotoxin problems.

Heat treatment is a well-known method for reduction of fungal infections, and previous studies have shown that drum drying reduces fungal abundance on the grain (Jacobsen, 1979; Kristensen, 1998; Kristensen and Søgaard, 2001). The aim was to establish the drying regime best suited to kill fungal propagules without reducing the baking quality of the grain. Special attention was paid to the ochratoxin A (OTA) producing fungus, *Penicillium verrucosum*, which is regarded a major health problem in food and feed production in countries with a temperate climate (Jørgensen *et al.*, 1996; Larsen *et al.*, 2001). Rye was chosen in the present study because it is known to be more sensitive to OTA contamination than wheat and oats (Jørgensen *et al.*, 1996; Schwarz, 2002).

The microbiological analyses consisted of fungal counts (dilution plating). The principle of plating is to prepare a dilution series from a hydrous suspension of grain and water. A specific volume of appropriate dilutions is plated onto solid nutrient agar and the fungi enumerated as colony forming units (cfu)/g grain. Two different agar substrates were used. V8 agar is a general medium, which was used to assess yeast fungi. Dichloran Yeast extract Sucrose 18% Glycerol agar (DYSG) is selective for xerophilic fungi (ability to tolerate low water activity), among which many mycotoxin producers (Frisvad *et al.*, 1992). Furthermore, it has diagnostic properties in enabling the detection and quantification of the ochratoxin A producing *P. verrucosum* (Elmholt *et al.*, 1999).

## 2. Materials and method

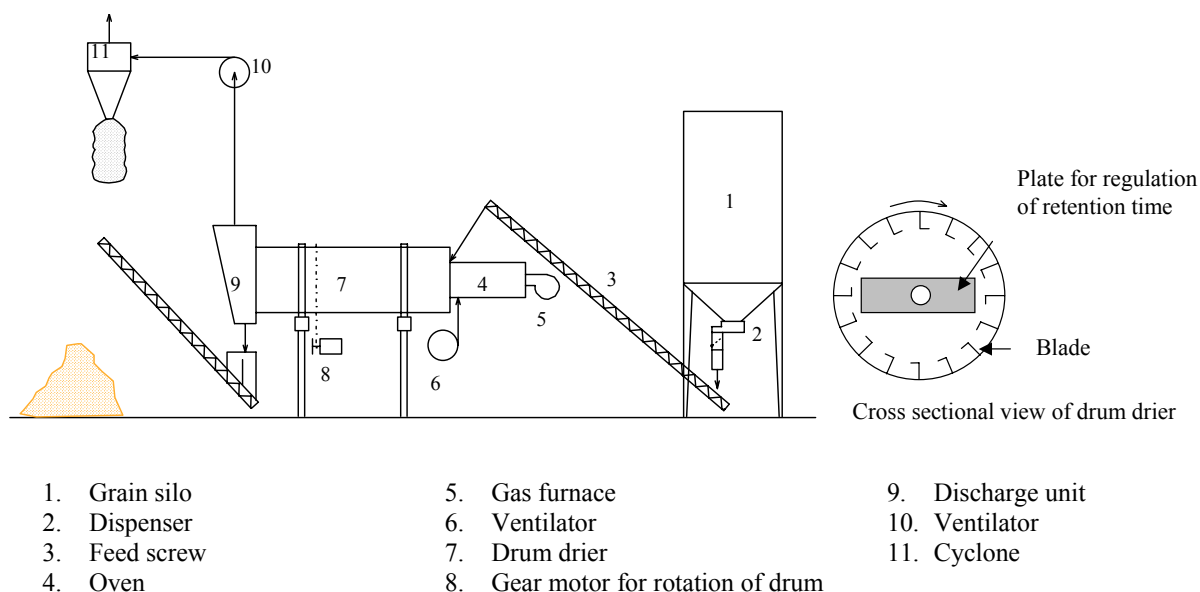
### 2.1 Experimental plant

An experimental drum drier was built up at Research Centre Bygholm. A natural gas furnace is used to heat the drying air. The gas furnace permits variations of the temperature of the inlet air from unheated to about 1000°C. A number of adjustable plates at the centre of the drum enable regulation of the time for the grain to remain in the drum (retention time). The drier is equipped with sensors for registration of temperatures and moisture content, etc. The sensors are connected with a data collection module.

