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# The use of modern fermentation techniques in the production of traditional wheat bread

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**Abstract:** A traditional Czech bakery white bread product is commonly made with directly mixed dough with the addition of yeast. In order to be able to reduce the salt content of the final product without compromising its acceptability to consumers, the dough was prepared with the addition of pre-ferments and wheat sourdoughs. The aim was to deepen the flavour background of the product and thus have a positive effect on its sensory properties. Wheat sourdoughs were prepared and tested using starter cultures and matured under different conditions. The resulting products were tested both sensorially and by means of precise physical methods.

**Keywords:** sourdough; yeast; lactic acid bacteria; sensory properties; volatile substances

The dough for wheat bread can basically be made in three ways. Directly, i.e. in one-step, flour is mixed with all the recipe ingredients, bakery yeast and water and the dough is kneaded to the optimum consistency. Fermentation then takes place in the maturation and dough rising stages. Another variant is the indirect method of preparation, where fermentation takes place in the first stage in a thin suspension of just flour in water with a yield of 200–300%. The yield being the ratio of the total weight of the mixture to the weight of flour expressed as a percentage. Only then is the actual dough made up by adding flour and other ingredients, usually with a yield of 165–175%, depending on the type of bread (Van Kerrebroeck et al. 2017).

Depending on the type of fermentation and the method by which it is initiated, we distinguish between a pre-ferment, in which ethanol fermentation initiated by yeast takes place, or a sourdough. In the sourdough lactic acid fermentation takes place, initiated by a starter culture of lactic acid bacteria (LAB), or spontaneously by the natural microflora contained in the flour, which includes both LAB and natural yeast cultures, or both can occur simultaneously (Chavan and Chavan 2011; Kavitake et al. 2018). Pre-ferments are usually aged for one to three hours, while sourdoughs are aged for ten hours or more. In a pre-ferment, only ethanol fermentation takes place, while lactic acid fermentation is present or completely dominates in a sourdough. Thus a sourdough ultimate-

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ly contains lactic acid fermentation products, i.e. lactic, acetic, and other acids, and other products, and is therefore much more varied in its composition (Sadeghi 2008; Banu et al. 2011; Walton 2011; Weckx et al. 2018).

A pre-ferment provides a more homogeneous, better hydrated dough compared to direct dough guidance, which is reflected in the quality of the bread. However, sourdough brings many other properties to the system. Thus, the use of sourdough has an impact on both sensory and physicochemical properties as well as on the nutritional value of bread. Sourdough can have a positive effect on stability, as the main lactic acid products (lactic and acetic acids have proven antifungal activity) and the lower pH contribute to the higher stability of the bread (Sadeghi 2008; Poutanen et al. 2009).

The sensory properties of the sourdough were essential for our work (Pétel et al. 2017). Indeed, the salt content of bread and baked goods is a major concern in terms of health impacts on the population. In all European countries, salt consumption exceeds the desirable intake, often severalfold, and bread and pastries account for 30–40% of this intake, despite there being a long known causal link between high $Na^+$ ion intake and hypertension, one of the main causes of cardiovascular disease.

A certain salt content in the dough is essential for the formation of the correct consistency of the dough and for the fermentation process. This is approximately 0.5% (*w*/*w*) salt by weight of flour. In reality, however, the salt dosage is in the range of 1.5–2% salt by weight of flour, sometimes more. Higher doses than is technologically necessary are the result of a longstanding habit of a certain intensity of salty taste in bread. From a health point of view, any reduction in the salt content, even by a few tenths of one per cent, is desirable. However, it has an impact on the intensity of the salty taste of bread that consumers are accustomed to (Quilez and Salas-Salvado 2012; Sluková and Skřivan 2019; Dunteman et al. 2021).

The use of sourdough has a significant impact on both the taste and aroma of bread in terms of sensory properties (Pétel et al. 2017), and our aim was to investigate whether the use of sourdough would enrich the flavour sufficiently to make the bread attractive to consumers even after reducing the salt content.

## MATERIAL AND METHODS

### Laboratory preparation of experimental wheat sourdoughs

Experimental laboratory-prepared sourdoughs were prepared using spontaneous fermentation, without a starter culture.

The flours used were smooth light (white) wheat flour (T530), smooth bread wheat flour (T1000), finely ground wholemeal wheat flour (granulation up to 160 μm).

