# Effect of dry heat treatments on survival of seed borne Bipolaris sorokiniana and germination of barley seeds<sup>1</sup>

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In dry heat treatments of barley seeds naturally infected with B. sorokiniana, as the temperature and the time of exposure to treatment increased survival of both fungus and seeds decreased but not at the same rate. A severe reduction in viability of barley was produced under conditions required to eliminate the fungus from the seeds. The best seed survival associated with a treatment giving complete elimination of the fungus was only 42% (60 h at  $90^{\circ}C$ ).

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Dans une experience portant sur des traitements à la chaleur seche de semences d'orge contaminbes naturellement par le 8. sorokiniana, la survivance du champignon et celle des graines ont toutes deux diminubes avec l'augmentation de la temperature et de la duree du traitement, mais à des rythmes diffbrents. Les conditions requises pour l'elimination du champignon des graines ont provoqub une rbduction marqube de la viabilité de l'orge. La meilleure survivance de graines reliée à un traitement conduisant à une élimination complète du champignon n'a ete que de 42% (60 h à 90°C).

## Introduction

Barley (*Hordeum vulgare* L.) seeds harvested from a crop severely affected by spot blotch may contain more than 95% seeds infected by *Bipolaris sorokiniana* (Sacc. in Sorok.) Shoem. (Couture and Sutton, 1978 a). The seed borne fungus serves as primary inoculum causing seedling blight. Eventually this may also provide secondary inoculum produced on above-ground affected parts to cause spot blotch on the foliage (Couture and Sutton, 1978 b).

For many years mercurial fungicides have helped control the seedling blight phase but their use was discontinued. There are reported cases from Great Britain of highly infected seed samples being harvested following the omission of organomercurial seed treatment (Hewett, 1975). However, several fungicides currently used for seed treatment of cereals are not as effective as mercury in controlling seedling blight (Wallace and Mills, 1968). Furthermore, because **B.** sorokiniana may persist in barley seeds for 10 years or more (Machacek and Wallace, 1952), ageing is not a practical method for obtaining seeds free from the pathogen.

There is some indication that **B.** sorokiniana may be eliminated from barley by exposure of seeds to a temperature of 100°C for 15 - 30 hours (Atanasoff and Johnson, 1920). For barley grown in the field, Skachkova (1975) reported that heat treatment (not defined) of seeds substantially reduced infection by B. sorokiniana in the crop. Heat treatment of seeds has also been used to control other

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pathogens of barley. The hot water treatment at 54°C for ten minutes has long been recommended for the control of loose smut, *Ustilago nuda* (Jens.) Rostr. (Fushtey, 1969). *Alternaria* sp. disappeared from barley seeds after a seven month period of storage at 41°C (Brown and Robert, 1943).

As an alternative to chemical control, the possibility of using dry heat for controlling seed borne *B. sorokiniana* was examined.

## Materials and methods

Seed at 5% moisture content and 89% germination of the 2-row barley cultivar Fergus was used. All the seed was naturally infected by *B. sorokiniana* and the sample was from a field harvested 18 months earlier at Elora, Ontario.

For heat treatments, seed batches contained in 50 ml glass beakers were placed in a thermostatically controlled hot air oven at 70, 80, 90, 100, 110, 120, 130, or 140°C. The samples were removed at time intervals ranging from 15 minutes to 72 hours and allowed to cool.

To determine the percentage of seeds colonized internally by B. *sorokiniana,* 100 seeds from each temperature-time treatment were immersed in 70% ethanol from 20-30 seconds, in 1% sodium hypochlorite for 10 minutes, then rinsed in sterile water and placed in petri dishes on weak carrot agar (Tuite, 1969), supplemented with 50 ppm each of streptomycin-sulphate and chlortetracycline hydrochloride. The seeds were examined at 40X through a stereomicroscope after one week of incubation in the laboratory.

To determine the percentage germination of seeds, 100 seeds from each temperature-time treatment were placed on moist filter paper in petri dishes and incubated for one week in the laboratory. The average of the sample plated on filter paper and of the one previously plated on agar, was used to determine percentage germination.