The drying drum is of the single-drum type. The length of the drum is 5000 mm, and the diameter is 600 mm. On the inside the drum is equipped with blades, which serve to lift the grain and shower it down through the heated drying air. The speed of rotation of the drum can be controlled, and in order to obtain an even distribution of the grain across the entire cross section of the drum during the operation of the drier, a speed of 8.5 rpm was chosen. The drier has an hourly capacity of 0.5 – 1.5 tonnes grain.

Drying treatments have been carried out at different drying air temperatures (75-375°C), different grain temperatures after drying (32-79°C) and different grain retention times in the drum drier. Three retention times, short, medium and long,

corresponding to 4.5, 10.5 and 15 minutes respectively were used. Two different control methods were used, i.e. either a Fixed constant drying Air Temperature (FAT) or a fixed constant Maximum Grain Temperature (MGT). Organically grown rye from an early harvest date (15 Aug., 2001) and a late harvest date (30 Aug., 2001) was treated. The rye was of the variety Dominator. Before the drying, the moisture content rate was 17-18%.



**Figure 1.** Schematic diagram of drying plant.

## 2.2 Microbiological analyses

From the early and the late harvest dates, a representative sample of non-drum dried grain and samples from 24 different drying regimes were forwarded to the laboratory for microbiological analyses within two weeks after drying. At the laboratory the samples for fungal analyses were stored at 2°C in air-tight plastic containers and analysed within two months after receipt.

Viable propagules of fungi were enumerated by dilution plating on V8 and DYSG agar. From grain samples of each drying regime, 10.0 g was added with tap water to 50 g, and the suspensions were rotated for 60 min at 250 rpm. The original suspensions ( $10^0$ ) were used to prepare appropriate ten-fold dilution series (three replicate Petri dishes of each dilution). All plates were incubated at 20°C in the dark for five days. Of the different dilutions, the most appropriate one was chosen for enumeration, i.e. for V8 it was the one giving 50-150 colony forming units (cfu) of yeast per plate and for DYSG the one giving 25-100 colony forming units (cfu) of filamentous fungi per plate. In some of the most intensively dried grain samples, lower counts had to be accepted. *P. verrucosum* colonies were enumerated after another two days of incubation, as described by Elmholt *et al.* (1999). All enumerations are given as cfu g<sup>-1</sup> dried grain (drying time: 2 h at 130°C, according to ISO 712:1998).

### 2.3 Baking Quality

To evaluate the baking quality of the treated rye analyses of representative samples were made. From the early harvest date, a sample of non-drum dried grain and samples from 14 different drying regimes were analysed.

The samples were forwarded to the laboratory (Drabæks Mill) for analyses within two weeks after drying. The samples were analysed within three months after receipt. The analyses included amylograph test, falling number and test baking. The 14 chosen treatments ensured that the main effects of control method, retention time and treatment temperatures plus interaction of retention time and temperature could be determined. The results were treated statistically by a principal component analysis, and a relative baking quality number was calculated for each different treatment (Kristensen and Søgaard, 1995).

### 3. Results and discussion

An overview of the drying regime is shown in table 1.

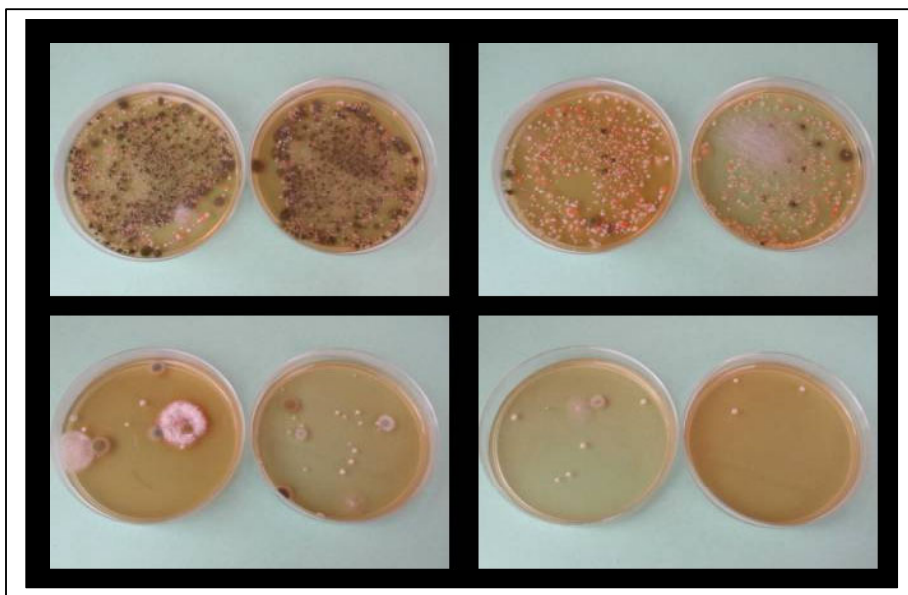
**Table 1.** Drying regime. When a drying air temperature is stated, control method FAT was used. For the other treatments control method MGT was used

