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Highlights

- Sprouting coupled with fermentation led to dextran production
- Sprouting coupled with fermentation improved free phenolic content and antioxidant activity
- Dextran presence markedly enhanced dough rheology and bread quality parameters
- Total and soluble fibers were significantly increased by the dextran produced
- Bioprocessing improved the aroma profile, estimated by GC/MS, of the bread

1	Sourdough fermentation of whole and sprouted lentil flours: <i>in situ</i> formation of dextran and
2	effects on the nutritional, texture and sensory characteristics of white bread
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24 Abbreviations

CWB, control wheat bread; DPPH, 2-2-diphenyl-1-picrylhydrazyl radical; d.w., dough weight; DY, 25 dough yield; EPS, exopolysaccharides; FQ, fermentation quotient; f.w., flour weight; GI, glycemic 26 index; HI, starch hydrolysis index; IDF, insoluble dietary fibers; LAB, lactic acid bacteria; L, lentil; 27 L-SLA4, lentil sourdough fermented with Weissella confusa SLA4; LSWB, lentil sourdough wheat 28 bread; ME, methanolic extract; Mw, molecular weight; MRS, De Man Rogosa and Sharpe medium; 29 OPA, o-phthaldialdehyde; PCA, Principal Component Analysis; RS, resistant starch; SDF, soluble 30 dietary fibers; SL, sprouted lentil; SL-SLA4, sprouted lentil sourdough fermented with W. confusa 31 SLA4; SLSWB; sprouted lentil sourdough wheat bread; TPA, texture profile analysis; TDF, total 32 dietary fibers; TFAA, total free amino acids; TTA, total titratable acidity; VOCs, volatile organic 33 compounds. 34

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37 Abstract

Exopolysaccharides produced in situ by lactic acid bacteria during sourdough fermentation are rec-38 ognized as bread texture improvers. In this study, the suitability of whole and sprouted lentil flours, 39 added with 25% on flour weight sucrose for dextran formation by selected strains during sourdough 40 fermentation, was evaluated. The dextran synthesized in situ by Weissella confusa SLA4 was 9.2 and 41 9.7% w/w flour weight in lentil and sprouted lentil sourdoughs, respectively. Wheat bread supple-42 mented with 30% w/w sourdough showed increased specific volume and decreased crumb hardness 43 and staling rate, compared to the control wheat bread. Incorporation of sourdoughs improved the 44 nutritional value of wheat bread, leading to increased total and soluble fibers content, and the aroma 45 profile. The integrated biotechnological approach, based on sourdough fermentation and germination, 46 is a potential clean-label strategy to obtain high-fibers content foods with tailored texture, and it can 47 further enhance the use of legumes in novel foods. 48

49 Keywords

- 50 Dextran; Fermentation; Fibers, Germination; Lentil; Prebiotic, Sourdough; Wheat bread
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According to the World Health Organization, legumes are essential components of the daily 54 diet, providing an inexpensive, sustainable source of proteins and other key nutrients (World Health 55 Organization, 2019). Lentils (Lens culinaris) have a relatively higher protein, carbohydrate, and 56 energy content than other legumes (Lee, Htoon, Uthayakumaran, & Paterson, 2007). Lentils are a 57 good source of lysine and arginine, important in balancing the deficiency of these essential amino 58 acids in cereal-based diets (Asif, Rooney, Ali, & Riaz, 2013). They are also rich in dietary fibers 59 (DF), minerals and are cholesterol-free, contain low in saturated fat, and very low sugar (Melini, 60 Melini, Luziatelli, & Ruzzi, 2017). Consumption of lentils has been linked to the reduction of several 61 health risks such cardiovascular diseases and cancer prevention (Asif et al., 2013). Lentils, as all dried 62 legume seeds, due to high proportion of non-digestible carbohydrates, generally promote slow and 63 moderate postprandial blood glucose increase (García Alonso, Goñi, & Saura-Calixto, 1998) which 64 is of nutritional benefit to lentil consumption. Despite of these potential benefits, the world 65 consumption of legumes is below the recommended amount (defined as 50-70 g daily) (McCrory, 66 Hamaker, Lovejoy, & Eichelsdoerfer, 2010) although national recommendations vary considerably 67 across the Region as do the definitions of portions and servings. One of the most suitable option to 68 increase the consumption of legumes in the daily diet is to use their flours in the formulas for making 69 baked goods and to improve their sensory and functional features through sourdough fermentation 70 (Gobbetti, De Angelis, Di Cagno, Calasso, Archetti, & Rizzello, 2019). Bread is a staple food 71 consumed in variable forms throughout the world. Fortifying a widely consumed food product like 72 bread can exert a positive significant impact in term of plant-protein consumption that, in turn, could 73 contribute to the reduction of water and carbon-foot print associated with the animal-protein 74 consumption (meat/dairy) (Boukid, Zannini, Carini, & Vittadini, 2019). Several nutritional and 75 functional advantages have been associated to sourdough biotechnology applied to legumes. 76 Compared to legume-based doughs without bacterial inoculum, the concentration of free amino acids, 77

78 soluble fibers and total phenols and the antioxidant activities increased. On the other hand, almost all fermented legumes decreased raffinose, phytase and condensed tannins content (Curiel, Coda, 79 Centomani, Summo, Gobbetti, & Rizzello, 2015), these latter considered as anti-nutritional factors 80 (ANF). The presence of ANF in lentil is indeed considered one of the major limiting factor for their 81 dietary exploitation (Hall, Hillen, & Garden Robinson, 2017). Germination (also known as sprouting) 82 is a green food engineering method, occurring at the beginning of the development of seeds into 83 plants, during which they sprout (Rumiyati, James, & Jayasena, 2012). This process involves changes 84 in the nutritional, biochemical, and sensory characteristics which may improve the quality of legumes. 85 It is used in processing of legumes to improve nutritional quality as it results in improving digestibility 86 (Oghbaei & Prakash, 2016), reduction of ANF (Laxmi, Chaturvedi, & Richa, 2015; Oghbaei & 87 Prakash, 2016), improving fibre content (Rumiyati et al., 2012) and to mitigate beany or bitter flavors 88 89 (Xu, Jin, Simsek, Hall, Rao, & Chen, 2019). In addition to being used for nutritional enrichment or modification of products, germinated or malted grains and legumes may have technological 90 functionalities in food systems due to their increased enzyme activities (Mäkinen and Arendt, 2015). 91 Despite the nutritional advantages (Kohajdová, Karovicova, & Magala, 2013), the fortification of 92 wheat bread with lentils were reported to cause detrimental effects on texture and flavor, due to the 93 presence of fiber and non-gluten proteins (Gobbetti et al., 2019). Besides the absence of gluten 94 proteins the type found in wheat, pulses proteins can compete for water absorption with cereal 95 proteins, thus negatively affecting loaf volume and crumb firmness (Portman et al., 2018; Bresciani 96 & Marti, 2019). One possible means to counteract these negative effects is the use of natural 97 hydrocolloids such as exopolysaccharides (EPS) (Wang, Sorvali, Laitila, Maina, Coda, & Katina, 98 2018; Lynch, Zannini, Coffey, & Arendt, 2018). EPSs produced by different lactic acid bacteria 99 (LAB) (e.g. Weissella, Leuconostoc, Streptococcus, Pediococcus and Lactobacillus genera), and in 100 particular homopolysaccharides (HoPS), are long-chain, high Mw polysaccharides, which can 101 counteract for the absence of wheat gluten and water absorption by lentil proteins weakening the 102 starch structure due to the inhibition of amylose leaching and crystallization, and amylopectin 103

104 retrogradation, thus modifying the water distribution and moisture retention in the bread crumb105 (Biliaderis, Arvanitoyannis, Izydorczyk, & Prokopowich, 1997).

Several studies have focused on the nutritional and technological benefits of the fortification
with sprouted grains (Marti, Cardone, Pagani, & Casiraghi, 2018; Montemurro, Pontonio, Gobbetti,
& Rizzello, 2019) or fermentation for *in situ* EPS synthesis in native flour by legumes (Xu et al.,
2017; Wang et al., 2018), but very limited information on EPS *in situ* synthesis in sprouted/malted
grains are available (Zannini et al., 2013).

111 Therefore, the objective of this work was set up a fermentation process started by selected LAB on whole and sprouted lentil flours, aiming at obtaining sourdoughs. Starter LAB were selected 112 based on the capability to produce EPS in situ. The effects of fermentation on the main 113 microbiological, biochemical and functional features of the sourdoughs with and without added 114 sucrose were investigated. The impacts of 30% lentil or sprouted lentil sourdoughs, with and without 115 EPS, on the textural, volatile aromatic compounds (VOCs) and sensorial quality of composite wheat 116 bread were also determined. Furthermore, predicted glycaemic index and fibers content of the final 117 bread were evaluated. 118

119 **2.** Materials and methods

120 2.1. Microorganisms and culture conditions

Fifteen LAB strains belonging to Pediococcus pentosaceus, Lactobacillus spp., Weissella spp. and 121 Leuconostoc spp. were screened for their EPS-producing abilities (Table S1). Strains belongs to the 122 culture collection of the Department of Soil, Plant and Food Sciences (DiSSPA), University of Bari, 123 except Leuconostoc pseudomesenteroides DSM 20193, and Weissella cibaria DSM 20194, purchased 124 from Leibniz Institute DSMZ-German Collection of Microorganisms and Cell Cultures GmbH 125 (Braunschweig, Germany) and chosen for their confirmed EPS-producing capacity (Xu et al., 2017). 126 L. plantarum DPPMAB24W was used as a non EPS producing control (Xu et al., 2017). Strains were 127 previously isolated from different sources (Table S1) and subjected to taxonomic strain identification 128

by analysis of the 16S rRNA sequence according to Rizzello, Calasso, Campanella, De Angelis, &
Gobbetti (2014). All LAB strains were maintained frozen at -25 °C as 20% (v/v) glycerol stocks and
propagated in De Man, Rogosa and Sharpe (MRS) broth (Sharpe, Fryer, & Smith, 1966) at 30°C
(Oxoid, Basingstoke, England).

133 2.2. Screening for EPS producing strains

Each strain was grown on agar plates containing modified MRS medium with 20 g L⁻¹ of sucrose. 134 Cell suspension (OD600 = 0.3) was prepared in MRS broth by growing strains 24 h at 30°C. Cells 135 from 0.5 mL of culture were harvested by centrifugation $(10,000 \times g, 5 \text{ min}, 4^{\circ}\text{C})$, washed with 1 mL 136 of sterile water and resuspended in 0.2 mL of sterile water. Then, inoculation was performed by spot-137 ting 2 µL of bacterial suspension (ca 7 log cfu ml⁻¹ bacteria and 10 spots/plate) on MRS-sucrose agar 138 media. After incubation at 30°C for 24 to 48 h, the strains which produced slimy colonies were rec-139 orded as capable of producing EPS and classified according the visual appearance (compact, creamy 140 or liquid slime). The best slime producers were selected (Table S1) and used for dough fermentation. 141

142 2.3. Materials

The materials used in this study included lentil grains (*Lens culinaris*, Caporal Grani s.a.s., carbohydrate 50.4%, protein 30.0%, fat 0.66%, fibers 23%, moisture 11%); wheat flour (*T. aestivum*, commercial wheat flour type "0", Puratos Italia s.a.s., protein 13%, fat 1.9%, fiber 3.3%, moisture 13.6%), fresh yeast (Puratos Italia), sucrose (Sigma Aldrich) (used to induce the synthesis of dextran during fermentations) and salt. Lentil grains were sprouted according to Montemurro et al. (2019) with some modifications (Fig. S1) Raw lentil (L) and sprouted lentil (SL) flours were obtained from whole or sprouted grains through the same laboratory mill. The particle size of the flour was < 500μm.</p>

150 2.4. Dough fermentation

151 For fermentation, L or SL flour and tap water were mixed in 1:5 ratio corresponding to a dough yield
152 (DY, dough weight×100/flour weight) of 500. Best EPS-producing LAB cells were harvested from
153 an overnight culture in MRS broth supplemented with 2% (w/v) sucrose at 30°C by centrifugation

 $(10,000 \times g \text{ for } 10 \text{ min at room temperature})$ and washed twice in 50 mM phosphate buffer, pH 7.0 154 (Xu, Coda, Holopainen-Mantila, Laitila, Katina, & Tenkanen, 2019). Cell pellets were re-suspended 155 in 1ml of tap water needed for making the dough and added at an initial cell density of ca. 7.0 log cfu 156 (colony forming unit) g⁻¹ (referred to as EPS NEG). To enable *in situ* dextran formation, 5% w/w 157 dough weight (corresponding to 25 % w/w flour weight) of sucrose was added (referred to as EPS 158 POS) (Galle, Schwab, Dal Bello, Coffey, Gänzle, & Arendt, 2012). L and SL doughs without sucrose 159 and without inoculum were prepared as described above to be used as controls (referred to as CT). 160 Additionally, for each flour, a dough with sucrose and inoculated by L. plantarum DPPMAB24W as 161 non EPS producing control (referred to as B24W) (Xu et al., 2017) was prepared. All fermentations 162 were carried out at 20 and 25 °C for 24 h. pH and viscosity were determinate at 0 h and after 24 h of 163 fermentation as the means to select the best EPS producing conditions (Wang et al., 2018; Wang, 164 Compaoré-Sérémé, Sawadogo-Lingani, Coda, Katina & Maina, 2019). The measurement of pH was 165 using a Foodtrode electrode (Model HI 99161, Hanna Instruments, Woonsocket, RI, USA). Viscosity 166 values were measured on 60 g of doughs at 20°C with a RheolabQC rheometer (Anton Paar, Austria) 167 at different shear rates from 2 to 100 1s⁻¹ (up and down sweeps) (Xu et al., 2017). All tests were done 168 in triplicate. 169

170 2.5. Enumeration of cultivable bacteria and yeasts

Microbial cell densities were determined according to methods previously described (Rizzello et al., 171 2014) using culture media and supplements purchased from Oxoid. Mesophilic aerobic microorgan-172 isms were determined using Plate Count Agar after incubating at 30°C for 48 h. LAB were enumer-173 ated using modified MRS (containing 1% w/v maltose, 5% v/v fresh yeast extract, pH 5.6) agar plates 174 with cycloheximide (0.1 g liter⁻¹) at 30 °C for 48 h under anaerobiosis (AnaeroGen and AnaeroJar, 175 Oxoid). The number of yeasts cells was estimated at 30°C by using Wort agar supplemented with 176 chloramphenicol (0.1 g l⁻¹) for 48 h. The microbiological counts were confirmed by taking repre-177 sentative colonies for each medium, which were analysed for morphology, motility, Gram staining 178 reaction and catalase test. 179

