Arabinoxylan content and grain tissue distribution are good predictors of the dietary fibre content and their nutritional properties in wheat products

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Abstract

Wheat millstreams and wheat-based foods (pasta, biscuits and bread) enriched or not in dietary fibre with fractions extracted from wheat grains, have been characterized either for their total dietary fibre content (TDF) and their arabinoxylan (AX) content. A strong correlation (r2 = 0.98) is observed between the AX and TDF contents indicating that AX can be used to estimate TDF content in wheat products. Moreover, by adding a previous step including enzymatic hydrolysis with a xylanase, a functional evaluation of DF is proposed based on the amount of AX released by the enzyme. Xylanase hydrolysable AX are likely also released by microbiota's enzymes in the gut and therefore an indicator for the proportion of fermentable DF in grain fractions and wheat-based foods (pasta, biscuits and bread). This assay opens the door for simple characterization of qualitative attribute of cereal DF.

Keywords:

bread; biscuits; pasta; dietary fibre; enzymatic hydrolysis; xylanase

Abbreviations

AX: arabinoxylan; AX_{enz}: arabinoxylan released by xylanase; DF: dietary fibre; TDF: total dietary fibre; HM_wDF: high molecular weight dietary fibre; IDF: Insoluble dietary fibre; LM_wSDF: low molecular weight soluble dietary fibre; HM_wSDF: high molecular weight soluble dietary fibre

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1. Introduction

Epidemiological studies have shown that high dietary fibre (DF) diets and whole grain consumption are associated with diminished risk of coronary heart disease, colon cancers, inflammatory bowel disease, and metabolic syndrome (Stephen et al., 2017). Thus, health authorities around the world recommend that adults consume 25 g to 35 g of fibre per day (Stephen et al., 2017). Bread, pasta and non-bread product such as wheat flour tortillas are staple foods in many countries and wheat flour is the main component of many food products such as pizzas, biscuits and cakes. Obviously, an increase in the DF content of these wheat-based foods might help to better meet dietary recommendations for DF intake. However, DF are a complex mixture of chemically heterogeneous carbohydrate components, and the quantity and nature of DF in wheat-based foods vary, in particular, depending on the "refining" of the flours (white flour vs whole grain flour) and the food manufacturing processes that can affect for example the amount of resistant starch. Although some of the physicochemical properties of DF (solubility, viscosity, bulking) are clearly related to important nutritional outcome (McRorie & McKeown, 2017), the mechanisms by which the consumption of DF modulates health are multiple and not fully elucidated. However, their role in maintaining diversity and stability of the microbiota probably plays a decisive role in the regulation of the immune and metabolic systems and therefore on health (Sonnenburg & Bäckhed, 2016). In this respect, a key feature of DF is their fermentation pattern which is related to their chemical structure and to the ability of microorganisms to produce the glycosyl-hydrolases allowing their degradation in the gut (Flint, Bayer, Rincon, Lamed, & White, 2008).

As previously stated, DF are a complex mixture of chemically heterogeneous carbohydrate components and their measurements have been based on sequential enzymatic-gravimetric assays mimicking human digestion. These methods have undergone numerous evolutions (Macagnan, da Silva, & Hecktheuer, 2016) but the AOAC method 985.29 (Prosky, Asp., Schweizer, DeVries, & Furda, 1988) was widely used for the determination of total dietary fibre (TDF) in foods. Prosky's method distinguished soluble DF from insoluble DF but did not include the low-molecular-weight non-digestible oligosaccharides and neglected part of the resistant starch. Integrated methods were therefore developed for the determination of TDF, including non-digestible oligosaccharides and subsequently resistant starch by adopting physiological conditions for the removal of starch. The AOAC method 2009-01 (McCleary et al., 2010) and its extension the AOAC method 2011.25 (McCleary et al., 2012) measures the fraction of high molecular weight dietary fibre (HMwDF) by enzymatic–gravimetric techniques, and low molecular weight soluble dietary fibre (LMwSDF) by high-performance liquid chromatography (HPLC). HMwDF are actually formed of insoluble dietary

fibre (IDF) and high molecular weight soluble dietary fibre (HMwSDF); HMwDF = IDF + HMwSDF. An evolution of the AOAC 2009.01 method (McCleary, 2014) was introduced to correct an overestimation of the LMwSDF fraction in starch-rich foods and particularly in cereal-based foods (Brunt & Sanders, 2013; McCleary, Sloane, & Draga, 2015). Despite their successive improvements, these methods made of several steps are time-consuming and results obtained are still under debate (Hell, Kneifel, Rosenau, & Böhmdorfer, 2014). Moreover they do not take into account the chemical and physico-chemical diversities of DF that is far more complex than proposed by a classification in HMwDF/LMwSDF and insoluble/soluble fractions (Gidley & Yakubov, 2019).

In view of the diversity of DF and of their mechanisms of action, reliable method to quantify their amount is crucial to characterize cereal products. Such method could help to monitor the development of DF enriched wheat-based foods and could also help to better assess some of their important nutritional property such as their fermentation ability. The wheat grain contains about 12–14% of DF (Gebruers et al., 2008) but this complex organ is composed of different tissues composed of cell wall polysaccharides exhibiting various properties and composition (Barron, Surget, & Rouau, 2007; Saulnier, Guillon, & Chateigner-Boutin, 2012). In wheat grain, arabinoxylans (AX) are the major polymers in the cell wall and therefore a major component of DF. In the starchy endosperm cell walls represent 3 to 5% of the tissue and are essentially constituted by AX with a lower proportion of (1,3)(1,4)-linked beta-d-glucans (Saulnier et al., 2012) and cellulose (Gartaula et al., 2018). In this tissue, AX are partly water soluble and exhibit an arabinose to xylose ratio (A/X) of about 0.6. In the most outer layers cell wall polysaccharides represent up to 50% of the tissue and are mainly composed of cellulose and complex AX that are mostly water insoluble and exhibit a structure more heavily substituted by arabinose than in starchy endosperm (A/X close to 1) (Antoine et al., 2003; Barron et al., 2007; Parker, Ng, & Waldron, 2005). In addition, hydroxycinnamic acids that are ester-linked to some of the arabinose side-chains of AX (Harris & Trethewey, 2010) play an important role for polymer interactions and cell wall properties, as they can dimerize under oxidizing conditions and act as bridging agent interconnecting AX chains together and with lignins (de Oliveira et al., 2015). The susceptibility to enzyme attack is clearly related to AX structure (Bonnin et al., 2006) and is deeply impacted by the interactions of AX within the cell wall. Outer tissues of the grain (pericarp) are not degraded by pure microbial xylanases (Beaugrand, Crônier, Debeire, & Chabbert, 2004; Ordaz-Ortiz, Devaux, & Saulnier, 2005), whereas AX from starchy endosperm or aleurone layer are extensively hydrolysed by these enzymes (Ordaz-Ortiz et al., 2005). The resistance is mainly explained by the covalent interactions through ferulic acid bridges between AX chains and with lignin. These interconnections are much more numerous in the outer pericarp that is resistant to enzyme attack than

in cell walls of endosperm tissues (including aleurone layer) (Chateigner-Boutin et al., 2018; Saulnier et al., 2012) that are degraded by microbial xylanases. Actually, the ability to be degraded by microbial enzymes (e.g. microbiota) differs largely between isolated polymers and polymers as present in the cell wall of the different tissues (Rose, Patterson, & Hamaker, 2010). The susceptibility to enzyme degradation of the different tissues of the grain has potentially important health impact as isolated AX polymers and arabino-xylo-oligosaccharides (AXOS) have been shown to favour the development of beneficial bacteria (Broekaert et al., 2011; Neyrinck et al., 2011) with an increase in Bifidobacteria/Lactobacilli as well as in Bacteroides/Prevotella and Roseburia groups (Despres et al., 2016)

In this work, the tissue composition of different millstreams has been measured (Hemery et al., 2009) and compared to their total DF amount measured with the AOAC 2009-01 method. Assuming that the most peripheral tissues are mainly constituted of DF, the tissue composition could be used to estimate the total DF content in millstreams. However, such approach could not be extended to wheat-based foods. Therefore, knowing that AX are the main components of cell walls in the different tissue of the wheat grain, biochemical determination of AX content has been investigated as a possible predictor of DF content. In addition, adding an enzymatic hydrolysis step with a xylanase and quantifying the amount of AX released was carried out in order to evaluate the potential fermentation capacity of various wheat millstreams.

