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Techniques and technologies for the breadmaking process with unrefined wheat flours

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Abstract

Background: In recent years there has been an increasing interest in the production of wholegrain products owing to the positive effects shown on human health. Although refined flour still represents the standard reference in breadmaking technology, consumer demand for unrefined breads has grown greatly. The different chemical composition of unrefined wheat flours (UWFs), which includes specific fractions of milling by-products (i.e., wheat bran and wheat germ), favours the nutritional value, but it has a negative effect on technological performance. Therefore, it is useful to develop new strategies specifically designed to improve the quality of UWF breads. Scope and approach: The present review aims to set out the techniques and technologies that have been reported in the literature for the breadmaking process with UWFs, that is, from raw material processing to bread formulation and breadmaking methods. Key findings and conclusion: The evaluation of UWF quality is still based on the tests developed for refined flour, which cannot properly estimate UWF technological properties. The greatest efforts to improve the breadmaking performance of UWF have been focused on modifying the bread formula, mainly with the addition of improvers. Conversely, very little investigation has been carried out on adapting the breadmaking process to the different characteristics of the raw material. Overall, the use of UWF in breadmaking may require further investigations into processing strategies to improve the quality of the end product, hence increasing the consumption of healthy foods.

Keywords	bread; wholewheat flour; milling by-products; wheat germ; wheat bran
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Dr. Paul Finglas Executive Editor Trends in Food Science & Technology

January 27, 2020

Dear Dr. Paul Finglas:

I am pleased to submit an original research article entitled "Techniques and technologies for the breadmaking process with unrefined wheat flours" for consideration for publication in *Trends in Food Science & Technology*.

This manuscript aims to report the processing strategies that have been developed until now for the breadmaking process with unrefined wheat flours, while suggesting some processing innovations to improve the exploitation of unrefined wheat flours. The main chapters of this manuscript include: (i) the introduction section, (ii) the evaluation of bread quality, (iii) the breadmaking process (reporting the scientific literature about breadmaking with unrefined wheat flours), (iv) the carbon footprint of unrefined wheat flour bread and (v) the conclusion.

We believe that this manuscript is appropriate for publication by *Trends in Food Science & Technology* since the use of unrefined flours for bread production has recently gained attention of the scientific research as well as of consumers. Moreover, reporting the processing strategies that improve the bredmaking process of unrefined wheat flours, may increase their consumption, which is associated to positive effects on human health.

To the authors' best knowledge, the use of unrefined wheat flour in breadmaking has never been reviewed before. The relevance of this study is a literature review about the breadmaking with unrefined wheat flours, from the raw materials to the storage phase.

This manuscript has not been published and is not under consideration for publication elsewhere. We have no conflicts of interest to disclose.

Thank you for your consideration!

Sincerely,

Dr. Lorenzo Guerrini Department of Agricultural, Food and Forestry Systems Management (DAGRI) University of Florence, Italy The authors would like to thank the Reviewers for the time spent in improving the paper, and for the important suggestions and corrections proposed. We hope to have addressed all the issues the reviewers outlined. Here following, a point by point reply to the comments received.

-Reviewer 1

- I really enjoyed reading the article, it is written in a clear way and is easy to follow. The different aspects of bread making are discussed in relation to the different flours that can be used and the challenges one faces when shifting from refined to unrefined flour. Mostly, the authors refer to recent literature, outlining the up to date literature available on the topic.

There were a few items that I would like to draw attention to:

Lines 263-265. It is mentioned that the fact that bran addition goes along with increase in ash content is contributing to a decrease in flour properties. Is that what the authors, Banu (2012) concluded? could it also relate to addition of bran fibres which affect flour properties? when you speak about technological properties, are there specific ones you are referring to? if yes, I recommend clarifying that.

Lines 263-265: Banu et al. (2012) studied the rheological properties and bread physical parameters of white flour enriched with different bran streams (3-30%). As reviewer observed, the conclusion of the study is that ash content, a measure of the bran and germ "contamination" on the white flour produced with the only starchy endosperm, significantly decreased the rheological properties of the dough and physical characteristics of breads. However, although they observed a negative impact of ash content on dough and bread quality, the authors identified in the 25% sample a level of ash content equal to that in wholewheat sample, but associated with a significant improvement of dough and bread quality.

According to reviewer observation we added some specifications about the study by Banu et al. (2012) (L 268-273).

L268: This sentence was a little unclear to me. was wheat pearled to 2-25% and was that used to make bread? or, was wheat pearled to X% and added to refined flour at 5-25% level?

Blandino et al. (2013) added 5 different levels of pearled fractions from wheat kernels (5-25%) on refined flours and tested the flour mixtures on dough rheology, bread quality and nutritional properties. We clarified the sentence in the Manuscript (L274-275) according to reviewer observation.

L339. I feel increase needs to be changed into decrease

The sentence has been rephrased (L346).

L400, 429 and 540. Are these percentages additions to the total recipe or expressed on flour weight? Also table 2 mentions percentages of addition and it would be good to clarify whether these are on flour weight or total recipe.

In accordance with reviewer's observation we specified the percentages of addition in L414, 444-445, 556-557. We also modified Table 2 adding the required specifications. L417: Why does a farinograph not work for UWF?

The farinographic official method was developed to evaluate the water absorption (WA) and the behaviour during the mixing step of refined flour. In refined flour the main components responsible for the WA are the gluten network proteins, which can adsorb up to three time their weight in water (Zaidul, I.S.M., Karim, A.A., Manan, D.M.A., Ariffin, A., Norulaini, N.A.N. & Omar, A.K.M. (2004). A farinograph study on the viscoelastic properties of sago /wheat flour dough. *Journal ofScience and Food Agriculture*, 84, 616–622). In unrefined products the presence of soluble and insoluble fibres as flour constituents significantly increases the flour water absorption (Schmiele et al., 2012). In fact, the hydroxyl groups in fibre structure bind to water molecules and compete with starch and proteins for hydration, altering the water distribution in the dough (Khalid et al., 2017; Curti et al., 2013). Hence, the farinographic WA in unrefined wheat flours does not reflect the amount of water required for the optimal hydration of the gluten proteins to reach the 500BU of consistency, but this value results also from the fibre water absorption.

The authors also added the reference Schmiele et al. (2012) to support the statement in L431 and in the reference list L1149.

L473: Which studies report and increase in stabilisation by DATEM? Tebben (2018) seems to indicate a decrease in stability

Tebben et al. (2018) reported that monoglycerides and diglycerides decreased the final proof time, while DATEM increased the final proof time as measured by Mathurograph. It means that monoand di- glycerides allowed the dough to perform a faster leavening, whereas DATEM reduced the dough rise. Hence, the authors reported DATEM to increase the fermentation stability in terms of a larger window of time during which the dough can be moved to the leavening to the baking step without loosing its optimal volume (Hrušková, M., Švec, I., & Jirsa, O., 2006. Correlation between milling and baking parameters of wheat varieties. *Journal of Food Engineering*, 77, 439–444; Mettler, E., & Seibel, W., 1993. Effects of emulsifiers and hydrocolloids on whole wheat bread quality - a response-surface methodology study. *Cereal Chemistry*, 70, 373–377). Converseley, a study by Armero, E., & Collar, C., 1996b. Antistaling additives, flour type and sourdough process effects on functionality of wheat doughs. *Journal of Food Science*, 61, 299–303, found that DATEM decreased the fermentation stability and final proof time; for this reasons in the present review we reported that controversial results about the effect of DATEM on fermentation stability are still present in the current literature.

L 474.: You speak about studies looking at emulsifiers, but in the whole paragraph only one reference is used. was there only one study? if yes, mention that you could identify just one study. That indicates a gap in the research simultaneously.

The authors agree with reviewer's observation. As we stated at the beginning of the paragraph 3.2.3 "UWF bread improvers" (L442-443), the addition of common bread improvers on the breadmaking performance of wholewheat flours has been recently reviewed by Tebben et al. (2018). In a previous version of the Manuscript we included all the references cited in the review Tebben et al. (2018), but unfortunately the final number of references exceeded those allowed by the Journal (at maximum 100 ref). Hence, we had to remove some of the scientific studies, and we limited to cite the review article where the reader could find all the specific references that are briefly summarized in the present review.

At the end of the paragraph we reported that the mentioned studies tested the effects of bread improvers on wholewheat flour, whereas information on the impacts of the same improvers on unrefined flour performances are still missing in the current literature (L519-521). We specify in the text that Tebben et al is a review.

L706-707: The sentence is a little confusing. were 13 or 14 attributes tested by a sensory panel?

We corrected the sentence in L723 according to reviewer's observation. In the mentioned study (Jensen et al., 2011a) a total of 13 sensory attributes was chosen for each of the tested bread samples.

Reference list: the reference to Tebben (2018) is mentioned in the text and tables but missing in the list. Please add this one

We added Tebben et al. (2018) in the reference list (L1173).

Table 2: treatments on whole grain, mainly germination are given. This is what Richter (2014) did as well. I see it in the reference list but not in the table. could it be added?

Table 2: There was an error in the reference name of Table 2, but the reference Ritcher et al. (2014) was already included in the Table. We corrected the mistaken reference, hence Ritcher et al. (2014) is now present in reference of the Table as well as in the reference list of the Manuscript.

Table 4 and **5** show '?' in the last column. What is meant with these? It would be good to clearify that in the foot note of the table. Or, use a - if no real conclusions can be drawn from the study.

Table 3: the authors added some specification about the article by Zilic et al (2016) in the colums "Measurements" and "Main Results".

Table 4: the authors made an in-depth research to include additional information in Table 4,studying the specific literature references of the cited review by Hemdane et al. (2016) and Boukidet al. (2018).

Table 5: the authors added some specification as suggested by the reviewer. **Table 6**: the authors added some specification consistently with Table 5.

-Reviewer 2

The topic is interesting and current, the paper is well written and well argued. Some suggestions:

- **Page 3 line 67**: In 2019 Whole Grain Initiative sets the following definition of whole grains also approved by ICC, Healthgrain and Cereal & Grain Association: "Whole grains shall consist of the intact, ground, cracked, flaked or otherwise processed kernel after the removal of inedible parts such as the hull and husk. All anatomical components, including the endosperm, germ, and bran must be present in the same relative proportions as in the intact kernel". I suggest referring to this definition because is the most recent.

The authors added the reference suggested by the reviewer in L67-68.

- page 3 line 79, page 4 lines 126 and 127, page 5 line 132..... : I suggest changing the term sensory, throughout the text, with sensorial.

As reviewer suggested we changed the word "sensory" with "sensorial" (L80, L127, L128, L133).

- **page 11 line 336**: the term gelatinizing should be changed with gelatinization.

We changed the term "gelatinizing" with "gelatinization" according to reviewer's suggestion (L342-343)

- **page 14 line 439-442**: other authors than Tebben et al, 2018 reported about the positive role exerted by xylanase:

Katina K., Heiniö R.L., Autio K., Poutanen K. Lebensm. Wiss. Technol., 2006, 39, 1189-1202. Messia M.C., Reale A., Maiuro L., Candigliota T., Sorrentino E., Marconi M. J. Cereal Sci., 2016, 69, 134-144.

Santala O., Lehtinen P., Nordlund E., Suortti T., Poutanen K. J. Cereal Sci., 2011, 54 187-194. However, Tebben et al., 2018 (page 14 line 444) is missing from the references !!

The reference list of the Manuscript already included "Katina, K., Heiniö, R. L., Autio, K., & Poutanen, K. (2006a). Optimization of sourdough process for improved sensory profile and texture of wheat bread. *LWT - Food Science and Technology*, 39, 1189–1202" in L1030 and in the Manuscript L138, L147, L614, L624. However, the aim of the paper was to (i) investigate the effect of sourdough process parameters (ash content of flour, fermentation temperature and fermentation time) on bread quality parameters performing two different types of fermentations (a single strain of lactic acid bacteria or a combination of lactic acid bacteria + yeasts) and (ii) to improve flavour, volume and shelf life of breads by optimizing the fermentation variables. Was the reviewer referring to this or to another paper for the missing reference about xylanase positive effects?