180 2.6. Determination of free sugars, dextran and resistant starch

For dextran analysis, an enzyme-assisted method based on the enzymatic activity of the dextranase 181 and transglucosidase was used (Katina et al., 2009). Selected sourdoughs, before and after fermenta-182 tion, were frozen (-20 °C) and subjected to freeze drying, milling and sieving through a 0.5mm screen 183 to obtain powder samples. To remove free sugars and short oligosaccharides, 100 mg of each freeze-184 dried sourdough were washed with 3ml of aqueous ethanol (50% v/v) and vigorously vortexed en-185 suring that large aggregates were not formed. Then samples were boiled for 5 minutes and added with 186 other 3 ml of ethanol, vigorously vortexed again to break down all aggregated to ensure that free 187 sugars and oligosaccharides were completely dissolved. The mixture was centrifuged at 10000 rpm, 188 10 min and the supernatant were discarded. For the hydrolysis, the pellet was re-suspended in 4.5 ml 189 190 sodium citrate buffer (pH 5.5) and placed in a boiling water for 5 minutes. After cooling, dextranase 191 from *Chaetomium erraticum* (10000 nkat/g) (Sigma-Aldrich, Germany) and α -glucosidase from Aspergillus niger (1000kat/g) (Megazyme, Ireland) were added. A control sample for the correction of 192 glucose background was carried out adding only α -glucosidase enzyme. The efficiency of the en-193 zymes in hydrolysing dextran was evaluated using commercial dextran (from Leuconostoc spp., GE 194 Healthcare, Sweden). The samples were hydrolysed for 48h at 30°C with constant shaking, then 195 placed in a boiling water bath for 10 minutes to inactivate the enzymes and centrifuged at 10.000 rpm 196 197 for 10 minutes. The supernatants were collected and used for the glucose analysis by high performance anion exchange chromatography with pulsed amperometric detection (HPAEC-PAD). Glu-198 cose (Merck, Germany) was used as standard and 2-deoxy-D-galactose (Sigma-Aldrich, UK) was 199 used as the internal standard for quantification. The glucose released from the commercial dextran 200 was quantified. The amount of dextran was calculated as the sum of anhydro-glucose using corrector 201 factor of 0.90. 202

For sugar profiles, 100 mg of each freeze-dried sourdough were washed twice with aqueous ethanol, mixed with 5 ml of bidistilled water, vortexed for 5 min, and boiled for 5 min to inactivate any enzymatic and microbial activities. After cooling, 500µl of each sample was filtered with Amicon ultra 0.5- centrifuge filters (Millipore, Billerica, MA), at 12,000 x g for 10 minutes in order to remove
any polymeric molecules. The samples were diluted and subsequently analyzed by HPAEC-PAD as
reported by Xu et al. (2017). Resistant starch (RS) was determined on freeze-dried doughs by using
the Resistant Starch K-RSTAR kit (Megazyme Int., Bray, Ireland), following the manufacturer's
instructions.

211 2.7. Physicochemical and biochemical analyses

Total titratable acidity (TTA) was measured at 0 h and after 24 h of fermentation and was determined as the amount of 0.1 M NaOH required to adjust the end pH of 10 g dough in sterile sodium chloride (0.9% w/v) solution to 8.3, as reported previously (Rizzello et al., 2014). Water/salt-soluble extracts (WSE) from sourdoughs at 0 h and after 24 h of fermentation were prepared according to the method described by Weiss et al. (1993) and used to analyse organic acids, peptides, and total free amino acids (TFAA).

The content of lactic acid and acetic acid in the extracts was determined with commercial kits, K-DLATE and K-ACET (Megazyme, Wicklow, Ireland) kits. The quotient of fermentation (QF) was determined as the molar ratio between lactic and acetic acids. The peptide concentration was determined by the *o*-phthaldialdehyde (OPA) method (Church, Swaisgood, Porter, & Catignani, 1983). A standard curve prepared using tryptone (0.25-1.5 mg ml⁻¹) was used as the reference. The total amino acids were quantified with the ninhydrin test (Friedmann et al., 2004).

224 2.8. Total phenols and antioxidant activity

Total phenols were determined on the methanolic extract (ME) of doughs. Five grams of each sample were mixed with 50 ml of 80% methanol to get ME. The mixture was purged with nitrogen stream for 30 min, under stirring condition, and centrifuged at 4600×g for 20 min. The supernatants (methanolic extracts, MEs) were transferred into test tubes, purged with nitrogen stream and stored at ca. 4°C before analysis. The concentration was determined as described by Slinkard and Singleton (1977) and expressed as gallic acid equivalent. MEs were used to determine the antioxidant properties. The

free radical scavenging capacity of MEs was determined using the stable 2,2-diphenyl-1-picrylhydrazyl radical (DPPH) as reported by Yu et al. (2002). The absorbance value was compared with 75 ppm
butylated hydroxytoluene (BHT) as the antioxidant reference.

234 2.9. Extraction, identification, and quantification of free and bound phenolic compounds

Free and bound phenolics were extracted as described by De Pasquale, Verni, Verardo, Gòmez-Cara-235 vaca & Rizzello (2021) and analyzed by an ACQUITY Ultra Performance LC system equipped with 236 photodiode array detector with a binary solvent manager (Waters Corporation, Milford, MA, United 237 States) series with a mass detector Q/TOF micro mass spectrometer (Waters) equipped with an elec-238 trospray ionization (ESI) source operating in negative mode. Briefly, 4 g of freeze dried samples were 239 extracted twice in an ultrasonic bath with ethanol/water (4:1 v/v) for 10 min. The supernatants were 240 241 collected, evaporated at 40°C in a rotary evaporator, and reconstituted with 2 mL of methanol/water (1:1 v/v). Residues of free phenolics extraction were digested with 300 mL of 2 M NaOH at room 242 temperature overnight by shaking under nitrogen gas and then acidified (pH 2-3) with hydrochloric 243 244 acid and extracted with diethyl ether/ethyl acetate (1:1 v/v). The organic fractions were pooled and evaporated to dryness at 40°C in a rotary evaporator and bound phenolic compounds were reconsti-245 tuted in 2 mL of methanol/water (1:1 v/v). The identification and quantification of free and bound 246 polyphenols were carried out as described in De Pasquale et al. (2021). Integration and data elabora-247 248 tion were performed using MassLynx 4.1 software (Waters Corporation, USA). For the quantification of phenolic compounds, solutions of gallic acid, chlorogenic acid, ferulic acid and quercetin in meth-249 anol were prepared and used as standard. 250

251 2.10. Bread making trials

Five types of breads were prepared: control wheat bread manufactured using wheat flour fermented by baker's yeast alone (CWB); lentil sourdough wheat bread (LSWB EPS NEG); dextran-containing lentil sourdough wheat bread (LSWB EPS POS); sprouted lentil sourdough wheat bread (SLSWB EPS NEG) and dextran-containing sprouted lentil sourdough wheat bread (SLSWB EPS POS) (Table 256 S2). The optimal water content for the breads was based on wheat flour as determined with a Brabender Farinograph (Brabender GmbH & Co. KG, Germany). Thus, the amount of flour and water 257 was same in the control bread and in the sourdough breads. All breads (DY 162) were manufactured 258 at the pilot plant of Puratos (Ceparana, La Spezia, Italy), according to the two-stage protocol com-259 monly used for sourdough bread making (Rizzello et al., 2014). In particular, L and SL flours obtained 260 from lentil grains were fermented for 24 h at 20 °C with the selected starter as described before (step 261 I); then, L and SL sourdoughs were mixed with wheat flour, water, and baker's yeast in a mixer bowl 262 (Sottoriva S.p.A Group) for 5 min at low speed and 5 min at fast speed. The doughs were divided into 263 pieces of 500 g, moulded mechanically and rested in pans for 20 min at 25 °C and relative humidity 264 (RH) of 75%, then were leavened in a fermentation cabinet (Zucchelli S.p.a) for 60 min at 30°C and 265 266 RH 85% (step II). Baker's yeast was added at the percentage of 1.1% w/w. The breads were baked in a rotating rack oven (Zucchelli forni S.p.a) at 220 °C for 30 min. After baking, the breads were cooled 267 for 2 h at room temperature before weighing. Baking was done on two different days (two independ-268 ent baking trails) and five breads were prepared for each type. Each bread was analysed twice. The 269 pH and TTA of the bread crumb were determined as reported earlier. 270

271 2.11. Bread technological characterization

The baking loss of the breads was evaluated (% baking loss = (dough weight-bread weight) * 272 100/dough weight). Bread volume was determined by rapeseed displacement method 10-05.01 273 (AACC, 2000). The specific volume of the bread was calculated as the loaf volume (mL)/ loaf weight 274 (g) ratio, after 2-6 h of cooling. Texture Profile Analysis (TPA) of bread crumbs packed in polypro-275 pylene micro perforated bags and stored for 24 h at room temperature was analysed with a texture 276 analyzer (TA, TA-XT2i, Stable Micro Systems Ltd., UK) using a 36mm radiused cylinder probe on 277 days 1 and 7 of storage. Samples for testing were prepared by cutting the breads into 2 cm slices. The 278 analysis was carried out applying two compression cycles, at a speed of 1 mm s⁻¹ and 30% defor-279

280 mation of the sample. Results were acquired with TPA analyzer Stable Micro Systems, software exponent (version 5.0.9.0), giving the following bread textural parameters: hardness, springiness, cohe-281 siveness and resilience. The staling rate was calculated as the increase in hardness during 7 d of 282 storage (staling rate = [hardness (day 7-day 1)/days of storage]). The crumb grain of breads was 283 evaluated after 24 h of storage using image analyses technology, as reported by Rizzello et al. (2014). 284 A threshold method was used for differentiating gas cells and non-cells, aiming at calculating gas cell 285 to total area ratio. Analysis was carried out on two sub-images of 500×500 pixels (field of view) 286 selected from within the bread slice. Two slices were analysed per treatment. 287

288 2.12. Breads nutritional characterization

289 2.12.1. In vitro starch hydrolysis

In vitro starch hydrolysis was determined as previously described (Liljeberg, Åkerberg, & Björck, 290 1996). The procedure mimicked the in vivo digestion of starch. Aliquots of breads, containing 1 g of 291 starch (determined in bread), were subjected to enzymatic process and the released glucose content 292 293 was measured with D-Fructose/D-Glucose Assay Kit (Megazyme). The degree of starch digestion 294 was expressed as the percentage of potentially available starch hydrolysed after 180 min. Control bread without sourdough leavened with baker's yeast was used as the control to estimate the hydrol-295 ysis index (HI=100). The predicted glycemic index (GI) was calculated using the equation: 296 297 GI=0.549×HI+39.71 (Goñi, Garcia-Alonso, & Saura-Calixto, 1997).

298 2.12.2. Total and Insoluble dietary fibers

Total (TDF) and Insoluble (IDF) dietary fibers were determined according to the AOAC 2011-25 enzymatic-gravimetric method (AOAC, 2011) and soluble (SDF) dietary fiber was calculated as a difference between TDF and IDF according to Tobaruela, Santos, de Almeida-Muradian, Araujo, Lajolo, & Menezes (2018).

303 2.13. Volatile organic compounds profile of breads

VOCs profile of samples was carried out by a GC-MS analyses according to Pico, Bernal, & Gòmez (2015). The GC-MS generated a chromatogram with peaks representing individual compounds. Each chromatogram was analyzed for peak identification using the National Institute of Standard and Technology (NIST) 2008 library. A peak area threshold of >1 000 000 and a match criterion of >85% was used for VOCs identification followed, when necessary, by manual visual inspection of the fragment patterns. Compounds were quantified in terms of arbitrary area units.

310 2.14. Bread sensory analysis

Sensory analysis of breads was carried out by 10 trained panellists (5 males and 5 females, mean age: 311 30 years, range: 18–54 years), as previously described by Rizzello et al. (2014). Sensory attributes 312 included: visual and tactual perception (color of crust and crumb, elasticity, consistency, friability); 313 taste (acidic taste, sweetness, salty, legume flavor, bitter flavour); smell perception (acidic odor, car-314 amel-like odour); chewing (chewiness, wetness), and overall aroma, using a scale from 0 to 10, with 315 316 10 the highest score. Samples were served in random order and evaluated in two replicates by all panellists. Before the sensory evaluation, the loaves were thawed at room temperature for 5-6h, then 317 318 cut into slices 1.5cm thick. Slices were cut into 4 pieces and each panellist received 2 pieces per sample. Final scores for each attribute were calculated as the means of the data collected in three 319 independent evaluations. 320

321 2.15. Statistical analysis

Experimental data as triplicates were used to determine significative variable multiple comparison in a two-way ANOVA, with a Turkey-Kramer post hoc test. Correction for multiple test was computed by using BH. Significative data were subjected to pair-comparison of treatment means using Tukey's procedure at P < 0.05 using a statistical software Statistica 7.0 (Statistica for Windows 7.0). The VOCs peak areas were analysed by principal component analysis (PCA) with SCAN software from Minitab (State College, PA, USA).

328 3. Results and Discussion

329 3.1. Screening for in situ EPS production

Fifteen LAB strains were screened for EPS production using sucrose as carbon source, at a concen-330 tration of 20 g L⁻¹ (Wang et al., 2019) (Table S1). The final screening was carried out considering the 331 ability to cause the decrease of the pH and the increase in viscosity of the liquid doughs after fermen-332 tation at the different conditions tested (Table S3) (Wang et al., 2018; 2019). Liquid doughs (contain-333 ing 80% of water) were obtained, to mimic the industrial production conditions, in which the use of 334 automatic bioreactors (intended for liquid sourdough propagation) is very common (De Pasquale et 335 al., 2021). Before fermentation, the pH of the doughs was approximately 6.15-6.38, and it decreased 336 to 4.0 to 5.6 after fermentation. No significant differences were observed between EPS POS and EPS 337 NEG (without sucrose addition) sourdoughs and between fermentation carried out at 20 or 25°C. 338 339 Almost all the controlled fermentations showed a significant change of viscosity compared to nonfermented doughs (0 h), except for the non EPS producing strain L. plantarum DPPMAB24W. This 340 increase happened only during controlled fermentation, clearly indicating that synthesis of dextran 341 occurred only through the starter activity and confirming the thickening ability of EPS (Table S3). 342 Viscosity is typically measured as indicator of EPS formation during fermentation (Lynch et al., 343 2018). Significantly (P < 0.05) higher viscosity values were reached after 24 h of fermentation at 20°C 344 than 25°C. Cerning, Bouillanne, Landon & Desmazeaud (1992) reported that an incubation tempera-345 ture below the optimum for growth resulted in greater production of EPS. The highest viscosity in-346 crease (up to 5.54 Pa \cdot s) (P < 0.05) was obtained in sourdoughs with sucrose addition fermented by 347 W. confusa SLA4, which was therefore selected as starter for L and SL sourdoughs fermentation, and 348 baking trials. This obligately heterofermentative LAB species is indigenous in many cereal raw ma-349 terials and in sourdough. Production of dextran, the best-known EPS formed by heterofermentative 350 LAB, has been recorded for Weissella species (Björkroth, Dicks, & Endo, 2014). The term "dextrans" 351 is given to a large class of α -glucans polysaccharides composed of α -1,6 glycosidic bonds in the main 352 chains and α -1,2, α -1,3 and α -1,4 branch linkages (Wang et al., 2019). As previously shown, dextran 353 produced by W. confusa had a linear structure of $\alpha 1 \rightarrow 6$ with up to 3% $\alpha 1 \rightarrow 3$ branching and 3.3 354

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 $\times 10^6$ g mol⁻¹ (Wang et al., 2019). Dextran form a network that strengthens the gluten network and binds water, thus improving dough stability and gas retention, further leading to improved loaf volume and crumb softness (Lynch et al., 2018). Dextran of different molecular structures has different efficiency in enhancing the quality of baking products. Dextran with high Mw and few branches has been extensively investigated to act as hydrocolloids and to achieve enhanced wheat bread quality (Rühmkorf et al., 2012, Zhang et al., 2018).