2. Material and Methods

2.1 Wheat grains and milling fractions

Different millstreams were produced from bread wheat (Caphorn cv; *Triticum aestivum* L.) and durum wheat (Miradoux cv; *Triticum durum* Desf). Fractions were obtained from conventional milling technology with specific diagrams to bread wheat in ENILIA-ENSMIC pilot mill (Surgères, France) or durum wheat in UMR IATE pilot plant (Montpellier, France). In addition, a PeriTech debranner (Satake, UK) was used to obtain debranning fractions differing in their extraction rate. Two specific fractions named F^- and F^+ were also produced from Caphorn grains. Fraction F^- corresponded to a debranning level of 3.8% in weight of the grain, and fraction F^+ was subsequently isolated at a higher debranning level (recovered between 6.4% and 12.9% in weight of the grain). The other debranning fractions (A to J) from both bread and durum wheats were also produced with this equipment. Different debranning levels and successive debranning steps were used in order to obtain fractions differing in their outer tissues content.

2.2 Wheat-based foods

Bread, pasta and biscuit were produced with white flour (regular product) or with flour enriched in DF by incorporating F^- and F^+ fractions.

Délifrance provided breads produced on an industrial line in Marquette-lez-Lille (France). Regular bread (R-bread) was made with white flour. The same flour that incorporated fraction F^- (20.5% weight basis) or fraction F^+ (50.8% weight basis) was used for F^--bread and F^+-bread, respectively. Precooked breads (55g) were used for analysis.

Mondelez provided biscuits produced on a pilot line at its R & D centre (Saclay, France). Regular biscuits (R-Biscuit) contained fat (15%), sugars (23%) and the rest of the formula was completed with cereals (2/3 wheat flour and 1/3 oat flakes) or incorporated fraction F^- (10.5%) or fraction F^+ (27.1%) completed with cereals for F^--biscuit and F^+-biscuit, respectively.

Pasta were provided by Panzani. Pasta was made from 100% durum wheat semolina for regular product (R-pasta) and incorporated fraction F^- (9%) or fraction F^+ (25.9%) for F^--pasta and F^+-pasta, respectively.

2.3 DF content and biochemical characterization

DF content in the different samples was determined according to the AOAC 2009-01 method (McCleary et al., 2010) on a dry matter basis. High molecular weight DF (soluble and insoluble fractions) are reported as HMwDF and low molecular weight soluble DF as LMwSDF, and TDF = HMwDF + LMwSDF. In addition, resistant starch was measured in wheat-based foods according to AOAC method 2002-02. In food, the relative maximum deviation between duplicates was 14%, 30% and 16% for HMwDF, LMwSDF and TDF, respectively. In wheat fractions coefficient of variation between duplicates was 2%, 7% and 3% for HMwDF, LMwSDF and TDF, respectively.

The AX content was calculated from neutral sugar analysis. Prior to neutral sugar analysis or enzymatic degradation, samples were ground in a Freezer/Mill 6700 cryogenic grinder (Spex, USA) then 0.8 g were treated with 5 mL of boiling 80% (w/v) aqueous EtOH for 10 min and the insoluble residue separated by centrifugation (6300 g, 10 min). The procedure was repeated once and the residue finally washed with 5 mL of 95% (v/v) EtOH and again separated by centrifugation (6300 g, 10 min). The supernatant was discarded and the residue dried in two steps: first in an oven at 40°C for 24 h and then in a vacuum-oven over P_2O_5 at 40°C for 24 h.

The neutral sugar composition of samples was identified, after acid hydrolysis, using gas chromatography of alditol acetates (Englyst & Cummings, 1988). Acid hydrolysis of samples was carried out as previously described (L Saulnier, Marot, Chanliaud, & Thibault, 1995): the samples

were pre-hydrolysed with 72% (w/w; 26 M) sulphuric acid for 30 min at 25°C and then hydrolysed into monomers at 100°C for 2 h in 1 M sulphuric acid. Analyses were performed in duplicate and AX content (AX_{tot}) was calculated by summing the amount of arabinose and xylose. The relative maximum deviation between duplicates was 3% or less.

Samples (100 mg) were incubated in water (1 mL) with an endoxylanase (EC 3.2.1.8) from *Trichoderma viride* (Xylanase M1, Megazyme, Ireland) at 40°C for 16 h under continuous stirring. Xylanase amount (2.4 U/mg of AX) was adjusted according to the amount of AX determined in the sample (Ordaz-Ortiz et al., 2005). The reaction mixture was centrifuged (6300 g, 10 min) and 0.7 mL of supernatant was removed and boiled for 10 min to inactivate the enzymes. Neutral sugar content in the supernatant was identified after acid hydrolysis (1 M H_2SO_4, 100°C, 2 h) using gas chromatography of alditol acetates (Englyst & Cummings, 1988). Analyses were performed in duplicate and AX amount released by the enzyme (AX_{enz}) was calculated by summing the amount of arabinose and xylose in the supernatant and expressed as weight % of the starting sample. Coefficient of variations between duplicates were 3% or less.

2.4 Tissue composition

Tissue composition was assessed by the biochemical markers methodology as previously described (Hemery et al., 2009). Reference values were obtained from the chemical analysis of pure dissected tissues except for starch amount in starchy endosperm, which was deduced from starch amount in high purity flour. For Miradoux cultivar, separation between the outer pericarp and the so-called "intermediate layer" could not be achieved by hand dissection. Therefore, both tissues were monitored at the same time by looking at the amount of alkylresorcinols.

3. Results & discussion

3.1 Determination of DF contents in wheat products

The TDF content was measured by the AOAC 2009-01 method and the amounts of HMwDF and LMwSDF are reported in Tables 1 and 2. TDF contents were determined in wheat-based foods (pasta, biscuit, bread) made from white wheat flour and enriched in DF with specific wheat milling fractions (Fractions F+ and F-) and over a wide range of wheat milling fractions obtained by classical milling and by debranning. This set of samples provided a large range of TDF contents ranging from 3% up to 70% in milling fractions. Within wheat-based foods (pasta, biscuits and bread) this range was only 3–15%. The higher contents (TDF > 50%) were obtained for bran and peeling fractions from common wheat and durum wheat that were especially rich in HMwDF. For milling fractions the variation coefficient of HMwDF values (70%) was much higher than for LMwSDF (32%), whereas the

variation coefficients were close for HMwDF and LMwSDF in wheat-based foods, 45% and 35%, respectively.