The sourdoughs were prepared as a suspension of flour in water in beakers and homogenized using a stick blender. For the smooth light (white) and bread flours, the proportions were 500 g of flour and 700 mL of water; for the wholemeal flour, which has a higher water absorption, the proportions were 500 g of flour and 900 g of water. After homogenisation of the suspension, the sourdough was ripened at 25 °C for up to 48 h.

### Preparation of experimental sourdoughs in the fermenter

A Diosna & IsernHäger AF Compact 100 HC fermenter (Germany) was used in the operational laboratory. Diosna & IsernHäger starter culture (StartGut®-Bio Bologna, Germany) was used for the preparation of experimental sourdoughs with starter culture. Its prescribed dosage is 0.5 g per 100 g of flour.

The sourdoughs were prepared spontaneously and with the starter culture as in the laboratory experiments. The sourdoughs were prepared with a yield of 220% for spontaneous fermentation and 230% with the starter culture. The yield is the ratio of the total weight of the sourdough at the start of fermentation to the weight of the flour used, expressed as a percentage. Yields are calculated on the basis of the sum of the weight of flour and water, the weight of the starter culture being neglected in the calculation. The sourdoughs were matured in the fermenter under controlled conditions at 30 °C for up to 20 h.

### Preparation of sourdoughs and pre-ferments under the operating conditions of the industrial bakery

A Diosna & IsernHäger AF Compact 100 HC low-volume fermenter was also used under these operating conditions, due to the required volumes of sourdough and fermentation stages.

On the basis of the results of laboratory and semi-operational experiments, industrial fermentation procedures were established using several types of starter cultures with different effects on the sensory properties (aroma and taste) of the sourdoughs and bakery products. The starter cultures were DIOSTart wheat classic (referred to as 'basic'), DIOSTart wheat fruit (referred to as 'fruit') and DIOSTart wheat strong (referred to as 'strong') produced by Diosna & IsernHäger (Germany).

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The starter cultures were used to prepare sourdoughs with a yield of 240% for the white wheat flour and 250% for the wholemeal flour, the starter culture was dosed at a ratio of 0.5% by weight of flour, and its weight was neglected in the calculation of the yield. The raw materials were stirred for 25 min before the actual fermentation, and the sourdough was matured in a single-stage mode at 30 °C for 20 h.

The preparation of the pre-ferments was carried out in the manner normally used in a bakery. For the white wheat flour, the pre-ferments were prepared at a yield of 200%, while for the wholemeal flour, the yield was 230%. The addition of liquid bakery yeast was 0.8% in both cases. Pre-ferments were matured at 25 °C for 3 h.

**Production of experimental bakery products**

Operational trials were carried out on a bakery production line to produce several types of reformulated wheat bread. The reformulation consisted of reducing the salt content to 1.2% by weight of flour, which meant reducing the salt content of the product to below 1%. The flavour of bakery products was modified by the addition of sourdough in a ratio of 8.5% by weight of flour. The amount of sourdough added was determined by several baking tests with additions of 5 to 10% sourdough, and the value of 8.5% was chosen as the optimum value in terms of the basic physical properties of the bread (shape, volume, uniform crumb porosity).

The addition of wheat sourdough (while reducing the recipe's salt content and hence the intensity of the salty taste of the product) was expected to improve the flavour profile (taste characteristics) of the reformulated dough by influencing the sensory components of the sourdough, and to have a partial effect on the physical properties of the crumb and the product as a whole.

The control product (standard, unreformulated) was a commonly produced high-quality Czech *veka* type wheat bread, which was produced with the aid of a pre-ferment (the use of a pre-ferment results in better physical and sensory properties of the product compared to direct preparation of the dough), which was added in the proportion of 20% by weight of flour. The amount of salt added in the standard recipe was 1.5% by weight of flour.

***Recipes for experimental bread.*** The recipes in both cases contained 4% oil and 1.5% sugar by weight of flour. The total dough yield (the sum of the weight of the flour of all the recipe ingredients and the water, i.e. the total weight of the dough relative to the weight of all the flour used) was 165–170%, depending on the water content. The weight loss through baking was 14–16%.

Both the reformulated loaves with reduced salt content and the addition of sourdough and the unreformulated bread with the standard recipe with a pre-ferment were also produced in a modified version (based on sourdoughs and pre-ferments based on wholemeal fine wheat flour).