361 **3.2** Cultivable microbiota of sourdoughs

A two-way ANOVA on triplicate readings of the data was conducted to determine significative 362 variable multiple comparison in order to examine the effect of sprouting and fermentation with in situ 363 dextran formation on sourdoughs (Table S4). Corrected P values < 0.05 were deemed statistically 364 365 significant and were considered in this study. Changes in cultivable microbiota were monitored in L and SL sourdoughs, before and after spontaneous fermentations for 24 h at 20°C, and in controlled 366 fermentation started by W. confusa SLA4, with and without sucrose addition (Table 1). Microbial 367 profiles changed as consequence of germination and fermentation. Overall, the addition of sucrose 368 did not significantly affect the final cell density and the cell density increase as already reported in 369 similar studies (Xu et al., 2017; Wang et al., 2018). Before fermentation, SL doughs were 370 characterized for the main microbial groups. After fermentation, the mesophilic bacteria count 371 showed very similar values between all the sourdoughs and reached over 9.0 log cfu g⁻¹. After 24 h, 372 *Enterobacteriaceae* grew in almost all sourdoughs, and increased up to 4 log cycles in spontaneously 373 fermented L and SL sourdoughs. In controlled fermentations, the initial cell count of presumptive 374 LAB was approximately 6.7 log cfu g⁻¹ (Table1), corresponding to the initial inoculum of the starter, 375 while it was ca 2.0 log cycles lower in non-inoculated doughs. After 24 h of fermentation, the cell 376 density of presumptive LAB in all the sourdoughs increased to 8.9 log cfu g⁻¹. Sourdough originates 377 from the spontaneous or starter culture-initiated fermentation of mixtures of flour and water (Gobbetti 378 et al., 2019) and in the first step redox potential decreases favouring the growth of facultative 379 anaerobes Enterobacteriaceae and LAB. The initial cell count of yeasts in all sourdoughs was lower 380

than 3 log cfu g⁻¹. After fermentation, yeasts number increased up to 1 log cycles only in not
inoculated sourdoughs and was almost completely inhibited in all the controlled fermentations.

383 3.3 Dextran, sugars, TTA and organic acids in sourdoughs

384 In this study, L and SL flours were used as a substrate for dextran formation in situ during LAB fermentation, with the aim of assessing the possibility to increase its applicability as baking ingredi-385 ent. Based on 25% w/w of flour weight (f.w.) of sucrose addition (corresponding to 5% w/w dough 386 weight), 12.5% w/w f.w. dextran could theoretically have been formed (Kothari, Das, Patel, & Goyal, 387 2015). In the conditions of this study, only ca. 73.6 and 77.6% of the theoretical dextran was synthe-388 389 sized in doughs with sucrose addition (referred as to EPS POS), corresponding to ca. 9.2 and 9.7% w/w f.w. in fermented L-SLA4 EPS POS and SL-SLA4 EPS POS sourdoughs, respectively (Table 390 2). This yield is comparable with that previously obtained in pearl millet fermented by W. confusa 391 392 A16 (3.5% dry weight) and shown to have a positive impact on bread quality when used as ingredient (Wang et al., 2019), in fava protein concentrate (10.0% dry weight) (Xu et al., 2019b) or other grains 393 (for review see Lynch et al., 2018). Differences in dextran production can derive by different factors, 394 including the type of flours, which affects the activity of dextransucrase and the presence of sugar 395 acceptor such as maltose, favouring the synthesis of oligosaccharides (Lynch et al., 2018). Based on 396 397 the sugar analysis, a lower theoretical amount of fructose released from sucrose through the activity of dextransucrase was accumulated in L-SLA4 EPS POS and SL-SLA4 EPS POS sourdoughs after 398 24 h of fermentation (8.8% and 9.2% w/w f.w., respectively, instead of 12.5%). It could be hypothe-399 400 sized that fructose was preferable to glucose for *Weissella confusa*, in agreement with Kajala et al., 401 (2015) which observed that different strains might have different carbon source preferences. Overall, lentil and sprouted lentil flour appeared to be good substrate for EPS production. Stredansky, Conti, 402 403 Navarini, & Bertocchi (1999) hypothesized that one important factor influencing the yield of synthe-404 sized EPS could be the similarity of the substrate to the natural habitat of the microorganisms. This could fit our experimental conditions, since SL flour is the natural habitat which W. confusa SLA4 405

406 was isolated (Perri, Calabrese, Rizzello, De Angelis, Gobbetti, & Calasso, 2020). During fermentation, a small amount of dextran was produced also in sourdoughs without sucrose addition (referred 407 as to EPS NEG) (0.88 and 1.17 % w/w f.w. in L-SLA4 EPS NEG and SL-SLA4 EPS NEG sour-408 doughs, respectively), probably due to the endogenous sucrose present in the native L and SL flours 409 (Table 2). In agreement with previous studies (Katina et al., 2009; Wang et al., 2018), a correlation 410 between viscosity increase (Table S3) and in situ formation of dextran during fermentation was ob-411 served. Viscosity increase is an index of dextran synthesis, but this correlation is not always linear. 412 The viscosity of a matrix containing dextran depends on other factors, including intrinsic character-413 istics of the biopolymer (e.g. Mw and structure) and environmental factors (e.g. pH, temperature) 414 (Rühmkorf, et al., 2012; Xu, Pitkänen, Maina, Coda, Katina, & Tenkanen, 2018). 415

Lentils, like other legumes also contain Raffinose Family Oligosaccharides (RFO) (raffinose, 416 417 stachyose and verbascose) which can have antinutritional effect (Bresciani and Marti, 2019). Germination caused a significant reduction of RFO in lupin, cowpeas and kidney bean (Kalpanadevi 418 & Mohan, 2013; Shimelis & Rakshit, 2007). Raffinose is not digested by pancreatic enzymes but 419 metabolized by gas-producing bacteria in the large intestine, thus causing disorders such as flatulence 420 (Bresciani and Marti, 2019). Their presence in significant amount prior and after the germination is 421 one of the major limitations to legumes use in animal and human nutrition (Wang et al., 2018). 422 Raffinose was significantly lower (P < 0.05) in SL-SLA4 sourdoughs compared to L-SLA4 423 sourdoughs. Sourdough fermentation caused a significant (P < 0.05) decrease of RFO concentration 424 (Curiel et al., 2015). Fermentation with W. confusa SLA4 significantly decreased the raffinose in both 425 L and SL sourdoughs, however an higher decrease was observed when L flour was used as substrate. 426 In SL sourdoughs, verbascose was not detectable while it was ca. 0.2 % f.w. in L sourdoughs. 427 Fermentation also significantly increased (P < 0.05) galactose, most probably due to α galactosidase 428 action on RFO, in all doughs after fermentation. 429

The total starch was 45.5% in L-SLA4 doughs, which is in accordance with the typically reported 430 starch content of lentil flours (Wang & Daun, 2006). Germination causes an increase of the α-amylase 431 activity (Pal et al., 2017) which lead to a total starch loss (ca. 28.6% for SL-SLA4 doughs) (Table 2). 432 Germination process in lentil seeds has been shown to have little effect on the physico-chemical 433 434 characteristics of lentil starch and a significantly increase of its digestibility (Frias, Fornal, Ring, & Vidal-Valverde, 1998). Resistant starch (RS) was ca. 5% of total starch. Among non-digestible food 435 components, RS represents a small fraction of starch that is resistant to hydrolysis by digestive en-436 zymes. Fermentation with the selected lactic acid bacterium, lead a considerable increase of RS con-437 tent, which is partially due to the acidification caused by the organic acids released during fermenta-438 tion (Verni, Verardo & Rizzello, 2019). Compared to T0, RS was ca. 1.6 time higher in all fermented 439 sourdoughs (Table 2), without significantly differences between sourdoughs with or without sucrose 440 addition. 441

Before fermentation, the TTA values of doughs varied between 2.1 (L-SLA4 EPS POS) and 3.8 (SL-442 SLA4 EPS POS) mL NaOH 0.1N (Table 2). Final TTA was higher in sourdoughs from SL flour 443 compared to the corresponding unsprouted. In addition, sucrose supplementation (EPS POS) leads to 444 higher TTA values compared with EPS NEG doughs, as reported earlier (Xu et al., 2017) (Table 2). 445 The concentration of lactic and acetic acid in sourdoughs plays an important role in the taste and 446 flavour of sourdough bread. In agreement with previous results, during L and SL sourdoughs fermen-447 tation by W. confusa, sucrose addition increased the lactic acid concentration (16.20 ± 0.8 and 20.10448 ± 1.00 mmol Kg⁻¹ in L-SLA4 EPS POS and SL-SLA4 EPS POS, respectively) compared to sour-449 doughs without sucrose addition (12.10 ± 0.50 and 17.10 ± 0.80 mmol Kg⁻¹ in L-SLA4 EPS NEG and 450 SL-SLA4 EPS NEG, respectively) (Xu et al 2019b) but did not affect the acetic acid concentration 451 (Wang et al., 2018). The resulting fermentation quotient (FQ, the molar ratio between lactic and acetic 452 acids) was higher in EPS POS sourdoughs. The FQ is a useful parameter for evaluating the balance 453

454 of acids produced in sourdough. *Weissella* spp. typically do not harbor mannitol dehydrogenase ac455 tivity and do not convert fructose to mannitol with concomitant acetate formation, which resulted in
456 less acidic doughs compared to those fermented with other LAB, e.g. *Leuconostoc* spp. (Katina et al.,
457 2009).

458 3.4 Protein derivatives, total phenols and antioxidant activity in sourdoughs

Aiming at investigating the proteolysis occurring on L and SL proteins during fermentation, the 459 profile of the organic nitrogen compounds was evaluated based on the analysis of peptides and TFAA 460 concentrations (Table 2). Germination leads an higher concentration of peptides and TFAA in SL 461 doughs, as already reported for cereal, pseudo-cereal and legume flours (Montemurro et al., 2019), 462 without significant differences (P > 0.05) by sucrose addition. As expected, fermentation led to an 463 increase of peptides and TFAA concentration, driven by a combined effect of the flour endogenous 464 465 proteases and LAB peptidases (Gobbetti et al., 2019). Overall, SL sourdoughs were characterized by higher concentration of TFAA compared to the corresponding L sourdoughs, slightly higher when 466 sucrose was added. TFAA was, in all the sourdoughs, in the range 1.4 - 2.6 g kg⁻¹ dough (Table 2). 467

Before fermentation, the concentration of total phenols, examined after methanolic extraction, was 468 significantly higher in SL doughs, probably due to a better extractability of phenolic compounds from 469 the seed altered tissues (Singh, Rehal, Kaur, & Jyot, 2015) (Table 2). In agreement with the higher 470 solubilisation of total phenols, also the antioxidant activity was higher (P < 0.05) in SL sourdough 471 (ca. 86% average value) compared to L sourdoughs (ca. 82.5%), without significant differences (P >472 0.05) for sucrose addition. Lactic fermentation increased the levels of bioactive compounds (e.g., 473 total phenolic compounds) compared to T0 but no significant differences were found between 474 sourdoughs with and without sucrose supplementation (Table 2). After 24h of fermentation, the total 475 phenols content in L sourdoughs was ca. 2 times higher, while was ca. 1.2 times in SL sourdoughs. 476 LAB fermentation was already suggested as tool to improve the total phenols bioavailability of 477 legumes (Curiel et al., 2015). Together with the increase of the total phenols concentration in 478

479 methanolic extracts, also the DPPH radical-scavenging activity increased (<10%) in all samples by
480 the fermentation (Table 2), which is consistent with data reported in literature (Wang et al., 2019).

Aiming at better understanding which compounds were responsible for the antioxidant activity of L 481 482 and SL sourdoughs, phenolic compounds were selectively extracted and analyzed by UPLC-PDA-ESI-QTOF (Fig. S2). Under the conditions of this study, eight free and bound phenolic compounds 483 were identified, all previously described in various lentil cultivars (Zhang et al., 2015; Singh et al., 484 2017; Ghummann, Singh & Kaur, 2020). Before fermentation, in free phenolic profile gallic acid, 485 chlorogenic acid and hydroxybenzoic acid derivatives were detected in both L-SLA4 and SL-SLA4 486 487 doughs, and a significant (P < 0.05) increase was observed with sprouting, as previously found by Ghummann et al. (2020) (Fig. S2 A). Sprouting led to a decrease of these phenolic compounds 488 identified in bound form, probably due to increased enzyme action on various cell wall components 489 490 to which these phenolics are bound (Fig. S2 B) (Ghummann et al., 2020). In agreement with Ghumann et al., (2020), hydroxycinnamic acid derivatives, such as ferulic and p-coumaric acids, increased 491 during sprouting. Fermented L-SLA4 and SL-SLA4 doughs, compared to unfermented ones, had a 492 concentration of total free phenolic compounds 9- and 8.7-fold higher, respectively, and a 493 concentration of total bound phenolic compounds ca. 20% lower (Fig. S2). Before fermentation, in 494 495 L-SLA4 and SL-SLA4 doughs, the concentration of bound phenolic compounds was 20- and 15times higher than free phenolics, respectively. In fermented sourdoughs, bound phenolics had a 496 concentration ca. 1.5 times higher than free phenolics. LAB metabolic activities can affect the 497 bioaccessibility of polyphenols bound to cell wall, glycosylated, or in polymeric forms, acting on 498 499 their release (e.g. feruloyl esterases, glycosyl hydrolases, tannases) or conversion (e.g. phenolic acid reductases and decarboxylases) into more active forms (Verni et al., 2019). Phenolic acids were the 500 501 most representative, reaching up to 45 and 39% of the total bound compounds in L-SLA4 and SL-SLA4, respectively, of which gallic and p-coumaric acids were the most abundant. Catechin 502 represents 42 and 47% of total bound phenolic compounds in L-SLA4 and SL-SLA4 doughs, 503

respectively. Fermentation allowed an increase up to 20% (SL-SLA4 sourdoughs) of the total phenols extractability from the matrix, and no significant differences were observed between EPS NEG and EPS POS doughs. Bacterial EPS have been proved to have antioxidant activity, that can be associated to physico-chemical properties or structural features (Mw and the number of hydroxyl and amino groups) (Wang, Hu, Nie, Yu & Xie, 2016). Bacterial purified EPS produced during fermentation have antioxidant activity *in vitro* and *in vivo* but few studies have confirmed it during food fermentation (Verni et al., 2019).