In the literature most of TDF content were obtained by the AOAC 985-29 method that essentially correspond to the HM_wDF fraction determined with the AOAC 2009-01 method (Stephen et al., 2017), although resistant starch was not fully measured by the AOAC 985-29 method. Resistant starch content was low in our wheat-based foods (Table 1) and was not measured in the milling fractions, as it is known to be very low in unprocessed cereal samples.

TDF values were linearly related to HM_wDF: $TDF_{AOAC\ 2009\text{-}01} = 0.989\ HM_wDF + 3.26$ and very highly correlated ($r^2 = 0.997$) (Figure 1). The value of the intercept is close to the average value of LM_wSDF calculated from the complete set of samples (3.0%). The measurement of the LM_wSDF fraction has been subjected to debate and an evolution of the AOAC 2009.01 method has been proposed to correct an overestimation of the LM_wSDF fraction in starch-rich food (Brunt & Sanders, 2013; McCleary et al., 2015). Using only the data from the wheat-based foods (Table 1; the richer in starch and with overall lower TDF content) the intercept value of the relationship (e.g average LM_wSDF content) was 2.06 while this value went up to 4.05 when only data from millstreams were used (Table 2. Supplementary Figure S1). Slopes and correlation coefficients of the relationships were not changed compared with the complete data set. In millstream fractions, starch content varied a lot compared to wheat-based foods, but the higher value of the intercept is not originating from an overestimation of the LM_wSDF fraction due to starch. It is more likely due to the presence of fructans that represents 2 to 4% of wheat grain. These components are more abundant in bran (Haskå, Nyman, & Andersson, 2008; Verspreet, Dornez, Delcour, Harrison, & Courtin, 2015) than in white flour. Knowing that most of the data found in the literature were obtained with the AOAC 985-29 method corresponding to the values of HM_wDF in this study, the TDF contents were in good agreement with values reported previously for similar wheat products and millstream fractions (Elleuch et al., 2011; Gebruers et al., 2008). Nevertheless values determined for fine/coarse bran were in a high range.

3.2 Dietary fiber content vs Arabinoxylans content

DF are a complex mixture of chemically heterogeneous carbohydrate components. In wheat grains arabinoxylans (AX) are the main polymers of the cell walls and therefore the main component of DF in wheat grain. Total AX contents varied on a quite large range between 1% and 30%, with similar trends within wheat product and millstream fractions as reported above for their TDF contents. Total AX content of wheat grain, and flour were comparable to previous reports (Barron et al., 2007;

226 Gebruers et al., 2008; Henry, 1987; L Saulnier, Peneau, & Thibault, 1995) but again values for
227 coarse/fine brans were in a high range compared to literature.

228 The relationship between total AX content in the samples and TDF (or HM_wDF) content is shown on
229 Figure 2. A strong linear relationship ($r^2 > 0.97$) was established with nearly the same slope close to
230 2 between AX and TDF (TDF=2.15AX + 2.61; r^2=0.975) or AX and HM_wDF (HM_wDF=2.17AX -
231 0.57; r^2=0.970). Using only the data from wheat-based foods (Table 1) or milling fractions (Table 2)
232 had very little impact on the values of the slope, intercept or correlation coefficient of these equations
233 (See supplementary Figure S2). Clearly, AX content corresponds to the HM_wDF part of TDF. It is
234 remarkable to note that, despite differences in AX structure and cell wall architecture within the
235 different tissues of wheat grain, the AX content (simply measured as the sum of arabinose and xylose
236 in the sample) accurately reflects the TDF contents of very different wheat products. Similar
237 relationship between AX content of wheat samples and their dietary fibre content was previously
238 reported: $TDF_{Englyst}$= 1.75AX - 0.71 (r^2= 0.994; 51 wheat samples)(Bell, 1985). In Bell's equation
239 TDF content was determined by Englyst's method, based on the determination of the non starch
240 polysaccharides and lignin content in the sample (Englyst, Wiggins, & Cummings, 1982). The use
241 of a different method for evaluating TDF content likely explains the differences in slope between the
242 equation obtained in this work and by Bell (Bell, 1985).

243 *3.3 Dietary fiber content and tissue distribution*

244 In millstreams, tissue composition was assessed through the biochemical marker methodology
245 (Hemery et al., 2009) (Table 2). Pericarp (outer and inner) is mainly constituted of secondary cell
246 walls (Chateigner-Boutin et al., 2018). The testa is also constituted of secondary cell walls surrounded
247 by a thick cuticular layer, whereas the nucellar epidermis is made of thin cuticular layer, and a tiny
248 layer of low substituted arabinoxylans (Barron et al., 2007). According to the dietary fibre definition,
249 roughly all of these tissues, recovered in the outer pericarp and the so-called "intermediate layer",
250 could be considered as dietary fibres. In the case of the endosperm tissues (the aleurone layer and the
251 starchy endosperm), the TDF content could be approximated by the amount of cell walls in these
252 tissues. In the aleurone layer approximately 45% of the dry mass is recovered in cell walls (Hemery
253 et al., 2009). Using the overall AX content in starchy endosperm or flour (around 2%) (Barron et al.,
254 2007; Gebruers et al., 2008) and the proportion of AX in starchy endosperm cell wall, the amount of
255 cell wall in this tissue was assumed to be 3%. TDF (expressed according to the dry matter) was
256 calculated from tissue composition considering the following equation assuming equal distribution
257 of water within the whole grain tissues:

258 TDF (% dm) = %OP + %IL + 0.45*%AL + 0.03*%SE

The calculated TDF was linearly correlated (r^2=0.87) to the measured TDF with the following relationship $TDF_{calculated}$=1.07 x $TDF_{measured}$ + 0.25 (Figure 3). Regardless of the type of wheat or the millstreams a unique relationship was obtained in the whole TDF range (4-60%).

For a similar TDF content, variation in the proportion of the different grain tissue between milling fractions may affect their degradation by microbial enzymes and therefore their fermentation pattern e.g. aleuronic layer is easily degraded whereas the most outer layers of the grain are resistant to microbial attack (e.g. microbiota).

3.4 AX solubilisation by xylanase in relation with DF content and tissue distribution

The susceptibility to enzyme degradation of the different tissues of the grain has potentially important health impact as isolated AX polymers and AXOS are known to favour the development of beneficial bacteria (Broekaert et al., 2011; Neyrinck et al., 2011). Enzymatic hydrolysis with a xylanase is therefore proposed as a functional evaluation of DF in wheat fractions.

The amount of AX released by xylanase (AX_{enz}) is reported in Table 1 and 2 and relationship with AX content is displayed on Figure 4. The amount of AX_{enz} ranged from 0.7% up to 10% and was poorly related to the total amount of AX (r^2= 0.64; Figure 4B). Especially samples with a high AX content (AX>25%) exhibited clearly large differences in the amount of AX_{enz}. Figure 4A shows that the proportion of AX_{enz} is globaly negatively correlated with AX (TDF) content (r^2=0.57) e.g. the extent of AX solubilisation is lower for brans (see table 2; Figure 4A) and the peeling fractions and the highest for pure starchy endosperm fraction (flour T55 or semolina S3; see table 2). As expected, a better linear relationship (r^2=0.73) was observed between the proportion of starchy endosperm and aleurone layer in a fraction and the proportion of AX released by the enzyme (see Figure S3). Conversely, fractions very rich in outer pericarp tissue are little degraded by enzymes (e.g. fraction F^-, debranning fraction A of common wheat). Therefore, the amount of AX solubilized from different fractions by xylanase treatment may be similar although these fractions may have very different AX (TDF) contents (e.g. F^- fraction vs. F^+ fraction). In other words, the fermentation ability of wheat fractions is not simply related to AX/DF content and fractions with a high AX/DF content that have a lower degree of solubilization by xylanase may have more limited prebiotic effects. Such an assay can help to better understand the nutritional effects of wheat DF, which is not only related to their quantity. The ability of a fraction to modulate the microbiota is probably related to its fermentability (assessed by degradation with xylanase), but other characteristics such as particle size can also affect the microbiota, as recently demonstrated on wheat bran in a mouse model (Suriano et al., 2018).