**Evaluation of sourdoughs and pre-ferments**

All the prepared sourdoughs and pre-ferments were evaluated organoleptically. Aroma and taste, homogeneity and consistency were assessed.

Total titratable acidity (TTA) and pH (measured by digital pH-meter) were determined at several different time points (fermentation phases) for the experimental sourdoughs prepared both in the laboratory in beakers and in the experimental fermenter. TTA value was determined by The American Association of Cereal Chemists (AACC) Method 02-31.01.

Sensory volatiles were analysed by headspace solid-phase microextraction coupled to gas chromatography-mass spectrometry HS-SPME-GC-MS for sourdoughs produced under industrial operating conditions.

**Determination of volatile substances in fermented and final products by SPME-GC-MS**

Volatile compounds were determined using SPME-GC-MS analysis, specifically a Supelco 50/30 µm 24 Ga DVB/CAR/PDMS SPME fibre (Merck, Germany), GC-Autosampler Combi Pal system (CTC Analytics, Switzerland), 7890 A gas chromatograph with a VF-WAX MS 30 m × 0.25 mm capillary column, film thickness 0.25 µm and 5975 C mass detector (Agilent, USA). A 0.2-gram sample was placed into a 10 mL SPME vial, and 0.8 mL of distilled water and an internal standard (10 µL of aqueous 2-methyl-3-heptanone solution, 80 mg·L$^{-1}$) were added.

Isolation conditions were according to the methodology described by Xu et al. (2020): pre-incubation for 20 min, extraction for 30 min at 60 °C and stirring speed 250 rpm, in the GC inlet, analytes were desorbed at 250 °C for 7 min. The chromatographic conditions were as follows: the temperature gradient started at 40 °C (held for 3.5 min), followed by a temperature increase at a rate of 5 °C·min$^{-1}$ to 90 °C (5 min) and a further temperature increase at a rate of 12 °C·min$^{-1}$ to 250 °C (10 min). Helium grade 4.8 with a flow rate of 0.8 mg·min$^{-1}$ was used as the carrier gas. Electron ionization with an ionization energy of 70 eV, ion source

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temperature of 230 °C and a quadrupole analyser temperature of 290 °C, and a measuring mass range *m/z* of 50–550 was used for MS.

Specific volatile compounds were identified by spectrum comparison with the National Institute of Standards and Technology (NIST) 14 mass spectrum library and using retention indices. Peak areas were used for the quantification.

**Evaluation of final products**

The experimentally produced reformulated bread samples and comparative standard breads were subjected to sensory evaluation, crumb stiffness evaluation using a PNR 10 penetrometer (Petrotest Instruments, Germany). The stiffness of the crumb was also determined using the penetrometer on selected samples during five days of storage to determine the rate of ageing. Storage experiments were carried out under defined conditions (laboratory conditions, in original PE packaging, temperature 25 °C).

The produced breads were sensorially evaluated by a panel of trained evaluators from the industrial bakery and also by a panel of evaluators from the university. The mechanical properties of the crumb, the aroma and taste of the crumb, its softness and sensory properties, and the overall impression of the product were evaluated. The evaluation was carried out according to a set of criteria scored from 5 to 1 (5 best, 1 worst).

The selected samples were analysed for significant sensory volatile substances using HS-SPME-GC-MS.

Sodium content in breads was measured using an ion-selective electrode. Salt content was calculated by multiplying the determined sodium content by a factor of 2.5.

Microbiological evaluation of the stability of the selected samples was also carried out. The total number of microorganisms, the number of yeasts and moulds was determined on the first day after baking and three days after the end of the declared shelf-life in the packaging (5 days), i.e. after eight days of storage in the packaging. The shelf-life of breads was assessed on the basis of storage experiments under defined conditions (laboratory conditions, in the original PE packaging, temperature 25 °C).

Microbiological analyses were carried out according to the methodology of EN ISO 4833 (Determination of the total number of microorganisms by culture) and ISO 21527-1, 2 (Determination of the number of yeasts and moulds by culture).

**Statistical analysis**

Analytical determination, rheological and textural measurement, and microbial evaluation were performed in duplicate. The mean value and standard deviation (SD) were given. Data were assessed using one-way analysis of variance (ANOVA) at $P \leq 0.05$ (STATISTICA software version 12.0, StatSoft, Inc., USA).