511 3.5 Breads characterization

Nowadays consumers are more health oriented and conscious of the nutritional benefits of food. In 512 response to consumers' demands, formulation of pulse composite bread is a challenge. Pulses are 513 generally incorporated in common wheat flours to percentages below 10-15% (Bresciani and Marti, 514 2019), indeed it is known that the use of legume flours markedly affect the texture of baked goods 515 (Kohajdová et al., 2013; Gobbetti et al., 2019). A few studies only discuss the incorporation of 516 germinated legumes in bread for nutritional enhancement (for review see Bresciani & Marti, 2019). 517 Previous studies on composite breads have confirmed the positive effects of dextran on the 518 technological and sensory quality of baked goods fortified with pulses (Wang et al., 2018) but no 519 studies are yet available on bread produced with dextran containing germinated grains. To the best of 520 our knowledge, this study reports for the first time a detailed investigation on the effect of germination 521 and fermentation with *in situ* dextran formation on the technological and nutritional properties of 522 lentils/wheat composite bread. In this study, 30% w/w d.w. of L and SL sourdough, corresponding to 523 10% w/w of the wheat flour substitution (Table S2), was used in the bread recipe. Experimental data 524 as triplicates were used to determine significative variable multiple comparison in a two-way 525 ANOVA. Corrected P values lower than 0.05 were considered as significant (Table S5) and variables 526 further described. The lowest baking loss (7.74%) was measured in bread with addition of SL-SLA4 527 EPS POS sourdough. Breads containing sourdoughs were characterized by higher (from +7.4 to 528

+26%) (P < 0.05) specific volume than the control wheat bread (CWB) (Table 3). In particular, dextran containing sourdoughs led to higher specific volumes (3.4 and 3.3 cm³ g⁻¹ for LSWB EPS POS and SLSWB EPS POS breads, respectively) compared to breads fortified with the corresponding EPS NEG (3.0 and 2.9 cm³g⁻¹ for LSWB EPS NEG and SLSWB EPS NEG breads) and CWB (2.7 cm³g⁻¹). Nevertheless, slight differences of volume and specific volume among the breads were found when SL were added indicating that starch changes upon sprouting did not interfere with dough properties and improved its leavening properties (Montemurro et al., 2019).

After 1 day, the addition of 30% w/w d.w. L or SL sourdough resulted in a significant decrease of hardness of bread crumb, compared to CWB, except for SLSWB EPS NEG (Table 3). Notably, the inclusion of dextran containing sourdoughs, significantly decreased the crumb hardness up to 33% compared to CWB, and this effect persisted during the 7 days of storage.

Springiness represents how well a product physically springs back after deformation. The lowest 540 value of springiness was found for CWB and SLSWB EPS NEG (ca. 0.89 cm). Cohesiveness and 541 resilience parameter indicate, respectively, how well the bread withstands a second deformation (after 542 the second compression cycle) and how well a product fights to regain its original position. For these 543 two parameters, EPS NEG sourdough breads showed the lowest value (ca. 0.76 - 0.39 and 0.74 - 0.37, 544 respectively). Breads containing EPS POS sourdough had increased springiness and resilience values 545 compared to CWB. Bread crumb was evaluated by image analysis technology. Digital images were 546 pre-processed to estimate crumb cell-total area through a binary conversion (Table 3). Using L-SLA4 547 EPS POS and SL-SLA4 EPS POS sourdoughs significantly increased (P < 0.05) cell-total area from 548 22.5% (CWB) to 56.5 and 56.2%, respectively, which was similar to that of bread with L-SLA4 EPS 549 NEG and SL-SLA4 EPS NEG sourdoughs (57.2 and 56.3%). 550

551 The pH value of CWB crumb was ca. 5.92 (Table 3). As expected, pH values of L and SL crumbs 552 were significantly (P < 0.05) lower in both the EPS NEG (ca. 4.87 and 4.83, respectively) and EPS 553 POS (ca. and 4.91 and 4.88, respectively) formulations. All bread added of sourdoughs showed TTA 554 values for crumbs significantly higher (P < 0.05) than the control bread.

It is known that the use of legume flours is usually associated to a weak structure and baking quality 555 556 of the dough, to a decreased volume of the bread and elasticity of the crumb, and to an increased hardness of the loaves (Kohajdová et al., 2013). However, sourdough biotechnology can be tailored 557 to obtain increased bread quality (Wang et al., 2018). Previously, a good acceptability of breads made 558 with 15% w/w of legume flours (chickpea, lentil and bean) was obtained as consequence of sourdough 559 fermentation (Rizzello et al., 2014). In this study, the presence of dextran further enhanced the quality 560 of the composite bread. Acting as hydrocolloid, dextran addition during bread making results in 561 enhanced stability of the dough film around the expanding gas cells during proofing, leading to 562 decreased foam losses, thus contributing to a higher loaf volume (Lynch et al., 2018). The positive 563 564 impacts of dextran on bread properties depends also on its concentration. In this study, the final breads contained 0.56-0.58 % w/w f.w. dextran, which was in the range (0.1-2%) of commercial 565 hydrocolloids such as carboxymethylcellulose, Guar gum and κ -carageenan applied in baking 566 567 (Ferrero, 2017). However, because these commercial hydrocolloids are typically added as ingredients, they are designated as additives in food and this requires labelling on the product 568 569 packaging, unlike in the case of dextran synthetized in situ. In addition, they hold a negative perception with consumers, demanding more natural products containing fewer additives. Previously, 570 571 it was shown that optimal baking quality of wheat-lentil flour was obtained using concentrations of 572 lentil flour up to 5% or up to 20% with the addition of gluten which maintained a superior loaf and crumb quality (Portman et al., 2018). This study showed good technological properties of breads 573 made by using 10% w/w f.w. of lentil flours as the consequence of the sourdough fermentation. 574

575 The use of dextran containing sourdoughs (EPS POS) led to a slight but significant reduction of the 576 staling rate (P < 0.05), which resulted in values up to 8% lower than those of CWB (205 g). In con-

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trast, no positive effect was observed by the addition of EPS NEG sourdoughs which staled significantly faster than the others. Bread staling is an irreversible and complex process involving multiple mechanisms through which starch amylopectin recrystallization and water redistribution occurs (Ferrero, 2017). The anti-staling effect of dextran could be partially attributed to dextran polymers competing for water, thus fewer water molecules are available for the formation of amylopectin crystallites resulting in an increased bread shelf life (Wang et al., 2019).

In agreement with a previous study (Wang et al., 2018), this result shows that sourdough fermentation alone in composite pulses wheat breads has no effect on crumb hardness and staling rate of stored breads. In contrast, the use sourdough with *in situ* dextran formation considerably modified all the textural attributes of the composite breads, resulting in higher volume, softer crumb, higher springiness and cohesiveness.

588 3.7 Dietary fibers and starch hydrolysis index

Many health benefits related to the regular consumption of whole cereals and legumes have been 589 attributed to their high content of DF, and their inclusion in diet is recommended to increase the daily 590 DF intake. During fermentation, an increase in the fibers composition of fermented pulses occurs 591 (Vidal-Valverde, Frias, Prodanov, Tabera, Ruiz, & Bacon, 1993). In this study, the composite bread 592 containing 10% w/w f.w. of L or SL flour was set up considering the TDF content of lentil, to obtain 593 a 3% dietary fibers content which allows the nutrition claim "source of fibers" (European Parliament 594 and Council, 20/12/2006). TDF concentration in sourdough breads was significantly higher (P < 0.05) 595 than that of CWB and was affected by the pre-treatment of the flour used as ingredient. During ger-596 mination, IDF and TDF decreased due to the activity of endogenous enzymes (Xu et al., 2019a). As 597 a result, among the breads fortified with sourdough EPS NEG, those containing SL had the lowest 598 values of TDF (3.17 %) compared to LSWB EPS NEG (3.37%). Inclusion of sourdough EPS POS 599 led to a further significant increase of TDF up to 23% compared to the EPS NEG counterparts. Fur-600 thermore, a significant increase of SDF in EPS POS bread samples was found, corresponding to 32 601 and 57% in LSWB and SLSWB, respectively. EPSs are dietary fibers, and their presence in cereal-602

based foods lead to a better bioavailability of dietary fibers in the product (Lynch et al., 2018). Dietary non-digestible oligosaccharides modulate the composition and activity of intestinal microbiota, and may exert human health benefits based on improved bowel functions, prevention of overgrowth of pathogenic bacteria, through the stimulation of probiotic members of the intestinal microbiota, and increased synthesis of short-chain fatty acids (Galle et al., 2012).

Bread is one of the most relevant sources of available carbohydrates in the diet and since lowering 608 dietary glycaemic index (GI) is considered favourable to health (Scazzina, Siebenhandl-Ehn, & 609 Pellegrini, 2013), in this study we investigate the effect of the addition of L or SL sourdoughs with 610 or without dextran on the GI of wheat bread. The hydrolysis index (HI) and predicted GI for bread 611 samples are summarized in Table 3. With 10% w/w L and SL flour substitution, GI changed 612 613 significantly (P < 0.05) compared to CWB, except for LSWB. Due to the appreciable increase of sugars during germination (Frias et al., 1996), a significant increase of GI was found in bread samples 614 containing SL flour. The HI and GI values of bread prepared with EPS POS sourdough significantly 615 differed from its EPS NEG counterpart. There are many studies showing the impact of dietary fibers 616 on the GI of bread. Viscous soluble dietary fibers are generally recognized for their effect on reducing 617 the GI of bread products which may slow the gastric emptying rate or the absorption of nutrients in 618 the small intestine (Scazzina et al., 2013). In this respect, the presence of high Mw dextran should 619 reduce the GI of breads. However, fibers effect on glycaemia depends on specific characteristics such 620 as the solubility and structure of the food which may lead to differences in nutrient bioavailability, 621 rates of absorption and post-prandial outcomes that might modify their potential health risks (Turgeon 622 & Rioux, 2011). Considering that GI is related to food structure, previous researches investigated the 623 impact of bread density on the glycemic index. A direct correlation between GI decreases and low 624 specific volume values was found (Burton and Lightowler, 2006). In this study, the GI value for CWB 625 was significantly lower (P < 0.05) than that of the higher volume LSWB EPS POS and SLSWB EPS 626 POS breads. These findings agree with those of Saulnier et al. (2013) showing a strong correlation 627 between GI index and the density of the bread, independently on the level of dietary fibre content. 628

629 3.8 Volatile organic compounds

Sixty-nine VOCs were found and grouped into nine different chemical classes (Table 4). To highlight
differences among bread samples, the results of VOC levels in the breads were elaborated in a PCA
(Fig. 1), with the principal components explaining ca. 83.2% of the total variance. The CWB and
sourdough breads were well separated from each other. LSWB and SLSWB, were also well separated.
Germination and sourdough fermentation can modify the flavour and texture of grain raw material
(Heinio et al., 2003).

636 Higher levels of furan and pyrazines compounds, pyrrolines, and aldehydes were found for SLSWB637 samples, while LSWB samples showed higher contents of alcohols and ketones compounds.

A total of 28 heterocyclic compounds were detected in the breads, being furan-2-pentyl, acetylfuran, 2-furancarboxaldehyde,5-methyl, and 2-methylpyrazine the most representative. SLSWB breads contained an high amount of heterocyclic compounds (Table 4). Pyrazines, mainly originating during baking (e.g., Maillard reaction), give roasted and burnt flavour to bread crust (Pico et al., 2015). In SLSWB breads the highest concentration of maltol was observed. This compound was abundant in roasted barley malt (Yahya, Linforth and Cook, 2014) and is most probably linked to the sprouting and subsequent drying process to which lentil was subjected before milling.

Among the 13 alcohol compounds, ethanol and 1-hexanol alcohol, followed by 3-nonen-1-ol and 645 benzylalcohol alcohol, were the most representative. Compared to CWB, the level of alcohols was 646 higher in all the sourdough breads, especially ethanol, 1-hexanol, 3-nonen-1-ol and benzylalcohol. 647 Ethanol is one of the end products of fermentation of carbohydrates operated by yeast and may 648 probably correlated to bakers' yeast activity. 1-Hexanol is an important VOC in the crumb, giving 649 green grass, flowery, woody, mild and sweet aroma notes (Pico et al., 2015). Benzylalcohol correlate 650 positively with the aroma of bread. Compounds like 3-nonen-1-ol, and benzyl alcohol, which have 651 been considered important for bread odour quality, were identified in bread whose sourdough was 652

started by selected LAB and yeasts but not in the bread made with spontaneous traditional sourdough
(wild microbiota) (Plessas, Mantzourani, & Bekatoru, 2020).

Besides alcohols, aldehydes greatly contribute to the VOC profile of bread (Pico et al., 2015). Furfural 655 was the most abundant among the 12 aldehydes found, followed by hexanal and nonanal. Furfural is 656 a volatile heterocyclic compound found in the crumb and/or crust of the wheat bread but also in in 657 sprouted and fermented rye (Heinio et al., 2003) known for the typical odors of soil, toasted. The 658 aldehydes 2-methyl butanal, 3-methyl butanal, benzaldehyde,2-nonenal were found at the highest 659 level in the bread produced with SL sourdough. 2-methyl butanal and 3-methyl butanal, giving malty 660 aroma to fresh bread, are among the so-called Strecker aldehydes; however, they may also derive 661 from the Ehrlich pathway of leucine and isoleucine, respectively, and are the direct precursors of 662 isoamyl alcohol and 2-methyl butanol (Pétel, Onno, & Prost, 2017). Bread crust color and the roasted 663 aroma are due to these Maillard compounds. 664

3-Hydroxy-2-butanone (acetoin) and 2-nonanone were the only two ketones identified in the breads.
They confer pleasant buttery and fruity odour (Pico et al., 2015). Overall, the level of ketones was
not affected by the addition of sourdoughs (Table 4).

