ADDITION OF FIBERS IN FRANKFURTERS MODIFIES THE READY-TO-SWALLOW FOOD BOLUS PROPERTIES AND ORAL BIOACCESSIBILITY OF NUTRIENTS AFTER IN VITRO MASTICATION

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Abstract – This work was designed to study the effect of fibers addition in frankfurters on food bolus characteristics after mastication and on oral release of nutrients. Frankfurters were prepared with addition of fibers (*psyllium* and tapioca starch) in reduced-fat formulations. Food boluses were prepared during normal mastication programmed on a mastication simulator (AM2). After liquid-solid phase separation, boluses were analyzed for physical (granulometry, rheological behavior) and chemical (protein/lipid oxidation, iron/peptides release in bolus liquid phase) properties. Results showed that physico-chemical characteristics of food bolus after mastication depended on presence and fiber type. Matrix disruption during chewing is more important with fiber addition but significance was observed for *psyllium* fiber only. The bolus of *psyllium*-enriched frankfurter was softer, less cohesive than no-fiber and tapioca starch-enriched ones. Iron release from food matrix was significantly lower and peptide release significantly greater in frankfurters containing *psyllium* fibers compared to no-fibers and tapioca starch-enriched ones. In presence of fibers, liquid part of boluses contained fewer particles, which were of larger diameter with tapioca starch. Fibers used as fat-replacers in frankfurters formulation lead to significant changes in food bolus characteristic after mastication.

Key Words – fat reduction, mastication, nutrients biochemical change and release

I. INTRODUCTION

Reduction of fat content in meat products is currently an important strategy to produce healthy foods. A number of studies have already been conducted to improve the fat profile of frankfurters with modification of lipids in formulation or replacement by oils, hydrocolloids, protein complexes or seaweeds [1]. Nevertheless, the analysis of impact of these formulation strategies on technological characteristics and sensory evaluation had never been extended to the masticatory process and bolus formation during consumption although this is an important step of mouthful transformation before digestion.

Dietary fibers are known for a long time of having a multitude of physiological effects including protection against several chronic diseases [2]. Psyllium fibers are a natural polysaccharide used for promoting the regulation of the large bowel and reducing blood cholesterol [3]. Tapioca starch is an efficient fat substitute and contains some resistant starch, hence could be considered as dietary fiber [4].

The purpose of this work was to analyze how the presence of two kinds of fibers (*psyllium* and tapioca starch) in the frankfurter formulation associated with fat reduction may have implication in the masticatory process, the food bolus formation and on free-iron and peptides bioaccessibility after the oral phase of food transformation.

II. MATERIALS AND METHODS

Frankfurters processing

Pork frankfurters were prepared with pig muscle (triceps brachii) and fat (backfat and throat fat) obtained from a local market. Sodium nitrite, lactose, spices, freeze-dried pig plasma and *psyllium* fibers or tapioca starch (Solina group, Bréal-sous-Montfort, France) were used. Prior to the cuttering step, visible fat and connective tissue were trimmed from the meat, and lean meat and fat were separately ground through a 4.5 mm plate and stored at 0°C ± 1°C. Minced meat was added into a chilled bowl cutter and homogenized for 1

min. Then sodium nitrite, spices and lactose were added and homogenized for 1 min. The plasma solution and crushed ice were then added and the mixture was homogenized for another minute. Finally, the fat and *psyllium* fibers or tapioca starch were added. Then all ingredients were mixed until a final temperature of 12°C. Each meat batters were stuffed by a piston stuffer into a 22-24 mm diameter sheep gut. Sausages were hand-linked at nearly 10 cm intervals. They were pre-cooked in a steam room for 15 min at 50°C, and then 60 min at 60°C. Then they were steamed using a water-bath at 67°C during 40 min. Finally sausages were cooled in an ice water-bath for 1 h. They were vacuum-packed and stored at 4°C ± 1°C until analysis.

Masticatory experiments

In vitro masticatory experiments were performed with a masticatory apparatus [5, 6] Boluses were collected after 27 masticatory cycles which was the mean number of chews observed *in vivo* in normal mastication to prepare a ready-to-swallow bolus of frankfurters. After *in vitro* mastication, the food bolus was immediately collected and solid and liquid phases were separated by centrifugation (2000 rpm) and characterized both for physical and biochemical properties.