4. Conclusion

In conclusion AX content can be used to estimate TDF content in wheat milling fractions and wheat-based foods. Moreover, the amount of AX released by xylanase is proposed as a functional evaluation of DF. AX solubilized by xylanase (AX_{enz}) are also potentially releasable by microbiota enzymes in the intestine and the AX_{enz} assay therefore indicates the proportion of fermentable DF in wheat milling fraction or in wheat-based foods. This assay provides a simple characterization of an important functional attribute of DF and could help to better understand the nutritional effects of different wheat milling fractions. The equation developed in this work is only valid for wheat products and cannot be applied to other cereal products such as corn, rye, barley, oat or rice… However we anticipate that the AX content could probably be used for the estimation of DF content in rye products, provided that an appropriate equation is developed. Indeed, in rye as in wheat, AX is the main component of cell walls and DF. The high amount of mixed linked–glucan (MLG) in oat and barley and the heterogeneity of the MLG distribution in the different parts of the grain are not favourable to the development of equations based on AX content to estimate the DF content, in these cereals.

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452

Figure Captions

Figure 1: TDF related to HM_wDF contents in the wheat products (Data table 1 and 2)

Figure 2: **A**) TDF content and **B**) HM_wDF content related to AX content in the wheat products (Data table 1 and 2)

Figure 3: TDF content calculated from tissue composition (by the following equation TDF= %OP + %IL+ 0.45x%AL + 0.03x%SE) related to TDF content measured by the AOAC 2009-01 method (expressed according to the dry matter) from milling fractions. .

%OP: proportion of outer pericarp , %IL: proportion of intermediate layer , %AL: proportion of aleurone layer , %SE: proportion of starchy endosperm.

Figure 4: **A**) AX amount released by xylanase (AX_{enz}) and **B**) proportion of AX released by xylanase (% AX_{sol}) related to AX content in the wheat products (Data table 1 and 2)

Supplementary figures

Supplementary figure S1: TDF content related to HM_wDF content **A**) in the wheat-based foods (bread. pasta and biscuits; data table 1) **B**) in the milling fractions (Data table 2)

Supplementary figure S2: TDF and HM_wDF contents related to AX: **A** and **B** in wheat-based foods (data Table 1); **C** and **D** in milling fractions (Data table 2).

Supplementary figure S3: proportion of AX released by xylanase (% AX_{sol}) related to the proportion of aleurone (AL) and starchy endosperm (SE) (% AL+SE) in the fractions.

Table 1 : Resistant starch and dietary fibre contents of wheat food products as determined by AOAC methods 2002.02 and 2009.01, respectively.

g/100g d.m.	Resistant Starch	HM_wDF	LM_wSDF	TDF	AX_{tot}	AX_{enz}	% AX_{sol}
Biscuit- R	0.14	1.8	1.3	3.2	1.0	0.7	67.4
Biscuit-F^+	0.17	7.2	1.8	9.0	3.5	1.4	40.1
Biscuit-F^-	0.17	6.4	1.3	7.7	3.2	0.9	27.3
Bread-R	1.21	4.3	2.4	6.7	2.1	1.7	83.4
Bread-F^+	0.55	9.9	2.0	11.9	4.7	2.6	55.7
Bread-F^-	0.80	11.8	2.2	14.0	5.6	2.0	35.5
Dry pasta-R	0.29	4.1	3.6	7.7	2.2	1.6	73.6
Dry pasta-F^+	0.21	8.0	2.9	10.9	4.1	2.2	54.4
Dry pasta-F^-	0.27	8.5	3.2	11.7	4.2	1.9	44.2
Mean	0.42	6.9	2.3	9.2	3.4	1.7	53.5
SD	0.37	3.10	0.80	3.3	1.4	0.6	18.6
RSD	87%	45%	35%	36%	42%	37%	35%

d.m.: dry matter

R: regular bread, pasta and biscuits

F^+ and F^-: products made with flour enriched in DF with F^+ and F^- fractions

SD : standart deviation ; RSD : relative standart deviation

AX_{tot}= sum of arabinose and xylose measured in the product

AX_{enz}= sum of arabinose and xylose released form the product after treatment with a xylanase

% AX_{sol} : = (AX_{enz}/AX_{tot}) x 100

Table 1 : Dietary fibre content, AX content and tissue proportion of various milling fractions from bread and durum wheats

g/100g dm	HM_wDF	LM_wSDF	TDF	AX_{tot}	AX_{enz}	% AX_{sol}	% OP	% IL	% AL	% SE	% EA
Grain common wheat	14.9	3.4	18.3	8.1	3.1	38.5	2.5	3.7	9.2	79.9	1.3
Coarse bran	52.4	4.6	57.0	25.0	9.9	39.6	13.3	29.4	36.0	15.5	0.5
Fine bran	47.0	4.6	51.5	25.6	8.8	34.5	11.4	27.0	29.6	18.2	2.8
Shorts	38.6	5.0	43.6	20.2	6.7	33.1	9.8	18.1	21.8	19.4	16.0
Middlings	12.2	4.4	16.5	6.2	2.3	37.3	2.0	2.7	14.3	61.7	15.4
Tail-end flour	7.6	3.8	11.4	4.0	1.6	39.3	1.2	1.3	11.5	76.7	8.7
Flour (T55)	3.7	2.0	5.7	2.2	1.5	67.4	0.2	0.1	2.0	100.7	0.2
Fibre fraction F^+	20.1	3.9	24.1	10.0	4.7	47.1	3.9	9.1	20.9	56.7	3.8
Fibre fraction F^-	57.9	1.9	59.9	25.0	4.8	19.3	26.5	12.8	18.2	16.7	3.6
Debranning fraction A	64.5	1.3	65.9	30.1	5.0	16.5	32.9	14.2	23.3	17.6	nd
Debranning fraction B	46.1	2.5	48.7	24.5	7.7	31.4	15.2	15.8	27.4	27.3	nd
Debranning fraction C	18.5	2.9	21.3	11.0	4.9	44.4	7.4	11.3	24.1	51.2	nd
Debranning fraction D	25.6	3.0	28.6	8.3	4.1	49.2	4.9	8.1	20.4	63.1	nd
Grain durum wheat	10.6	4.0	14.6	5.3	2.5	46.7	7.0		10.1	82.0	1.3
Coarse bran	53.9	2.0	55.9	24.1	7.5	31.0	54.2		31.0	15.6	5.1
Purified Fine bran	44.0	3.2	47.1	21.5	7.1	33.1	60.2		25.8	19.2	3.2
Sized fine bran	49.7	2.8	52.6	25.4	5.2	20.4	51.7		27.4	18.2	2.0
Shorts	18.0	4.1	22.2	8.7	4.7	53.7	16.9		21.8	56.1	5.1
Tail-end flour	7.1	4.0	11.1	3.7	2.2	58.4	4.7		18.9	73.4	3.1
Last break flour	6.7	3.8	10.5	3.4	1.8	52.1	4.0		18.3	78.7	0.8
Semolina S6	7.4	4.2	11.7	3.8	2.3	59.6	0.8		12.0	81.9	6.6
Semolina S3	3.1	3.6	6.7	1.8	1.5	83.0	3.6		2.4	100.0	0.0
Debranning fraction E	69.5	0.9	70.4	28.6	2.3	8.1	67.5		12.5	10.8	nd
Debranning fraction F	52.1	2.2	54.2	20.2	4.3	21.5	48.2		36.8	19.1	nd
Debranning fraction G	45.5	2.6	48.0	19.4	4.6	23.5	55.4		33.7	25.6	nd
Debranning fraction H	24.9	3.6	28.4	10.4	3.4	32.4	18.7		25.3	40.5	nd
Debranning fraction I	15.5	3.6	18.9	8.6	3.0	35.1	11.1		17.8	62.5	nd
mean	30.3	3.3	33.5	14.3	4.3	39.1	10.1	11.8	20.5	47.7	4.4
SD	21.1	1.1	20.6	9.5	2.3	16.6	10.0	9.2	9.3	29.5	4.7
RSD	70%	32%	61%	67%	54%	43%	99%	78%	45%	62%	106%