The authors added the reference proposed by the reviewer "Messia, M. C., Reale, A., Maiuro, L., Candigliota, T., Sorrentino, E., & Marconi, E. (2016). Effects of pre-fermented wheat bran on dough and bread characteristics. *Journal of Cereal Science*, 69, 138–144" in the reference list (L1078) and in the Manuscript Paragraph 3.1.2.2 L369. Furthermore, the authors included this reference in Table 4 and L372 since the study also tested the effect of bran pre-soaking on dough rheology and bread physical properties.

The authors added the reference "Santala, O., Lehtinen, P., Nordlund, E., Suortti, T., & Poutanen, K. (2011). Impact of water content on the solubilisation of arabinoxylan during xylanase treatment of wheat bran. *Journal of Cereal Science*, 54, 187–194" in the Manuscript Paragraph 3.1.2.2 L368-369 and in the reference list L1142.

The authors added Tebben et al. (2018) in the reference list (L1173).

- **References section**: page 32 line 938, page 32 line 947, page 35 line 1039, page 36 line 61, page 37 line 1095: in the references section Authors must pay attention to how the surnames and names of the authors are reported. Here are some correct underlined surnames: Fadda C, Sanguinetti A.M., Del Caro A., Collar C., Piga A (2014); Gobbetti M., Rizzello C.G., Di Cagno R., De Angelis M. (2014); Marti A., Cardone G., Nicolodi A., Qualgia L., Pagani M.A., (2017): Otoni C.G., Pontes S.F.O., Medeiros E.A.A., Soares Nde F. (2014).

We corrected the references in the reference list and throughout the Manuscript as reviewer suggested (L965, L1066, L322, L1101).

Furthermore, the authors added in the paragraph 3.1.2.1 (L352-356) "Pre-treatment of wheat kernels: germination" the interesting results recently showed in the paper by Cardone et al. (2020), which was published during the submission process of the present review. The article studied the application of a controlled germination process on rheological characteristics of doughs and physical quality of breads from wholewheat flour showing promising results.

Highlights

- Positive effects on human health shown by the consumption of unrefined wheat products
- Review of the use of unrefined wheat flours in the breadmaking process
- Discussion of the standard criteria for the evaluation of unrefined bread quality
- Description of modifications to the bread formula and process for unrefined flours
- Proposal for new research to improve the employment of unrefined flours

Abstract:

Background: In recent years there has been an increasing interest in the production of wholegrain products owing to the positive effects shown on human health. Although refined flour still represents the standard reference in breadmaking technology, consumer demand for unrefined breads has grown greatly. The different chemical composition of unrefined wheat flours (UWFs), which includes specific fractions of milling by-products (i.e., wheat bran and wheat germ), favours the nutritional value, but it has a negative effect on technological performance. Therefore, it is useful to develop new strategies specifically designed to improve the quality of UWF breads.

Scope and approach: The present review aims to set out the techniques and technologies that have been reported in the literature for the breadmaking process with UWFs, that is, from raw material processing to bread formulation and breadmaking methods.

Key findings and conclusion: The evaluation of UWF quality is still based on the tests developed for refined flour, which cannot properly estimate UWF technological properties. The greatest efforts to improve the breadmaking performance of UWF have been focused on modifying the bread formula, mainly with the addition of improvers. Conversely, very little investigation has been carried out on adapting the breadmaking process to the different characteristics of the raw material. Overall, the use of UWF in breadmaking may require further investigations into processing strategies to improve the quality of the end product, hence increasing the consumption of healthy foods.

- 1 Techniques and technologies for the breadmaking process with unrefined wheat flours
- 2

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11

12 Abstract:

13

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31

32 Keywords: bread; wholewheat flour; milling by-products; wheat germ; wheat bran

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34 Abbreviations: unrefined wheat flour (UWF), wheat germ (WG), wheat bran (WB)

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36 **1. Introduction**

Wheat bread is the staple food in many diets, representing one of the main sources of the daily
energy intake. Bread composition is the result of several factors including wheat genotypes,
agronomic treatments, environmental conditions, flour composition, breadmaking conditions and
product storage.

The wheat species most widely used for breadmaking is *Triticum aestivum* L. or "common" wheat, accounting for 95% of wheat production, followed by *Triticum durum* or "durum" wheat, which is widely used in Mediterranean cuisine for making special breads, couscous, pasta and bulgur.

44 For most of human history, flour was produced using stone mills, which simultaneously crushed and ground wheat kernels in a single millstream, giving wholewheat flour. The milling process was 45 completely revolutionized during the second half of the 19th century with the introduction of the 46 47 roller mill, which allowed the three fractions of the caryopsis (i.e., starchy endosperm, bran and 48 germ) to be separated at the beginning of the process, resulting in different millstreams (Jones, 49 Adams, Harriman, Miller, & Van der Kamp, 2015). The refined flour obtained from the starchy 50 endosperm alone shows better technological quality and gives breads with sensory properties that 51 are widely appreciated by consumers. Hence, refined flour has become the standard for the further 52 technological developments in the bakery industry up to the present day. This means that all the 53 knowledge on bread formulation and process implementation has been made by considering 54 refined wheat flour, characterized by a chemical composition mainly composed of starch (80%-85%) 55 and proteins (8%-14%). The other millstreams (i.e., bran, germ) are instead considered milling by-56 products and mainly used as animal feed.

However, in recent years there has been renewed interest in unrefined wheats, since several studies have shown that regular consumption of these products is associated with health benefits such as a lower risk of chronic-degenerative diseases and improved body weight regulation (Ye, Chacko, Chou, Kugizaki, & Liu, 2012; Hauner et al., 2012) The increased interest in healthy and functional foods has led to a consequent growth in the demand for high nutritional value breads (Gani, & Hameed, 2012). As a result, it has been necessary to re-interpret bread quality, also including nutritional value. Unrefined wheat flour (UWF) is a composite class which includes flours supplemented with milling by-products at the same or a different relative proportion compared to that of the intact caryopsis (Fig. 1). The official definition for flours containing the same components in the same relative proportions as the wheat kernel is "wholewheat" flour (AACC International, 1999 Whole Grain Initiative approved by ICC, Healthgrain and Cereal & Grain Association, 2019), although the modern milling process does not allow for the inclusion of wheat germ (WG), but only recovers wheat bran (WB). Hence, the resulting "wholewheat" flour is no more than flour enriched with bran.

Despite the nutritional benefits, the introduction of milling by-products in the breadmaking process
has some drawbacks, the solutions to which are still open challenges for bread makers.

73 First, the outer layers are the most susceptible to contaminants, i.e., mycotoxins and heavy metals 74 (Sovrani et al., 2012). Hence, the possibility of using milling by-products to produce high nutritional 75 value breads requires an integrative approach from the field to the breadmaking. Therefore, appropriate agronomical and/or post-harvest strategies must be adopted to reduce the safety risk 76 77 and preserve the high nutritional value of the outer layers. Secondly, WG and WB negatively affect 78 the technological quality of doughs and breads (Boukid, Folloni, Ranieri, & Vittadini, 2018; Hemdane 79 et al., 2016). Finally, since the sensorial characteristics of unrefined bakery products are little 80 appreciated by consumers, several scientific studies have dealt with the sensory sensorial profile to 81 increase their acceptability (Gani at al., 2012; Heiniö et al., 2016).

The present review aims to report the processing strategies that have been developed until now for the breadmaking process with UWFs (Fig.2), while suggesting some processing innovations to improve the exploitation of UWFs.

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109 **2. Bread quality**

110 According to the common good manufacturing practices, "bread" must be prepared by baking a 111 dough which consists of flour, yeast, and a moistening ingredient, usually water. In the present 112 review this term refers to all typologies of leavened breads, not including flat breads obtained 113 without any leavening. Bread is one of the most ancient and widespread foods of all over the world. 114 Progressive technical developments over many thousands of years have led to a high diversification 115 of the product. Hence, a "good bread" presents different features depending on cultural 116 background, individual experiences, and personal likes and dislikes. Moreover, quality features 117 change over space, in different regions, and over time, together with food technology innovations. 118 However, despite the large diversity of bread characteristics, in the literature bread quality is mainly 119 evaluated as: (i) bread specific volume, (ii) crumb characteristics and (iii) crust colour (Zhou et al., 120 2014; Cauvain, 2015). These features are the results of the raw materials and processing conditions 121 adopted during bread production. Almost all of the quality characteristics are related to the gluten 122 network, since it traps the gas produced during the leavening and contributes to the formation of a 123 cellular crumb structure which confers the bread's volume, texture and eating qualities. The 124 addition of the "right" quantity of water is another key factor affecting dough rheology and the 125 development of gluten; too much or too little water means that the right gluten network cannot be 126 properly developed (Cauvain, 2015).

Although there are not any standard sensory sensorial attributes of wheat bread, they are all considered of maximum importance in the evaluation of the product. Sensory Sensorial features affect consumers' preferences and drive their choices towards the different bread typologies proposed by the current bakery market.

The flour used in the bread formulation plays a critical role for the product characteristics. Refined 131 132 bread presents a high loaf volume, a light colour, homogeneous crumb porosity and soft crumb. 133 Refined flour results in a fairly bland sensory sensorial attribute with only a slight grain-like flavour 134 or malted note, since most of these sensory contributions come from the WG oils and from the WB 135 particles removed during milling (Callejo et al., 2015; Challacombe, Abdel-Aal, Seetharaman, & 136 Duizer, 2012; Eckardt et al., 2013; Hayakawa, Ukai, Nishida, Kazami, & Kohyama, 2010; Heenan, 137 Dufour, Hamid, Harvey, & Delahunty, 2008; Jensen, Oestdal, Skibsted, Larsen, & Thybo, 2011a; 138 Katina, Heiniö, Autio, & Poutanen, 2006a; Lotong, Edgar-Chambers, & Chambers, 2000). The sensory 139 attributes of refined breads are largely appreciated by consumers, who, despite the proven health 140 benefits of wholewheat consumption, still prefer refined products (Ye, Chacko, Chou, Kugizaki, & 141 Liu, 2012).

The distinctive characteristics of UWF breads include a low loaf volume, coarse and hard texture,
dark colour and "speckled" appearance. Moreover, they are characterized by a nutty odour,
bitter/sour taste, a grain-like, "seedy" flavour, malted note and musty attribute (Curti, Carini,
Bonacini, Tribuzio, & Vittadini, 2013; Callejo et al., 2015; Challacombe, Abdel-Aal, Seetharaman, &
Duizer, 2012; Eckardt et al., 2013; Heenan, Dufour, Hamid, Harvey, & Delahunty, 2008; Jensen,
Oestdal, Skibsted, Larsen, & Thybo, 2011a; Katina, Heiniö, Autio, & Poutanen, 2006a; Katina,
Salmenkallio-Marttila, Partanen, Forssell, & Autio, 2006b).

The sensory profile of UWF breads is one of the major obstacles to increasing their consumption. The most challenging attribute is probably the bitter taste, associated with the presence of bioactive compounds (such as phenolic compounds, amino acids, small peptides, fatty acids and sugar) which are highly concentrated in the outer layers of the wheat kernel (Heiniö, 2009; Zhou et al., 2014; Van Gemert, 2011; Mattila, Pihlava, & Hellström, 2005); rancid sensory defects could also occur relating to oxidation of the WG oil (Heiniö et al., 2016).

The quality characteristics were created for refined breads, but in the literature they are also used for UWF bread. However, well-informed consumers appreciate the better nutritional value of UWF and they have different expectations about the product characteristics.

5

This can lead implicitly to some questions on bread quality. Does the different chemical composition of UWF require new qualitative standards or are the quality criteria of refined flours still suitable for UWF? Is it right to have the same expectations about the characteristics of the final product? It is hard to answer these questions and they require an in-depth discussion from a wide perspective.