Characterization of solid phase of food bolus

Particle size distribution (PSD) in boluses was obtained by manual sieving after drying (30 min at 37°C) onto a stack of sieves (10, 7.1, 6.3, 4, 2.5, 2, 1.4, 1, 0.8 and 0.4 mm aperture). Particles retained on each sieve were weighted. The median values of particle size of boluses (d50) were extracted from PSD curves. For rheological analysis of the solid phase, a Texture Profile Analysis (TPA) test was carried out with an Instron machine equipped with a flat piston head and a cylindrical cup. The food bolus was submitted to a double compression at a deformation of 65% of its initial height realized at 50 mm/min. Elasticity, cohesiveness and hardness were extracted from the force-deformation curves.

One g of solid phase of bolus was homogenized in 10 mL KCl 0.15 M + BHT 0.1 mM in a Ultra Turrax system. For lipid oxidation, the homogenate (0.5 mL) were incubated with 1% (w/v) 2-thiobarbituric acid in 50 mM NaOH (0.25 mL) and 2.8% (w/v) trichloroacetic acid (0.25 mL) for 10 min in a boiling water bath. After cooling at room temperature for 30 min, the pink chromogen was extracted with n-butanol (2 mL) and its absorbance was measured at 535 nm against a n-butanol blank. TBA-RS concentrations were calculated using 1,1,3,3 tetraethoxypropane (0-0.8 µM) as standard, and expressed as mg of MDA per kg of meat (TBA units). For protein oxidation, carbonyl groups were detected by reactivity with 2.4-dinitrophenylhydrazine (DNPH) leading to formation of protein hydrazones. Carbonyl content was expressed as nanomoles of DNPH fixed per mg of protein. Schiff bases were measured by fluorescence front face using an excitation wavelength of 380 nm and an emission wavelength of 475 nm.

Characterization of liquid phase of food bolus

Particles in liquid phase were analyzed by laser granulometry using a Sysmex FPIA-3000 flow particle image analyzer (Malvern Instruments Ltd, UK). The number of particles was measured, and particle size and form were evaluated by equivalent circle diameter (EC Diameter), circularity, and Feret aspect ratio. Free iron was measured using the ferrozine formation [7] and peptides release was quantified with the optical density measured at 280 nm.

Statistical analysis

Statistical analysis was performed using Statistica software. After verification of data normality, Analyses of Variance were carried out to test if differences exist in measured variables between boluses collected after mastication of the different Frankfurters (no-fiber, psyllium-enriched and tapioca starch-enriched). When a significant difference was observed ($p<0.05$), a multiple means comparison was realized with the Student Newmann-Keuls-test.

RESULTS AND DISCUSSION

Characterization of solid phase of food bolus

The addition of fibers in the frankfurter formulation increased the disintegration level observed in the swallowable food in normal masticatory conditions. P*syllium* fibers added in the matrix lead to a significantly greater proportion of small particles illustrated by a smaller value of median particle size (d50 of 5.01

mm for no-fiber frankfurter and 4.43 mm for frankfurter enriched in *psyllium* fibers; p=0.0103). The addition of tapioca starch fibers also decreased d50 value of boluses after mastication but not significantly (d50=4.77 mm).

Rheological characteristics of boluses were significantly modified by the addition of *psyllium* fibers. The swallowable bolus of *psyllium*-enriched frankfurter was softer and less cohesive than the others (p<0.05; Fig 1). The addition of tapioca starch lead to a more elastic bolus (p<0.05; data not shown).

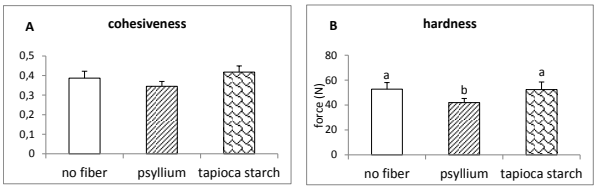

Figure 1: Values (mean and SD) of cohesiveness (A) and hardness (B) extracted from force-displacement curves obtained from Texture Profile Analysis tests performed on bolus collected after complete mastication of Frankfurter containing no-fiber, *psyllium* or tapioca starch fibers. Different small letters indicate a significant difference between boluses (p>0.05).

The effect of chewing on the oxidative parameters of frankfurters formulated with or without fibers showed no increase of oxidation for the carbonyles content (data not shown). For lipid oxidation an increase up to 30% was observed with frankfurter enriched with *psyllium* fibers. However this oxidation remains below the threshold of detection by a panel (MDA 0.5mg per kg of meat, [8]).

Characterization of liquid phase of food bolus

Adding fibers (*psyllium* or tapioca starch) did not modify the release of free iron compared to the no-fiber control. However, the free iron in the liquid phase was lower in frankfurter with *psyllium* compared to that with tapioca starch (Fig 2A). The possible chelation of iron by these polysaccharides that are both soluble and non-soluble can explained this difference. The literature did not mentioned any effect for iron, but the fact that solid meals enriched with psyllium fiber strongly modified postprandial signals arising from the GI tract. Adding *psyllium* fibers increased significantly the OD at 280 nm. This result showed that the release of peptides was favored in frankfurters formulated with *psyllium* (Fig 2B).