d.m.: dry matter

SD : standart deviation ; RSD : relative standart deviation

AX_{tot}= sum of arabinose and xylose measured in the product

AX_{enz}= sum of arabinose and xylose released form the product after treatment with a xylanase

% AX_{sol} : = (AX_{enz}/AX_{tot}) x 100

OP: outer pericarp; IL: intermediate layers; AL: aleurone layer; SE: starchy endosperm; EA: embryonic axis

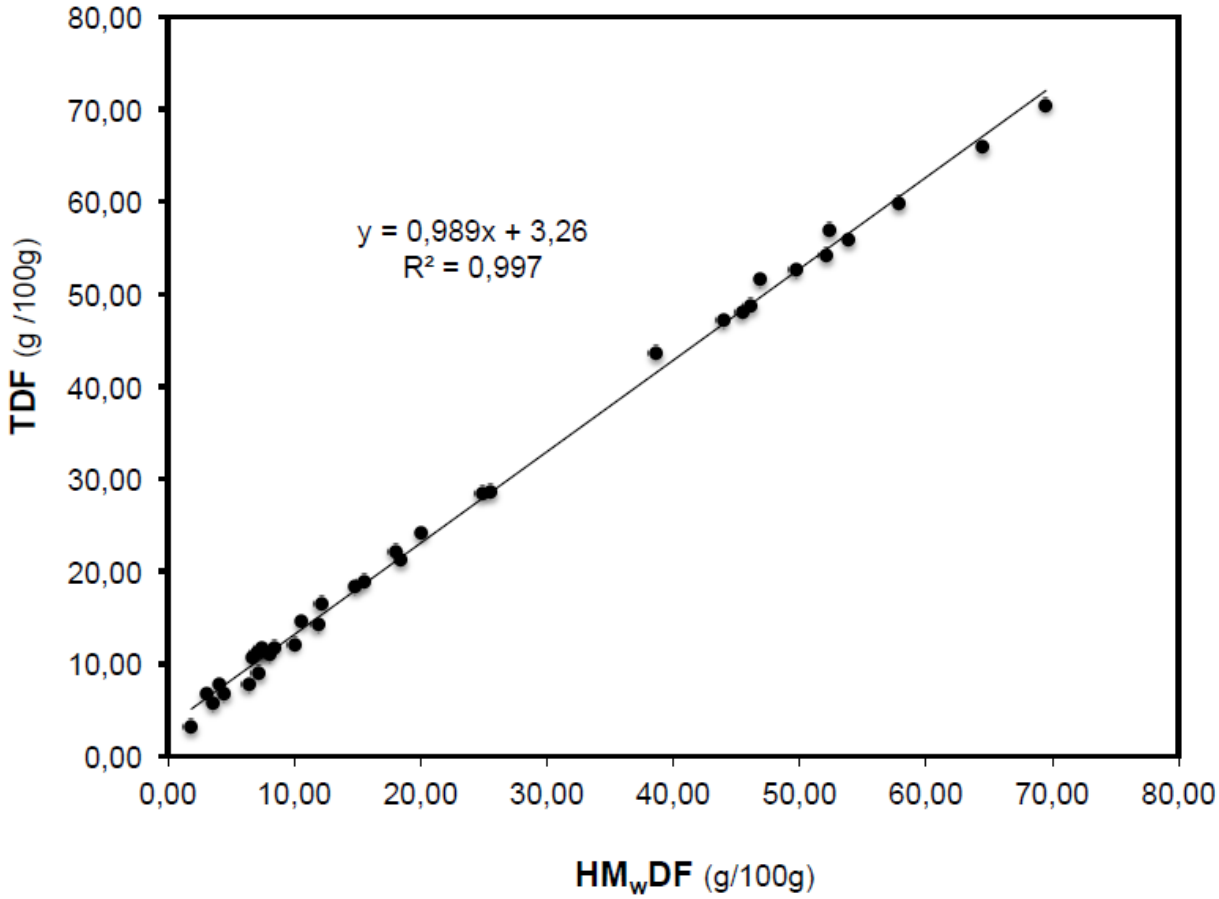

Figure 1

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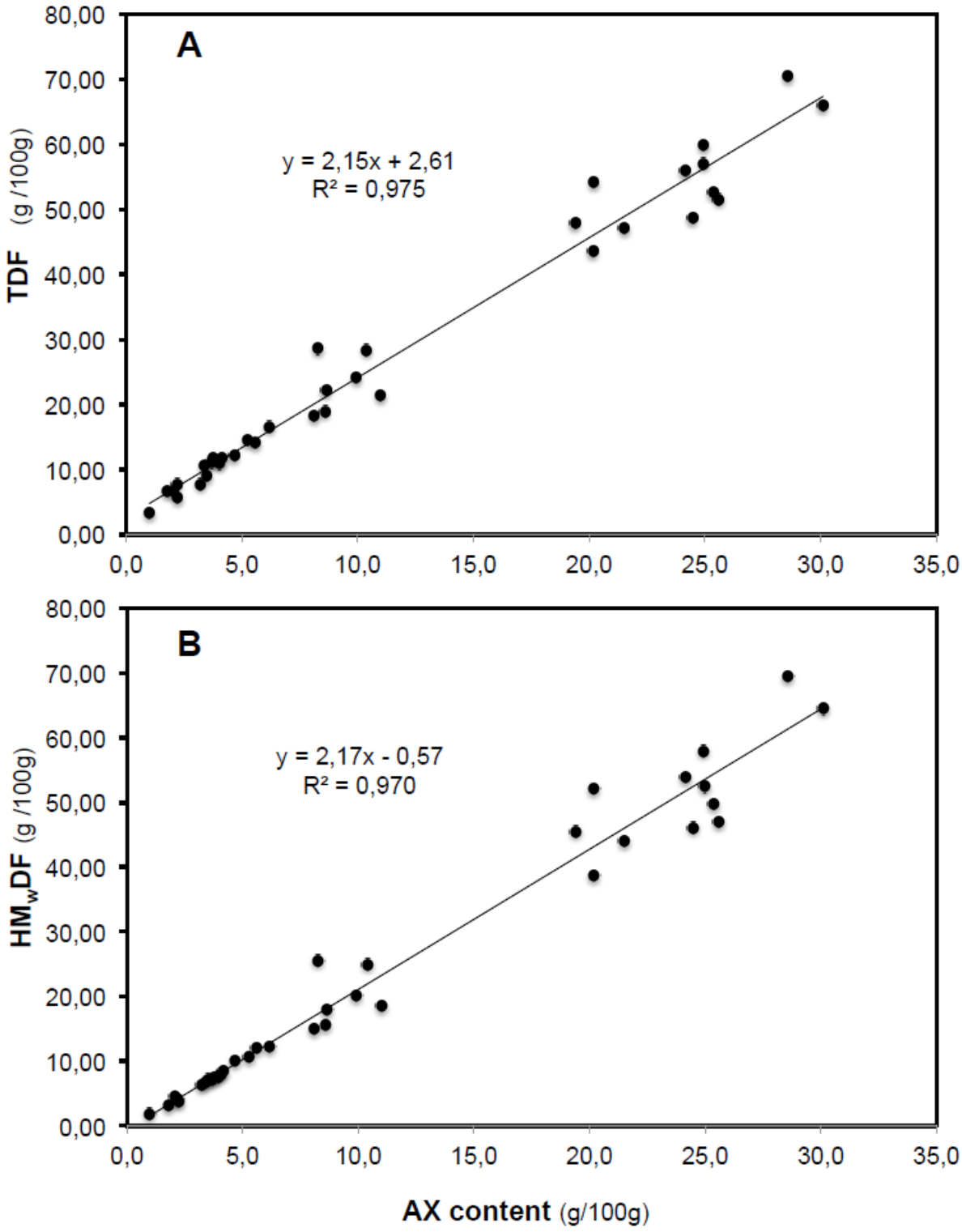