Ethyl acetate, characterized by low odour activity and high volatility, is very important in wheat bread
crumb aroma since it possess pleasant, sweet, fruity odours (Pico et al., 2015). Overall, the addition
of sourdough did not affect the level of ethyl acetate in breads.

Acetic acid was the most abundant carboxylic acid (6 compounds), followed by hexanoic acid. Acetic
acid is the major volatile compound resulting from lactic acid fermentation (Pico et al., 2015).
Hexanoic acid was abundant in bread made by SL flour, and was associated with sweaty, cheesy,
fatty, goat-like odour.

675 **3.9 Breads sensory aspects**

The addition of sourdough caused an increase of the score of several attributes such as elasticity, 676 color, acidic taste and legume flavour (Fig. 2). The score for crust and crumb color was higher for all 677 sourdough breads, especially SLSWB both EPS NEG (7.6 ± 0.6 and 5.6 ± 0.7 , respectively) and EPS 678 POS (7.7 \pm 0.6 and 6.8 \pm 0.7) than CWB bread (1.1 \pm 0.50 and 1.2 \pm 0.50). Bread crust color is due 679 to Maillard compounds and some of those (2-methylbutanal, 3-methylbutanal) were more abundant 680 in SLSWB (Table 4). Moreover, during sprouting, reducing sugars and amino acids are released, 681 which subsequently react during heating, originating Maillard products. The scores for the acidic 682 attributes and legume taste were significantly higher in sourdough breads compared to CWB. In 683 particular, a markedly higher perception of legume taste was observed in LSWB compared to SLSWB 684 and CWB. Some beany flavours markers were employed for germinated lentil and included hexanal, 685 686 1-hexanol, and 2-pentylfuran which can be developed by lipolysis, lipid oxidation, and amino acids 687 degradation during pulse seeds germination (Xu, Jin, Lan, Rao, & Chen, 2019). In the breads studied, these compounds were higher in L sourdough breads than in the corresponding from SL. This can be 688 explained by the loss of beany flavour carriers which would result in the easy removal of beany 689 flavour in post-processing (Xu et al., 2019c). In addition, we can hypothesize an effect of LAB 690 fermentation although additional evidences are needed. SLSWB breads were sweeter than the others. 691 The unique flavour profile of sprouted grains is due to the activation of endogenous amylolytic 692 693 enzymes that transform starch into oligosaccharides and sugars, conferring sweetness. Caramel-like taste was perceived in all the sourdough breads, especially when SL flour was used. Germination was 694 shown to promote the formation of compounds yielding a caramel-like odour, and it is a well-known 695 process for adjusting the flavor of grains, especially after subsequent heat treatment process (Heinio 696 et al., 2003). The wetness of the crumb of the LSWB and SLSWB breads (average value 6.1) was 697 significantly different (P < 0.05) from that of CWB (4.8 \pm 0.6), especially for breads containing 698 dextran. The overall taste, considered as a global index of palatability, was higher in dextran 699 containing breads, especially LSWB EPS POS bread. In previous studies (Rizzello et al., 2014; 700

701 Montemurro et al., 2019), breads fortified with sourdough made by legume or fermented sprouted702 legumes and cereals flours have shown peculiar sensory profiles compared to wheat sourdough bread.

703 4. Conclusion

In conclusion, this study showed that germination of lentil flour may be combined with sourdough 704 705 biotechnology for improving structural, nutritional and sensory attributed of bread. The bioprocess resulted in an enough dextran production, which effectively counteract the quality deficiencies in-706 duced by gluten network disruption and balanced the negatives effects of wheat flour substitution by 707 lentil flour in the composite sourdough bread. The best results were obtained using W. confusa SLA4, 708 a key-species of the sourdough microbiota. As well known for breads in which the use of dextran 709 710 producing lactic acid bacteria improve nutritional and functional features, the use of lentil and sprouted lentil sourdoughs dextran-containing at 30% of the dough weight in wheat bread baking 711 showed potential advances with respect to enhanced nutritional (i.e. high fibers content) and sensory 712 713 (i.e. synthesis of bread key-aroma compounds) quality of the final products. These results open the way to future researches about the use of germinated grains and *in situ* dextran-producing LAB in 714 bakery industry. 715

716 Declaration of Competing Interest

717 The authors declared that there is no conflict of interest.

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971 Figure captions

Figure 1. Score (A) and loading (B) plot of the first and second principal components (PC) after PC 972 analysis based on VOC that mainly (P < 0.05) differentiated breads containing wheat flour added of 973 lentil flour fermented by Weissella confusa SLA4 sourdough (LSWB EPS NEG), wheat flour added 974 of lentil flour fermented by W. confusa SLA4 - dextran containing sourdough (LSWB EPS POS), 975 wheat flour added of sprouted lentil flour fermented by W. confusa SLA4 sourdough (SLSWB EPS 976 NEG), wheat flour added of sprouted lentil flour fermented by W. confusa SLA4 - dextran containing 977 sourdough (SLSWB EPS POS). All strains were inoculated at ca. 7 log cfu g⁻¹ and sourdough 978 fermented at 20 °C for 24 h. Wheat flour bread started with baker's yeast (CWB) was used as control. 979 Doughs for bread making had DY 162. Data are the means from three independent experiments 980 981 analysed in triplicate.

982 Figure 2. Spider web chart of the sensory analysis data for breads containing wheat flour added of lentil flour fermented by Weissella confusa SLA4 sourdough (LSWB EPS NEG), wheat flour added 983 of lentil flour fermented by W. confusa SLA4 - dextran containing sourdough (LSWB EPS POS), 984 wheat flour added of sprouted lentil flour fermented by W. confusa SLA4 sourdough (SLSWB EPS 985 NEG), wheat flour added of sprouted lentil flour fermented by W. confusa SLA4 - dextran containing 986 sourdough (SLSWB EPS POS). All strains were inoculated at ca. 7 log cfu g⁻¹ and sourdough 987 fermented at 20 °C for 24 h. Wheat flour bread started with baker's yeast (CWB) was used as control. 988 Doughs for bread making had DY 162. Data are the means from three independent evaluations 989 analysed in triplicate. Panel A, visual and tactual perception attributes; panel B, taste attributes; panel 990 991 C, smell perception, chewing attributes and overall aroma.

992

Figure 1



Α



Factor 1:66.75%

В

Figure 2



Figure S1. Sprouting process for lentil grains. The optimal germination time was established based on the length of rootlets (ca. 3/4 of the medium seed length), before the seedling development.



Figure S2. Concentration, expressed as mg Kg⁻¹ dough, of free (A) and bound (B) phenolic compounds in lentil and sprouted lentil doughs estimated by UPLC-PDA-ESI-QTOF before (0 h) and after fermentation for 24 h at 20 °C. L-SLA4 EPS NEG, lentil dough fermented by *Weissella confusa* SLA4 without sucrose addition; L-SLA4 EPS POS, lentil dough fermented by *W. confusa* SLA4 with sucrose addition; SL-SLA4 EPS NEG, sprouted lentil dough fermented by *Weissella confusa* SLA4 without sucrose addition; SL-SLA4 EPS POS, sprouted lentil dough fermented by *Weissella confusa* SLA4 without sucrose addition; SL-SLA4 EPS POS, sprouted lentil dough fermented by *W. confusa* SLA4 without sucrose addition; SL-SLA4 EPS POS, sprouted lentil dough fermented by *W. confusa* SLA4 with sucrose addition. All doughs had DY of 500. Data are the mean of three independent fermentations twice analyzed.





A

Table 1. Cell count of total mesophilic aerobic bacteria, Enterobacteriaceae, lactic acid bacteria, and yeast (log cfu g⁻¹), in sourdoughs (DY 500)

	L	SL	L-SLA4	L-SLA4	SL-SLA4	SL-SLA4
Microbial group			EPS NEG	EPS POS	EPS NEG	EPS POS
				0 h		
Total mesophilic aerobic bacteria	$4.52\pm0.009^{\text{c}}$	5.81 ± 0.145^{ab}	5.01 ± 0.192^{b}	5.03 ± 0.162^{b}	6.45 ± 0.081^a	6.51 ± 0.119^{a}
Enterobacteriaceae	2.49 ± 0.063^{b}	5.04 ± 0.152^a	2.12 ± 0.044^{b}	$2.16\pm0.051^{\text{b}}$	5.14 ± 0.147^{a}	5.25 ± 0.059^a
Lactic acid bacteria	$3.62 \pm 0.110^{\circ}$	4.92 ± 0.091^{b}	6.87 ± 0.113^{a}	6.55 ± 0.242^a	6.66 ± 0.232^a	6.72 ± 0.171^{a}
Yeast	2.31 ± 0.095^{b}	2.84 ± 0.042^{a}	2.47 ± 0.054^{ab}	2.57 ± 0.028^{ab}	2.76 ± 0.068^{a}	2.88 ± 0.044^a
			,	24 h		
Total mesophilic aerobic bacteria	8.96 ± 0.158^{b}	8.87 ± 0.342^{b}	9.11 ± 0.158^a	9.18 ± 0.259^{a}	9.15 ± 0.231^{a}	9.22 ± 0.263^a
Enterobacteriaceae	6.47 ± 0.123^{b}	7.06 ± 0.053^{a}	3.59 ± 0.094^{d}	3.65 ± 0.082^d	5.48 ± 0.074^{bc}	5.08 ± 0.054^{c}
Lactic acid bacteria	8.92 ± 0.213^{b}	8.89 ± 0.143^{b}	9.28 ± 0.221^{ab}	9.45 ± 0.151^a	9.61 ± 0.082^{a}	9.32 ± 0.311^{ab}
Yeast	2.56 ± 0.021^b	3.05 ± 0.081^a	<1 ^c	<1 ^c	<1 ^c	<1 ^c

before (0 h) and after fermentation for 24 h at 20 °C.

L, lentil sourdough fermented spontaneously; SL, sprouted lentil dough fermented spontaneously; L-SLA4 EPS NEG, lentil dough fermented by

Weissella confusa SLA4 without sucrose addition; L-SLA4 EPS POS, lentil dough fermented by *W. confusa* SLA4 with sucrose addition; SL-SLA4 EPS NEG, sprouted lentil dough fermented by *Weissella confusa* SLA4 without sucrose addition; SL-SLA4 EPS POS, sprouted lentil dough fermented by *W. confusa* SLA4 with sucrose addition.

Data are the mean of three independent fermentations twice analyzed.

^{a-d} Means within a row with different superscript letters are significantly different (P < 0.05).

Table 2. Dextran, sugars, acidity (TTA), concentration of organic acids, peptides, total free amino acid (TFFA), total phenols and radical

Chemical parameter	L-SLA4	L-SLA4	SL-SLA4	SL-SLA4
	EPS NEG ¹	EPS POS ²	EPS NEG ³	EPS POS ⁴
			0 h	
Dextran (% flour weight)	nd	nd	nd	nd
Sugars (% flour weight)				
Glucose	nd	nd	4.01 ± 0.11^{a}	$4.11 \pm 0.0^{\mathrm{a}}$
Sucrose	$1.77\pm0.02^{\rm b}$	23.48 ± 0.70^{a}	$1.95\pm0.01^{\mathrm{b}}$	$23.26\pm0.44^{\rm a}$
Fructose	0.17 ± 0.0^{b}	0.22 ± 0.09^{b}	$0.28\pm0.01^{\text{a}}$	0.31 ± 0.05^a
Melibiose	Nd	$0.12\pm0.09^{\rm b}$	nd	0.30 ± 0.09^{a}
Raffinose	$1.34\pm0.0^{\rm a}$	$1.36 \pm 0.0^{\mathrm{a}}$	$0.21\pm0.01^{\mathrm{b}}$	$0.26\pm0.04^{\rm b}$
Stachyose	$1.87\pm0.08^{\rm a}$	$1.90\pm0.2^{\mathrm{a}}$	$0.18\pm0.01^{ m b}$	0.11 ± 0.11^{b}
Verbascose	0.65 ± 0.01^{a}	0.77 ± 0.03^{a}	$0.06\pm0.04^{\rm b}$	$0.11 \pm 0.0^{\mathrm{b}}$
Maltose	nd	Nd	nd	nd
Galactose	nd	$0.06 \pm 0.0^{\mathrm{a}}$	nd	$0.08\pm0.0^{\mathrm{a}}$
Starch	$45.08\pm0.75^{\mathrm{a}}$	$45.48\pm0.44^{\rm a}$	$28.30 \pm 1.04^{\text{b}}$	29.04 ± 0.66^b
Resistant Starch	2.32 ± 0.01^{a}	$2.41 \pm 0.08^{\mathrm{a}}$	$1.58\pm0.02^{\rm b}$	1.61 ± 0.03^{b}
Non-Resistant Starch	42.76 ± 1.14^a	43.07 ± 0.85^a	$26.72\pm0.55^{\mathrm{b}}$	27.42 ± 0.26^{b}
TTA	2.20 ± 0.045^{b}	2.10 ± 0.052^{b}	$3.70\pm0.083^{\rm a}$	3.81 ± 0.085^a
Lactic acid (mmol Kg ⁻¹)	Nd	nd	nd	nd
Acetic acid (mmol Kg ⁻¹)	Nd	nd	nd	nd
FQ	Nd	nd	nd	nd
Peptides (g Kg ⁻¹ dough)	10.3 ± 0.2^{b}	$9.8\pm0.3^{\mathrm{b}}$	15.5 ± 0.56^{a}	16.7 ± 0.79^{a}
TFAA (mg Kg ⁻¹ dough)	754.8 ± 18.0^{b}	766.7 ± 38.3^{b}	1444.4 ± 68.0^{a}	1462.3 ± 73.1^{a}
Total phenols (mmol GA Kg ⁻¹	2.64 ± 0.01^{b}	2.84 ± 0.22^{b}	$5.67\pm0.09^{\rm a}$	$5.81 \pm 0.10^{\mathrm{a}}$
dough)				
Radical scavenging activity (%)	83.3 ± 1.03^{b}	82.0 ± 1.83^{b}	$86.2\pm2.5^{\mathrm{a}}$	$87.0 \pm 3.94^{\rm a}$
			24 h	
Dextran (% flour weight)	$0.88\pm0.01^{\text{b}}$	9.20 ± 0.21^{a}	1.17 ± 0.04^{b}	$9.70\pm0.38^{\rm a}$
Sugars (% flour weight)				
Glucose	nd	nd	Nd	Nd

scavenging activity in sourdoughs¹ (DY 500) before (0 h) and after fermentation for 24 h at 20 °C.

