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### ▶ To cite this version:

Chantal C. Brossard, Maud Petitot, Cecile Barron, Marie-Anne Legoux, Evelyne Paty, et al.. Changes in pasta protein networks induced by drying and their relationship to protein digestibility and allergenicity. 4. International Symposium on Molecular Allergology, Oct 2010, Munich, Germany. 2010. hal-02818282

### HAL Id: hal-02818282 https://hal.inrae.fr/hal-02818282

Submitted on 6 Jun2020

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# Changes in pasta protein network induced by drying and their relationship to protein digestibility and allergenicity

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Pasta is a popular food which possesses interesting nutritional quality but may trigger allergic reaction in sensitized people. Many questions remain open for research area, including the relationship between pasta processing, pasta structure and resulting nutritional properties. The purpose of this study was to characterise the structure of pasta dried at different conditions and to relate it to the in vitro digestibility and allergenicity of proteins. Four drying profiles were studied: Low Temperature 55°C (LT), High Temperature 70°C(HT), Very High Temperature 90°C applied either from the beginning of the cycle, when the moisture content of spaghetti was high (20%) (VHT) or at the end of the drying cycle, when the moisture content of pasta was low (12%) (VHT\_LM).

#### **Methods**

- Proteins: Size Exclusion HPLC (SE-HPLC) after protein extraction with SDS (detergent), then with DTE (reducer) in dried and cooked pasta (Fig.1)
- Microstructure: Confocal Laser Scanning Microscope of cross sectioned cooked pasta after protein staining with fuchsine acid (Fig. 2).
- Protein digestibility : In vitro digestion of cooked pasta composed of a buccal phase ( $\alpha$ -amylase, pH7), a gastric phase (pepsin, PH2) and an intestinal phase (pancreatin, pH7). Protein hydrolysis was evaluated by measuring the increase in free amine groups in protein extracts (Fig. 3).
- Protein allergenicity : Juices from in vitro bucco-gastric or pancreatic digests were used to inhibit recognition of wheat proteins by IgE from a pool of allergic patients (table 1).

#### **Microstructure of cooked Pasta**



- Fig.2: Example of CLSM images of LT cooked pasta at the central (A), intermediate (B) and external (C) zones of pasta strand.
- ⇒ Localisation effect: External zone: looser protein network with swollen starch granules: creation of a moisture gradient during cooking

 $\Rightarrow$  **Drying profile:** LT  $\neq$  other drying profiles

### Allergenicity of Digests from Cooked Pasta

Table 1. Competitive ELISA with digestion juices from cooked pasta and a pool of sera from allergic patients to wheat. Percentage of inhibition obtained with digestion juices at the end of the gastric phase (5 minutes by  $\alpha$ -amylase and 3 hours by pepsin) and at the end of the intestinal phase (end of gastric phase + 3 hours by pancreatin) are presented. Distinct letters by protein indicate significant difference between mean values(p<0.05)

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	Digestion juice from the end of	LT	НТ	VHT	VHT_LM	
<b>α</b> -gliadin	gastric phase	66 <sup>a</sup>	64 <sup>a</sup>	44 <sup>b</sup>	40 <sup>b</sup>	
	intestinal phase	16 <sup>cd</sup>	14 <sup>cd</sup>	10 <sup>d</sup>	19 <sup>c</sup>	
γ-gliadin	gastric phase	88 <sup>a</sup>	79 <sup>bc</sup>	80 abc	81 <sup>ab</sup>	
	intestinal phase	78 <sup>bc</sup>	75 <sup>bc</sup>	72 <sup>c</sup>	77 <sup>bc</sup>	
Low MW glutenins	gastric phase	93 <sup>a</sup>	91 <sup>ab</sup>	90 <sup>ab</sup>	87 <sup>b</sup>	
	intestinal phase	51 <sup>d</sup>	66 <sup>c</sup>	46 <sup>d</sup>	63 <sup>c</sup>	
Albumin/ globulin fraction	gastric phase	28 <sup>a</sup>	26 <sup>ab</sup>	24 <sup>ab</sup>	24 <sup>ab</sup>	
	intestinal phase	14 <sup>c</sup>	16 <sup>c</sup>	14 <sup>c</sup>	21 <sup>b</sup>	

- $\Rightarrow$  Wheat fraction IgE reactive peptides from different wheat fractions are present in all tested digestion juices.
- $\Rightarrow$  **Digestion step**: digests from gastric phase are richer in IgE reactive peptides than those from intestinal phase, difference depends on wheat fraction
- $\Rightarrow$  **Drying process**: compared to LT process, increasing the drying temperature led to a reduction in gastric digests and an increase in intestinal digests of IgE reactive peptides, specially for VHT\_LM.



Fig. 1: Peak surfaces of SE-HPLC elution profiles of SDS-soluble, DTE-soluble and calculated unextractable protein fractions in semolina and dried pasta (A) and in cooked pasta (B)

#### Dried pasta are different Cooked pasta are similar but

Increasing drying temperatures  $\rightarrow$ led to increased protein aggregation (lower protein solubility in SDS).

protein aggregation, especially for LT and HT pasta Aggregation during cooking

 $\Rightarrow$ Aggregation probably occurred through disulphide bonds (increased DTE-soluble fraction) and through other covalent bonds (presence of insoluble proteins) with VHT drying profiles.

 $\Rightarrow$  Bucco-gastric phase

 $\Rightarrow$  Intestinal phase

to other drying processes.

VHT\_LM

drying profile (data not shown)

drying

probably occurred through disulphide bonds (increase in DTE-soluble fraction) and other covalent bonds (increase in insoluble fraction)

 $\Rightarrow$  Cooking led to an increased

#### **Protein digestibility in Cooked Pasta**



VHT\_LM decreased significantly protein digestibility (by 10 %) and increased allergenicity of intestinal digests. This could not be explained by a different protein spatial distribution at a microscopic level. VHT LM cooked pasta presented a higher proportion a high molecular weight protein aggregates (data not shown) which may have contributed to this lower digestibility and higher allergenicity. Both gluten and soluble proteins seemed to be involved. It appears that applying VHT at the end of the drying cycle led to the formation of specific protein aggregates.

Acknowledgement: PASTALEG was carried out within the framework of Programme National de la Recherche and financially supported by the Agence Nationale de la Recherche.

**Diapositive 1** 

**d3** remplacer 20 ppm gluten par 20 ppm dj161659; 01/09/2009