Figure 2

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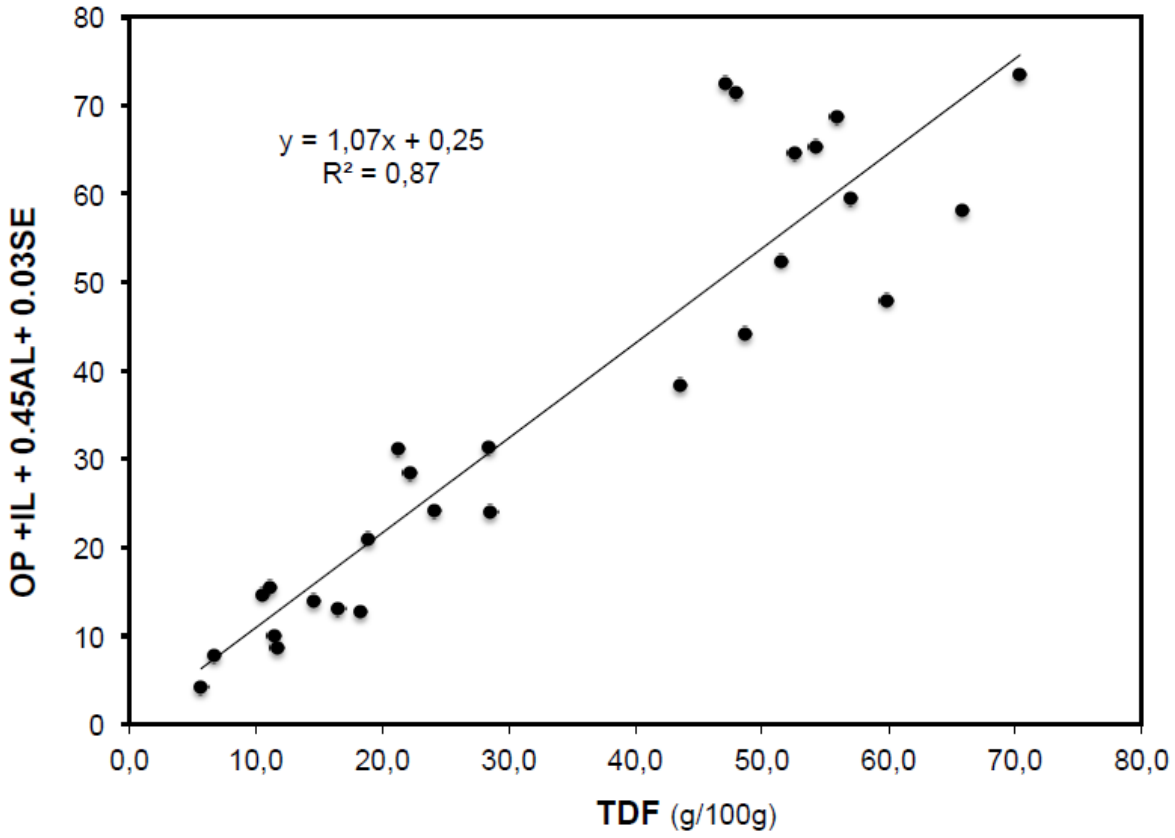

Figure 3

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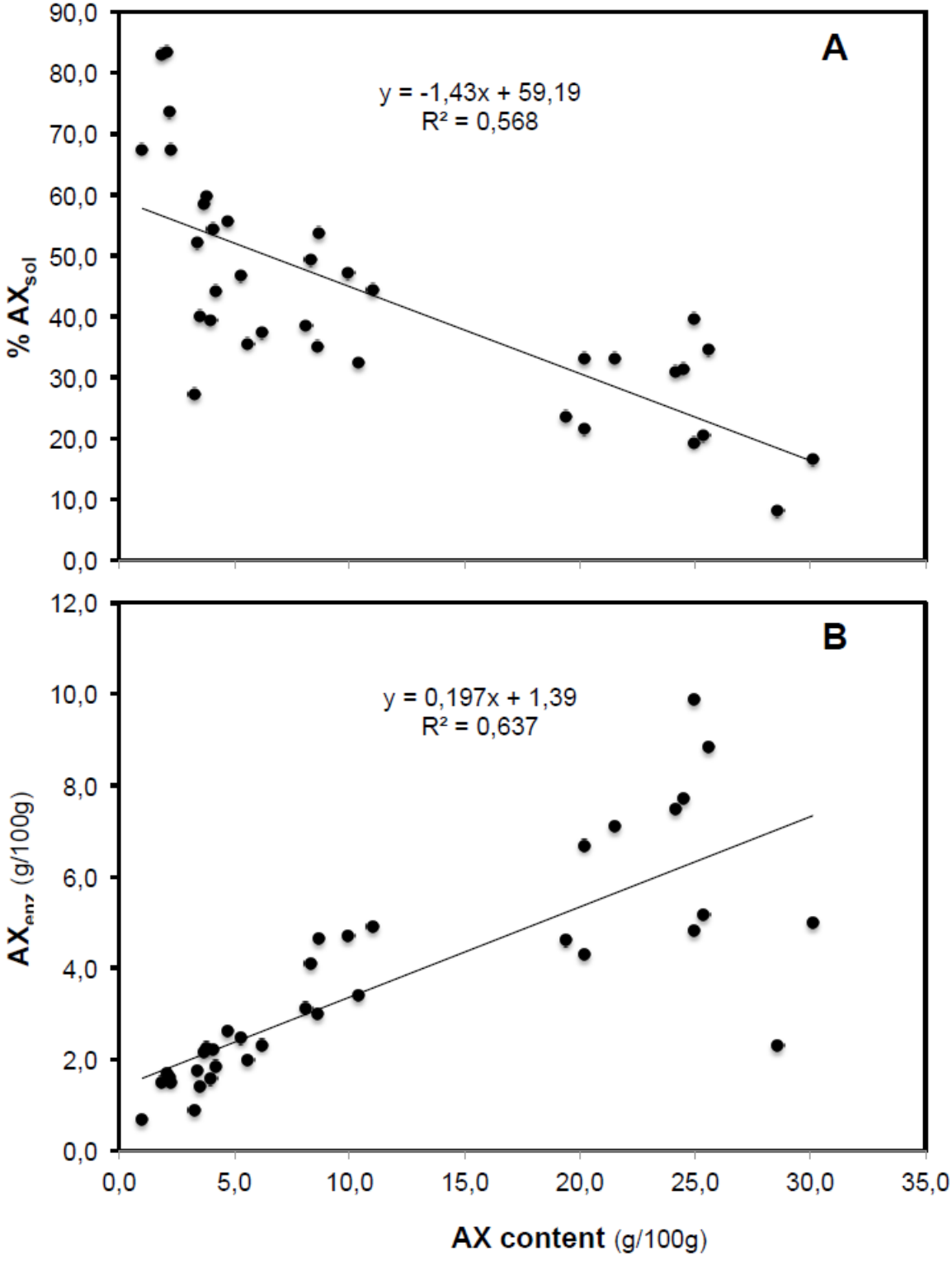