162 Besides bread quality criteria, some important methods have also been developed for refined flours. 163 This is the case of the optimum water absorption (WA) of the flour, officially determined as the 164 amount of water required to reach 500BU in the farinographic test. However, it has been reported 165 that 500BU underestimates the absorption capacity of UWF (Hemdane et al., 2016). Improper 166 hydration of the dough could lead to i) the wrong evaluation of UWF bread quality, especially when 167 compared to refined bread, and ii) biased results if a new process setting is desired and the tested 168 variables significantly interact with water. Nevertheless, indications regarding the correct amount 169 of water for UWFs are still missing in the literature.

170

171 **3. Breadmaking process**

The literature has shown controversial results regarding breadmaking with UWFs, making it difficult
to interpret or compare experimental data in the literature. Indeed, several operating factors have
been reported to have a significant effect on bread quality (Table 1):

- 175 (i) The composition of UWFs is extremely variable; beside the starchy endosperm they
 176 include the presence of specific fractions of milling by-products, WG and WB. The
 177 different chemical compositions of UWFs produce great variability in the breadmaking
 178 performance.
- 179 (ii) The milling method. It is widely known that the milling method has a marked impact on
 180 the technological performance of flour (Jones, Adams, Harriman, Miller, & Van der Kamp,
 181 2015). Standard indications about the milling method, and particle size and composition
 182 of milling by-products could be useful for a better comprehension of their impacts and
 183 could help overcome their adverse effects in breadmaking. It is important to point out
 184 that the milling method strongly affects the UWF composition.
- 185 (iii) Bread formulation. In the literature there is no official bread formula, but different
 186 recipes are adopted. Besides the basic components (i.e., flour, water and the leavening
 187 agent), other ingredients are often added to the bread dough, such as salt (NaCl), sugar,
 188 shortening and oxidizing agents, which affect the flour performance. This variability of
 189 recipes could hinder the comparison and understanding of the effects and results.

(iv) Breadmaking procedure. Owing to the absence of a standard process in breadmaking,
operating conditions cannot be standardized, making it difficult to compare and
understand the literature data.

The paragraphs below set out to improve the comparability between the different studies in the literature by promoting both a standardization of methods and some technological innovations for UWF processing.

196

197 3.1 UWF quality and composition

In recent years several studies have reported the nutritional value, technological impact and sensory
profile of the milling by-products WG and WB in the breadmaking process (Hemdane et al., 2016;
Boukid, Folloni, Ranieri, & Vittadini, 2018; Heiniö et al., 2016).

WG (2%-3% of the caryopsis) is composed of the embryo and scutellum, the latter being discarded with the bran during the milling process (Boukid, Folloni, Ranieri, & Vittadini, 2018). It is considered the most nutritious part of the wheat kernel, providing 381 cal/100 g: 54% carbohydrates, 23% proteins and 23% lipids (Boukid, Folloni, Ranieri, & Vittadini, 2018; Nutrition data, 2014).

205 The limited utilization of WG in the bakery industry is primarily due to the high presence of 206 unsaturated fats and hydrolytic and oxidative enzymes (i.e., lipoxygenase and lipase) which favour 207 WG degradation (Boukid, Folloni, Ranieri, & Vittadini, 2018). Several treatments have been 208 developed to improve WG stability while preserving the high nutritional value (Boukid, Folloni, 209 Ranieri, & Vittadini, 2018). The first crucial step is to perform an efficient separation of the WG from 210 the other flour components. Two different approaches have been developed: (i) direct 211 degermination, which is performed before the milling process and (ii) indirect degermination, which 212 is realized through gradual separation phases during the milling process (Boukid, Folloni, Ranieri, & 213 Vittadini, 2018). A high recovery of the WG fraction enables the subsequent adoption of stabilization 214 strategies through the deactivation of the oxidative enzymes or/and removal of the oil fraction. 215 These strategies are based on physical, chemical or biological methods, as extensively revised by 216 Boukid, Folloni, Ranieri, & Vittadini (2018).

WB (17% of the caryopsis), together with the aleurone layer and remnants of the starchy endosperm and WG, produces a range of milling by-products which are recovered at different stages in the mill (Hemdane et al., 2016). The bran fractions can be classified as different by-products (i.e., coarse bran, coarse weatings, fine weatings and low-grade flour), which are roughly distinguishable based on two main characteristics: particle size and endosperm content (Hemdane et al., 2016).

7

222 The bran streams recovered further down the milling process consist of finer bran particles and 223 contain relatively more endosperm (Hemdane et al., 2016). Coarse bran mainly consists of non-224 starch carbohydrates, with 17%-33% arabinoxylan (AX), 9%-14% cellulose, 3%-4% fructan and 1%-225 3% mixed-linkage β -D-glucan as major components (Hemdane et al., 2016). In order to study the 226 mechanisms through which bran affects breadmaking, multiple approaches which change bran 227 functionality (physical properties, chemical composition, and/or enzymatic load) have been 228 developed. These strategies (i.e., (i) particle size reduction, (ii) (hydro-)thermal treatments, (iii) pre-229 soaking, (iv) enzymatic treatment, (v) fermentation, and (vi) chemical treatment, reviewed by 230 Hemdane et al., 2016), treat the bran before its re-addition to the flour, and investigate both bran 231 properties and breadmaking performance.

232 In the literature, great efforts have been made to try to understand the functionality of milling by-233 products or to develop new strategies to stabilize them. Conversely, far fewer efforts have been 234 made to try to improve the breadmaking with stabilized milling by-products. The combination of 235 stabilizing treatments with roller milling, followed by flour re-combination/re-constitution and/or 236 enrichment, could provide UWFs with a better nutritional and technological quality. Moreover, 237 although it seems that the roller milling method could ensure better preservation of the wheat's 238 nutritional value (Jones, Adams, Harriman, Miller, & Van der Kamp, 2015), the consumer is still 239 highly attracted by the "stone mill" label on flours or baked products, considering this method 240 better than roller milling (Jones, Adams, Harriman, Miller, & Van der Kamp, 2015). This points out 241 that it is pressing to improve the dissemination of scientific knowledge to the final target.

The presence of WB and WG significantly changes the chemical composition of flour. Regardless of the amount of milling by-products added, UWFs show a low endosperm fraction and include specific flour constituents, such as fibre, oil, enzymes, reactive components and anti-nutrients. The amount of the above constituents can change markedly, depending on various factors: (i) level of addition, (ii) milling by-product treatment (if any), (iii) milling process, (iv) bran stream fraction (v) wheat species and cultivar, and (vi) environmental conditions.

248 Considering WG, the main constraints associated with its utilization in the baking industry are 249 represented by its poor chemical stability, the presence of reducing compounds (glutathione) that 250 degrade breadmaking ability, and the presence of non-polar lipids which tend to destabilize gas cells 251 (Tebben et al., 2018; Boukid, Folloni, Ranieri, & Vittadini, 2018).

On the other hand, flour supplementation at different levels and/or with different bran fractions
has shown deleterious impacts on the technological properties of doughs and breads (Hemdane et

al., 2016). Overall, flours containing bran produce a poor loaf volume, dark colour, dense and firm
texture, and bitter taste (Hemdane et al., 2016). The following reasons account for these negative
effects: fibre-gluten interactions, dilution of gluten proteins by non-endosperm proteins, fibre
competition for water resulting in insufficient hydration of gluten and starch, physical effects of
bran particles and bran constituents on the gluten network and high level of ferulic acid (Tebben et
al., 2018).

260

261 3.1.1 Unprocessed UWFs

The incorporation of raw milling by-products in flour without introducing some modification of the product formula and/or processing method gives "unprocessed UWF". It generally shows a poor breadmaking performance. However, some studies have introduced raw milling by-products into the flour while maintaining an acceptable bread quality (Table 2).

266 Banu, Stoenescu, Ionescu, & Aprodu (2012) investigated the addition of bran streams (3%, 5%, 10%, 267 15%, 20%, 25%, 30% expressed on total flour weight) to refined flour on dough rheology and bread 268 quality. Despite the negative effect of ash on the technological rheological properties of flour and 269 on bread quality, the incorporation of 25% bran streams showed the same ash content as UWF, but 270 with an improvement in bread quality dough rheology and bread physical parameters were 271 significantly improved. Therefore, this level can be used to obtain high nutritional quality bread 272 without improvers in the formulation of incorporation can be used to increase the nutritional value of the breads with less damage on the bread quality compared to wholewheat flour. 273

Blandino et al. (2013) tested refined flour enriched with selected fractions of the kernel obtained by sequential pearling of wheat kernels and added at 5 different levels (5%, 10%, 15%, 20%, 25% expressed on total flour weight) on dough rheology, bread quality and nutritional properties. The presence of a 10% pearled fraction enhanced the nutritional value of the bread, revealing only a slightly increase in deoxynivalenol contamination and showing a technological quality comparable to the control.

Bagdi et al. (2015) evaluated the breadmaking potential of an aleurone-rich flour (ARF) (40g/100g, 75g/100g) in comparison with refined bread. The ARF was suitable for breadmaking without any flour additives. Bread made with aleurone-rich flour showed better nutritional properties than refined bread, but a low technological quality. The optimal blending ratio for the sensory quality resulted 40g/100g since it showed a similar acceptability to the control sample. Sun, Zhang, Hu, Xing, & Zhuo (2015) tested different levels (0%, 3%, 6%, 9%, 12% expressed on total flour weight) of WG flour to improve the quality of Chinese steamed bread (CSB). WG negatively affected both the dough and bread properties, and the steaming performance, in proportion to the level of addition. The incorporation of up to 6% WG showed fewer negative effects and gave CSB with acceptable sensory characteristics, suggesting that this blend could be used for the production of functional breads.

291 Pasqualone et al. (2017) compared the effect on breadmaking ability of two substitution levels 292 (100g/1kg, 200g/1kg) of three different durum wheat milling by-products: i) residuals of the second 293 and third debranning steps (DB), ii) the micronized and air-classified thin fraction from the same 294 residuals (MB), or iii) coarse bran from roller milling of non-debranned durum wheat (B). MB and 295 DB did not alter the textural properties compared to B. Furthermore, the addition of MB (100g/1kg) 296 improved the nutritional value of the bread without reducing its quality. Hence, debranning 297 followed by micronization could represent an interesting strategy for UWF breads from durum 298 wheat.

299

300 3.1.2 Processed UWFs

In the literature several treatments have been developed for wheat kernels and milling by-products,
 namely "processed UWF". The processing treatments that showed technological improvements for
 the use of processed UWFs in breadmaking are reported below (Table 3).

304

305 3.1.2.1 Pre-treatment of wheat kernels: germination

306 In recent years, several studies have investigated the impact of the germination process on the 307 technological and nutritional quality of cereals, pseudo-cereals and pulses (Lemmens et al. 2019; 308 Bellaio, Kappeler & Bühler, (2013); Richter, Christiansen, & Guo, 2014; Benincasa, Falcinelli, Lutts, 309 Stagnari, & Galieni, 2019). In fact, by inducing the activation of hydrolytic enzymes for plant growth 310 and development, germination leads to a considerable improvement in the product nutritional 311 value, which makes this process attractive for healthy and functional foods. Applications of the 312 germination process in breadmaking are reported in the literature, but most of these studies used 313 refined wheat flours in the bread formula (Lemmens et al., 2019). The main issue of this approach 314 is optimizing the processing conditions: longer germination times (>72 h) are required to improve 315 the nutritional value of the flour, but they negatively impacted the flour technological performance; 316 on the other hand, only shorter times (20-36 h) or low substitution levels (10%-20%) improved the technological properties of doughs and breads, although they showed lower effects on their nutritional value (Lemmens et al., 2019). These results reflect the different degrees of enzyme activities in wheat kernels as a function of the germination time. Activation of the proper alfaamylase activity can promote yeast fermentation, carbon dioxide production and gas cell expansion, thus determining a higher oven spring and improving bread volume (Lemmens et al., 2019). Moreover, optimized alfa-amylase activity can also improve the product's shelf life and sensory quality (Lemmens et al., 2019).