Time of exposure	Survival (%)* of <i>Bipolaris sorokiniana</i> ( <i>Bip.</i> ) and barley seeds (seed)															
to treatment (h)	70 C <i>Bip.</i> seed		80 C <i>Bip.</i> seed		90 C <i>Bip.</i> seed		100 C <i>Bip.</i> seed		110 C <i>Bip.</i> seed		120 C <i>Bip.</i> seed		130C <i>Bip.</i> seed		140C <i>Bip.</i> seed	
0.25			100	100	100	99	98	90	96	89	41	59	36	9	0	0
0.50			100	98	95	97	90	84	57	73	17	40	4	2	0	0
0.75											2	5	1	0		
1			100	98	92	96	62	48	17	44	0	1	0	0		
2			100	91	90	90	18	40	4	10	0	0				
3							15	37	2	1	0	0				
4	100	91	100	91	87	90	6	30	0	0						
6							5	14								
8	100	90	99	90	61	90	0	10								
16	99	89	97	88	57	85										
24	98	88	96	86	50	84										
36	98	86	92	83	32	78										
48	98	83	91	82	5	73										
60					0	42										
72					0	34										

Table 1. Effect of dry heat treatments on survival of seed borne Bipolaris sorokiniana and germination of barley seeds.

\*Survival as a percentage of checks: 100% infected seeds and 89% germination.

# Results

Survival of both fungus and seeds decreased as the temperature and the time of exposure to treatment increased but not at the same rate (Table 1). The seeds died more rapidly than *B. sorokiniana* at 70, 80, and 130°C and the fungus was differentially more affected at temperatures ranging from 90 to 120°C. Both fungus and seeds were killed in less than 15 minutes at 140°C. The most differential effect beneficial to the barley was at 90"C where, after 8 hours or more of exposure, the viability of the fungus declined much more rapidly than the viability of the seeds.

*B. sorokiniana* tolerated exposure periods of 48, 4, 0.5, and 0.25 hours respectively at 70 and 80, 90, 100, and 110°C. Exposure periods of 60, 8, and 4 hours were required for complete elimination of the fungus at temperatures of 90, 100, and 110°C. A large difference was observed in the time required, at a given temperature, to kill the fungus in a few seeds as compared to that needed to kill it in all seeds (e.g. at 90°C, a 5% kill was obtained in half an hour but it took 48 hours for a 95% kill). The fungus was killed rapidly (<1 hour) at temperatures of 120°C or more.

It was also observed that other seed borne fungi such as *Alternaria* sp. and *Fusarium* sp. were eliminated at temperatures of  $110^{\circ}$ C or more while *B. sorokiniana* survived.

#### Discussion

Exposure periods at various temperatures required to eliminate *B. sorokiniana* from the barley seeds produced a severe reduction in viability of the seeds. The best seed survival associated with a treatment giving complete elimination of the fungus was only 42% (60 h at  $90^{\circ}$ C).

However, this figure might possibly be higher at some temperature-time combinations around 90°C.

Atanasoff and Johnson (1920) reported good control of 6. sorokiniana at 100°C and an average of about 89 and 73% germinated seeds compared to their check after exposures of 15 and 30 hours respectively at 100°C. Although, this is a much higher survival than the results shown in Table 1, the germination of some of their seed samples was also cut down severely. These differences may be related to differences in moisture content of the seeds, to differences in seed vigor or to the approximate control of temperature indicated by Atanasoff and Johnson (1920).

Grain drying is an increasing practice in Canada. To avoid overheating damage to the grain, the maximum allowable air temperatures to dry barley grain are 45°C for seed or malting, 55°C for commercial use and 80°C for feed (Friesen, 1976). According to results in Table 1, none of these temperatures is likely to reduce the proportion of 6. *sorokiniana* infected seeds.

Because the temperature required to kill the pathogen also severely reduces seed viability, it is obvious that dry heat treatment of barley seeds is impracticable for a barley grower. Despite the concomitant high incidence of barley kill, heat treatment may however be useful to obtain *B*. *sorokiniana* - free barley seeds for experimental purposes. In other respects, on account of our observation that some seed-borne fungi, namely *Alternaria* sp. and *Fusarium* sp., that might interfere with 6. *sorokiniana*, can be selectively eliminated by means of heat treatment, the procedure can also be used to facilitate isolation of 6. *sorokiniana* from barley seeds as has been done for *Drechslera avenacea* (Curt. ex Cke.) Shoem. in oat seeds (Malone, 1962).

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