Varietal differences in protein polymer built-up of wheat at different temperature and nitrogen regimes during grain filling

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ABSTRACT

The amount and size distribution of polymeric proteins, influenced by cultivar, temperature and nitrogen timing, is an important factor in determining gluten strength in wheat. In mature wheat grains, the distribution of the monomeric and polymeric proteins as well as their solubility play a critical role in governing wheat flour properties and uses. Four varieties of wheat (two early and two late), differing in high-molecular-weight glutenin subunit composition (2+12 versus 5+10) were grown at four nitrogen regimes and two temperature regimes in order to determine the manner in which differences in mature protein composition were the result of differences in accumulation of proteins during grain filling. Combination of temperature and nitrogen regimes leads to changes in amount and size distribution of polymeric proteins in mature grains of wheat. A relationship is present between polymerisation of proteins during grain maturation and amount and size distribution of polymeric proteins in the mature grains.

INTRODUCTION

In wheat grains percentage of proteins are known as a significant determinant of both baking and nutritional qualities but protein composition is also a crucial factor which influences both of these properties. Therefore, proteins are recognized as the most important components governing bread-making quality in cereal grains[1]. A strong correlation exists between baking quality and size distribution of glutenin polymers[2]. Specific composition of protein subunits is genetically determined[3] while extractability, concentration of proteins and amount of polymeric proteins are both genetically and environmentally determined[4-5]. Influencing environmental factors are e.g. nitrogen levels and temperature conditions. The aim of the present study was to evaluate varietal differences in built-up of polymeric proteins in wheat. Effects of temperature and nitrogen regimes were investigated together with the cultivars to view changes in amount and size distribution of polymeric proteins at maturity.

MATERIAL AND METHODS

Four wheat cultivars Vinjett, Soljett, Springjett and Kronjett, were grown in pots, three plants in each pot, in the greenhouse. These cultivars were selected as being two early maturing and two late maturing in field. Furthermore, one of the early and one of the late maturing cultivars had high molecular weight (HMW) glutenin subunits 2+12 and 5+10, respectively. The soil used in the pots was krukväxtjord, teca (Weibull Trädgård AB, Landskrona, Sweden), with the nitrogen amount 980 g m^{-3}. All plants were grown at day/night temperatures of 18/14 °C, with 17 h day until spike formation, decimal code (DC) 45-49. Thereafter, the plants were divided into two separate chambers with day/night temperature regimes 17/14 (T1) and 24/21 °C (T2). Different nitrogen regimes were used in order to create differences in timing of nitrogen applications. Mid-flowering times (when half of the plants were at anthesis) were determined. Thereafter plants were harvested 4, 8, 12, 18, 26, 36 and 50 days after mid-flowering (anthesis). Three plants were harvested randomly at each harvest occasion of each plant treatment (including both temperature and nitrogen regimes). The spikes were immediately threshed by hand directly after harvesting and grains were weighed, frozen at -20°C, and then lyophilized and weighed again. Grain water content was calculated. Grains harvested 4 and 8 days after anthesis were crushed manually, and the rest of the grains were milled in a Yellow Line, A 10, IKA-Werke, and Staufen, Germany commercial Blender. Amount and size distribution of polymeric protein were analysed applying size exclusion-high performance liquid chromatography (SE-HPLC) with a two-step extraction procedure[2] with modifications according to Johansson et al[6]. The first step in this method extracts the proteins soluble in dilute sodium dodecyl sulphate (SDS), whilst the second extract contains proteins soluble only after sonication. Proteins were detected by UV absorbance at 210 nm. The SE-HPLC chromatograms were divided into five parts and the following protein parameters were calculated; %UPP (percentage of unextractable polymeric protein in total polymeric protein), TOTE (total amount SDS-extractable protein) and TOTU (total amount SDS-unextractable protein) [7-8].