- Marti, Cardone, Nicolodi, Quaglia, & Ambrogina Pagani (2017) proposed the addition of a low amount of germinated wheat flour (1.5%) for a long amount of time (72-90 h) as a natural improver in breadmaking with refined flour. Germinated flour produced similar effects to common improvers (i.e., 0.5% malt or 0.5% enzymatic improver). It could be interesting to test the effect of this natural improver on the technological performance of UWF.
- Richter, Christiansen, & Guo (2014) developed a 100% germinated white spring wholewheat flour for bread applications. The authors compared 100% white wholewheat flour (control) with 100% germinated white wholewheat flour, including different additions of vital gluten (0%-5%) in the bread formula (Richter, Christiansen, & Guo, 2014). Germinated wholewheat flour significantly increased loaf volume (5%-9%), independently of the presence of gluten (Richter, Christiansen, & Guo, 2014). Moreover, germinated flour significantly improved the sensory quality of the bread, reducing the bitter taste.
- Zilic et al. (2016) investigated the effect of germination on wholewheat flour proteins. Intensive protein hydrolysis was revealed by an increase in free SH groups and a decrease in albumin + globulin polypeptides with a molecular weight of over 85.94 kDa and between 85.94 and 48.00 kDa. Although this modification affected the dough's viscoelastic properties, germinated wheat flour is proposed as a potential food ingredient owing to the high antioxidant capacity and reduced antigenicity of the glutenin fraction (Zilic et al., 2016).
- Ding et al. (2018) tested germination time on the functionality of wholewheat doughs. Controlled germination for 5-15 h (T= 28±2°C and RH=95±3%) produced wholewheat flour with improved functionality: enhanced glucose content, reduced starch retrogradation during gelatinizing gelatinization, improved gluten quality, and increased dough stability during mixing (Ding et al., 2018).
- Johnston et al. (2019) applied controlled germination in the production of wholewheat. Controlled germination (t=24 h, T=21°C, FN=200 s, excess of water) to increase the activity of α -amylase by

decreasing the Falling Number (FN) from 350 s to 200 s reduced dough mixing time and increased bread specific volume (Johnston et al., 2019). Sensory analysis revealed a higher acceptability for germinated wholewheat breads, thanks to their lower degree of bitterness, greater sweetness and moisture (Johnston et al., 2019). Hence, germinating wholewheat flour to an FN value of 200 s proved to be a good strategy to improve flour functionality and consumer preference for wholewheat breads (Johnston et al., 2019).

A very recent work by Cardone, Marti, Incecco, & Pagani (2020) showed promising results on the application of a controlled germination to enhance the quality of wholewheat breads. In fact, under the conditions applied in this study (48h, 20°C, 90% relative humidity), although the decrease in

dough rheological features, a significant improvement in gluten stretching ability and bread physical
 properties was obtained.

Further studies on the application of germination to improve the UWF breadmaking performance could be an interesting field of research. The substitution of refined flour with UWF may not require additional enhancement of the nutritional quality, since the raw material is naturally characterized by a high nutritional value.

364

365 3.1.2.2 Pre-treatment of milling by-products

366 Over time several pre-treatments have been developed for milling by-products to stabilize their 367 chemical composition and better understand their effects on the breadmaking performance 368 (Hemdane et al., 2016; Boukid, Folloni, Ranieri, & Vittadini, 2018). The present review reports the 369 results of the most promising treatments on technological quality (Table 4).

370 Hemdane et al. (2016) extensively reviewed wheat bran pre-treatments. Chemical treatments did 371 not improve bread quality; very little work has been performed on enzymatic treatments (Santala, 372 Lehtinen, Nordlund, Suortti, & Poutanen, 2011; Messia et al. 2016), while the effects of bran particle 373 size reduction and (hydro)thermal treatments on breadmaking still remain unclear. Pre-soaking 374 appears to be a promising strategy for the incorporation of bran in breadmaking since it generally 375 improved the bread quality (Messia et al. 2016); however, a complete understanding of this 376 approach has yet to be established. In a recent work following the method proposed by Wang et al. 377 (2015a, b), Zhang et al. (2019) developed an arabinoxylan-enriched flour (AXF) (ash content 378 between 39.2%-55.8%) as a fibre supplement (2%, 5%, 10%) for refined bread. AXF pre-soaking 379 positively affected flour functionality, resulting in bread with comparable properties to the control

380 (Zhang et al., 2019). Fermentation treatment has been reported to enhance breadmaking ability: it
381 was effective in improving bread volume and crumb softness.

382 The impact of WG supplementation on breadmaking is reviewed by Boukid, Folloni, Ranieri, & 383 Vittadini (2018). The rheological properties of doughs were affected by the different WG 384 treatments. Fermentation by lactic acid bacteria appeared to be the most promising treatment, 385 showing a significant improvement in the dough properties. In all cases, the addition of more than 386 20% WG severely damages dough quality. Considering bread quality, the addition of up to 5% 387 extruded WG increased bread volume and decreased bread firmness. Sourdough fermentation of 388 WG positively affected bread quality, by decreasing crumb firmness, resilience and fracturability, 389 and enhancing bread shelf life without reducing product acceptability (Boukid, Folloni, Ranieri, & 390 Vittadini, 2018).

These results revealed that the use of sourdough fermentation on milling by-products enhanced the performance of UWF in breadmaking. Indeed, Gobbetti, Rizzello, Di Cagno, & De Angelis (2014) extensively reported the positive effects of using this approach for wholegrain products. Prefermentation allowed modification of the techno-functionality of the milling by-products, showing improved technological quality in terms of dough retention capacity, loaf volume and crumb softness during storage. In addition, it decreased the anti-nutritive factors and enhanced the sensory properties.

Recently, Pontonio et al. (2020) proposed an integrated biotechnological approach, combining LAB fermentation with xylanase treatment on milling by-products (Pontonio et al., 2020). Biochemical and nutritional analysis revealed that fortified breads had higher protein digestibility and a lower glycemic index combined with a better sensory quality (Pontonio et al., 2020). Therefore, a significant improvement could be achieved in UWF breads by applying an integrated approach, suggesting new strategies for the exploitation of UWF.

404

The reviewed studies identified effective technological strategies for producing UWF with a high technological quality. The possibility of supplementing bread with even low amounts of milling byproducts, without decreasing the bread quality, should be regarded as a technological success.

408

409 3.2 Bread formulation and improvers for UWF

13

In the literature the most common solution to improve the breadmaking performance of UWF and,
consequently, the quality of UWF breads is to modify the bread formula; the literature results are
outlined in Table 5.

413

414 3.2.1 Optimization of water amount for UFW breads

415 Some improvements in breadmaking with UWF can be obtained by optimizing the amount of water in the bread recipe (Table 5). Cappelli et al. (2018) examined the effect of water (70%, 76%, 82%, 416 417 88%, 94% expressed as a percentage of the dry weight of the flour) and degree of flour refinement 418 (refined, brown and wholewheat flour) on the dough rheology. Significant differences in rheological 419 properties were found for refined flour compared to UWF, showing that alveographic analysis 420 cannot be extended to unrefined doughs. Addition of the optimal amount of water, modelled in 421 function of the degree of flour refinement, could be a strategy to optimize the rheological 422 parameters relating to product quality: flour strength "W" and the ratio between tenacity "P" and 423 extensibility "L", P/L (Cappelli et al., 2018). This approach could improve UWF dough quality, 424 without introducing additional ingredients to the recipe.

Similar results were reported in a survey conducted by Guerrini, Parenti, Angeloni, & Zanoni (2019) on the breadmaking process with UWF. The creation of highly hydrated doughs improves the flour workability and bread quality (Guerrini, Parenti, Angeloni, & Zanoni, 2019). Indeed, in the literature it is known that the inclusion of bran significantly affects the water adsorption capacity of the flour and causes a competition for the water uptake with the other flour constituents (Hemdane et al., 2016). High water quantities could allow proper hydration of the gluten matrix even in the presence of bran, resulting in a better P/L balance as well as a higher W.

All these results may derive from the wide utilization of the Farinographic test as the official
predictor of the water absorption of the flour: this evaluation works well for refined flour, but it is
not suitable for UWF (Bruckner et al., 2001; Schmiele, Jaekel, Patricio, Steel, & Chang, 2012;
Hemdane et al., 2016).

436

437 3.2.2 Modification of the flour for UWF bread

In the literature, few process strategies have tested a more "natural approach" to the use of
improvers: the addition of UWF flour in a modified form, as reported in Table 5. Parenti et al. (2019)
reported an improvement in breadmaking performance with the use of pre-gelatinized brown flour
(6%). Pre-gelatinized UWF was obtained by heating some of the bread dough flour to 85°C in water;

the product was cooled to room temperature and tested on the dough and bread properties. The
addition of the flour in a different physical form increased the water absorption capacity, improved
the alveographic parameters, and increased the bread volume, crumb softness and shelf life (Parenti
et al., 2019).

Hung, Maeda, & Morita (2007) tested the addition of whole waxy flour in order to improve the quality of high-fibre bread. Different levels of whole waxy flour were used to substitute refined flour (10%, 30%, 50% expressed on total flour weight); the resultant flour mixtures were tested on the breadmaking performance of wholewheat flour compared to refined flour. This strategy improved crumb softness during bread storage (Hung, Maeda, & Morita, 2007). It could be interesting to investigate the use of whole waxy flour compared to 100% wholewheat flour for breadmaking, in order to evaluate the impact of this ingredient on the quality of wholewheat bread.

453

454 3.2.3 Improvers for UWF bread

455 The use of improvers in breadmaking with UWF is summarized in Table 5. Tebben et al. (2018) 456 reviewed the effects of common bread improvers, namely enzymes, emulsifiers, hydrocolloids, 457 oxidants and other functional ingredients on the performance of wholewheat flour. A positive role 458 is outlined for some enzymes: (i) by hydrolysing arabinoxylans (AX), xylanase was reported to 459 decrease the water absorption of the flour, increase the concentration of fermentable sugars in the 460 dough, the rate of fermentation and the dough proof height; moreover, xylanase improved the gas 461 retention capacity, loaf volume, crumb softness and crumb staling; (ii) alfa-amylase appeared 462 beneficial under certain conditions; (iii) G4-amylase showed promising effects on loaf volume, 463 crumb hardness and staling (Tebben et al., 2018).

464 Considering hydrocolloids, a general improvement in dough rheology is reported in the literature 465 (Tebben et al., 2018; Farbo et al., 2020). The effects of hydrocolloids change in function of their 466 typology and level of addition. With regard to bread dough, the use of carboxymethylcellulose 467 (CMC) decreased the final proof time and resistance to extension. Guar gum (GG) combined with an 468 emulsifier (diacetyl tartaric esters of monoglycerides, DATEM) was reported to increase the 469 fermentation stability and slightly increased bread volume. However, both CMC and GG reduced 470 the elasticity of wholewheat dough. Hydroxypropyl methylcellulose (HPMC) increased dough 471 elasticity, proof height, and decreased resistance to extension (Tebben et al., 2018).

15

Farbo et al. (2020) studied the effect of methylcellulose (MC), GG, psyllium gum (PG), xanthan gum
(XG) and tara gum (TG) on the quality of dough made with old durum wheat. They found that 1% of
PG or XG improved dough extensibility, while all hydrocolloids increased gas retention.

475 Considering bread quality, GG was able to increase the specific volume of wholewheat bread.
476 Furthermore, a non-significant effect of HPMC, XG and dextran was reported on bread volume.
477 Conversely, HPMC proved effective in increasing the specific volume of both refined and
478 wholewheat bread, while CMC did not improve the loaf volume of either variety of bread (Tebben
479 et al., 2018).