Sample No.	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24
Time, min.	5	5	5	5	11	11	11	11	15	15	15	15	5	5	5	5	11	11	11	11	15	15	15	15
Drying air, °C	149	181	216	244	152	179	215	247	147	180	216	255	-	-	-	-	-	-	-	-	-	-	-	-
Grain, °C	51	54	58	63	53	57	61	66	45	51	55	59	53	61	71	77	51	56	64	74	53	60	71	79
Sample No.	25	26	27	28	29	30	31	32	33	34	35	36	37	38	39	40	40	42	43	44	45	46	47	48
Time, min.	5	5	5	5	11	11	11	11	15	15	15	15	5	5	5	5	11	11	11	11	15	15	15	15
Drying air, °C	118	237	248	287	126	222	244	286	132	192	248	263	-	-	-	-	-	-	-	-	-	-	-	-
Grain, °C	32	47	51	60	29	50	48	57	28	49	45	59	50	59	67	74	48	59	63	77	43	52	62	76

Dilution plating assesses only living fungi (viable counts). It is not an estimate of fungal biomass, but rather an indicator of the number of living fungal propagules (spores, conidia, *etc.*) on the grain surface. Figure 2 shows an example of how method MGT with medium retention time affected the grain fungi. The dark colonies are dematiaceous fungi, mainly belonging to the genera of *Cladosporium* and *Alternaria*. These fungi will be almost totally eliminated at a grain temperature of 56°C. The remaining colonies are primarily yeast. At the two highest temperatures, the fungal colonies are larger due to reduced competition.

Fungal abundance increased in the grain from the early to the late harvest. For the yeast, the reference grain showed  $4.81 \times 10^5$  cfu g<sup>-1</sup> and  $5.11 \times 10^5$  cfu g<sup>-1</sup> at the early and late harvest date, respectively. For the filamentous fungi, the corresponding values were  $3.38 \times 10^4$  cfu g<sup>-1</sup> and  $7.37 \times 10^4$  cfu g<sup>-1</sup> (Figure 3). The yeast thus increased by 6%, and the filamentous fungi increased by 118%. It should be noted that DYSG agar, which was used for the enumeration of filamentous fungi, is a selective

medium, and therefore it underestimates the total number of surface dwelling grain fungi to some degree.

