



## Analytical Methods

## Quantitative and qualitative study of spelt and wheat fibres in varying milling fractions

E. Escarnot<sup>a,\*</sup>, R. Agneessens<sup>b</sup>, B. Wathélet<sup>c</sup>, M. Paquot<sup>c</sup><sup>a</sup> Walloon Agricultural Research Centre, Department of Biological Control and Plant Genetic Resources, Rue de Liroux, 4, 5030 Gembloux, Belgium<sup>b</sup> Walloon Agricultural Research Centre, Department of Agricultural Systems, Rue de Serpont, 100, 6800 Libramont, Belgium<sup>c</sup> University of Liège, Gembloux Agro-Bio Tech, Department of Industrial Biological Chemistry, Passage des déportés, 2, 5030 Gembloux, Belgium

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## ABSTRACT

The fibre composition of four spelt genotypes and of three wheat genotypes was studied on three grindings: bran, whole bare grains flour for all genotypes, and whole hulled grains flour for spelt only. Insoluble fibre and soluble fibre contents were measured after removal of proteins, starch and ashes from the sample (Lee, Prosky, & De Vries, 1992). Cellulose, hemicellulose and lignin contents were measured according to the same principles with different chemical degradations of the sample (Van Soest & Wine, 1967). Spelt and wheat bran and whole grain flour displayed significant statistical differences for hemicellulose and cellulose contents. Variability amongst the spelt genotypes was much higher than amongst the wheat genotypes. The study also highlighted the special profile of the true baking variety Ressac and the richness in fibres of the landrace 140. Finally, various methods of measurement were compared and a combination of these methods was proposed for cereal grains dietary fibre analysis.

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## 1. Introduction

Spelt (*Triticum aestivum* ssp. *spelta*) is an ancient subspecies of common wheat (*Triticum aestivum* ssp. *aestivum*). Until the beginning of the 20th century, spelt was the main grain used for bread production in south-western Germany and parts of Switzerland and Austria. Since then, however, modern wheat has largely replaced spelt. This is due to spelt's lower yield, to its sensitivity to lodging and to the hulls which account for 21–32% of the spikelet (Percival, 1921). In agronomic terms, spelt might be more resistant to disease, and perform better than wheat under less advantageous growing conditions, such as wet, cold soils and high altitudes (Campbell, 1997). Besides, with the hull covering the seed, chemical treatment before sowing might not be necessary and due to its long straw, it does not support a high level of nitrogen fertilisation (Bonafaccia et al., 2000). In addition, it contributes to agrobiodiversity. Spelt meets the European Union guidelines on growing practices for more environmentally friendly cereal production and is

also suitable for organic farming. Therefore, it continues to be cultivated in several central and middle European countries. In Belgium, it covers 10,000 ha, in Germany 23,000 ha (Statistisches Bundesamt Deutschland, 2008), while in Switzerland it represents 10,000 T of grains (Office fédéral de la statistique Suisse, 2008).

Spelt is believed to possess valuable nutritional qualities, differing from those of wheat. It has a higher protein content than wheat (Abdel-Aal, Hucl, & Solsulski, 1995; Bonafaccia et al., 2000; Grela, 1996; Marconi, Carcea, Graziano, & Cubadda, 1999; Ranhotra, Gelroth, Glaser & Lorenz, 1995; Ranhotra, Gelroth, Glaser, & Stallknecht, 1996), a higher lipid content, especially in  $\Delta^7$ -avenasterol (Abdel-Aal et al., 1995; Grela, 1996; Marconi et al., 1999; Ranhotra, Gelroth, Glaser, & Lorenz, 1995; Ranhotra et al., 1996; Ruibal-Mendieta, Delacroix, & Meurens, 2002) and higher magnesium, phosphorus, iron, copper and zinc contents (Ruibal-Mendieta et al., 2005). Spelt has long been used mainly in cattle feed especially for calves. It also suits growing and fattening animals and can be used to supplement forage for rearing animals (Lecomte, Boreux, Agneessens, Beckers, & De Keyser, 1996). Over the past few decades spelt has attracted renewed and increasing interest as human food due to its image as a “healthier, more natural, less over-bred” cereal than modern wheat (Schober, Bean, & Kuhn, 2006). It falls into the niche product and is important in specialty breads, organic food and food products with characteristics that differ from regular

Abbreviations: ADL, acid detergent lignin; ADF, acid detergent fibre; C, cellulose; DF, dietary fibre; H, hemicellulose; IF, insoluble fibre; L, lignin; LR, landrace; NDF, neutral detergent fibre; SF, soluble fibre; TF, total fibre.