Chemical parameter	L-SLA4	L-SLA4	SL-SLA4	SL-SLA4
-	EPS NEG ¹	EPS POS ²	EPS NEG ³	EPS POS ⁴
Sucrose	nd	0.04 ± 0.0	nd	nd
Fructose	nd	$9.20\pm0.20^{\mathrm{a}}$	$0.01 \pm 0.0^{\mathrm{b}}$	8.80 ± 0.3^a
Melibiose	nd	nd	nd	Nd
Raffinose	0.54 ± 0.04^{a}	0.46 ± 0.06^{a}	0.11 ± 0.02^{b}	0.15 ± 0.1^{b}
Stachyose	0.99 ± 0.01^{a}	0.95 ± 0.02^{a}	$0.06 \pm 0.02^{\mathrm{b}}$	$0.06\pm0.01^{\text{b}}$
Verbascose	0.26 ± 0.02^{a}	$0.22 \pm 0.2^{\mathrm{a}}$	nd	Nd
Maltose	nd	nd	nd	nd
Galactose	0.42 ± 0.03^{a}	0.38 ± 0.03^{a}	0.16 ± 0.02^{b}	$0.17\pm0.01^{\mathrm{b}}$
Starch	25.42 ± 1.15^{ab}	29.86 ± 0.12^a	17.98 ± 1.69^{b}	28.86 ± 0.24^a
Resistant Starch	3.66 ± 0.01^{a}	3.81 ± 0.13^{a}	2.25 ± 0.09^{b}	$2.83\pm0.02^{\text{b}}$
Non-Resistant Starch	21.76 ± 0.85^{b}	26.05 ± 1.01^{a}	$15.73 \pm 0.65^{\circ}$	26.03 ± 0.55^a
TTA	6.61 ± 0.156^{bc}	7.25 ± 0.182^{b}	9.91 ± 0.148^{a}	10.22 ± 0.155^a
delta TTA	4.40	5.10	6.20	6.40
Lactic acid (mmol Kg ⁻¹)	$12.10 \pm 0.5^{\circ}$	16.20 ± 0.8^{b}	17.10 ± 0.8^{b}	$20.10\pm1.0^{\rm a}$
Acetic acid (mmol Kg ⁻¹)	$3.60\pm0.07^{\mathrm{b}}$	3.40 ± 0.10^{b}	5.50 ± 0.21^{a}	$5.20\pm0.15^{\rm a}$
FQ	3.36 ± 0.12^{bc}	4.76 ± 0.11^{a}	$3.11 \pm 0.09^{\circ}$	3.87 ± 0.06^{b}
Peptides (g Kg ⁻¹ dough)	12.7 ± 0.26^{b}	12.9 ± 0.43^{b}	22.1 ± 0.34^{a}	22.6 ± 0.62^a
TFAA (mg Kg ⁻¹ dough)	$1447.3 \pm 33.2^{\circ}$	$1424.9 \pm 31.2^{\circ}$	2486.0 ± 61.3^{b}	2644.1 ± 54.1^{a}
Total phenols (mmol GA Kg ⁻¹	$5.71\pm0.08^{\mathrm{b}}$	5.62 ± 0.24^{b}	6.96 ± 0.04^{a}	$7.03\pm0.05^{\rm a}$
dough)				
Radical scavenging activity (%)	90.1 ± 2.92^{b}	89.1 ± 2.54^{b}	91.2 ± 2.57^{a}	93.2 ± 3.13^{a}

¹L-SLA4 EPS NEG, lentil dough fermented by *Weissella confusa* SLA4 without sucrose addition; L-SLA4 EPS POS, lentil dough fermented by *W*.

confusa SLA4 with sucrose addition; SL-SLA4 EPS NEG, sprouted lentil dough fermented by Weissella confusa SLA4 without sucrose addition;

SL-SLA4 EPS POS, sprouted lentil dough fermented by W. confusa SLA4 with sucrose addition.

FQ, fermentation quotient, molar ratio between lactic and acetic acids.

GA, gallic acid equivalent.

nd, not detected.

Data are the mean of three independent fermentations twice analyzed.

^{a-d} Means within a row with different superscript letters are significantly different (P < 0.05).

Table 3. Characteristics of breads containing wheat flour added of lentil flour fermented by *Weissella confusa* SLA4 sourdough (LSWB EPS NEG), wheat flour added of lentil flour fermented by *W. confusa* SLA4 - dextran containing sourdough (LSWB EPS POS), wheat flour added of sprouted lentil flour fermented by *W. confusa* SLA4 sourdough (SLSWB EPS NEG), wheat flour added of sprouted lentil flour fermented by *W. confusa* SLA4 sourdough (SLSWB EPS NEG), wheat flour added of sprouted lentil flour fermented by *W. confusa* SLA4 sourdough (SLSWB EPS NEG), wheat flour added of sprouted lentil flour fermented by *W. confusa* SLA4 - dextran containing sourdough (SLSWB EPS POS). All strains were inoculated at ca. 7 log cfu g⁻¹ and sourdough fermented at 20 °C for 24 h. Wheat flour bread started with baker's yeast (CWB) was used as control. Doughs for bread making had DY 162.

Samples	CWB	LSWB	LSWB	SLSWB	SLSWB
_		EPS NEG	EPS POS	EPS NEG	EPS POS
Structural characteristics					
Baking loss (%)	$8.76\pm0.33^{\text{b}}$	10.24 ± 0.41^a	8.04 ± 0.33^{bc}	9.66 ± 0.23^{ab}	$7.74 \pm 0.23^{\circ}$
Total volume (cm ³)	$1233 \pm 104^{\circ}$	1360 ± 52^{b}	1566 ± 57^{a}	1316 ± 76^{b}	1533 ± 57^{a}
Specific volume	2.7 ± 0.31^{b}	3.0 ± 0.20^{ab}	3.4 ± 0.13^{a}	$2.9\pm0.11^{\text{b}}$	3.3 ± 0.20^{a}
Hardness/ day 1 (g)	1377 ± 47^{a}	1104 ± 29^{b}	919 ± 14^{c}	1442 ± 94^{a}	$1087 \pm 51^{\mathrm{b}}$
Hardness/ day 7 (g)	2610 ± 250^{b}	2698 ± 166^{b}	2056 ± 335^{d}	3312 ± 87^{a}	$2263 \pm 330^{\circ}$
Springness (%)	0.89 ± 0.01^{b}	0.91 ± 0.01^{ab}	0.93 ± 0.01^{a}	$0.89\pm0.01^{\text{b}}$	0.91 ± 0.01^{ab}
Cohesivness (%)	0.77 ± 0.01^{ab}	0.76 ± 0.01^{ab}	0.79 ± 0.01^{a}	0.74 ± 0.01^{b}	0.78 ± 0.01^{a}
Resilience (%)	0.40 ± 0.01^{ab}	0.39 ± 0.01^{ab}	0.45 ± 0.00^{a}	0.37 ± 0.00^{b}	0.41 ± 0.02^{ab}
Staling rate (g day ⁻¹)	$205.5 \pm 11^{\circ}$	265.7 ± 15^{b}	189.5 ± 9.5^{d}	311.7 ± 15^{a}	196.0 ± 9^{cd}
Image analysis					
Black pixel area %	22.5 ± 0.03^{b}	57.2 ± 0.05^a	56.5 ± 0.04^a	56.3 ± 0.07^a	56.2 ± 0.01^{a}
Chemical characteristics					
pH (bread crumb)	5.92 ± 0.111^a	4.87 ± 0.19^{b}	4.91 ± 0.24^{b}	4.83 ± 0.14^{b}	4.88 ± 0.18^{b}
TTA (bread crumb)	2.20 ± 0.04^{c}	5.5 ± 0.15^{b}	5.1 ± 0.22^{b}	6.1 ± 0.28^{ab}	6.9 ± 0.33^a
Nutritional characteristics					
TDF (%)	$2.12\pm0.082^{\rm c}$	$3.37{\pm}0.056^{ab}$	3.68 ± 0.097^a	3.17 ± 0.028^b	3.63 ± 0.072^a
IDF(%)	$1.09 \pm 0.018^{\circ}$	2.20 ± 0.043^a	2.21 ± 0.066^{a}	2.14 ± 0.077^{b}	2.16 ± 0.021^{b}
SDF(%)	1.04 ± 0.061^{b}	1.17 ± 0.06^{ab}	1.47 ± 0.051^{a}	1.03 ± 0.014^{b}	1.44 ± 0.077^{a}
HI	49.6 ± 0.122^{cd}	47.4 ± 0.017^{d}	58.9 ± 0.031^{b}	53.9 ± 0.032^{c}	67.4 ± 0.023^a
GI	66.9 ± 0.074^{cd}	65.7 ± 0.089^d	72.1 ± 0.021^{b}	69.3 ± 0.012^{c}	76.7 ± 0.076^a

Baking loss = (dough weight–bread weight) * 100/dough weight)

Staling rate = (hardness (day 7-day 1)/days of storage)

TDF, total dietary fibers.

IDF, Insoluble dietary fibers.

SDF, soluble dietary fibers.

HI, hydrolysis index.

GI, predicted glycemic index.

nd, not detected.

Data are the mean of three independent baking tests twice analyzed.

^{a-f} Means within a row with different superscript letters are significantly different (P < 0.05).

Table 4. Concentration of volatile organic compounds (VOCs) (peak area) normalized with the internal standard area, quantified in terms of arbitrary area units and identified in breads containing wheat flour added of lentil flour fermented by *Weissella confusa* SLA4 sourdough (LSWB EPS NEG), wheat flour added of lentil flour fermented by *W. confusa* SLA4 - dextran containing sourdough (LSWB EPS POS), wheat flour added of sprouted lentil flour fermented by *W. confusa* SLA4 sourdough (SLSWB EPS NEG), wheat flour added of sprouted lentil flour fermented by *W. confusa* SLA4 sourdough (SLSWB EPS NEG), wheat flour added of sprouted lentil flour fermented by *W. confusa* SLA4 sourdough (SLSWB EPS NEG), wheat flour added of sprouted lentil flour fermented by *W. confusa* SLA4 sourdough (SLSWB EPS NEG), wheat flour added of sprouted lentil flour fermented by *W. confusa* SLA4 - dextran containing sourdough (SLSWB EPS POS). All strains were inoculated at ca. 7 log cfu g⁻¹ and sourdough fermented at 20 °C for 24 h. Wheat flour bread started with baker's yeast (CWB) was used as control. Doughs for bread making had DY 162.

Compound	Odour	CWB	LSWB EPS NEG	LSWB EPS POS	SLSWB EPS NEG	SLSWB EPS POS
Furans						
furan,2-pentyl-	Butter, green bean, floral	0.10 ± 0.024^{b}	0.27 ± 0.019^{ab}	0.43 ± 0.008^{a}	0.33 ± 0.048^{ab}	0.24 ± 0.011^{ab}
acetylfuran	Smoky, roasty	nd	0.28 ± 0.014^{b}	nd	0.44 ± 0.04^{ab}	0.96 ± 0.009^{a}
2(3h) -furanone, dihydro-5- pentyl-	nf	nd	nd	nd	nd	$0.01\pm0.011^{\text{a}}$
4-hydroxy-2,5-dimethyl- 3(2h) -furanone	Caramel, strawberry	nd	nd	nd	nd	$0.01\pm0.010^{\text{a}}$
2-furancarboxaldehyde,5- methyl	nf	0.11 ± 0.015^{c}	0.26 ± 0.007^{b}	0.19 ± 0.038^{bc}	0.23 ± 0.039^{b}	1.3 ± 0.021^{a}
Total		0.21 ± 0.02	0.81 ± 0.03	0.62 ± 0.00	1.00 ± 0.00	2.52 ± 0.29
Percentage (%)		2.69 ± 0.051^{c}	4.89 ± 0.121^{b}	5.64 ± 0.105^{b}	8.76 ± 0.167^a	14.29 ± 0.271^a
Pyrazines						
2-methylpyrazine	Roasted, burnt, sweet	0.23 ± 0.004^{c}	0.38 ± 0.002^{bc}	0.22 ± 0.030^{c}	0.45 ± 0.034^{b}	0.74 ± 0.011^a
2,5-dimethylpyrazine	Crust-like, popcorn	0.10 ± 0.01^{c}	0.22 ± 0.003^{b}	0.15 ± 0.033^{bc}	$0.22\pm0.01^{\text{b}}$	0.41 ± 0.003^{a}
2,6-dimethylpyrazine	Roasted	0.07 ± 0^{c}	0.18 ± 0.005^{b}	0.11 ± 0.031^{bc}	$0.17\pm0.01^{\text{b}}$	0.37 ± 0.001^a
2-ethylpyrazine	Popcorn, nutty	0.10 ± 0.006^{b}	0.18 ± 0.002^{ab}	0.11 ± 0.034^{b}	0.18 ± 0.01^{ab}	0.23 ± 0.007^a
2,3-dimethylpyrazine	Popcorn, roasted	0.05 ± 0.004^{b}	0.10 ± 0.00^{ab}	0.04 ± 0.018^{b}	0.08 ± 0.005^{ab}	0.14 ± 0.002^{a}
2-ethyl-6-methylpyrazine	Nutty	0.05 ± 0.007^{c}	0.17 ± 0.009^{b}	0.05 ± 0.008^{c}	0.19 ± 0.011^{b}	0.29 ± 0.013^a