480 Oxidants are commonly added in breadmaking to increase dough strength by forming disulphide 481 bonds through the oxidation of free sulfhydryl groups on the gluten proteins (Zhou et al., 2014). The 482 presence of reducing compounds in wholewheat flour counteracts the effect of oxidants, which 483 must be added at higher levels (Tebben et al., 2018). Hence, higher amounts of oxidants will also 484 presumably be required for other UWF typologies. Potassium bromate and ascorbic acid improved 485 the dough rheology, dough strength and gas retention ability. The addition of rosehip as a source of 486 ascorbic acid increased the resistance to extension and reduced the extensibility of the wholewheat 487 dough. The best effect on bread volume was reported for ascorbic acid, added at 200 ppm; 488 accordingly, rosehip resulted effective in enhancing bread volume. This latter improver also 489 improved the sensory score of the crumb, increasing the acceptability of the wholewheat bread. 490 Conversely, potassium bromate showed little effect on loaf volume (Tebben et al., 2018).

Emulsifiers in breadmaking cause dough strengthening and/or crumb softening (Tebben et al., 2018). The addition of DATEM was reported by some studies to increase the fermentation stability, whereas the opposite effect was observed by others (Tebben et al., 2018). However, these studies are consistent in showing that DATEM improved dough elasticity, a valuable property for the breadmaking performance. Another emulsifier, sodium stearoyl lactylate (SSL), improved the handling properties of the wholewheat dough (Tebben et al., 2018).

The specific volume of wholewheat bread was generally improved by the addition of emulsifiers (Tebben et al., 2018). DATEM was reported to produce positive effects. Furthermore, the combined addition of DATEM and oxidants improved the gas-holding ability of the dough during the proofing and baking phases. DATEM and SSL had the greatest effect on volume increase, but ethoxylated monoglycerides, succinylated monoglycerides and lecithin significantly increased loaf volume too. On the other hand, polysorbate and monoglycerides did not affect the parameter. Similar results were reported for the inclusion of DATEM, SSL, soy lecithin, polyoxyethylene sorbitan monostearate 504 (polysorbate-60), poly-oxyethylene sorbitan monopalmitate (polysorbate-40) and glycerol-505 monostearate: all these emulsifiers increased the volume of the wholewheat bread. Conversely, the 506 addition of monoglycerides, DATEM and SSL was not effective in improving wholewheat or refined 507 bread specific volume (Tebben et al., 2018).

508 DATEM and mono- and diglycerides were reported to improve the crumb structure of wholewheat 509 bread; a similar effect was observed with SSL as well as an increase in the eatability score (Tebben 510 et al., 2018).

511 The supplementation of vital gluten is effective in overcoming the multiple problems related to 512 wholewheat bread (Tebben et al., 2018).

513 Parenti, Guerrini, Cavallini, Baldi, & Zanoni, (2020) tested the addition of 7 improvers (i.e., sucrose, 514 sodium chloride, extra virgin olive oil, gelatinized flour, GG, ascorbic acid and ice) to optimize the 515 quality of wholewheat bread. The optimized sample resulted from the combination of sucrose (2%) and extra virgin olive oil (3%), disclosing the interesting role that these improvers can play in the 516 517 quality of wholewheat bread (Parenti, Guerrini, Cavallini, Baldi, & Zanoni, 2020). Furthermore, the 518 authors proposed a two-step optimization approach for improving the use of UWF in breadmaking: 519 (i) the Screening Design method revealed the most relevant factors affecting bread quality; (ii) the 520 Full Factorial Design gave an in-depth evaluation of the selected variables and allowed identification 521 of the optimized sample (Parenti, Guerrini, Cavallini, Baldi, & Zanoni, 2020).

All these results concerned the use of improvers on wholewheat flour. However, due to the very different composition of the raw materials which probably changes the effects of the improvers, they should be further tested before extending these findings to all UWF breads.

525

526 3.3 The breadmaking process with UWFs

527 Only a few studies have investigated the possibility of modifying the breadmaking operating 528 conditions. The breadmaking process has been designed to maximize the quality of refined bread. 529 Therefore, the substitution of refined flour with UWF may require an adaptation of the process to 530 the different characteristics of the raw material. Processing conditions, such as the type of mixer, 531 mixing time and speed, resting period etc., may require modifications from the standard procedure. 532 This latter area of research appears poorly investigated in the literature, since the greatest efforts 533 have been made in modifying the bread formulation, while the breadmaking variables were kept 534 almost unchanged. Studies about modifications of the breadmaking process with UWFs are 535 reported in Table 6.

536

537 3.3.1 Mixing

The mixing is one of the most important phases in the breadmaking process since most of the characteristics of the final product are determined during this phase (Zhou et al., 2014). Considering the different composition of UWF, modification of the mixing variables (type of mixers, mixing speed, mixing time...) could represent a good strategy to be explored, despite being poorly investigated in the current literature.

Angioloni & Rosa (2006) tested the effect of mixing time (10-15-20 s) combined with an improver (cysteine, 20 mg/kg) on the rheological properties of refined and wholewheat dough obtained at high-speed revolutions (1600 rpm). Dough viscoelastic behaviour was affected by both cysteine and kneading conditions. Cysteine significantly reduced the mixing time (optimum = 15 s) by decreasing the elastic component of the dough and aiding dough relaxation in both refined and wholewheat flour. Therefore, the use of high-speed mixing combined with cysteine could be useful to improve UWF doughs.

Parenti et al. (2013) tested different mixing times on the breadmaking of brown flour. Two trials evaluated different mixing times ((i) 12, 17, 22 min; (ii) 17, 22, 27 min). Mixing time significantly affected loaf increase during proofing: the samples mixed for 17 min showed the highest value in both trials. Furthermore, doughs obtained at the optimum mixing time (17 min) were characterized by a better water retention capacity during storage.

555 The control of the mixing time also proved to be extremely important in the survey by Guerrini, 556 Parenti, Angeloni, & Zanoni (2019); short times, between 10 and 20 min, represented one of the 557 most effective strategies for the breadmaking process with UWF.

A recent work by Cappelli, Guerrini, Cini, & Parenti (2019) investigated the delayed addition of bran and middlings during the mixing step. Three bran and middlings incorporation substitution levels (10%, 20%, 30% expressed on refined flour weight) and five times of addition (0, 2, 3.5, 5, 6.5 min) were tested on the dough rheology and bread quality. The addition of bran and middlings at 2 min into the mixing step improved the dough rheology and increased the bread specific volume. Furthermore, the combination of 10% bran and middlings with time of 2 min produced bread of a better quality than the control bread (i.e., without delayed addition).

565 A specific laboratory test, developed to predict the optimized mixing time, could boost the research 566 on the mixing step, but, to the best of the author's knowledge, no such test currently exists.

567

568 3.3.2 Leavening

569 During leavening, the bread loaf develops its final structure and several modifications of its 570 constituents occur as a function of the different leavening agents used in the recipes. Sourdough 571 fermentation represents one of the oldest biotechnologies in cereal food production; however, when industrial-scale baking was developed in the 19th century, baker's yeast - Saccharomyces 572 573 cerevisiae - became the most common leavening agent (Zhou et al., 2014). In recent years, the 574 increasing interest in healthy and functional foods has led to a rediscovery of sourdough bakery 575 products, which are characterized by positive health benefits and unique flavours (Zhou et al., 2014). 576 Chavan & Chavan (2011) made an exhaustive review of this ancient biotechnology. In the present 577 review, only the issues related to the technological performance of UWF are discussed. In the 578 literature it is largely reported that the substrate, mainly flour, used for sourdough production 579 deeply influences its properties (Chavan & Chavan, 2011; Decock, & Cappelle, 2005). The presence 580 of bran, increasing the ash content of wheat flour, promotes the growth of lactic acid bacteria (LAB) 581 and increases the acidification of the sourdough system. LAB are responsible for the production of 582 several organic acids, which are reported to improve the swelling of gluten and increase gas 583 retention, while functioning as natural dough conditioners and reducing bread staling. Furthermore, 584 the acid enhances the solubility of the glutenin fraction, improving the swelling power of the gluten 585 (Chavan & Chavan, 2011). Hence, the use of UWF for sourdough production seems to improve the 586 breadmaking performance, thanks to a better development of the gluten matrix. Studies on UWF 587 performance are reported in Table 6.

588 In the survey by Guerrini, Parenti, Angeloni, & Zanoni (2019), all of the bakers use sourdough as the 589 leavening agent for breadmaking with UWF: they perceive that this method improves the quality of 590 the final product.

591 Komlenić et al. (2010) showed the positive effects of biological acidification on the quality of bread 592 obtained with refined and wholewheat flour. They investigated the effect on dough and bread 593 properties of three different acidifications: chemical (lactic acid) and biological (dry sourdough and 594 *Lactobacillus brevis* pre-ferment) acidification. The bread specific volume was only significantly 595 increased by the biological acidifiers, whereas the acidifier typologies improved the crumb hardness 596 (Komlenić et al., 2010). Therefore, dry sourdough, characterized by a longer shelf life and better 597 stability, could be an interesting strategy for breadmaking with UWF.

Taccari et al. (2016) reported the possibility of applying the back-slopping technique to produce
type I sourdough from wholewheat flour. Wholewheat sourdough improved the quality of high fibre

breads, overcoming the detrimental effect of bran on bread volume. Moreover, sourdough
fermentation improved bread texture, flavour, nutritional value and shelf life. The study outlines
the suitability of wholewheat flour for sourdough production, encouraging further research for its
application in UWF breadmaking.

604 Choi, Kim, Hwang, Kim, & Yoon (2005) evaluated the application of *Leuconostoc citreum* HO12 and 605 *Weissella koreensis* HO20 isolated from kimchi as starter cultures for sourdough wholewheat bread. 606 The sourdoughs fermented with the selected LAB had an optimal Fermentation Quotient (FQ), a 607 criterion for good bread quality. Although no significant improvement was observed on bread 608 specific volume, the LAB reduced crumb hardness on both fresh and stored breads (Choi, Kim, 609 Hwang, Kim, & Yoon, 2005). Hence, the study presented the potential application of LAB isolated 610 from kimchi for the improvement of UWF bread quality.

Didar et al. (2011) observed positive effects on bread quality (95% extraction rate) and sensory properties upon performing sourdough fermentation with *Lactobacillus plantarum* (PTCC 1058) and *Lactobacillus reuteri* (PTCC 1655). Different dough yields (DY, 250 and 300) and different levels of sourdough addition (10%, 20%, 30%) were also tested. *Lb. plantarum* sourdough with a DY of 250 and 30% addition produced the greatest effect on the overall quality score of the breads (Didar et al., 2011).

617 Katina, Heiniö, Autio, & Poutanen (2006a) studied the influence of sourdough conditions on bread 618 flavour and texture. Ash content (0.6-1.8 g/100 g), fermentation temperature (16-32°C), and 619 fermentation time (6-20 h) were considered independent factors and different starter cultures (i.e., 620 Lactobacillus plantarum, Lactobacillus brevis, Saccharomyces cerevisiae or a combination of yeast 621 and LAB) were tested. Ash content and lactic acid fermentation were the main factors affecting the 622 intensity of the sensory attributes. The greater the ash content, the higher the intensity of both 623 desired and undesired flavour attributes. An optimization of the process conditions, according to 624 the ash content and the specific LAB strain, improved the sensory quality of UWF breads. However, 625 the improvement of bread volume and texture required different optimized conditions than those 626 required for bread flavour. Hence, an efficient use of sourdough fermentation has to consider its 627 end use in wheat baking (Katina, Heiniö, Autio, & Poutanen, 2006a).

The results from the use of sourdough as a leavening agent showed positive effects on UWF bread quality. The drawback of this procedure is primarily represented by the great variability of the sourdough composition, which makes the process difficult to standardize. Further research is

20

631 needed to find new solutions to combine the use of this leavening agent with a standardization of632 bread features.