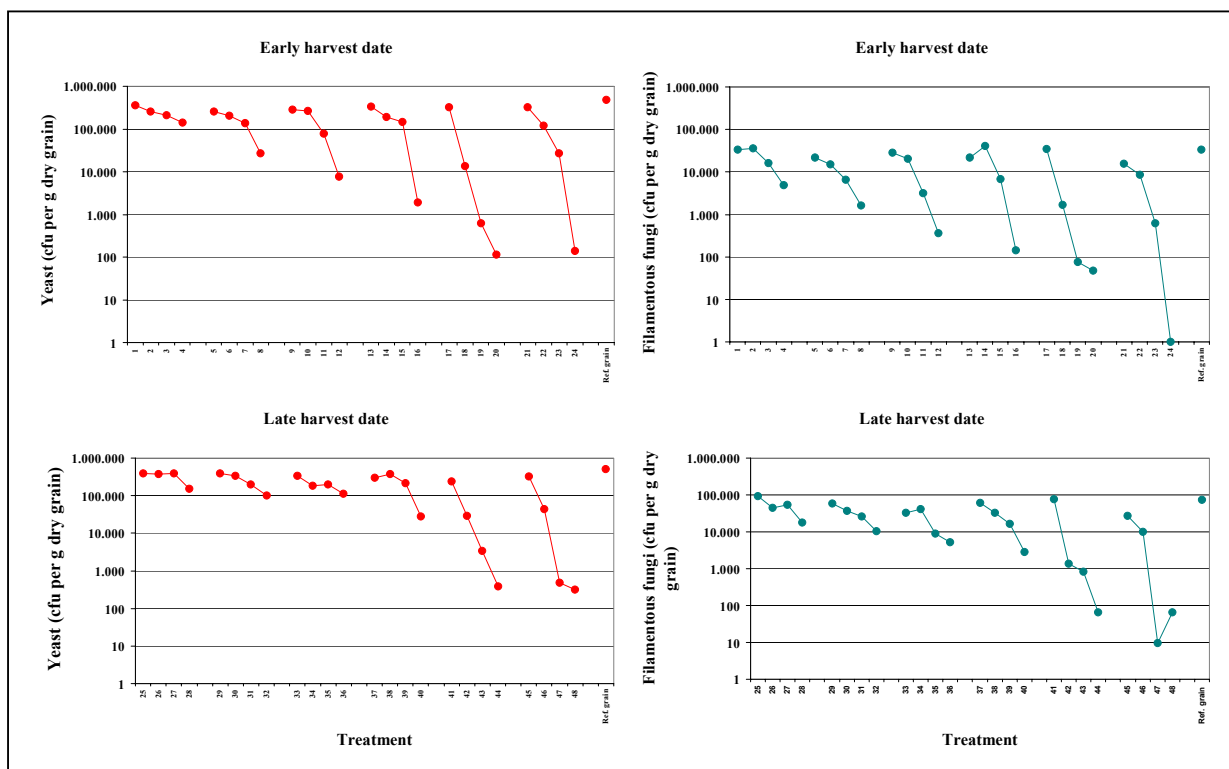


**Figure 2.** Fungi isolated from drum-dried grain by dilution plating. The Petri dishes have been plated with the  $10^0$  suspension (see text) and show treatments at grain temperatures of 51 (upper left), 56 (upper right), 64 (bottom left) and 74°C (bottom right).

The time for the grain to be treated in the drum – the retention time – affected the drying result, and the general trend shows a decrease in fungal abundance when the time is increased (Figure 3). This increased effect is most clearly seen at the high temperatures.

Increasing the temperature will have a more profound effect than increasing the retention time (Figure 3), and for both yeast and filamentous fungi, high temperatures when using the control method MGT will be more efficient in reducing fungal numbers than high temperatures when using the control method FAT. It should be noted, however, that results had to be depicted on a log scale to illustrate the very dramatic reductions in living fungal propagules at MGT, and that reductions at FAT are also profound, especially at an early harvest date. The reason for this higher effect at the early harvest date might be interaction between grain moisture content, drying air temperature and grain temperature. At the early harvest date, the moisture content was 0.4 % lower than at the late harvest date. At fixed constant drying air temperatures, lower inlet moisture content will lead to higher grain temperatures and lower outlet moisture contents.

*P. verrucosum* was not found in any of the samples, and the effect of drum drying on this important species cannot be deduced from the present experiment.



**Figure 3.** Fungi isolated from drum-dried grain by dilution plating and calculated as colony forming units (cfu)  $g^{-1}$  dry grain. Results from different drying regimes show yeast at the early harvest date (upper left), filamentous fungi at the early harvest date (upper right), yeast at the late harvest date (bottom left), and filamentous fungi at the late harvest date (bottom right).

The baking quality of the grain will also be affected by drum drying. As regards falling number, no evident effects have been seen, except for the treatments at very high grain temperatures (74 -79°C). The amylograph test showed a higher viscosity for the grain treated at high temperatures than for untreated grain. The ability of water absorption was highest for the grain treated at the highest temperature. Only for the rye exposed to very high grain temperatures (74-79°C), a visual judgement of dough and bread showed quality changes. For the reference grain and grain from all the other different drying regimes, the baking quality – on a visual basis - was judged to be good compared to common bread rye from the same year and from the same region.