Compound	Odour	CWB	LSWB EPS NEG	LSWB EPS POS	SLSWB EPS NEG	SLSWB EPS POS
2-ethyl-5-methylpyrazine	Baked	$0.02\pm0.017^{\rm c}$	0.09 ± 0.007^{bc}	$0.03\pm0.007^{\rm c}$	0.1 ± 0.007^{b}	0.16 ± 0.026^a
2-ethyl-3-methylpyrazine	Nutty, roasted, sweety	$0.07\pm0.011^{\rm c}$	0.17 ± 0.017^{b}	0.05 ± 0.009^{c}	0.16 ± 0.008^{b}	0.32 ± 0.008^a
3-ethyl-2,5-dimethylpyrazine	Baked, potato-like, earthy	0.03 ± 0.001^{c}	0.13 ± 0.005^{b}	0.04 ± 0.007^{c}	0.12 ± 0.006^{b}	0.25 ± 0.017^a
2-ethenyl-6-methylpyrazine	nf	nd	nd	nd	0.01 ± 0.012^{a}	0.03 ± 0^{a}
2-isobutyl-3-methylpyrazine	Caramellic	nd	nd	nd	0.03 ± 0.007^{b}	0.06 ± 0.002^{a}
3,5-Diethyl-2-methylpyrazine	Roasted, peanut butter	nd	0.03 ± 0.003^{b}	nd	0.03 ± 0.001^{b}	0.05 ± 0.004^a
2-methyl-6-(1-propenyl) - pyrazine	nf	nd	nd	nd	0.04 ± 0.003^a	0.04 ± 0.009^{a}
2-butyl-3-methylpyrazine	Caramellic	nd	0.03 ± 0.013^{ab}	0.01 ± 0.011^{b}	0.05 ± 0.002^a	0.04 ± 0.003^{a}
2,6-diethylpyrazine	Green, spicy	0.03 ± 0.001^{b}	0.09 ± 0.002^{ab}	0.03 ± 0.005^{b}	0.08 ± 0.007^{ab}	0.1 ± 0.001^{a}
Total		0.72 ± 0.01	1.68 ± 0.06	0.81 ± 0.18	1.83 ± 0.12	3.13 ± 0.04
Percentage (%)		9.21 ± 0.274^{c}	10.14 ± 0.301^{c}	7.37 ± 0.219^{d}	16.04 ± 0.402^{b}	17.75 ± 0.503^a
Pyridines						
2-acetylpyridine	Biscuit, cracker-like, crust- like, roasted	nd	nd	nd	nd	0.02 ± 0.023
pyrazinamide	nf	0.02 ± 0.003^{b}	0.06 ± 0.003^{a}	0.02 ± 0.004^{b}	0.04 ± 0.003^{ab}	0.07 ± 0.003^a
6-Acetyltetrahydropyridine	Roasty	0.01 ± 0.008^{b}	0.02 ± 0^{ab}	nd	nd	0.03 ± 0.001^a
pyridine, 1-acetyl-1,2,3,4-tet	Vegetable	nd	nd	nd	0.01 ± 0.009^{b}	0.04 ± 0.008^a
Total	-	0.03 ± 0.01	0.02 ± 0.00	0.00 ± 0.00	0.04 ± 0.01	0.09 ± 0.02
Percentage (%)		0.13 ± 0.015^{b}	0.12 ± 0.008^{ab}	$0.00\pm0.007^{\rm c}$	0.09 ± 0.003^{b}	0.51 ± 0.016^a
Pyrrolines						
1h-pyrrole,1-(2- furanylmethyl)	Corn chip, roasty, crust-like, sweet, cereal, popcorn-like, bread	nd	0.04 ± 0.004^{b}	$0.01\pm0.009^{\text{c}}$	0.04 ± 0.005^{b}	0.10 ± 0.010^{a}
2-acetyl-1-pyrroline	Cracker-like	0.01 ± 0.010^{b}	0.07 ± 0.005^{ab}	0.02 ± 0.006^{b}	0.05 ± 0.008^{ab}	0.09 ± 0.008^a
1h-pyrrole-2-carboxaldehyde	Warmy-fruity,caramellic- sweet	nd	nd	nd	nd	0.01 ± 0.012
3-Hydroxy-2-methyl-4- pyrone (maltol)	Warmy, fruity, caramellic- sweet	0.06 ± 0.014^{c}	$0.15\pm0.024^{\text{b}}$	$0.04\pm0.014^{\rm c}$	$0.13\pm0.013^{\text{b}}$	0.20 ± 0.033^{a}
Total		0.07 ± 0.01	0.26 ± 0.00	0.07 ± 0.01	0.22 ± 0.02	0.40 ± 0.03

Compound	Odour	CWB	LSWB EPS NEG	LSWB EPS POS	SLSWB EPS NEG	SLSWB EPS POS
Percentage (%)		0.90 ± 0.013^{bc}	1.57 ± 0.022^{b}	0.64 ± 0.015^c	1.93 ± 0.007^{ab}	2.27 ± 0.028^a
Alcohols						
ethanol	Alcohlic	2.18 ± 0.096^{b}	6.02 ± 0.286^a	2.33 ± 0.286^b	2.43 ± 0.043^{b}	2.69 ± 0.236^{b}
1-propanol,2-methyl-	Glue, alcoholic, wine-like, malty	0.08 ± 0.005^a	0.07 ± 0.003^a	nd	0.02 ± 0.023^{b}	0.06 ± 0^{a}
1-butanol,3-methyl-	Balsamic, alcoholic, malty	0.65 ± 0.014^{a}	0.63 ± 0.049^a	0.38 ± 0.066^{c}	0.47 ± 0.012^{b}	0.48 ± 0.007^{b}
1-hexanol	Green grass, flowery, woody, mild, sweet	0.22 ± 0.014^{c}	1.88 ± 0.010^a	1.96 ± 0.266^a	0.65 ± 0.008^{b}	0.53 ± 0.040^{b}
1-heptanol	Green	nd	0.05 ± 0.001^{ab}	0.05 ± 0.008^{ab}	0.03 ± 0^{b}	0.07 ± 0.001^{a}
2-ethylhexanol	Green, vegetable	0.32 ± 0.026^{b}	nd	0.41 ± 0.066^a	nd	nd
furfurylalcohol	Burnt, warmy oil,	0.33 ± 0.045^a	0.33 ± 0.013^a	0.11 ± 0.02^{c}	0.19 ± 0.02^{b}	0.36 ± 0.018^{a}
3-nonen-1-ol, (z)-	Waxy	0.04 ± 0.006^{c}	0.48 ± 0.008^a	0.48 ± 0.071^a	0.28 ± 0.003^{b}	$0.27\pm0.012^{\text{b}}$
2furanmethanol,5-methyl-	Honey, sweet	nd	nd	nd	0.02 ± 0^{b}	0.04 ± 0.002^a
benzylalcohol	Pleasant, aromatic	0.44 ± 0.135^{b}	0.88 ± 0.119^{a}	1 ± 0.066^{a}	0.44 ± 0.113^{b}	0.95 ± 0.218^{a}
phenylethylalcohol	Rose-honey-like, wilted rose	$0.11{\pm}0.006^a$	0.09 ± 0.002^{a}	0.06 ± 0.01^{b}	0.1 ± 0^{a}	0.12 ± 0.002^{a}
2,4-bis(1,1-dimethylethyl) phenol	Weak aromatic	nd	0.02 ± 0.002^b	$0.01\pm0.01^{\text{b}}$	nd	0.04 ± 0.005^{a}
1-nonanol	Citrus	0.03 ± 0.003^{c}	0.14 ± 0.004^{b}	0.11 ± 0.016^{bc}	0.18 ± 0.002^{a}	0.19 ± 0.012^{a}
Total		4.40 ± 0.13	10.59 ± 0.22	6.90 ± 0.92	4.86 ± 0.01	5.80 ± 0.03
Percentage (%)		56.27 ± 1.28^{ab}	63.95 ± 1.18^a	62.78 ± 0.57^{a}	42.16 ± 0.74^{b}	$32,9 \pm 1.19^{b}$
Aldehydes						
2-methylpropanal	Malty	0.1 ± 0.026^{b}	0.15 ± 0.012^{bc}	0.11 ± 0.02^{b}	0.17 ± 0.007^{a}	0.18 ± 0.018^{a}
2-methylbutanal	Almond, malty	0.03 ± 0.001^{c}	0.07 ± 0.002^{b}	0.06 ± 0.009^{b}	0.18 ± 0.006^{a}	0.14 ± 0.008^{ab}
3-methylbutanal	Malty, roasty cucumber-like	0.03 ± 0.003^{c}	0.14 ± 0.005^{bc}	0.14 ± 0.029^{bc}	0.26 ± 0.015^a	0.18 ± 0.013^{b}
hexanal	Green, grassy, tallow	0.49 ± 0.017^a	0.45 ± 0.004^{ab}	0.44 ± 0.077^{ab}	0.4 ± 0.003^{ab}	0.33 ± 0.031^{b}
octanal	Citrus, flowery	0.06 ± 0.029^{c}	0.14 ± 0.005^{b}	0.06 ± 0.059^c	0.15 ± 0.022^{b}	0.17 ± 0.002^a
nonanal	Citrus,soapy	$0.43\pm0.036^{\rm a}$	0.37 ± 0.024^{ab}	0.25 ± 0.026^{b}	$0.22\pm0.001^{\text{b}}$	$0.21\pm0.008^{\text{b}}$
furfural	Almond, soil, burnt, roasted, sweet, toasted	0.47 ± 0.020^b	0.53 ± 0.002^{b}	0.68 ± 0.108^{b}	0.47 ± 0.034^{b}	2.57 ± 0.010^{a}

Compound	Odour	CWB	LSWB EPS NEG	LSWB EPS POS	SLSWB EPS NEG	SLSWB EPS POS
benzaldehyde	Almond, caramel	0.12 ± 0.009^{c}	0.22 ± 0.006^{b}	0.16±0.026 ^{bc}	0.26 ± 0.012^{b}	0.34±0.004 ^a
2-nonenal	Fatty, tallowy, green	0.07 ± 0.006^{c}	0.14 ± 0^{ab}	0.11 ± 0.018^{b}	0.12 ± 0.007^b	0.17 ± 0.001^{a}
acetaldehyde	Fruity	0.02 ± 0.003^{d}	0.11 ± 0.005^{c}	0.04 ± 0.016^d	0.17 ± 0.006^{b}	0.26 ± 0.002^a
2,4-decadienal	Deep, fat, fried, waxy	nd	0.02 ± 0^{a}	0.02 ± 0.002^a	nd	0.02 ± 0.001^{a}
benzeneacetaldehyde	flowery, honey-like	nd	nd	nd	nd	0.02 ± 0.001
Total		1.85 ± 0.03	$2.34{\pm}0.03$	2.07 ± 0.40	2.40 ± 0.11	4.59 ± 0.02
Percentage (%)		23.27 ± 0.92^{ab}	14.13 ± 0.56^{c}	18.84 ± 0.248^{bc}	21.03 ± 0.132^{b}	26.4 ± 0.428^a
Ketones						
acetoin	Butterscotch, butter, yogurt, cream	0.14 ± 0.005^a	0.09 ± 0.001^{b}	0.07 ± 0.014^{bc}	0.04 ± 0.003^{c}	0.07 ± 0.003^{bc}
2-nonanone	Fruity	0.09 ± 0.011^b	0.19 ± 0.013^a	0.13 ± 0.023^{b}	0.11 ± 0.002^{b}	0.1 ± 0.013^{b}
Total		0.23 ± 0.01	0.28 ± 0.01	0.20 ± 0.03	0.15 ± 0.00	0.17 ± 0.01
Percentage (%)		2.91 ± 0.106^a	1.69 ± 0.016^{b}	1.82 ± 0.022^{b}	1.30 ± 0.012^{bc}	0.95 ± 0.018^{c}
Esters						
ethylacetate	Sweet, fruity, pineapple	0.11 ± 0.003^a	0.10 ± 0.005^{ab}	0.08 ± 0.011^{b}	0.11 ± 0.001^{a}	0.1 ± 0.006^{ab}
decanoicacid, ethylester	nf	nd	0.04 ± 0.001^{a}	0.02 ± 0.019^b	nd	nd
Total		0.11 ± 0.00	0.10 ± 0.00	0.08 ± 0.01	0.11 ± 0.00	0.10 ± 0.00
Percentage (%)		1.41 ± 0.002^a	$0.85\pm0.0^{\rm c}$	0.91 ± 0.01^{bc}	0.96 ± 0.0^{b}	$0.57\pm0.0^{\rm c}$
Hydrocarbons						
d-limonene	Citrus	0.08 ± 0.032^{ab}	0.06 ± 0.003^{b}	0.05 ± 0.048^b	0.09 ± 0.005^a	0.08 ± 0.002^{ab}
styrene	Pungent	nd	nd	nd	0.19 ± 0.019^a	0.03 ± 0.002^{b}
nonadecane	nf	nd	0.03 ± 0.001^{a}	nd	nd	nd
pentadecane	nf	nd	nd	nd	nd	0.03 ± 0.004
4h-pyran-4-one,2,3-dihydro- 3,5	Caramelised	nd	0.03 ± 0.011^{a}	0.01 ± 0.015^{b}	nd	0.03 ± 0.003^a
Total		0.11 ± 0.03	0.21 ± 0.00	0.09 ± 0.06	0.36 ± 0.03	0.27 ± 0.00
Percentage (%)		1.02 ± 0.015^{bc}	$0.54{\pm}0.012^{bc}$	0.45 ± 0.011^{c}	2.45 ± 0.114^a	$0{,}79\pm0.021^{b}$
Organic Acids						

Compound	Odour	CWB	LSWB EPS NEG	LSWB EPS POS	SLSWB EPS NEG	SLSWB EPS POS
Acetic acid	Sour, acid, pungent	0.03 ± 0.004^{c}	0.13 ± 0.009^{b}	0.07 ± 0.013^{c}	0.22 ± 0.019^{ab}	0.27 ± 0.004^{a}
Butanoic acid	Sweaty, rancid	nd	0.04 ± 0.002^{b}	nd	nd	0.14 ± 0.02^{a}
Hexanoic acid	Sweaty, cheesy, fatty, goat-like	0.06 ± 0.005^{d}	0.13 ± 0.003^{c}	0.07 ± 0.015^{d}	0.19 ± 0.008^{b}	0.23 ± 0.005^a
propanoic acid, 2-methyl-	Sweaty, butter, fatty, sour, rancid	0.02 ± 0.001^{b}	0.05 ± 0.001^a	0.03 ± 0.005^{ab}	0.02 ± 0^{b}	0.03 ± 0.006^{ab}
Octanoic acid	Cheese, fatty, sweaty, soapy	0.04 ± 0.006^{b}	nd	nd	0.1 ± 0.01^{a}	0.02 ± 0.002^{ab}
n-decanoic acid	Rancid, fatty, citrus, sweaty, cheesy	0.02 ± 0.001^{b}	nd	nd	0.07 ± 0.014^{a}	nd
Total		0.17 ± 0.02	0.35 ± 0.03	0.17 ± 0.04	0.60 ± 0.06	0.69 ± 0.17
Percentage (%)		2.17 ± 0.076^{bc}	2.11 ± 0.036^{bc}	$1.55\pm0.012^{\rm c}$	5.26 ± 0.108^{a}	3.91 ± 0.054^b
1 . 1 1						

nd, not detected.

nf, not found.

Data are the mean values calculated as ratio peak area/total peak area percent of three independent baking tests analyze in triplicate.

^{a-d} Values in the same row with different superscript letters differ significantly (P < 0.05).

Normalized data with the internal standard are reported.