## RESULTS AND DISCUSSION

**Evaluation of experimental sourdoughs prepared under laboratory conditions**

These sourdoughs were mainly used to verify the fermentation capabilities of the individual flours. Their preparation consisted of homogenising the suspension in a beaker, and fermentation was carried out at a lower temperature than is usually the case for standard fermentor preparation, and also without continuous stirring. The conditions for fermentation were thus more complicated and the fermentation took longer. Experiments have shown that not all the flours tested are capable of fermenting spontaneously on the basis of the natural microflora. This is clearly mainly possible with wholemeal flour, and to some extent with bread flour. The light (white) flour (T530) comes from the inner regions of the endosperm and therefore does not contain enough natural microflora, which is why the TTA values remain low, even with longer fermentation times (see Table 1).

Table 1. Parameters of spontaneously prepared sourdoughs

| Type of flour | Yield (%) | Fermentation time (h) | pH | TTA (mmol·kg$^{-1}$) |
|---|---|---|---|---|
| Wholemeal | 280 | 24 | 4.5 | 100 ± 2 |
| | | 48 | 4.1 | 139 ± 4 |
| T530 | 240 | 24 | 4.3 | 46 ± 0 |
| | | 48 | 4.0 | 62 ± 1 |
| T1000 | 240 | 24 | 4.2 | 82 ± 2 |
| | | 48 | 4.0 | 108 ± 2 |

TTA – total titratable acidity

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**Evaluation of experimental sourdoughs prepared in the fermenter**

The fermentations were made under semi-operating conditions. During the fermenter fermentation, the process was maintained at a constant temperature in a stable environment. The fermenting sourdough was automatically stirred at regular intervals.

***Spontaneous fermentation.*** These sourdoughs were prepared using only wholemeal flour and bread flour (T1000) for which previous experiments had shown the ability to ferment spontaneously. The pH and, in particular, the TTA values are shown in the tables below (Tables 2 and 3).

The sourdough yield (220% in both cases) is a summary quantity, but at the same time it is related to the consistency of the suspension that forms the yeast. The higher the yield, the higher the water/flour weight ratio. In addition to the water/flour ratio, the consistency is determined by the water content. With sourdoughs, a lower yield, i.e. a denser consistency, tends to favour the development of LAB. Sourdough based on wholemeal flour has a clearly faster fermentation rate, which may be due both to the higher concentration of natural microflora and to the denser consistency caused by the higher water binding capacity of wholemeal flour. After 16 h, the sourdough from bread flour had optimal organoleptic properties – smoothness, homogeneity, foaminess, pleasantly sour taste, whereas the sourdough from wholemeal flour was already more acidic after 14 h.

***Sourdoughs prepared with* a *starter culture.*** The starter culture consists of LAB and yeast (a mixture of strains of bacteria of the genus *Lactobacillus* and yeast of the genus *Saccharomyces*). The activity of LAB predominates. In this case, the yield was chosen to be slightly higher than with spontaneous culture (230%), i.e. sourdoughs with a slightly looser consistency. When using a starter culture, experimental sourdoughs were prepared based on bread and wholemeal flour as well as white flour (T530). A uniform fermentation time of 16 h was chosen (Table 4).

It is clear that despite the assumed predominance of microorganisms supplied by the starter culture, the natural microflora also play a role. Sourdough from light (white) flour, which contains only a minimal concentration of natural microflora, ferments significantly more slowly than bread and wholemeal flour (where the denser consistency may also play a role).

The sourdough initiated with starter cultures always had a pleasant aroma. The sourdoughs were frothier, and the volume of the sourdough was noticeably higher than the volume of the sourdough without starter culture. This is due to the presence of yeast and therefore a higher proportion of ethanol fermentation in the overall fermentation. The sensory properties of all the sourdoughs prepared using the starter culture were more favourable than those of the spontaneous sourdoughs, due to fermentation controlled by purposely selected populations of microorganisms. The acidity

