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Short communication

Digestibility of pasta made with three wheat types: A preliminary study

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ABSTRACT

The aim of this study was to assess the digestibility of the protein and starch in pasta made with different cereals, i.e. *Triticum durum*, *Triticum polonicum* and *Triticum dicoccum*, and to measure the glycemic index (GI) of the different types of pasta.

The digestibility of the starch in *T. polonicum* pasta differed significantly from the others. It seemed to be less digested than *dicoccum* and *durum* wheat pasta. *T. polonicum* pasta also had a lower glycemic index, while there were no significant differences in the protein digestibility of the three types of pasta.

1. Introduction

Pasta is a wheat-based food consumed worldwide for its flavour, low cost and nutritional value. Durum wheat semolina (*Triticum turgidum* ssp. *durum*) is the preferred and most often used raw material for the production of good-quality pasta.

Pasta processing, and especially the temperature at which it is dried and the subsequent cooking process, gives the end product its rheological characteristics due to starch gelatinization and protein coagulation (mainly gluten proteins). Industrial production (drying and extrusion in particular) can modify the pasta's structure, influencing starch digestibility and protein hydrolysis (De Zorzi, Curioni, Simonato, Giannattasio, & Pasini, 2007; Petitot et al., 2009). The protein network depends strictly on the drying temperature and influences the action of the proteolytic enzymes due to the formation of highly-aggregated proteins linked by covalent bonds (De Zorzi et al., 2007; Petitot et al., 2009).

It is well known that a more compact structure of the pasta, due to starch granules being trapped in a strong protein network, can reduce or delay access by amylolytic enzymes, thereby delaying starch digestion (Fardet et al., 1999). The pasta protein network depends strictly on the protein content (Del Nobile, Baiano, Conte, & Mocci, 2005). and the drying temperature and influence the action of the proteolytic enzymes due to the formation of highly-aggregated proteins linked by covalent bonds (De Zorzi et al., 2007; Petitot et al., 2009).

Pasta's structure may play an important part in determining the rate of starch digestion, making it more or less readily digestible (Monge, Cortassa, Fiocchi, Mussino, & Carta, 1990), and influencing the glycemic response to its ingestion (Björck, Liljeberg, & Ostman, 2000).

Many microstructural factors affect the rate of starch digestion, including the size of the granules and the surface area accessible to digestive enzymes (Petitot, Barron, Morel, & Micard, 2010), the granules' encapsulation by fibres (Brennan, Blake, Ellis, & Schofield, 1996) and proteins (Petitot et al., 2010), and the physical properties of the starch, such as the degree of gelatinization and the amylose/amylopectin ratio (Petitot et al., 2009). Microscopic studies have been conducted on the starch gelatinization rate during cooking (Khongsak, Furst, Ashton, & Hosken, 2006). The structure of the starch in processed foods, such as bread and pasta, and its disruption during amylolytic digestion following physical degradation as well as the protein network surrounding the starch granules has also been studied at a microscopic level (Autio & Salmenkallio-Marttila, 2001; Cunin, Handschin, Walther, & Escher, 1995).

Nowadays, there is a renewed interest in using ancient wheat species, such as *Triticum monococcum*, *Triticum dicoccum* (Acquistucci, Aureli, Codiann, Colonna, & Galterio, 2004), *Triticum polonicum* (Gauthier, Gélinas, & Beauchemin, 2006) and *Triticum aestivum* ssp. *spelta* (Abdel-Aal & Rabalski, 2008) to make pasta or baked products. Despite the numerous studies conducted to assess protein and starch digestibility in baked and extruded products (De Zorzi et al., 2007; Englyst, Vinoy, Englyst, & Lang, 2003; Petitot et al., 2009), there is still little information on the digestibility of foods made from ancient wheats.

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T. dicoccum seems to have a hypoglycemic property due to a slower starch digestibility than common wheat (Mohan & Malleshi, 2006). Starch digestion index of spelt biscuits and bread proved similar to those of *T. aestivum* products (Abdel-Aal & Rabalski, 2008). Marques et al., 2007 found no significant difference in the glycemic index of *T. aestivum* ssp. *spelta* and *T. aestivum* breads. In contrast, Bonafaccia et al., 2000 found bread made with spelt wheat more digestible than bread made with common wheat (*T. aestivum*).

The present study focused on pasta made with three types of wheats namely *Triticum durum* (typical for modern pasta), *T. dicoccum* and *T. polonicum* (which is present on the market with the commercial name of Kamut®). The aim of the paper was to determine their starch and protein digestibility based on *in vitro* and *in vivo* assays.

2. Materials and methods

2.1. Materials

Commercial pasta samples (spaghetti) prepared with *T. durum* (durum wheat), *T. polonicum* (Kamut®) and *T. dicoccum* ("Farro"), supplied by the same Italian manufacturer, were used. All samples, produced with refined semolina from organically cultivated wheats, were processed in identical technological conditions. Briefly, pasta samples were obtained by mixing water and semolina (particle size mainly from 200 to 300 μm) to form a dough, that was then extruded to give the pasta the required shape. According to the technology commonly used in Italy, shaped pasta was subjected to a brief pre-drying step in a tunnel at 80 °C (air temperature) to reach a water content to about 23%. Then predried pasta was rapidly transferred to the dryer and treated with decreasing air temperatures (from 90 °C to 60 °C) to reduce the moisture content to 12.5% (about 8 h). Finally, pasta was air-cooled to room temperature. The "Resistant starch kit" (K-RSTAR 05/2008) was from Megazyme (Milan, Italy).