Figure 4

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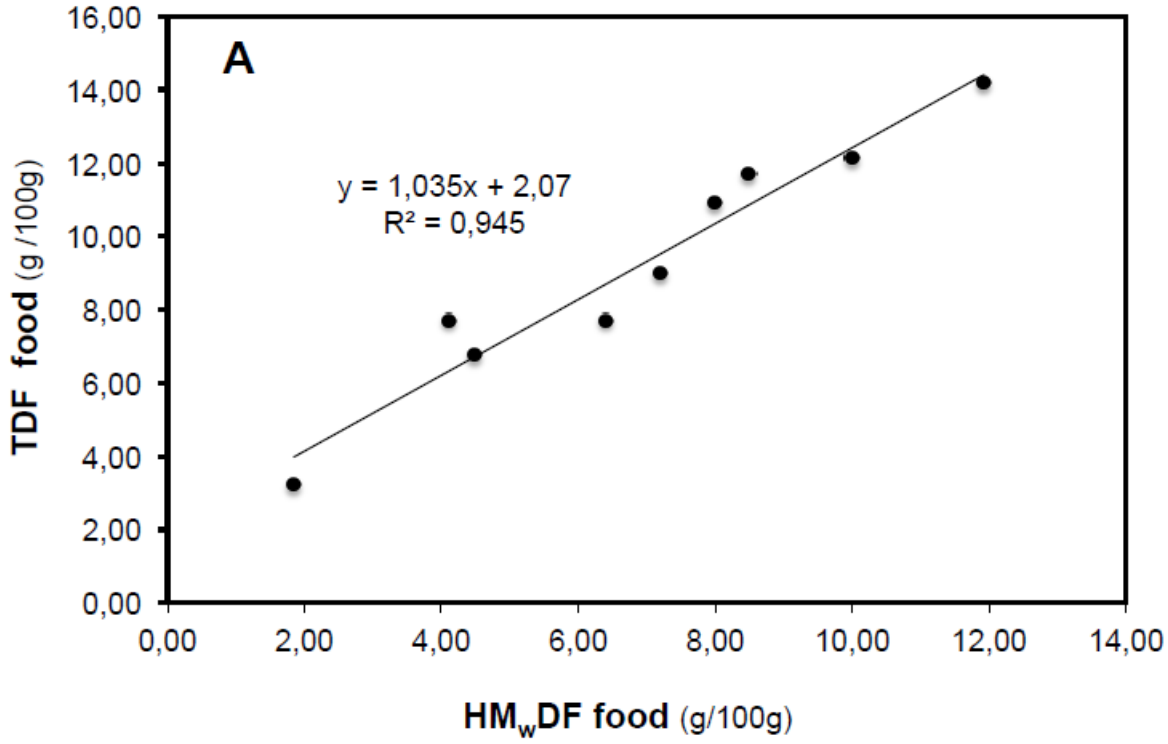