Table 2. Parameters of spontaneously prepared sourdoughs from bread flour (T1000)

| Fermentation time (h) | pH | Temperature during measurement pH (°C) | TTA (mmol·kg$^{-1}$) |
|---|---|---|---|
| 14 | 3.9 | 23.2 | 106 ± 3 |
| 15 | 3.9 | 23.4 | 108 ± 2 |
| 16 | 3.9 | 22.6 | 114 ± 2 |
| 17 | 3.9 | 24.7 | 120 ± 3 |
| 18 | 3.8 | 23.7 | 122 ± 4 |
| 19 | 3.8 | 24.3 | 126 ± 3 |

TTA – total titratable acidity

Table 3. Parameters of spontaneously prepared sourdoughs from wholemeal flour

| Fermentation time (h) | pH | Temperature during measurement pH (°C) | TTA (mmol·kg$^{-1}$) |
|---|---|---|---|
| 14 | 4.2 | 23.0 | 153 ± 3 |
| 15 | 4.1 | 22.6 | 161 ± 2 |
| 16 | 4.1 | 24.3 | 164 ± 2 |
| 17 | 4.1 | 22.8 | 167 ± 1 |
| 18 | 4.1 | 24.1 | 170 ± 2 |
| 19 | 4.0 | 23.1 | 175 ± 2 |

TTA – total titratable acidity

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Table 4. Parameters of sourdoughs prepared with a starter culture

| Type of flour | Fermentation time (h) | pH | Temperature during measurement pH (°C) | TTA (mmol·kg$^{-1}$) |
|---|---|---|---|---|
| Wholemeal | 16 | 4.0 | 22.7 | 190 ± 2 |
| T530 | 16 | 3.7 | 21.6 | 117 ± 0 |
| T1000 | 16 | 3.7 | 22.0 | 153 ± 2 |

TTA – total titratable acidity

of the samples was pleasant, and no aftertaste was detected. The starter culture sourdoughs exhibited lower pH and higher TTA than spontaneous fermentations and started fermentation earlier. From the point of view of industrial production, starter culture sourdoughs are preferable to spontaneous fermentations. They are more stable in terms of sensory and technological properties. Therefore, starter culture sourdoughs were mostly used for the production of experimental bakery products (Chavan and Chavan 2011; Kavitake et al. 2018).

**Evaluation of the preparation and use of sourdoughs and pre-ferments under the operating conditions of an industrial bakery**

Based on the results of 14 technological tests, the optimum yield of wheat sourdough (addition of water to the sourdough made from T530 wheat flour and using 3 different starter cultures – DIOSTart wheat classic, DIOSTart wheat fruit and DIOSTart wheat strong) was determined to be 240%. Furthermore, the optimum addition of wheat pre-ferment (addition of about 8.5% wheat sourdough per flour, compared to 20% wheat pre-ferment in the production of unreformulated breads) to the reformulated *veka* recipe was verified with minimal impact on product quality (appearance, shape, volume, texture, aroma, and taste). The consistency of the dough with the addition of the sourdough was satisfactory, the dough was slightly sticky (compared with the dough for the production of unreformulated breads), but technologically satisfactory and easy to process on a conventional line.

The production of wheat sourdough, the mixing of doughs with sourdough and the further processing of doughs of reformulated *veka* loaves (maturation, cutting, shaping, proofing, and baking) were similar to the production of unreformulated *veka* loaves. The aroma and flavour of the prepared wheat sourdough depended on the type of starter culture used, with only slight variations in the time taken for the dough to rise. The resulting doughs and finished products were comparable to the quality of the unreformulated products.

From the production point of view, and on the basis of the experience of the operational technologists of industrial bakery, the wheat sourdough prepared under optimum conditions using the DIOSTart wheat fruit starter culture was the best from the technological point of view. It was also experimentally confirmed that a high-quality and technologically suitable pre-ferment and wheat sourdough can also be produced from special finely ground wholemeal flour.

The main objective was to reduce the recipe salt addition to 1.2%, which corresponds to a reduction of the salt content of the finished product to 0.9% (with a dough yield of 168% and a baking loss of 15%). This reduction can be considered a significant reduction. The basic requirement was to achieve, through the use of sourdough, sufficiently pronounced sensory characteristics in the products to compensate for the reduction in the intensity of the salty taste for the consumer.

**Evaluation of sensory analysis results**

The produced *veka* loaves were evaluated by a panel of trained evaluators from the industrial bakery and also by a panel of evaluators from the university.