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634 3.3.3 Baking

Baking is the final step in the breadmaking process. The phenomena occurring during this phase
include gas evaporation, starch gelatinization, modification of the bread loaf from a sponge-like to
a porous structure, and water evaporation. The most significant factors of the baking step are
represented by temperature, time and moisture (Zanoni, Peri, & Pierucci, 1993; Zanoni, Pierucci, &
Peri, 1994; Zhou et al., 2014).

640 In the literature there appears to be a lack of information on the baking step specifically developed 641 for UWF breads. Guerrini, Parenti, Angeloni, & Zanoni (2019) reported that bakers create high 642 temperatures at the beginning, followed by a temperature decrease, to improve the quality of UWF 643 breads. Moreover, the majority of bakers check the moisture during this step, since it represents 644 another critical factor affecting bread quality. In fact, especially in the first phase of baking, the 645 addition of moisture improves loaf expansion. Therefore, modification of the baking conditions, 646 such as temperature and moisture, in function of the characteristics of the raw material could 647 represent another interesting field of exploration.

648

649 3.4 UWF bread storage

650 The most important phenomena limiting the shelf life of breads are bread staling and microbial 651 growth (Fernandez, Vodovotz, Courtney, & Pascall, 2006). Bread staling is a complex phenomenon, 652 whose mechanism has not been well established yet; however, the most important factors seem to 653 be starch retrogradation, starch-gluten interaction and moisture redistribution (Fadda, Sanguinetti, 654 Del Caro, Collar, & Piga 2014; Curti, Carini, Tribuzio, & Vittadini, 2015). Bread microbial spoilage is 655 generally caused by moulds, bacteria and yeasts (Melini, & Melini, 2018). Different approaches have 656 been developed to reduce bread staling and microbial spoilage, which generally achieve positive 657 effects, allowing the production of breads with a shelf life of up to 4 weeks (Fadda, Sanguinetti, Del 658 Caro, Collar, & Piga 2014; Sargent, 2008). Hence, bread flavour and aroma have become the new 659 limiting factors for bread shelf life.

660

661 3.4.1 Improving the shelf life of UWF bread

662 Different strategies can be applied to extend bread shelf life: (i) direct approach on the food matrix;

663 (ii) indirect approach through packaging systems.

Within the direct approaches, Gobbetti, Rizzello, Di Cagno, & De Angelis (2014) reviewed the importance of fermentation of the raw material for wholegrain products. Specifically, the application of this method on milling by-products before their incorporation in the bread formula was reported to improve crumb softness during bread storage.

Furthermore, the germination process also showed positive effects on bread storage, linked to theactivation of alfa-amylase activity (Lemmens et al., 2019).

670 The supplementation of pre-gelatinized UWF in the bread formula delayed bread staling, in terms671 of crumb specific volume and texture parameters (Parenti et al., 2019).

With regard to improvers, different enzymes reduce the staling of UWF bread: (i) xylanase and (ii)
alfa amylase result the most effective enzymes; furthermore, one study has reported that (iii) G4amylase showed a positive outcome, but further research is necessary to confirm this result (Tebben
et al., 2018).

The effects of emulsifiers on wholewheat breads were reported by Tebben et al. (2018). DATEM showed anti-staling properties. A reduction in hardness was also reported for wholewheat bread with 0.4% DATEM or 0.6% monoglycerides. Similarly, 0.5% SSL was able to decrease the staling rate of wholewheat bread over 4 days of storage. It is interesting to note that DATEM and SSL only acted as crumb softeners in wholewheat breads but not in refined breads (Tebben et al., 2018).

The use of hydrocolloids in UWF breads led to controversial results. Both CMC and GG reduced the staling rate of wholewheat bread; HPMC softened the crumb of both wholewheat bread and refined breads, while another study reported that CMC inclusion was ineffective for both bread typologies. Furthermore, the literature reported that dextran and HPMC produced a non-significant reduction in the initial loaf hardness and delay in bread staling. Hence, further research is necessary to better understand the role of emulsifiers on UWF bread staling (Tebben et al., 2018).

Malted wholewheat flour in breadmaking reduced the staling of wholewheat bread (Tebben et al.,2018).

Some efforts have been made to increase oxidative stability during the storage of UWF breads,delaying rancidity phenomena.

Jensen, Ostdal, Skibsted, & Thybo (2011b) tested three antioxidants, alfa-tocopherol and fat-soluble
and water-dispersible rosemary extracts, on the sensory profile and antioxidant capacity of
wholewheat bread during storage. These antioxidants did not improve the sensory quality or

stability of the wholewheat bread (Jensen, Ostdal, Skibsted, & Thybo, 2011b). Furthermore, alfatocopherol produced fresh wholewheat bread with higher concentrations of hydroperoxides and
secondary lipid oxidation products, similarly to the stored control sample (Jensen, Ostdal, Skibsted,
& Thybo, 2011b). Hence, lipid oxidation is responsible for less favourable sensory notes like a rancid
aroma and flavour, bitter taste and astringency, attributes most often associated with low product
acceptability (Jensen, Ostdal, Skibsted, & Thybo, 2011b).

700 Ning, Hou, Sun, Wan, & Dubat (2017) tested green tea powder (GTP) on the quality and antioxidant 701 activity of wholewheat dough and bread. Five levels of GTP were tested (0 g, 1 g, 2 g, 3 g, 4 g/100 g 702 flour): the higher the amount of GTP included, the worse the bread quality, while the antioxidant 703 activity showed a reverse trend (Ning, Hou, Sun, Wan, & Dubat, 2017). The best result was obtained 704 with GTP 1 g/100 g, since it did not affect bread quality while enhancing the antioxidant capacity 705 (Ning, Hou, Sun, Wan, & Dubat, 2017). Hence, 1 g/100 g GTP resulted an effective improver in 706 reducing the rate of peroxide accumulation in wholewheat bread during storage (Ning, Hou, Sun, 707 Wan, & Dubat, 2017).

708 Lu, & Norziah (2011) studied the effect of substituting shortening with different levels of 709 microencapsulated n-3 polyunsaturated fatty acid (PUFA) powder (1%, 1.75%, 2.5% of total dough 710 weight) on the sensory and oxidative stability of UWF bread during storage. The flour used was a 711 blend of wholewheat and refined flour (Lu, & Norziah, 2011). Breads containing PUFA were no 712 different to the control containing shortening, revealing that PUFA had a similar effect on bread 713 quality (Lu, & Norziah, 2011). The lowest PUFA addition (1%) resulted in bread with the best sensory 714 acceptability for up to 3 days of storage, suggesting that this improver could be an effective 715 substitute for shortening (Lu, & Norziah, 2011).

With regard to the breadmaking process, the most effective variable in enhancing the shelf life of
UWF bread was sourdough fermentation (Chavan & Chavan, 2011; Taccari et al., 2016; Choi, Kim,
Hwang, Kim, & Yoon, 2005).

The evaluation of the sensory profile of wheat bread during shelf life has been little investigated in the literature. Significant changes in the flavour, aroma and taste of refined and wholewheat bread have been reported by Jensen, Oestdal, Skibsted, Larsen, & Thybo (2011a). The sensory characteristics of refined bread and wholewheat bread during storage were studied by measuring volatile and non-volatile compounds and performing a descriptive sensory profiling (Jensen, Oestdal, Skibsted, Larsen, & Thybo, 2011a). Refined and wholewheat bread showed distinctive flavours, revealing two different sensory profiles (Jensen, Oestdal, Skibsted, Larsen, & Thybo, 726 2011a). Storage time affected 8 out of 13 of the tested attributes of refined bread, while all 44 13 727 attributes of wholewheat bread were significantly impacted by storage time (Jensen, Oestdal, 728 Skibsted, Larsen, & Thybo, 2011a). The fresh wholewheat samples were characterized by higher 729 concentrations of fermentation products; after one week of storage, dough and bran aroma were 730 the predominant attributes, while breads stored up to 2-3 weeks were defined by rancid and fatty 731 aromas, and a bitter taste (Jensen, Oestdal, Skibsted, Larsen, & Thybo, 2011a). The formation of off-732 flavours in bread could be related to the formation of secondary lipid oxidation products during 733 storage together with a reduction in compounds from Maillard reactions (Jensen, Oestdal, Skibsted, 734 Larsen, & Thybo, 2011a). Since UWFs are characterized by higher enzymatic activity and lipid and 735 antioxidant contents than refined flours, the development of specific strategies for the control of 736 oxidative reactions represents a key factor for improving bread storage (Doblado-Maldonado, Pike, 737 Sweley, & Rose 2012).

738

739 3.4.2 Packaging of UWF breads

540 Several packaging strategies have been developed to preserve bread freshness. The main objectives 541 of these methods are to prevent microbial spoilage and bread staling. Bread packaging has been 542 studied on refined bread, while no techniques have been specifically developed to preserve UWF 543 bread.

- Therefore, here we discuss the packaging strategies that appear promising to us for extending theshelf life of UWF breads.
- Packaging methods are classified as conventional and active packaging. The former includes traditional packaging methods, aimed at preserving the food from chemical, physical and biological damage without interacting with it. Conversely, active packaging is based on the interaction between the packaging material and the food matrix by absorbing or releasing specific substances (Melini, & Melini, 2018).
- WG makes UWF particularly susceptible to lipid oxidation (Boukid, Folloni, Ranieri, & Vittadini, 2018), and the fibre component also impacts the product moisture during storage time (Hemdane et al., 2016). Hence, the critical aspects of UWF storage could be identified as rancidity phenomena and higher moisture retention, linked to microorganism spoilage.
- Active Packaging with Antimicrobial Releasing Systems could be useful in preventing UWF bread spoilage. These methods release antimicrobial agents (organic acids, fungicides, alcohols and

antibiotics) into the food surface, thus inhibiting or delaying microbial growth and spoilage (Melini,
& Melini, 2018).

Active Packaging with oxygen absorbers could be even more interesting for UWF (Alhendi & Choudhary, 2013; Melini, & Melini, 2018). In fact, Nielsen, & Rios (2000) observed that oxygen absorbers combined with essential oils prevented microorganism spoilage. Furthermore, Latou, Mexis, Badeka, & Kontominas (2010), by combining oxygen absorbers with an alcohol emitter, and Tian, Decker, & Goddard (2012), using metal chelating carboxylic acids, reported effective prevention against microorganism spoilage and lipid peroxidation.

765 The innovative trend in Active Packaging includes the application of nanotechnology, a fusion of 766 traditional packaging polymers with nanoparticles. These methods are able to extend a product's 767 shelf life, while reducing the addition of preservatives in the food formulation (Melini, & Melini, 768 2018). Being a "natural" approach, nanotechnology could be applied to UWF bread, since it could 769 preserve the high nutritional value of the product. Silver nanoparticles included in polypropylene 770 food containers were reported to keep bread fresher over 3 or 4 times longer and to reduce 771 bacterial growth by 95% compared to conventional food containers (Bumbudsanpharoke et al., 772 2015). Moreover, nanoencapsulation applied to essential oils, which show potent antimicrobial 773 and/or antioxidant properties, may represent another promising technique for UWF bread. This 774 method protects the compound against chemical reactions and undesirable interaction with the 775 food matrix (Melini, & Melini, 2018). Nanoencapsulated essential oils extend the shelf life and 776 maintain the sensory properties of breads (Otoni, Pontes, Medeiros, & Soares, 2014; Gutiérrez, 777 Batlle, Andújar, Sánchez, & Nerín, 2011; Souza, Goto, Mainardi, Coelho, & Tadini, 2013). However, 778 nanoparticles are not inert materials: they may interact with food, its surroundings and negatively 779 impact human health. Therefore, there is an urgent need to assess the risks of this innovative 780 method (Alhendi & Choudhary 2013; Melini, & Melini, 2018).