The statistical analyses on falling number and amylograph values for max. vis-

osity and max. temperature show that a relative quality number can be calculated as follows:

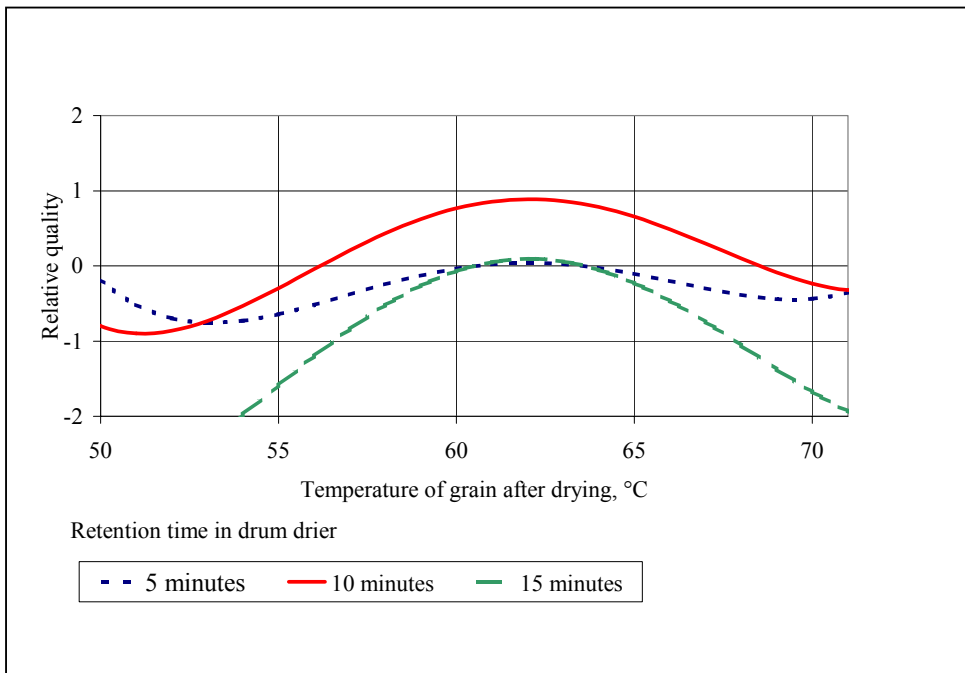
$$Q = 3.56 \times 10^{-2} \times F + 4.73 \times 10^{-3} \times Av + 1.39 \times At - 96.4$$

- Q = Relative baking quality
- F = Falling number
- Av = Amylograph max. viscosity (B.U.)
- At = Amylograph max. temperature (°C)

On the basis of the statistical analyses the baking quality as a function of the grain temperature and the retention time in the drum dryer can be expressed as follows:

$$Q = 1.20 \times 10^3 - 131 \times t - 7.10 \times T + 2.50 \times 10^{-1} \times T \times t + 3.21 \times t^2 - 3.28 \times 10^{-2} \times T^2 - 3.49 \times 10^{-2} \times t^3 - 2.02 \times 10^{-3} \times t^2 \times T + 1.42 \times 10^{-4} \times t^4 \quad (R^2 = 0.91)$$

- Q = Relative baking quality
- T = Grain temperature, °C
- V = Retention time in drum drier, min.



**Figure 4.** Baking quality of rye treated at different drying temperatures and at short, medium and long retention times in the drum drier.

The results are shown graphically in Figure 4. In general, the highest baking quality in rye was obtained at grain temperatures of about 62°C and a retention time in the drum drier of about 10 minutes. In proportion to the confidence interval (95% significance) the estimated differences were small. Thus, at grain temperature of 62°C the difference in quality caused by different retention time was not statistically certain.

#### 4. Conclusion

In conclusion, drum drying has very efficiently reduced the fungi colonising the grain. When using a fixed constant maximum grain temperature (MGT), a retention time of, e.g., 10.5 min and a temperature of 64°C (treatments 19 and 43) resulted in less than 1% of the yeast propagules and less than 2% of the filamentous fungi that were found in the reference grain. At these treatments the moisture content of the grain was reduced from about 17% to about 12%. This reduction of living fungal propagules will significantly reduce the risk of grain deterioration during storage.

The highest baking quality in rye was obtained at grain temperatures of about 62°C. Only in samples where the grain temperature had been higher than 70°C, the baking tests showed visual quality changes. Thus, it is possible to achieve a high reduction in fungi colonising the grain and at the same time to maintain a high baking quality.

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