RESULTS AND DISCUSSION

At maturity, the cultivar Springjett, having HMW glutenin subunits 2+12, was found to have the highest %UPP (Table 1). This is not in accordance with earlier findings which indicated higher %UPP within cultivars having HMW glutenin subunits 5+10 compared to those having 2+12[9]. However, in a large screening investigation, comparing protein compositions with %UPP in wheats of different origins, no relation was found between protein composition and %UPP in some of the wheat collections (unpublished results). In the

present investigation the two cultivars having HMW glutenin subunits 5+10 were found to have higher %UPP compared to one of the 2+12 cultivars (Kronjett; Table 1). Percentage of UPP has also been related to length of anthesis time as well as to maturity time[6,9]. The cultivars in the present study were selected not only in accordance to composition of HMW glutenin subunits but also in relation to maturity time. However, in the present study, the cultivars did not totally follow the found differences in maturity time from the field trials. Thus, relations between maturity time and %UPP has to be further evaluated.

The main reason for the cultivar Springjett having high %UPP was found to be a low amount of SDS-extractable protein (TOTE) and not a high amount of SDS-unextractable protein (TOTU; Table 1). Low amount of TOTE has in previous study been related to a short time to anthesis, thereby leading to early maturity[9]. Kronjett, the cultivar with the lowest %UPP showed both a significantly low amount of TOTU and a significantly high amount of TOTE.

Table 1. Means of different protein parameters in wheat cultivars at maturity

Cultivar	HMW	%UPP	TOTE	TOTU
Vinjett	5+10	33bc	7.25b	2.56ab
Soljett	5+10	38b	7.65ab	2.86a
Springjett	2+12	42a	6.22c	2.61ab
Kronjett	2+12	30c	8.11a	2.42b

HMW=High molecular weight glutenin subunits, %UPP=percentage of unextractable polymeric protein in total polymeric protein, TOTE=total amount SDS-extractable protein, TOTU=total amount SDS-unextractable protein. Numbers followed by the same letter is not found to differ significantly (LSD 0.05).

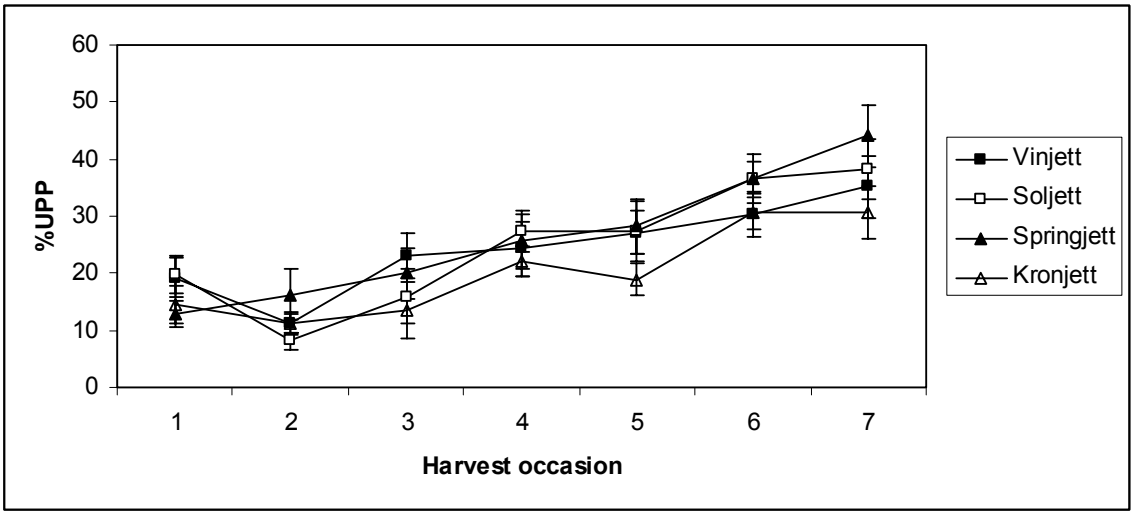

Figure1. Percentage of UPP during grain development in four different varieties. Bars indicate standard deviation.

Investigations about cultivar differences in %UPP during grain maturation showed no consistent relationship between the %UPP at maturity and during grain maturation. (Fig 1) The %UPP generally increased for all cultivars during grain maturation,(Fig). However, the differences between cultivars were mostly not significant for the different harvest occasions. Generally, the cultivar Kronjett was found having relatively low %UPP during the whole maturation time, while the cultivar Springjett was found to have high %UPP mainly during the last part of the grain maturation time (Fig 1).

Further investigations and evaluations of the material is needed before it will be possible to totally understand the background and reason for cultivar variation in polymeric protein composition at maturity.

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