781

782 4. The carbon footprint of UWF bread

Technological innovations sometimes have negative environmental effects, such as the emission of greenhouse gases (GHG) and waste as a result of manufacturing activities. The Carbon Footprint (CFP) is a useful tool to quantify GHG emission during the life cycle of a product/service, allowing an estimation of its environmental impact. The food industry, including food production, preservation and distribution, consumes a considerable amount of energy which contributes to total CO₂ emission (Roy et al., 2009). Furthermore, consumers in developed countries require safe foods of a high quality, produced with a minimal impact on the environment (Boer, 2002), showing that
sustainability will soon become a primary factor in making food choices a part of food quality criteria
(Andersson, Ohlsson, & Olsson, 1994; Pattara, Russo, Antrodicchia, & Cichelli, 2016).

792 In the literature several papers have analysed the CFP associated with the life cycle of wheat bread 793 (Holderbeke, Sanjuán, Geerken, & Vooght, 2003; Braschkat, Patyk, Quirin, & Reinhardt, 2003; Rosing 794 & Nielsen, 2003; Roy et al., 2009; Pattara, Russo, Antrodicchia, & Cichelli, 2016; Meisterling, 795 Samaras, & Schweizer, 2009; Notarnicola, Tassielli, Renzulli, & Monforti, 2017; Laurence, Hartono, 796 & Christiani, 2018). These studies have shown that the main hotspots in the bread supply chain are 797 the agricultural phase, primarily due to the use of pesticides and fertilizers, followed by the baking, 798 mainly performed with an electric source of energy. The consumption of bread, including 799 refrigerated storage or toasting, has an important environmental impact too (Holderbeke, Sanjuán, 800 Geerken, & Vooght, 2003; Rosing & Nielsen, 2003; Meisterling, Samaras, & Schweizer, 2009; 801 Espinoza-Orias, Stichnothe, & Azapagic, 2011; Notarnicola, Tassielli, Renzulli, & Monforti, 2017; 802 Laurence, Hartono, & Christiani, 2018). Conversely, the CFP associated with the phases of packaging 803 and transport still deserve discussion (Roy et al., 2009).

804 To the best of the authors' knowledge, only one paper has considered flour composition as a 805 variable for CFP estimation (Espinoza-Orias, Stichnothe, & Azapagic, 2011). This paper reported the 806 hot spots in the life cycle of packaged sliced breads from refined, brown and wholewheat flours 807 produced and consumed in the UK (Espinoza-Orias, Stichnothe, & Azapagic, 2011). The key findings 808 showed that the CFP of bread ranges from 977 to 1244 g CO₂eq per loaf of bread (defined as 800 g), 809 and that thick-sliced wholewheat bread packaged in plastic bags has the lowest CFP while medium-810 sliced refined bread in a paper bag has the highest. The degree of refinement of the flour used in 811 the bread recipe made a significant environmental impact: the higher the milling extraction rate, 812 the lower the CFP (Espinoza-Orias, Stichnothe, & Azapagic, 2011). This means that bread with higher 813 degrees of refinement is more ecologically sustainable. However, the reported results could be 814 attributed to UWF breads produced with raw materials not subjected to additional processing. On 815 the other hand, the current literature does not evaluate the environmental impact associated with 816 those techniques designed to increase the technological quality of UWF bread. In our opinion, this 817 topic deserves deeper investigation, considering that environmental impact has becoming an 818 essential quality criterion for a food product (Pattara, Russo, Antrodicchia, & Cichelli, 2016). 819 Therefore, in evaluating the best processing techniques for breadmaking with UWF, computation 820 of the CFP should be included as a quality requirement.

26

822 **5. Conclusions**

The present review reported the main techniques and technologies that have been specifically developed for the use of UWF in the breadmaking process. Although the consumption of UWF breads characterized the greatest part of human history, the introduction of the roller mill in the 19th century led to the use of refined flour with a better technological performance, longer shelf life and sensory quality largely appreciated by modern consumers. Hence, refined flour has become the standard in the development of the quality tests, bread formulation and processing methods applied in each phase of the breadmaking process.

830 In recent years, studies about the positive effects of wholegrain consumption have led to a renewed 831 interest in the employment of UWF in breadmaking. However, although the presence of various 832 amounts of wheat bran and/or WG enhances the flour's nutritional value, these supplementations 833 significantly change its composition. As a result, a different raw material, that is, UWF, can be used 834 as a substitute for refined flour in bread production. The following points summarize the main 835 consequences that the re-introduction of UWF have brought:

- i) The standard tests to predict the breadmaking attitude of flour have remained
 unchanged, often giving an improper evaluation of the potentiality of UWF (i.e., water
 absorption capacity of the flour).
- 839 ii) The main efforts to improve the quality of UWF bread have been focused on optimizing840 the bread formula with the inclusion of various improvers.
- 841 iii) Little research has been conducted on modifying the processing variables of the 842 breadmaking phases (i.e., mixing, resting, leavening and baking); practically the same 843 methods developed for refined flours are adopted for the production of UWF bread too. 844 In our opinion, the different composition of UWF requires specific adaptation of the quality tests, 845 so that this may improve both the technological evaluation and the use of UWF in bread production. 846 Furthermore, new processing methods specifically adapted for the chemical characteristics of UWF 847 may require further investigation as strategies to both preserve the high nutritional value and 848 increase the technological quality of the final products, hence promoting the consumption of 849 healthy foods.

FIGURE CAPTIONS

Fig. 1 Schematic representation of the production of unrefined wheat flours (UWFs): flour enriched with wheat germ, flour enriched with wheat bran, and flour enriched with both wheat germ and bran in the same (i.e., wholewheat flour) or in a different relative proportion to the wheat kernel.

Fig. 2 Schematic representation of the main techniques and technologies reported in the literature for the breadmaking process with unrefined wheat flours (UWFs). (i) Treatments on the raw material (i.e., wheat kernels) before the milling step; (ii) treatments on milling by-products (i.e., wheat germ and bran); (iii) modification of the bread formulation; (iv) modifications of processing variables: mixing, leavening and (v) improvement of bread storage.

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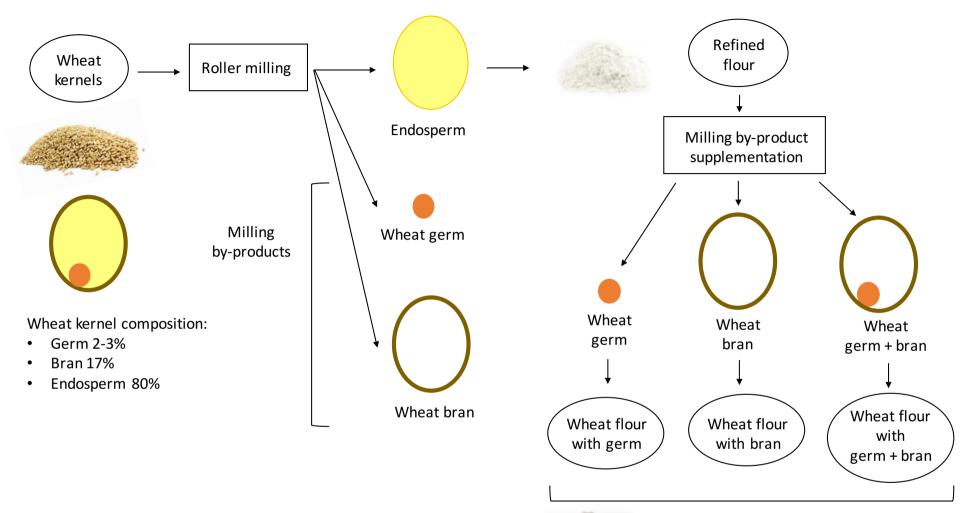
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Unrefined wheat flours (UWFs)

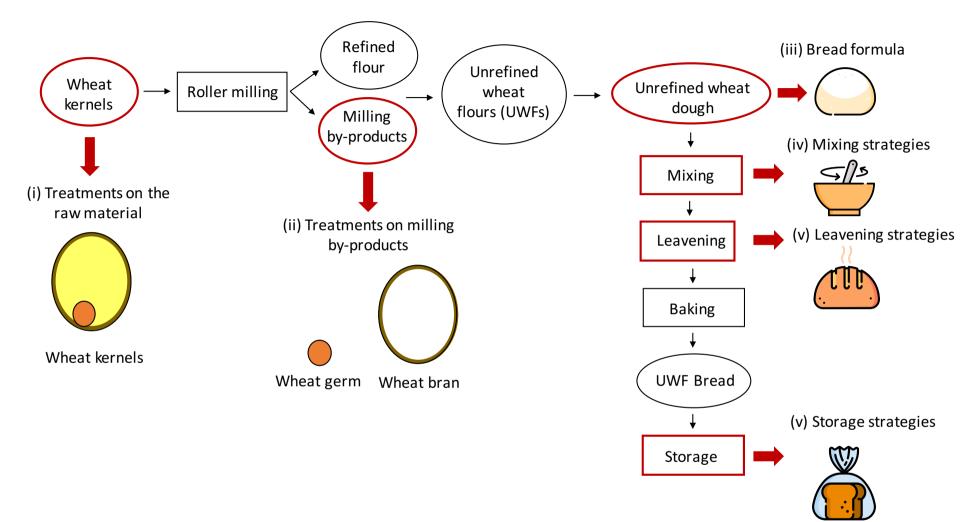


Fig.2

Food matrix/ Process step	Processing strategy			
Wheat kernels	Germination			
Milling Bran pre-soaking				
by-products	Bran/germ fermentation			
Bread	Bread formulation			
dough	-Optimization of water amount			
	-Addition of modified flour			
	Pre-gelatinized flour			
	Waxy wholewheat flour			
	-Addition of improvers			
	• Enzymes			
	, (xylanase, alfa-amylase, G4 amylase)			
	Hydrocolloids			
	(carboxymethylcellulose CMC, guar gum GG, hydroxypropyl methylcellulose			
	HPMC, methylcellulose MC, psyllium gum PG, xanthan gum XG, tara gum TG)			
	Oxidants			
	(ascorbic acid, rosehip, potassium bromate)			
	Emulsifiers			
	(diacetyl tartaric esters of monoglycerides DATEM, sodium stearoyl lactylate			
	SSL, ethoxylated monoglycerides, succinylated monoglycerides, lecithin			
	polyoxyethylene sorbitan monostearate, polyoxyethylene sorbitar			
	monopalmitate, glycerol-monostearate)			
	Vital gluten			
Mixing	Mixing time			
	Delayed addition of milling by-products			
Leavening	Sourdough fermentation			
Bread	Treatments on milling by-products (fermentation)			
storage	Bread formulation			
	-Addition of modified ingredients			
	Pre-gelatinized flour			
	-Addition of improvers			
	Enzymes			
	(xylanase, alfa-amylase, G4 amylase)			
	Emulsifiers			
	(diacetyl tartaric esters of monoglycerides DATEM, sodium stearoyl lactylate			
	SSL, monoglycerides)			
	Hydrocolloids			
	(carboxymethylcellulose CMC, guar gum GG, hydroxypropyl methylcellulose			
	HPMC, dextran)			
	Malted flour			
	Anti-oxidants			
	(alfa-tocopherol, rosemary extract, green tea powder, microencapsulated n-3			
	polyunsaturated fatty acids PUFA powder)			

Table 1. The main processing strategies for breadmaking with unrefined wheat flours (UWFs).

Literature reference	Wheat flour	Milling by-product supplement	Supplementation with good bread technological and sensory quality
Banu et al. (2012)	Refined flour	Bran streams (3%, 5%, 10%, 15%, 20%, 25%, 30% <mark>expressed on</mark> <mark>total flour weight</mark>)	25% bran stream
Blandino et al. (2013)	Refined flour	Pearled fractions (5%, 10%, 15%, 20%, 25% <mark>expressed on total</mark> <mark>flour weight</mark>)	10% pearled fraction
Bagdi et al. (2015)	Refined flour	Aleurone (40%, 75% <mark>expressed as percentages of refined flour</mark>)	40% (only on sensory profile)
Sun et al. (2015)	Refined flour	Wheat germ (0%, 3%, 6%, 9%, 12% <mark>expressed on total flour</mark> <mark>weight</mark>)	6% wheat germ
Pasqualone et al. (201 <mark>7</mark>)	Refined flour (re- milled semolina)	 3 fractions of durum wheat milling by-products (10%, 20% expressed as percentages of refined flour): i) bran obtained from non-debranned wheat ii) second and third debranning fractions mixed together iii) thin subfraction obtained by micronization and air classification of the second and third debranning fraction mix 	10% second and third debranning step

Table 2. The use of unprocessed unrefined wheat flours (UWFs) for breadmaking. Reporting literature reference, wheat flour, type of milling by-products used for supplementation and level of supplementation that gave good bread technological and sensory quality.