Although the unreformulated bread had the most elastic crumb, the breads with wheat sourdough were softer and fluffier, while the wholemeal sourdough breads were slightly stiffer with a slight bitter aftertaste. As for the flavour and aroma profile, the wheat sourdough bread fermented with DIOSTart wheat fruit was judged to be the most palatable. A milder intensity of salty flavour was also evident in this loaf.

The whole wheat sourdough was experimentally developed with the starter culture DIOSTart wheat strong. The sourdough yield of this whole wheat flour was 10% higher than the yield of the wheat sourdough from T530 white wheat flour.

The model batch of breads with the addition of wholemeal wheat flour sourdough and reduced salt content was judged to be less satisfactory. The wheat sourdough bread was less domed to slightly flat and had a lower volume, but a more supple crumb compared to the standard unreformulated bread, which had a more balanced appearance, higher volume, and a saltier taste.

Therefore, from a sensory point of view, the loaves with DIOSTart wheat fruit sourdough produced using

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Table 5. Results of sensory evaluation of fresh breads with the DIOSTart wheat fruit starter culture

| Evaluated parameter | Points – average | | |
|---|---|---|---|
| | standard bread | reformulated bread with sourdough | reformulated bread with wholemeal sourdough |
| Mechanical properties of the crumb | 4.8 | 4.3 | 3.5 |
| Smell of crumb | 4.2 | 4.7 | 4.4 |
| Taste of crumb | 4.1 | 4.4 | 3.9 |
| Crumb smoothness and tactile properties | 4.8 | 4.7 | 3.2 |
| General impression | 4.6 | 4.6 | 3.8 |

only white wheat flour (T530) were selected as the optimal reformulated variant with reduced salt content.

The results of the evaluation of the sourdough breads from the experimental batches under operating conditions are presented in the following table (Table 5). Final values (points) are the arithmetic averages of the scores recorded by the individual evaluators from the two panels.

It is significant that it has been verified at this stage that the reduction in the salt content of the reformulated breads from 1.5% to 1.2% per flour, and thus the reduction in the intensity of the salty flavour when using sourdough, was sensorially assessed as generally imperceptible or not significant.

**Determination of flavour profiles by SPME-GC-MS**

Aromatic profiles of the prepared wheat pre-ferment, sourdoughs and breadcrumbs were determined using the SPME-GC-MS technique. The following sensory active substances with specific sensory perceptions (Pétel et al. 2017) were detected in high quantities in all three sourdoughs: ethyl heptanoate (sensory perception: fruity, pineapple, cognac), 1-butanol-3-methyl acetate (sweet, fruity, banana) and ethyl lactate (butter, caramel, fruit pie). The sourdough samples analysed had richer volatile profiles than the crumb samples. The wheat pre-ferment had the poorest volatiles profile.

**Determination of change in crumb stiffness during storage and microbial evaluation of breads**

The results of the assessment of the stiffness of the bread crumb on a PNR 10 penetrometer over a selected time period of 5 days are shown in Figure 1. Bread with wheat pre-ferment was chosen as the standard bread and sourdough bread prepared by the DIOSTart wheat fruit starter culture was labelled as reformulated.