\* Corresponding author. Tel.: +32 81 62 03 36; fax: +32 81 62 03 49.

E-mail address: [escarnot@cra.wallonie.be](mailto:escarnot@cra.wallonie.be) (E. Escarnot).

wheat products (Ranhotra et al., 1995). Making bread from spelt flour requires adapted baking methods. Spelt dough is less stable and has less elasticity and a higher extensibility than common wheat dough. It is very soft and sticky after kneading; handling it is therefore more difficult and the loaf volume is generally lower than with modern wheat cultivars (Schober, Clarke, & Kuhn, 2002). However, the technological potential of spelt for milling, bread making and pasta production seems promising (Bonafaccia et al., 2000; Ranhotra et al., 1995; Schober et al., 2002; Schober et al., 2006). From the health point of view, spelt is recommended for the treatment of colitis ulcerosa, neurodermitis and other allergies, as well as for high blood cholesterol (Strechlow, Hertzka, & Wueffen, 1991), but it is forbidden for people suffering from celiac disease, and it can provoke wheat allergy and gluten enteropathy (Kasarda & D'Ovidio, 1999).

Cereal foods constitute a major part of the daily diet in Europe and are one of the main dietary sources of fibre. The recommended current fibre consumption is 25 g/day/person (Poutanen, 2006). There is increasing agreement that a sufficient amount of whole grain consumption protects against the development of diet-related disorders such as cardiovascular disease and type 2 diabetes (Jones, 2006), as well as colorectal cancer (Larsson, Giovanucci, Bergkvist, & Wolk, 2005). A greater intake of whole grain foods is also associated with less obesity (Koh-Banerjee et al., 2004). Concerning colon cancer risk, whole grains and wheat bran might be the most protective fibre sources (Kritchevsky, 2001). Wheat bran is very effective in increasing laxation and therefore is used in the treatment of constipation and might prevent colorectal cancer (Ferguson & Harris, 1999). The addition of large amounts of wheat bran to the diet will also significantly reduce cholesterol saturation (McDougall, Yakymyshyn, Walker, & Thursten, 1978).

Several studies (Abdel-Aal et al., 1995; Bonafaccia et al., 2000; Marconi et al., 1999; Ranhotra et al., 1995; Ranhotra et al., 1996) have analysed IF and SF from spelt whole grain flour in comparison to wheat. No clear difference was found except for a slightly higher SF/IF ratio for spelt (Abdel-Aal et al., 1995; Bonafaccia et al., 2000; Marconi et al., 1999; Ranhotra et al., 1995; Ranhotra et al., 1996). However, most of the studies compared one wheat variety against 1–5 spelt varieties; often from different locations and/or years; and without reference to the growing conditions. Only one study concentrated on L, H and C without reference to the growing locations and conditions (Grela, 1996). As noted earlier, bran holds great interest for human nutrition and is regarded as a 'high-fibre ingredient' in the food industry. However, there is a gap of knowledge regarding spelt bran composition. As the first outlet of spelt production, whole spikelet flour should not be neglected. One thorough study in this area analysis 24 spelt varieties for C, H and L but not SF (Lecomte et al., 1996). Consequently these three milling fractions were analysed for insoluble (IF) and soluble fibres (SF), lignin (L), hemicellulose (H) and cellulose (C) for three wheat varieties and four spelt genotypes grown and stored in one location under the same conditions.

