INVESTIGATION OF THE PHYSIOLOGICAL BASIS OF MALTING QUALITY OF GRAIN DEVELOPING UNDER HIGH TEMPERATURE CONDITIONS

by

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LIST OF ABBREVIATIONS

ABARE	Australian Bureau of Agriculture and Resource Economics
ABB	Australian Barley Board
α-amylase	alpha amylase
ADPG	adenosine diphospho glucose
ADPGp	adenosine diphospho glucose pyrophosphorylase
ANOVA	analysis of variance
β-amylase	beta-amylase
β-glucan	beta-glucan
β-glucanase	beta-glucanase
BE	branching enzyme
CCL	crushed cell layer
°C	degrees Celsius
°Cd	degree day
CLSM	confocal laser scanning microscope
cP	centipoise
daa	days after anthesis
DP	diastatic power
et al.	and others
FAN	free amino nitrogen
Fig.	figure
GBSS	granule bound starch synthase
h	hour
HWE	Hot Water Extract
K	potassium
λ	wavelength
LM	light microscope
MBIBTC	Malting and Brewing Industry Barley Technical Committee
min	minute
N	nitrogen
nm	nanometre
P	phosphorus
PAS	periodic acid/Schiffs
SEM	scanning electron microscope
SSS	soluble starch synthase
TBO	Toluidine blue O
μm	micrometre
UDPG	uridine 5'-diphospho glucose
UDPGp	uridine 5'-diphospho glucose pyrophosphorylase
WBMQEL	Waite Barley and Malt Quality Evaluation Laboratory

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(a) Fluorescence micrograph of a longitudinal section through a malted control grain. The pattern of endosperm modification was essentially parallel to the scutellum, with some loss of fluorescence adjacent to the surrounding aleurone cells (arrows). Embryo (em).
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(a) SEM image of the central endosperm region of a malted control grain with open and friable appearance due to loss of protein and most of the B-type starch granules. Very little cell wall material remained (arrows) but there was no evidence of degradation of A-type starch granules.
(b) SEM image of the central endosperm region of a malted heat treated grain. Considerable loss of B-type starch granules, protein and cell wall material was apparent. Extensive pitting (arrows) was observed in A-type starch granules as a result of malting.

Abstract

Short periods (up to 5 days) of high temperatures have been shown to reduce grain weight in barley although the specific effects on malting quality are still not clearly understood. Controlled environment conditions were used in this study to investigate the physiological and biochemical responses in Schooner barley to 5 days of elevated temperatures during grain filling. Control plants were maintained at 21/16°C (14h day) from anthesis to harvest ripemess. Plants undergoing heat treatment experienced increasing temperatures for two days (from 16 days after anthesis) followed by 3 days at high temperatures (35°C day /25°C night). The period of high temperature reduced individual grain dry weight by 18%. Schooner barley endosperm (the grain storage tissue) exhibited greater sensitivity to high temperatures than the whole grain (25% reduction in dry weight) in response to high temperatures. Grain development was accelerated by exposure to high temperatures and a reduction of approximately 8% in the duration of grain filling was observed in heat treated compared with control grain.

Changes in endosperm composition provided evidence that exposure to high temperatures altered overall grain metabolism. Reduction in starch, the single most important grain component contributing to final grain weight was the major factor lowering final grain dry weight. b-glucan deposition was also reduced following high temperature exposure. Little change was observed in the absolute amount of nitrogen accumulated per endosperm, although nitrogen concentration was higher in heat treated grain, due mainly to reduced grain size and lower starch content.

The results of this study confirmed other reports that the conversion of sucrose to starch was limiting in grain exposed to a period of high temperature. The amount of substrate, sucrose, within heat treated endosperms was not found to be limiting starch accumulation. The reduction in starch synthesis appeared to result from the combined effects of diminished catalytic activity of several enzymes in the committed pathway of starch synthesis and/or delayed recovery of enzyme activity during the cooler conditions which followed the heating period. Soluble starch synthase (SSS) showed an

immediate loss of catalytic activity, even at moderate temperatures, while sucrose synthase and uridine 5'-diphosphoglucose pyrophosphorylase showed greatest reduction in activity only after plants were returned to cooler conditions. Individual enzymes showed variation in the level of recovery under the cooler temperature conditions which followed the heating period.

The high temperature conditions which induced changes in the metabolism of grains following high temperature exposure, were associated with alterations to the appearance of the starchy endosperm cells, including the distribution of starch granules and their growth. Under elevated temperatures the synthesis of endosperm starch did not appear to keep pace with cell division and cell enlargement and peripheral endosperm cells remained relatively empty of starch granules. A reduction in Calcofluor fluorescent cell wall material was observed in both the endosperm and the crushed cell layer of heat treated grains. Microscopic observations revealed that the lower levels of starch observed in heat treated barley grains were not only associated with reduced accumulation of starch, but also localised degradation of the barley endosperm. Increased embryo growth, which was observed to take place in heat treated grains, may have been due to the nutrients thus generated although further investigations are required to elucidate the mechanisms involved. Structural observations of harvest ripe grain developing under control conditions also revealed changes in the grain associated with early germination events, and the possibility of a continuum between development and germination has been put forward.

High temperature exposure resulted in an overall decrease in malt extract potential, but there was evidence that the effects of heat on individual malt parameters were both beneficial and detrimental. While DP, β-amylase deposition, β-glucan and viscosity were improved, heat treatment was associated with a reduced quantity of starch and increased protein concentration. Grain and malt β-glucan levels were reduced as a result of high temperature exposure and β-glucan degradation was enhanced in these grains. The overall reduction in the amount of 'maltable' grain (due to reduced grain size) represented one of the most significant effects of heat treatment.

The effects of a period of high temperature on grain growth and subsequent malting quality were examined for barley varieties Schooner and Arapiles and advanced breeding

line WI-2875*22. All showed reduced final grain dry weight in heat treated grains but the response to high temperature exposure with respect to starch, protein and β-glucan accumulation varied. The continued synthesis of starch by Arapiles and WI-2875*22 under high temperature conditions warrants further investigation. Evidence has also been presented indicating that water loss may not have a decisive role in the termination of grain filling.

The effect of high temperatures on the non-endosperm component of the grain showed both genotypic and environmental variation. Changes in the contribution made by the non-endosperm component to final grain weight was observed to lead to alterations in the relative concentration of important grain storage components. This was found to be crucial where potentially important heat stable quality characteristics, such as reduced accumulation of nitrogen, were masked by changes in the contribution to final grain dry weight by the non-endosperm component of the grain.