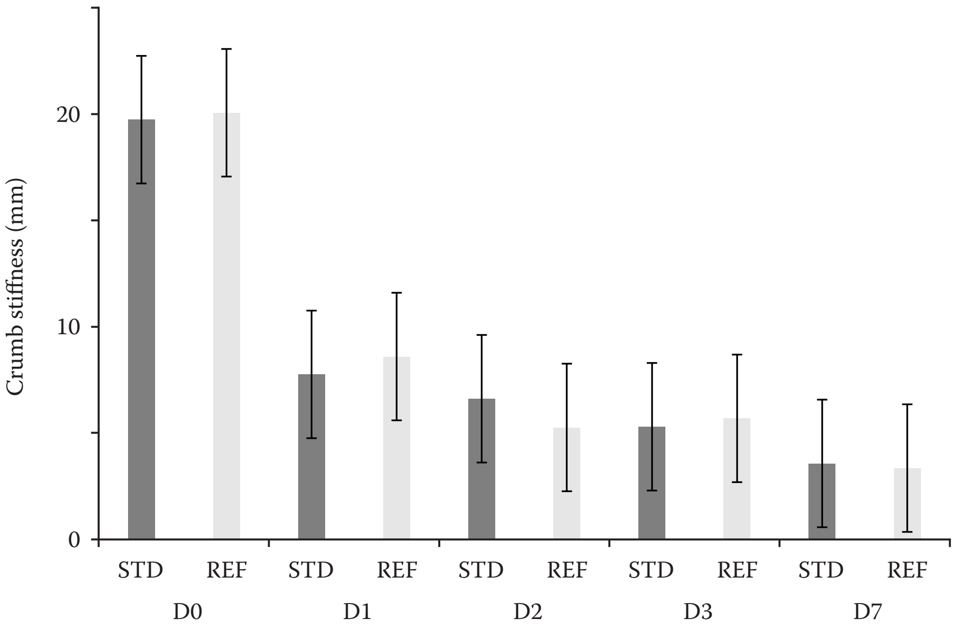


Figure 1. Penetrometric evaluation of bread stiffness (values and error bars)

STD – bread with wheat pre-ferment, standard, unreformulated; REF – sourdough bread prepared with the fruit starter, reformulated; D – day of storage

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Table 6. Determination of the number of microorganisms in breads (evaluation on the day after bread production)

| Sample | Total number of microorganisms* | Moulds* | Yeasts* |
|---|---|---|---|
| standard bread | $4.0 \times 10^1$ | < 10 | < 10 |
| reformulated bread | $4.2 \times 10^1$ | < 10 | < 10 |

* number of microorganisms/moulds/yeasts expressed in 1 g sample (CFU·g$^{-1}$); CFU – colony forming unit; standard bread – bread with wheat pre-ferment, unreformulated; reformulated bread – sourdough bread prepared with the fruit starter culture

Table 7. Determination of the number of microorganisms in stored breads after eight days*

| Sample | Total number of microorganisms** | Moulds** | Yeasts** |
|---|---|---|---|
| standard bread | $5.4 \times 10^3$ | $1.1 \times 10^4$ | < 10 |
| reformulated bread | $4.9 \times 10^3$ | $5.5 \times 10^5$ | < 10 |

* three days after the expiry date; ** number of microorganisms expressed in 1 g sample (CFU·g$^{-1}$); CFU – colony forming unit; standard bread – bread with wheat pre-ferment, unreformulated; reformulated bread – sourdough bread prepared with the fruit starter culture

It is clear from the results that the aging process of standard and reformulated breads is quite comparable in terms of the change in crumb stiffness.

Furthermore, a series of storage experiments were carried out to investigate the tendency of microorganisms to grow. It was found that the reformulated breads were subject to microbial deterioration in the same way as the unreformulated (standard) breads. The reformulated breads therefore did not demonstrate a stabilising effect of the addition of wheat sourdough, which is probably due to the relatively low addition of wheat sourdough, which, although beneficial in terms of sensory properties, is not sufficient in terms of lactic and acetic acid (or other acids) concentration.

The following tables (Tables 6 and 7) give examples of the parameters determined in relation to the microbiological changes in the analysed breads. The total number of microorganisms was determined one day after manufacture and at the end of the shelf-life indicated on the product packaging.

## CONCLUSION

With respect to the technology of sourdough production for use in the industrial production of wheat bread, it seems to be more advantageous to prepare sourdough with the use of LAB starter cultures. This is particularly true for sourdoughs from common (white) flours, where the content of natural microflora is very low. On the basis of sensory tests on experimental samples of wheat bread, it has been shown that the sourdough does indeed deepen the flavour of the bread to such an extent that a reduction of 0.3% in salt content by weight of the flour used does not have an undesirable effect on the consumer. The salt content of the sourdough bread was reduced to 0.9%, which is considered highly desirable from a health point of view (Quilez and Salas-Salvado 2012).

In our case, we used a relatively low sourdough addition (8.5% *w*/*w* per flour), as a higher acidity could have an undesirable effect in the chosen type of bread. Therefore, the addition did not significantly affect the microbiological stability of the bread, nor did it slow down the aging process, which potentially higher sourdough additions should have a positive effect on. However, it is important to note that the addition of sourdough, together with the reduction in salt content, appears to be sensorially and technologically quite comparable to the control bread samples prepared with a higher (20%) addition of pre-ferment, which in itself significantly improves the quality of the bread.

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