## 2. Experimental conditions

### 2.1. Materials

The genotypes were grown in one replicate in a single trial at Gembloux, Belgium. The crop was conducted to obtain safe grains free of disease. The plots were 4.5 m long and 1.5 m wide, and were sown in October 2005 (density: 300 grains/m<sup>2</sup>). At the end of the winter, a weed-killer was spread on the trial and during spring nitrogen (2 times), one growth regulator and one fungicide (2 times) were applied. Plots were harvested in August 2006. An auto-blowing combine harvester was used to prevent the blending

of samples. Grain sacks were dried at 30 °C for 1 day and stored for 1 month at room temperature in a dry place. Harvest per plot yielded about 4 kg. After homogenisation, 500 g were sampled for milling. Two milling fractions were chosen for wheat: whole grain flour (Cyclotec) and bran (Moulin Chopin CD1). Three milling fractions were chosen for spelt: whole grain flour (Cyclotec), bran (Moulin Chopin CD1) and whole spikelet flour (Cyclotec). All the milling fractions were ground to 0.5 mm. After milling, the samples were stored at 4 °C to prevent mould formation and enzyme activity. Prior to analysis, the samples were set at room temperature and homogenised.

Four spelt genotypes were selected for analysis: Cosmos, a Belgian variety with high yield potential used for feeding and secondly baking; Landrace (LR) 140, a Belgian landrace; Alkor, a new Swiss variety introduced in Belgium used for feeding; and Ressac, the only true baking Belgian variety. LR140 has never been bred and should be a pure spelt; Ressac and Cosmos were descendants from the Belgian breeding and contained respectively 9.5% and 25% of winter wheat in their genetic background. Indeed several ten years ago due to the lack of spelt genetic resources, Ardenne, a Swedish winter wheat, and Castell a Belgian winter wheat, were crossed with spelt (Clamot, 1978; Herman, 2007). Three winter wheat varieties were chosen: Centenaire, a Belgian feed wheat cultivated mainly in Belgium; Apache, the variety the most cultivated in Europe between 2000 and 2004; and Soissons, a successful European variety registered in 1987. These last two varieties are both baking cultivars.

### 2.2. Methods

The fibre content was measured using non-enzymatic gravimetric and enzymatic gravimetric methods.

#### 2.2.1. Non-enzymatic gravimetric methods

Two non-enzymatic gravimetric methods – acid detergent fibre (ADF) analysis and acid detergent lignin (ADL) analysis as described by Van Soest (1963) – were used to measure the cellulose and lignin content by removal; ADF includes cellulose and lignin. The samples (1 g = P) were boiled for 1 h with 100 mL acid detergent solution (sulphuric acid 0.2 N and 20 g/L of *N*-cetyl-*N,N,N*-trimethyl ammonium bromide). The residue was washed with distilled water and acetone. The crucibles were dried in drying oven at 103 °C during one night, cooled in a desiccator and weighed ( $P_1$ ). The remaining ADF residues were submerged with 72% sulphuric acid and then washed with hot distilled water. The crucibles are dried again, weighed ( $P_3$ ) and calcinated ( $P_2$ ) in a muffle furnace at 550 °C during 3 h or until ashes were white.

The fibre percentage formulae used were:

$$\text{ADF}\% = \frac{P_1 - P_2}{P} \times 100 \times \frac{100}{DM},$$

$$\text{ADL}\% = \frac{P_3 - P_2}{P} \times 100 \times \frac{100}{DM},$$

DM = sample dry matter.

#### 2.2.2. Enzymatic gravimetric method

Two enzymatic gravimetric methods were used. One was the Van Soest and Wine (1967), neutral detergent fibre (NDF) method, which consists of an  $\alpha$ -amylase treatment (Schaller, 1977) and provides data on L, H and C content by subtraction with ADF and ADL. The samples (1 g = P) were treated with  $\alpha$ -amylase (Termamyl, Novozymes) (0.1 mL) and were boiled for 1 h with 100 mL NDF solution, ~2 g sodium sulphite (Merck) and 50  $\mu$ L octanol (Merck). The residue was rinsed with hot water and acetone. The crucibles were dried ( $P_1$ ) and calcinated ( $P_2$ ).

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