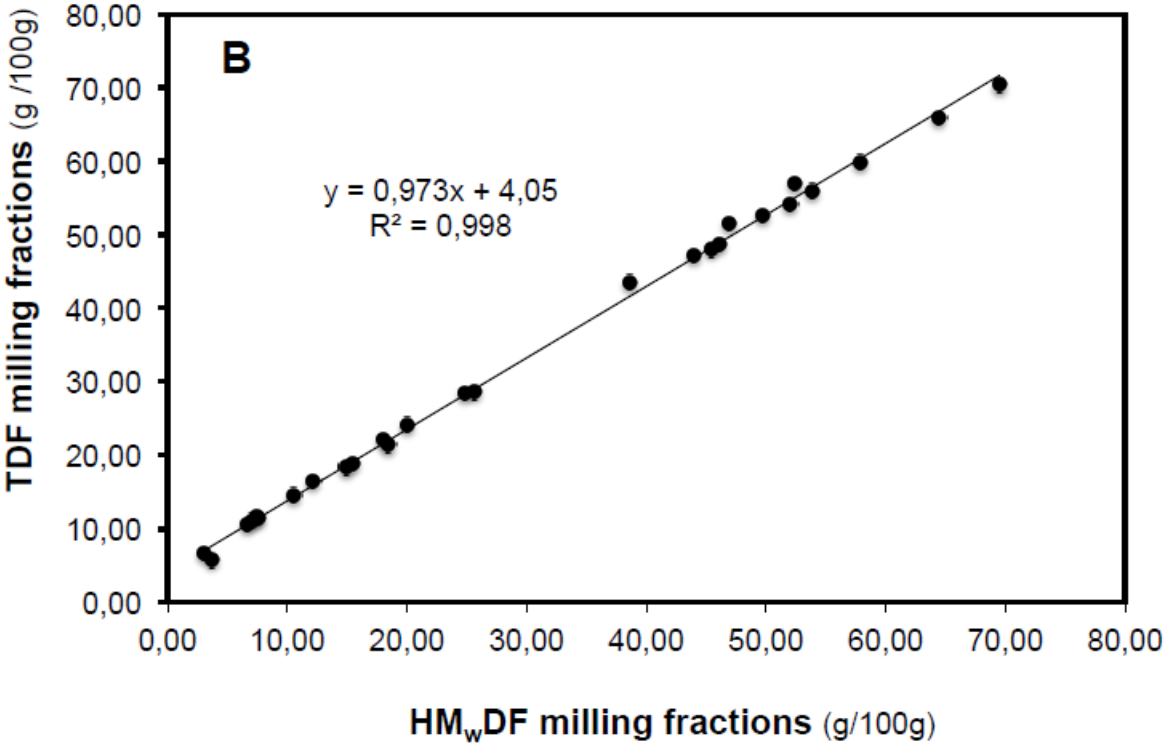

Figure S1

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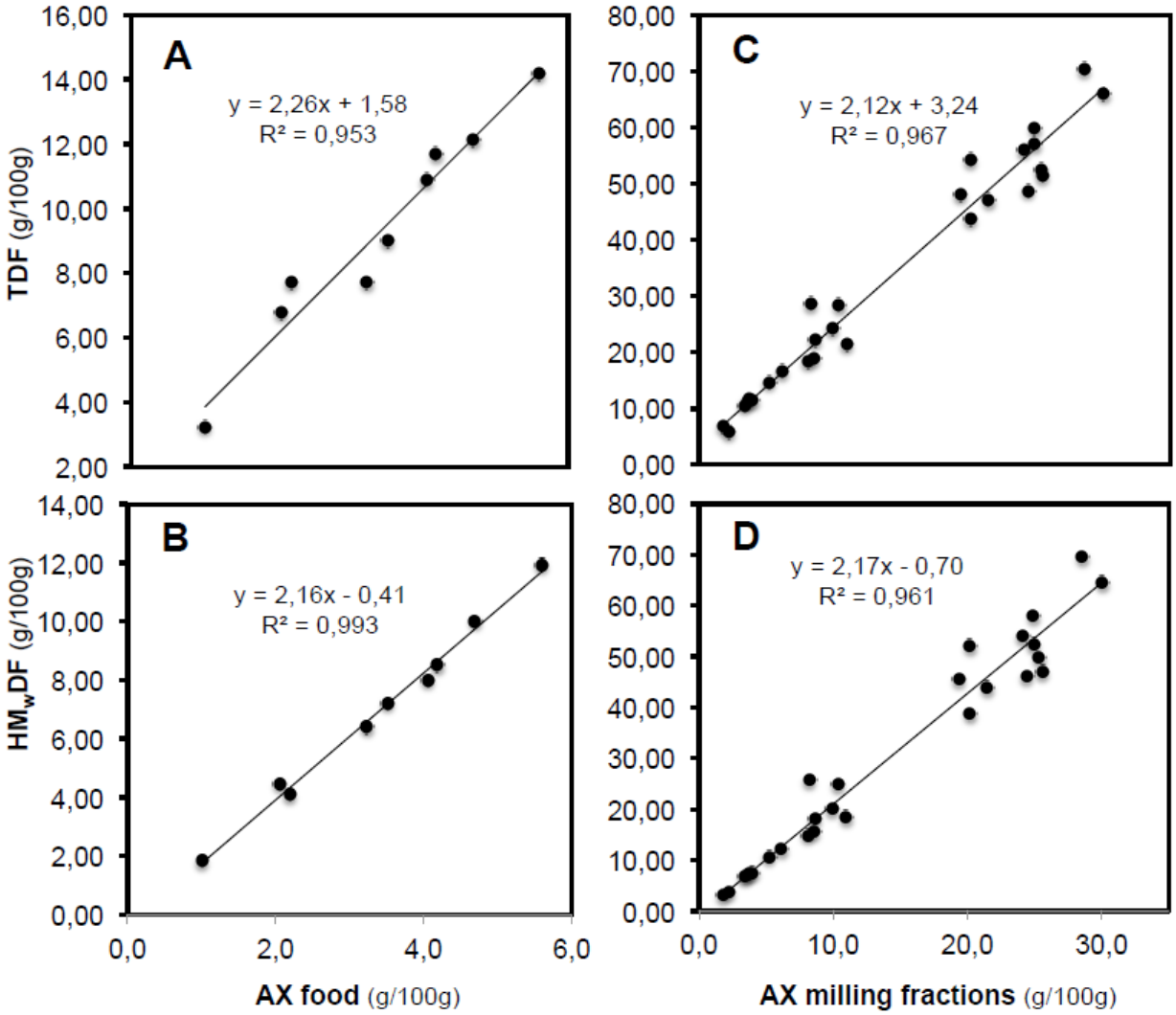
A
y = 2,26x + 1,58
R² = 0,953
TDF (g/100g)
B
y = 2,16x - 0,41
R² = 0,993
HMwDF (g/100g)
AX food (g/100g)
C
y = 2,12x + 3,24
R² = 0,967
D
y = 2,17x - 0,70
R² = 0,961
AX milling fractions (g/100g)

Figure S2

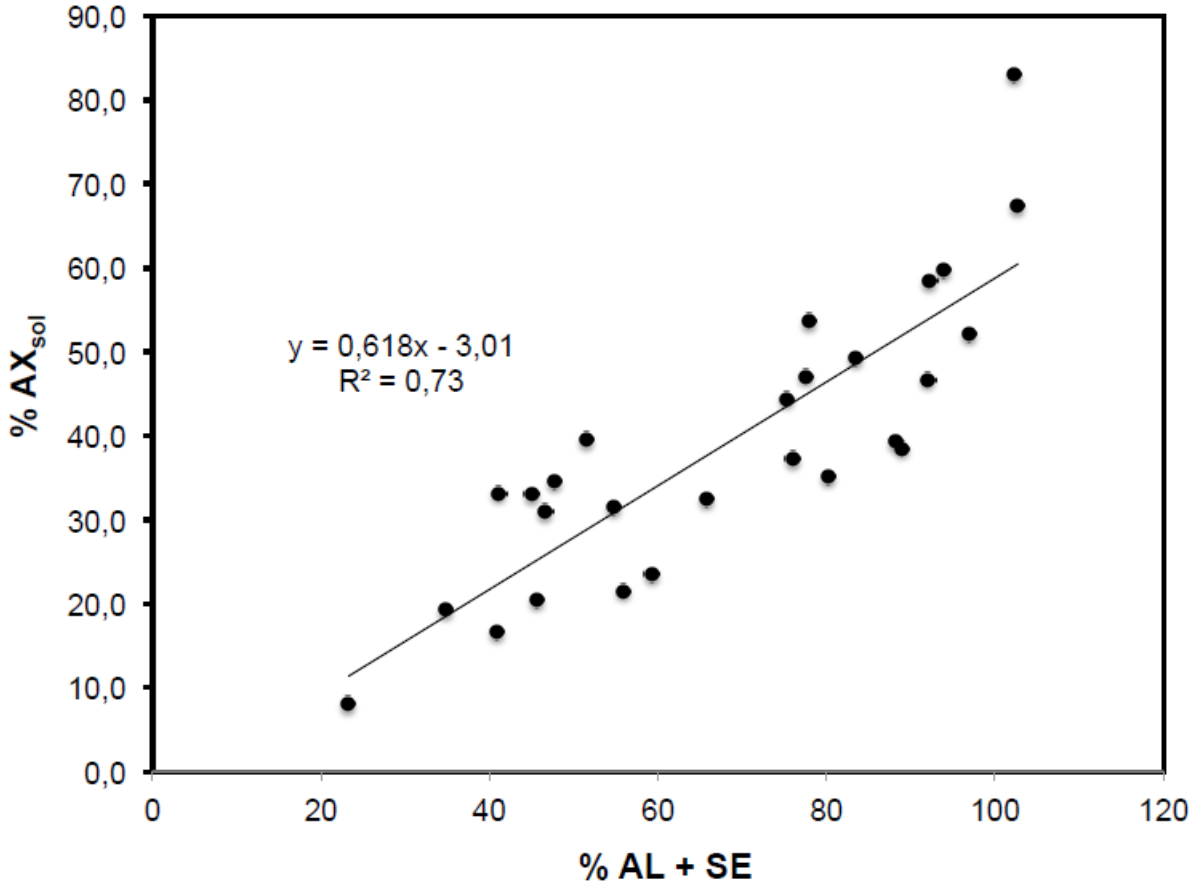

Figure S3

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