Literature reference	Treatment	Tested variables	Measurements	Main results
Ding et al. (2018)	Germination	Germination time	Hagberg falling number (FN)	Controlled germination (t=5-15 h, T=
			Rapid Visco Analyser (RVA)	28±2°C, RH=95±3%) improved wholewheat
			Starch pasting properties	flour functionality
			Mixolab mixing properties	
			Physicochemical analysis	
			γ-aminobutyric acid (GABA)	
			content	
<mark>Ritcher et al. (2014)</mark>	Germination	Germinated wholewheat flour +	Farinographic test	100% germinated wholewheat bread
		vital gluten (0%, 3%, 4%, 5%) for	Proof time	showed better technological and sensory
		breadmaking	Loaf volume	quality than control;
			Sensory analysis	Vital gluten did not improve bread quality
Zilic et al. (2016)	Germination	Germination effect on	Total and free sulfhydryl (-SH)	Total protein content did not change with
		wholewheat protein functionality	groups	germination
			<mark>Lipoxigenase (LOX) and</mark>	Intensive protein hydrolysis
			<mark>peroxidase (POX) activity</mark>	Increased antioxidant capacity of albumin
			SDS-PAGE gel electrophoresis	globulin fraction and
			Total antioxidant capacity of	reduced glutenin antigenicity
			albumin+globulin proteins	Potential health positive effects
			Gliadin and glutenin	
			immunogenicity	
			Analysis of pasting viscosty	
Johnston et al. (2019)	Germination	Germination effect on	Kernel hardness	Controlled germination (t=24 h, T=21°C,
		wholewheat flour functionality	Hagberg falling number (FN)	FN=200 s, excess of water) increased
		and flavour	Sodium dodecyl sulphate	wholewheat bread volume and flavour
			sedimentation analysis (SDS)	
			Starch content	
			Total dietary fibre	
			Alfa-amylase activity	
			Metabolite analysis	
			Mixograph analysis	
			Sensory analysis	

Table 3. References concerning the use of processed unrefined wheat flours (UWFs) produced through treatments on the raw material, i.e., wheatkernels. Outlining type of treatment, tested variables, measurements made and main results.

Literature reference	Milling by-product	Treatments	Best results on bread quality	Interpretation of results
Hemdane et al. (2016)	Wheat bran	Pre-soaking	14% pre-soaked bran	Reduction of bran water uptake during mixing
		Particle size reduction	22% pre-soaked, fine-ground	
			bran	
Hemdane et al. (2016)	Wheat shorts	Pre-soaking	Bread specific volume	Activation of endogenous lipoxygenase
Hemdane et al. (2016)	Wheat bran	Pre-soaking	Good bread quality	A complete understanding has not been
		(limited/excess water)		established yet. Some hypotheses proposed:
				-Saturating bran with water before mixing
				prevent the detrimental effects on
				dough/bread quality;
				-Activation of endogenous lipoxygenase
				oxidize components detrimental to bread
				quality
				-Washout effect when pre-soaking in excess of water
<mark>Messia et al. (2016)</mark>	Wheat bran	Pre-soaking	Improved dough rheology	Modification of arabynoxylan solubility
		Enzyme addition (xylanase,	and bread physical properties	allowing a better redistribution of water
		<mark>amylase, cellulase)</mark>		
Zhang et al. (2019)	Arabinoxylan flour from wheat bran	Pre-soaking	Up to 10% pre-soaked	The positive effects associated to the pre-
		Enzyme addition (xylanase)	arabinoxylan flour	<mark>soaking of arabinoxylan flour was not</mark>
				explained
Hemdane et al. (2016)	Wheat bran	Fermentation	20% fermented bran from	Solubilization of arabinoxylans
	(native bran and bran from peeled kernel)	(20 h, yeast starter)	peeled kernel	Reduction of endogenous xylanase activity
Hemdane et al. (2016)	Wheat bran	Fermentation	15% 160 um bran fermented	Lactic acid fermentation (Lactobacillus brevis)
		(8 h, lactic acid bacteria and	8 h	
		yeast strain)		
		Particle size reduction		
Boukid et al. (2018)	Wheat germ	Sourdough fermentation	Up to 20% sourdough	Reduction of enzymatic activities (lipase,
		(bacteria and yeast)	fermented wheatgerm	lipoxygenase)
				Reduction of glutathione content
Boukid et al. (2018)	Wheat germ	Sourdough fermentation	Better nutritional, chemical	Reduction of pH
		(Lactobacillus plantarum LB1,	and stabilization properties	Reduction of enzymatic activities (lipase,
		Lactobacillus rossiae LB5)		lipoxygenase) Higher total amino acids
				Higher protein digestibility Inactivation of anti-nutritional factors
Boukid et al. (2018)	Wheat corm	Sourdough formantation	4% sourdough fermented	Higher antioxidant activity
Doukid et al. (2018)	Wheat germ	Sourdough fermentation (Lactobacillus plantarum LB1,		Reduction of enzymatic activities (lipase,
		(Lactobacillus plantarum LB1, Lactobacillus rossiae LB5)	wheat germ	lipoxygenase) Reduction of glutathione content
Boukid et al. (2018)	Wheat corm	Sourdough fermentation	4% sourdough fermented	Antifungal activity of sourdough fermented
Doukiu et al. (2010)	Wheat germ	(Lactobacillus plantarum LB1,	4% sourdough termented wheat germ	wheat germ (phenolic acids, organic acids)
		Lactobacillus piantarum LB1,	bread shelf life	
		Laciobacilius rossiae LB5	Dread Shell life	Lower pH values

Table 4. References concerning the use of processed UWF produced through treatments on the raw material, i.e., milling by-products (wheat bran and wheat germ). Summarizing milling by-product, type of treatment, best results on bread quality and interpretation of the main results.

Literature reference	Process	Tested variables	Wheat flour	Improvement of bread quality
	strategy			
Cappelli et al. (2019)	Optimization	Water amount (70%, 76%, 82%, 88%, and 94%)	Refined, brown and	Optimal water addition as a function of
	of water	Flour refinement degree (refined, brown, wholewheat)	wholewheat flour	degree of flour refinement
	amount			
Guerrini et al. (2019)	Optimization	Different variables used by bakers	Brown and wholewheat	Higher water amount
	of water		flour	
	amount			
Parenti et al. (2019)	Modification	Water amount (59%, 70%, 80%)	Brown flour	6% pre-gelatinized flour + high water
	of the flour	Pre-gelatinized flour (0%, 6%)		amount
Hung et al. (2007)	Modification	Waxy wholewheat flour (<mark>0%,</mark> 10%, 30%, 50%)	Wholewheat flour and	Waxy wholewheat breads showed softer
	of the flour		waxy wholewheat flour	crumb during storage
Tebben et al. (2018)	Improver	Enzyme, xylanase	Wholewheat flour	Optimization of xylanase usage level
Tebben et al. (2018)	Improver	Enzyme, alfa-amylase	Wholewheat flour,	Optimization of amylase usage level
			blends of refined and	
			wholewheat flours	
Tebben et al. (2018)	Improver	Enzyme, G4-amylase	Wholewheat flour	Optimization of G4-amylase usage level
Tebben et al. (2018)	Improver	Hydrocolloids	Wholewheat flour	0.5%-1% hydrocolloids
		(carboxymethylcellulose, CMC; guar gum, GG; methylcellulose, MC; psyllium gum, PG; xanthan		
		gum, XG; tara gum, TG)		
Tebben et al. (2018)	Improver	Oxidants	Wholewheat flour	Optimum amount of antioxidants
		(potassium bromate, ascorbic acid, rosehip as a source of ascorbic acid)		corresponds to higher quantities than
				refined flour; best results with 200 ppm
Tebben et al. (2018)	Improver	Emulsifiers	Wholewheat flour	Usage level 0.4%-0.5%
		(diacetyl tartaric esters of monoglycerides, DATEM; sodium stearoyl lactylate, SSL; ethoxylated		Positive results emulsifiers combined
		monoglycerides, succinylated monoglycerides, lecithin, polyoxyethylene sorbitan		with oxidants
		monostearate, poly-oxyethylene sorbitan monopalmitate, glycerol-monostearate, mono- and		
		diglycerides)		
Tebben et al. (2018)	Improver	Vital gluten	Wholewheat flour	Usage level 2%-2.5%

Table 5. Literature references about the modification of unrefined wheat flours (UWFs) bread formula to improve breadmaking performance. Reporting the main process strategies (i.e., optimization of water amount, modification of flour and addition of improvers), tested variables, type of UWF and strategies that improved bread quality. Percentages relate to flour base (ingredient/total flour %)D.

Literature reference	Breadmaking step	Tested variable	Wheat flour	Processing strategy
Angioloni et al. (2006)	Mixing	Mixing time (10, 15, 20 min) Cysteine (20 mg/kg)	Refined and wholewheat flour (T. aestivum L.)	Combination of high-speed mixer and cysteine addition
Parenti et al. (2013)	Mixing	Mixing time (12, 17, 22, 27 min)	Brown flour (T. aestivum L.)	Optimized mixing time (17 min)
Guerrini et al. (2019)	Mixing	Different variables used by bakers	Brown and wholewheat flour (T. aestivum L., T. durum)	Short mixing time (10-20 min)
Cappelli et al. (2019)	Mixing	Time for addition of bran and middlings during mixing (0, 2, 3.5, 5, 6.5 min) Levels of bran and middlings (10%, 20%, 30%)	Refined flour enriched with bran and middlings (T. aestivum L.)	10% bran and middlings added at t=2 min
Kolmenic et al. (2010)	Leavening	Biological acidification (dry sourdough, <i>Lactobacillus brevis</i> preferment) Chemical acidification (lactic acid)	Refined flour Wholewheat flour (commercial blends)	Biological acidification (dry form)
Taccari et al. (2016)	Leavening	Back-slopping technique for type I sourdough	Wholewheat flour (T. <i>aestivum</i> L.)	Application of back-slopping techniques for sourdough fermentation
Choi et al. (2005)	Leavening	Selected LAB isolated from kimchi as starter cultures (Leuconostoc citreum HO12 and Weissella koreensis HO20)	Wholewheat flour (T. <i>aestivum</i> L.)	Applicability of selected LAB (Leuconostoc citreum HO12 and Weissella koreensis HO20)
Didar et al. (2011)	Leavening	Sourdough fermentation with <i>Lactobacillus plantarum</i> (PTCC 1058) and <i>Lactobacillus reuteri</i> (PTCC 1655) Level of sourdough addition (10%, 20%, 30%) Dough yield (250 and 300)	Flour with 95% extraction rate (cv Alvand wheat)	30% <i>Lb. plantarum</i> sourdough with DY 250
Katina et al. (2006 <mark>a</mark>)	Leavening	Sourdough time = 6-20 h Sourdough temperature = 16-32°C LAB and yeast for sourdough fermentation (<i>Lactobacillus</i> <i>plantarum</i> , <i>Lactobacillus brevis</i> , <i>Saccharomyces cerevisiae</i> or a combination of yeast and LAB) Flour ash content (0.6-1.8 g/100 g)	Flours with different ash content (0.6-1.8 g/100 g) (commercial flours)	Different flour ash contents required different optimization strategies

Table 6. Literature references about processing strategies for breadmaking with unrefined wheat flour (UWF). Outlining breadmaking step, tested variable, types of UWF used and process strategy.