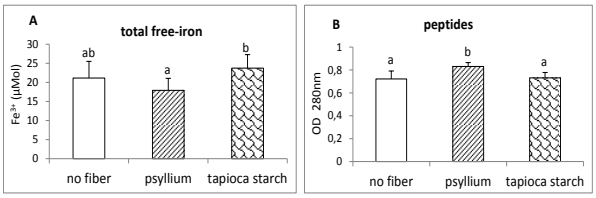

Figure 2: Mean values (and SD) of total free iron (A) and peptides (B) determined in liquid phase of boluses ready to swallow after mastication of different Frankfurters (no-fiber, *psyllium*- and tapioca starch-enriched). Different small letters indicate a significant difference between boluses (p<0.05).

On the liquid phase of the bolus, granulometry parameters of particles were analyzed: number of particles, circularity, diameter, ferret ratio (Fig 3). Addition of fibers (*psyllium* and tapioca starch) reduced the number of particles in the liquid phase of the bolus. In the bolus containing *psyllium* fibers the circularity was higher than that without fiber (0.9045 *vs* 0.8947). In contrast the ferret ratio (lengthening of the particle) did not change according to the formulation. Frankfurters with tapioca starch showed an average particle diameter greater than with *psyllium* or without fibers (8.57µm vs 8.23µm). This increase is possibly due to the specificity of tapioca starch granules. Indeed during heating, tapioca starch granules start to gelatinize and the diameter can increase from 4 µm to 35µm. In our work, the particle diameter varied between 5 and 37µm for tapioca starch granules (data not shown).

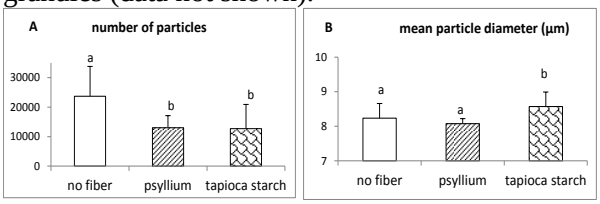

Figure 3: Mean values (and SD) of the number of particles (A) and the mean particle diameter (B) in the liquid phase of boluses ready-to-swallow after mastication of different frankfurters (no-fiber, psyllium-enriched and tapioca starch-enriched). Different small letters indicate a significant difference between boluses from different Frankfurters (p<0.05).

Discussion

The study of the impact of the formulation of emulsions, including the type of added fiber (*psyllium* or tapioca starch) compared to fiber-free sausage, was performed on the food bolus ready for swallowing. The boluses from *psyllium*- or

tapioca starch-enriched frankfurters had different physical and biochemical characteristics from that without fiber.

Firstly, the addition of fibers lead to a reduction in the number of particles in the liquid phase of the bolus. This is possibly due to retention of the particles in the solid phase by the gel formed by the fibers or starch.

For the formulation with tapioca starch, the bolus was at the time of swallowing more elastic, more cohesive and less sticky, and presented higher level of protein oxidation in the liquid phase as well as more oxidized free iron. This could be explained by the structure of the starch, composed by a succession of potentially oxidizing reducing sugars [9]. The bolus containing *psyllium* fiber has median particle size and was stickier, less hard and less cohesive than the control without fiber. The structure of the emulsion seemed to have an impact on the dispersion of the fractures within the matrix. In the same way, more peptides were released due after chewing, possibly due to a greater degradation of the matrix at the physical level.

Biochemically, the bolus containing psyllium fibers has significantly more peptides that the one without fiber, which might be due to the greater disruption of the matrix. It also presents more lipid oxidation that the fiber-bolus and the more numerous Schiff bases (greater accessibility of proteins, fats or sugars). Those related to the circularity of the particles from the liquid phase could allow to assume that the addition of psyllium fiber in the matrix involves a greater release of lipids during chewing. But nothing allows us to affirm this hypothesis. It would, for this measure the lipids in the liquid phase of the bolus and not only oxidation.

III. CONCLUSION

This work points up for the first time that the addition of fibers as fat-replacers in frankfurters formulation lead to significant physical et chemical changes in food bolus characteristics and different nutrient oral release due to mastication. Despite the low percentage of added fiber (about 3%) these have a significant influence on the physical and biochemical characteristics of the sausage and the bolus. These fibers therefore appear to be adapted to the formulation of new products.

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