The glucometer and "Accu-chek, Aviva" were supplied by Roche Diagnostics (Monza, Milan, Italy).

2.2. Chemical composition of cooked pasta

Protein, starch and ash content were quantified in freeze dried cooked pasta samples.

Protein was quantified using the Kjeldahl method (AOAC 976.05, 2000), the total starch content was measured according to AOAC 996.11 method and the ash were quantified according to AOAC method 923.03 (AOAC, 2000).

2.3. Cooking parameters

Spaghetti samples were cooked in deionised boiling water in a ratio 1:10. The "al dente" cooking time was determined by observing the disappearance of the white core of the pasta squeezed between two glass plates, according to AACC 16-50 method (AACC International, 2000).

2.4. Light microscopy

Short lengths of uncooked and cooked pasta were inserted lengthwise into the chuck of an ultramicrotome (Ultracut E, Reichert-Jung). Using a glass knife, the surface of the cross-section of the pasta was planed away until a polished mirror-like surface was obtained. Sections of pasta (nominally 2–3 μm thick) were planed from this polished surface, mounted in a drop of distilled water on a microscope slide and covered with a cover slip. These unstained sections were then examined and photographed using bright field microscopy, using an Olympus BX60 (Olympus, Japan) microscope with ProgRes® Capture Pro 2.1 software (Jenoptik, Germany). The autofluorescence in bran tissue was recorded using the UV filter cube (U-MWU, exciter filter BP330-385, barrier filter BA420) of the microscope. Further sections were stained with diluted iodine in potassium iodide (Lugol's solution) to highlight starch, amylose and amylopectin, and examined and photographed using bright field microscopy.

2.5. In vitro cooked pasta protein digestion

In vitro protein digestion was performed as described by Pasini, Simonato, Giannattasio, Peruffo, & Curioni (2001). After being cooked, pasta samples, were freeze dried and powdered by grinding in a mortar. 60 mg of each powdered sample were suspended in 4 ml of 0.2 N HCl (pH 2.2) containing 0.05 mg/ml of pepsin Fluka). After 30 min, 1.15 ml of 1 M boric acid, 0.5 N NaOH, adjusted to pH 6.8 with 5 N HCl and containing 0.25 mg/ml of pancreatin (Sigma), were added; the resulting pH was 7.6. The reactions were performed at 37 °C in a shaking water bath and stopped at various times (after 0, 15, 30 min of pepsin attack, and 30, 60, 90 and 150 min of pancreatic digestion) by adding 1 volume of 20% (w/v) trichloroacetic acid. After standing for 1 h, samples were centrifuged (6000*g*, 10 min) and pellets were analysed for protein content according to the Hach, Brayton, & Kopelove, 1985. Nitrogen was quantified using the AOAC method 976.05 (AOAC, 2000). For each pasta sample, the tests were performed in triplicate.

2.6. In vitro cooked pasta starch digestion

Starch digestion was measured *in vitro* with a modified version of the method used by Kim et al. (2008). 1 g of minced cooked pasta sample was placed in four separate glass vials, to measure starch digestion at four different time points (20, 120, 150 and 180 min). The samples underwent 30 min of incubation with pepsin (Fluka) to simulate the gastric phase. Then the pH was adjusted to 7.2 and 4 ml of (10 mg ml^{-1}) pancreatic amylase (Megazyme, Ireland) were added to obtain starch digestion. The samples were incubated at 37 °C in a shaking water bath, for 20, 120, 150 and 180 min and digestion was stopped by adding a volume of ethanol. The digesta were centrifuged at 5000*g* for 10 min at 8 °C and supernatants were diluted to 100 ml with 100 mM of sodium acetate buffer pH 4.5. A 100 μl aliquot was treated with 10 μl of α-amylase and amyloglucosidase (AMG, 300 U ml^{-1}) and incubated at 50 °C for 20 min. Then 3 ml of glucose oxidase/peroxidase reagent (GOPOD, Megazyme, Ireland) were added and the samples were incubated for 20 min at 50 °C before measuring the absorbance. The glucose released during digestion was quantified as starch hydrolysed during the given time period, multiplying the absorbance by 0.9.

2.7. In vivo glycemic response

The *in vivo* glycemic response was assessed in 10 healthy volunteers, i.e. 3 males and 7 females, aged from 24 to 34 years old, with a mean body mass index (BMI = kg/m^2) of 24 ± 3. All participants gave their informed written prior consent to the experiments and a standard protocol (Wolever, Jenkins, Jenkins, & Josse, 1991), accepted by the FAO/WHO (1998), was used to assess their glycemic response. Briefly, they attended 4 morning sessions after fasting overnight. They were fed with a portion of white bread (reference food) or *T. durum*, *T. polonicum* (Kamut®) or *T. dicoccum* pasta (test foods), containing 50 g of carbohydrates as determined using the official AOAC method 996.11 (AOAC, 2000). Blood