Strain	Source	EPS production		
Pediococcus pentosaceus SWA2	Sprouted wheat flour (DiSSPA ² collection)	-		
P. pentosaceus SWA8	Sprouted wheat flour (DiSSPA)	-		
Enterococcus faecium SBC2	Sprouted barley flour (DiSSPA)	-		
Lactobacillus fermentum SBB10	Sprouted barley flour (DiSSPA)	-		
L. fermentum QB6	Raw quinoa flour (DiSSPA)	-		
Weissella confusa SLA4	Sprouted lentil flour (DiSSPA)	+		
Weissella paramesenteroides SLA5	Sprouted lentil flour (DiSSPA)	+		
Lactobacillus rossiae LB5-CONV	Wheat flour (DiSSPA)	-		
L. plantarum 1A7-CONV	Wheat flour (DiSSPA)	-		
Lactobacillus sanfransciscensis DE9-CONV	Wheat flour (DiSSPA)	-		
Leuconostoc citreum PRO-17	Wheat flour sourdough (DiSSPA)	-		
P. pentosaceus OA1	Wheat flour sourdough (DiSSPA)	-		
P. pentosaceus S3N3	Wheat flour sourdough (DiSSPA)	-		
L. pseudomesenteroides DSM 20193	Cane juice (DSMZ ³ collection)	+		
Weissella confusa DSM 20194	Soured carrot mash (DSMZ)	+		
¹ L. plantarum DPPMAB24W	Cheese (DiSSPA)	-		

Table S1. Results for screening of EPS producing strains grown on agar plates containing modified MRS medium added of sucrose (20 g L⁻¹).

¹L. plantarum DPPMAB24W was used as non EPS producing control; ²DiSSPA, Department of Soil, Plant and Food Sciences, University of Bari Aldo Moro, Bari, Italy; ³DSMZ, Leibniz Institute DSMZ-German Collection of Microorganisms and Cell Cultures GmbH, Braunschweig, Germany.

Table S2. Recipes for different wheat bread doughs. LSWB EPS NEG, wheat bread added of lentil flour fermented by *Weissella confusa* SLA4 sourdough; LSWB EPS POS, wheat bread added of lentil flour fermented by *W. confusa* SLA4 - dextran containing sourdough; SLSWB EPS NEG, wheat bread added of sprouted lentil flour fermented by *W. confusa* SLA4 sourdough; SLSWB EPS POS, wheat bread added of sprouted lentil flour fermented by *W. confusa* SLA4 sourdough; SLSWB EPS POS, wheat bread added of sprouted lentil flour fermented by *W. confusa* SLA4 sourdough; SLSWB EPS POS, wheat bread added of sprouted lentil flour fermented by *W. confusa* SLA4 sourdough; SLSWB EPS POS, wheat bread added of sprouted lentil flour fermented by *W. confusa* SLA4 - dextran containing sourdough . All strains were inoculated at ca. 7 log cfu/g and sourdough fermented at 20 °C for 24 h. Wheat flour started with baker's yeast (CWB) was used as control. Doughs had DY 162.

Recipes				LSWB		LSWB		SLSWB		SLSWB	
		CWB		EPS NEG		EPS POS		EPS NEG		EPS POS	
		% d.w. ¹	% f.w. ²	% d.w.	% f.w.						
Wheat flour		61.7	100.0	55.7	90.2	55.7	90.2	55.7	90.2	55.7	90.2
Water		38.3	62.0	14.0	22.7	14.0	22.7	14.0	22.7	14.0	22.7
Sourdough (DY 500)				30.3	49.1	30.3	49.1	30.3	49.1	30.3	49.1
Sourdough recipes	Lentil flour			6.1	9.8	6.1	9.8				
	Sprouted lentil flour							6.1	9.8	6.1	9.8
	Water			24.3	39.3	24.3	39.3	24.3	39.3	24.3	39.3
	Sucrose					1.5	2.4			1.5	2.4
Fresh yeast		0.7	1.1	0.7	1.1	0.7	1.1	0.7	1.1	0.7	1.1
Salt		1.1	1.8	1.1	1.8	1.1	1.8	1.1	1.8	1.1	1.8
Flour sum ³		61.7	100.0	61.7	100.0	61.7	100.0	61.7	100.0	61.7	100.0
Water sum ⁴		38.3	62.0	38.3	62.0	38.3	62.0	38.3	62.0	38.3	100.0

¹d.w., dough weight

²f.w., flour weight

³Flour sum in the recipe is calculated as the sum of flour from sourdough and flour used in baking

⁴Water sum is calculated as the sum of water from sourdough and water used in baking.

For control and sourdough bread the total amount of water is the same, 62% of flour weight (f.w.) and the dough yield (DY) is same 162.

Table S3. Acidity (pH) and viscosity (Pa s⁻¹) of sourdoughs obtained from lentil (L) and sprouted lentil (SL) flours started with *Lactobacillus plantarum* DPPMAB24W (non EPS producing strain) (B24W), or with the selected lactic acid bacteria *Weissella confusa* SLA4 (SLA4), *Weissella paramesenteroides* SLA5 (SLA5), *Leuconostoc pseudomesenteroides* DSM 20193 (DSM20193) and *Weissella cibaria* DSM 20194 (DSM20194) (EPS NEG), and fermented at 20 and 25 °C for 24 h. To enable EPS formation, doughs were fermented with addition of 5% dough weight (25% flour weight) sucrose (EPS POS). Doughs prepared without sucrose and without inoculum were used as control (CT).

Stantan	Daugh Cada	Sugrage		20 °C	25 °C		
Starter	Dough Code	Sucros	^{se} pH	Viscosity	рН	Viscosity	
				0			
Lentil sourdough							
Not inoculated	L_CT	-	6.39 ± 0.07^{a}	$0.21\pm0.02^{\rm h}$	6.34 ± 0.01^{a}	0.19 ± 0.01^{1}	
Lactobacillus plantarum							
DPPMAB24W	L-B24W	+	6.35 ± 0.09^{a}	$0.18\pm0.01^{\rm i}$	$6.38\pm0.03^{\rm a}$	$0.20\pm0.02^{\rm l}$	
	L-SLA4_EPS POS	+	6.28 ± 0.10^{ab}	$0.23\pm0.03^{\rm h}$	$6.33\pm0.08^{\rm a}$	$0.18\pm0.01^{\rm l}$	
weissella confusa SLA4	L-SLA4_EPS NEG	-	6.25 ± 0.05^{ab}	$0.21\pm0.02^{\rm h}$	6.27 ± 0.07^{ab}	$0.19\pm0.02^{\rm l}$	
Waiggalla nangwagantanaidag SL A5	L-SLA5_EPS POS	+	6.36 ± 0.25^a	0.19 ± 0.02^{hi}	6.35 ± 0.11^{a}	$0.16\pm0.03^{\rm l}$	
weissella paramesenterolaes SLAS	L-SLA5_EPS NEG	-	6.34 ± 0.11^a	$0.17\pm0.02^{\rm i}$	6.32 ± 0.14^{a}	0.17 ± 0.01^{1}	
Leuconostoc pseudomesenteroides	L-DSM20193_EPS POS	+	6.37 ± 0.13^a	$0.22\pm0.01^{\rm h}$	$6.31\pm0.03^{\rm a}$	$0.22\pm0.01^{\rm i}$	
DSM 20193	L-DSM20193_EPS NEG	-	6.34 ± 0.07^a	0.24 ± 0.01^{h}	6.22 ± 0.14^{b}	0.26 ± 0.03^{i}	
W and a DSM 20104	L-DSM20194_EPS POS	+	6.39 ± 0.05^a	$0.18\pm0.03^{\rm i}$	$6.37\pm0.06^{\rm a}$	$0.18\pm0.01^{\rm l}$	
w. conjusa DSM 20194	L-DSM20194_EPS NEG	-	$6.42\pm0.08^{\:a}$	$0.25\pm0.01^{\rm h}$	6.30 ± 0.04^{a}	$0.25\pm0.04^{\rm i}$	
Sprouted lentil sourdough							
Not inoculated	SL_CT	-	6.19 ± 0.14^{b}	$0.14\pm0.04^{\rm i}$	6.24 ± 0.11^{ab}	$0.19\pm0.01^{\rm l}$	
L. plantarum DPPMAB24W	SL-B24W	+	6.17 ± 0.09^{b}	$0.15\pm0.01^{\rm i}$	6.22 ± 0.13^{b}	0.15 ± 0.01^{1}	
W confuse SI A4	SL-SLA4_EPS POS	+	6.15 ± 0.14^{b}	$0.17\pm0.02^{\rm i}$	6.29 ± 0.01^{ab}	$0.21\pm0.02^{\rm l}$	
w. conjusu SLA4	SL-SLA4_EPS NEG	-	6.14 ± 0.02^{b}	$0.15\pm0.01^{\rm i}$	6.23 ± 0.08^{b}	0.16 ± 0.01^{1}	
W nangu agantanaidag SL A 5	SL-SLA5_EPS POS	+	$6.15\pm0.05^{\mathrm{b}}$	$0.21\pm0.01^{\rm h}$	6.21 ± 0.05^{b}	$0.21\pm0.03^{\rm l}$	
w. paramesenterotaes SLAS	SL-SLA5_EPS NEG	-	6.19 ± 0.07^{b}	$0.20\pm0.02^{\rm h}$	6.26 ± 0.02^{ab}	$0.20\pm0.01^{\rm l}$	
L manufam agentancidas DSM 20102	SL-DSM20193_EPS						
L. pseudomesenieroides DSM 20195	POS	+	6.19 ± 0.15^{b}	0.19 ± 0.02^{hi}	6.21 ± 0.03^{b}	$0.18\pm0.02^{\rm l}$	

				20 °C	25 °C	
Starter	Dough Code	Sucros	e pH	Viscosity	pН	Viscosity
				ī	0 h	
	SL-DSM20193_EPS					
	NEG	-	6.17 ± 0.04^{b}	$0.21\pm0.02^{\rm h}$	6.28 ± 0.02^{ab}	$0.23\pm0.01^{\rm i}$
	SL-DSM20194_EPS					
W confuse DSM 20104	POS	+	6.15 ± 0.13^{b}	$0.23\pm0.01^{\rm h}$	$6.20\pm0.01^{\text{b}}$	$0.26\pm0.03^{\rm i}$
w. conjusa DSWI 20194	SL-DSM20194_EPS					
	NEG	-	$6.15\pm0.11^{\text{b}}$	$0.17\pm0.02^{\rm i}$	$6.22\pm0.02^{\text{b}}$	$0.25\pm0.01^{\rm i}$
				2	24 h	
Lentil sourdough						
Not inoculated	L_CT	-	4.80 ± 0.12^{c}	0.24 ± 0.01^{h}	$4.50\pm0.09^{\rm c}$	0.23 ± 0.03^{i}
L. plantarum DPPMAB24W	L-B24W	+	4.25 ± 0.13^{ef}	0.19 ± 0.03^{hi}	4.06 ± 0.02^{de}	0.21 ± 0.02^{1}
W confuse SLAA	L-SLA4_EPS POS	+	$4.56\pm0.18^{\text{d}}$	3.60 ± 0.01^{d}	$4.18\pm0.13^{\text{d}}$	$0.70\pm0.01^{\text{g}}$
W. confusa SLA4	L-SLA4_EPS NEG	-	4.40 ± 0.11^{de}	$0.49\pm0.02^{\text{g}}$	$4.13\pm0.04^{\rm d}$	0.33 ± 0.02^{hi}
W nanamagantanoidag SI A5	L-SLA5_EPS POS	+	4.35 ± 0.11^{e}	$0.20\pm0.01^{\rm h}$	4.05 ± 0.14^{de}	$0.20\pm0.03^{\rm l}$
W. paramesenteroides SLA5	L-SLA5_EPS NEG	-	4.23 ± 0.12^{ef}	$0.29\pm0.02^{\rm fh}$	4.10 ± 0.01^{de}	$0.27\pm0.01^{\rm i}$
L. pseudomesenteroides DSM 20193	L-DSM20193_EPS POS	+	4.34 ± 0.03^{e}	2.67 ± 0.01^{e}	$4.15\pm0.03^{\text{d}}$	1.70 ± 0.01^{e}
	L-DSM20193_EPS NEG	-	4.30 ± 0.11^{e}	$0.38\pm0.03^{\text{gh}}$	4.09 ± 0.01^{de}	0.36 ± 0.01^{h}
W = 0 DSM 20104	L-DSM20194_EPS POS	+	4.33 ± 0.12^{e}	$0.21\pm0.01^{\rm h}$	4.06 ± 0.04^{de}	$2.14\pm0.02^{\rm c}$
w. conjusu DSW 20194	L-DSM20194_EPS NEG	-	4.38 ± 0.13^{e}	$0.47\pm0.02^{\text{g}}$	4.10 ± 0.01^{de}	$0.41\pm0.01^{\rm h}$
Sprouted lentil sourdough						
Not inoculated	SL_CT	-	5.00 ± 0.21^{c}	$0.18\pm0.03^{\rm i}$	$4.61 \pm 0.01^{\circ}$	$0.19\pm0.04^{\rm l}$
L. plantarum DPPMAB24W	SL-B24W	+	$4.10\pm0.02^{\rm f}$	$0.15\pm0.01^{\rm i}$	3.92 ± 0.04^{e}	$0.16\pm0.02^{\rm l}$
W confuse SLAA	SL-SLA4_EPS POS	+	4.47 ± 0.02^{de}	5.70 ± 0.01^{a}	4.10 ± 0.03^{de}	$1.80\pm0.01^{\rm d}$
w. conjusu SLA4	SL-SLA4_EPS NEG	-	4.52 ± 0.04^{d}	$0.62\pm0.03^{\rm f}$	4.09 ± 0.01^{de}	0.31 ± 0.01^{hi}
W naramasantaroidas SI A5	SL-SLA5_EPS POS	+	4.31 ± 0.03^{e}	$0.44\pm0.01^{\text{g}}$	4.14 ± 0.07^{d}	0.23 ± 0.01^{i}
w. paramesenterolaes SLAS	SL-SLA5_EPS NEG	-	4.25 ± 0.01^{ef}	$0.22\pm0.03^{\rm h}$	$4.13\pm0.05^{\rm d}$	$0.21\pm0.02^{\rm i}$
	SL-DSM20193_EPS					
	POS	+	4.31 ± 0.11^{e}	4.74 ± 0.01^{b}	$3.80\pm0.06^{\rm f}$	3.00 ± 0.01^{a}
L. pseudomesenterotaes DSM 20195	SL-DSM20193_EPS					
	NEG	-	4.39 ± 0.01^{e}	$0.75\pm0.01^{\rm f}$	4.14 ± 0.04^{d}	$0.80\pm0.03^{\rm f}$
W confuse DSM 20104	SL-DSM20194_EPS					
w. conjusu DSWI 20194	POS	+	4.43 ± 0.02^{de}	4.46 ± 0.02^{c}	4.35 ± 0.01^{cd}	2.58 ± 0.01^{b}

Starter	Dough Code	Sugar	0	20 °C	25 °C		
		Sucrose	e pH	Viscosity	рН	Viscosity	
				(0 h		
	SL-DSM20194_EPS						
	NEG	-	$4.50\pm0.11^{\text{d}}$	$0.63\pm0.01^{\rm f}$	4.19 ± 0.02^{d}	$0.27\pm0.01^{\rm i}$	

Data are mean values of triplicate determination \pm standard deviation. ^{a-h}Means within a column with different superscript letters are significantly different (P < 0.05).