Research article

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Microbial communities and malt quality of durum wheat used in brewing

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Durum wheat (*Triticum durum* Desf.) has potential as an adjunct in brewing given its agronomic, chemical and technological properties. The aim of this work were to identify the cultivable microflora and evaluate the technological quality of the durum wheat variety 'Senatore Cappelli' grown and used by a craft brewery in Sardinia, Italy. The isolated bacterial strains were mainly rhizospheric (*Kocuria rizophila*, *Microbacterium aerolatum* and *Bacillus pumilus*) and associated with the microbiota of wheat (*Staphylococcus* spp.). None have been reported previously as spoilage species in brewing. The dominant yeast genera were *Cryptococcus* spp. and *Rhodotorula* spp., followed by *Saccharomyces cerevisiae*. The dominant filamentous fungus genera were *Alternaria* and *Rhizopus*. Low levels of mycotoxigenic *Fusarium* spp., *Aspergillus* spp. and *Penicillium* spp. were isolated. However, the levels of deoxynivalenol, T2-HT2, fumonisin, aflatoxin and ochratoxin detected in the malt and grain were below the thresholds defined by European law. Malt obtained from raw grain showed interesting technological properties, but required specific malting parameters different from those of common wheat and barley. These data suggest that the use of locally grown durum wheat in brewing can increase sustainability and reduce costs, while reinforcing the link with the terroir and promoting reduced mycotoxin levels. © 2019 The Institute of Brewing & Distilling

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Keywords: craft beer; locally grown cereals; microbiota; 'Senatore Cappelli' durum wheat; mycotoxins

Introduction

In recent years, the number of breweries has grown exponentially in response to the worldwide demand for beers that use fresh ingredients with rich flavours and aromas *(1,2)*. The use of specific microbial strains and locally grown cereals allows brewers to formulate original recipes to confer distinct sensorial qualities to their beers *(3–5)*. Wheat derivatives are common adjuncts in the production of specialty beers, such as Belgian white beers (40% unmalted wheat) and German '*weissbier*', or wheat beer (≥50% malted wheat). Over the last 20 years, the popularity of wheat beers has also increased. In particular, in Germany the consumption of wheat beers has almost doubled between 1990 and 2009 *(6)*.

Compared with lager beers, wheat beers have more unique aromatic compounds, such as those that confer the typically 'malty' flavour (e.g. maltol, furaneol). The wheat varieties used in brewing almost exclusively belong to common wheat (*Triticum aestivum* L.), although recent studies have noted the possibility of using durum wheat (*Triticum durum* Desf.) for the production of beers with interesting sensorial properties *(4,7)*. Durum wheat is grown mainly in the Mediterranean area, with Italy as the main producer (4.313 million tons/year) *(8)*. Thus, its use in beers produced in the Mediterranean region should reinforce the link with the *terroir* thereby enhancing the distinct characters of the beers.

'Senatore Cappelli' is an old Italian durum wheat variety that was originally released in 1915 by the agronomist and plant breeder Nazareno Strampelli. This hallmark variety was then widely grown in Italy and in most durum wheat growing areas all over the world, until the 1960s. Furthermore, most of the durum wheat varieties released in subsequent years in Italy were derived from 'Senatore Cappelli' in one way or another *(9)*. Nowadays, this has been largely superceded by the modern durum wheat varieties that have been selected for pasta production owing to their high gluten strength, although 'Senatore Cappelli' is still grown for bread making, owing to its high protein and soluble fibre contents *(10)*.

'Senatore Cappelli' is highly resistant to biotic and abiotic stresses, has a high content of free phenols, and particularly flavonoids, which contribute to its beneficial health potential *(11–13)*. Indeed flavonoids have strong antioxidant and anti-inflammatory activities and they inhibit hydrolytic oxidative enzymes *(14)*.

Owing to the long tradition of pasta and bread making in Italy, intense efforts have been made in the past to select durum wheat genotypes with the best agronomic and technical performance. However, these wheat varieties require specific evaluation when used in brewing. The high protein content, which is good for baking bread, may result in long lautering times, difficulties with filtration and problems with fermentation during beer production *(6)*. In contrast, the most important criteria for wheat varieties for brewing purposes are viscosity, soluble nitrogen, attenuation and the Kolbach index *(15)*. Also, the microbial composition of wheat requires careful evaluation to avoid problems during fermentation or beer stabilisation. Indeed, microorganisms that are important for some biotechnological processes might be detrimental to

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others. For example, the lactic acid bacteria (LAB) that are essential in the fermentation of sourdoughs are dangerous spoilage organisms in beer. In addition, microorganisms associated with the cereal grain might cause technical problems (e.g. gushing, premature yeast flocculation) or health issues, particularly in terms of the production of mycotoxins by filamentous fungi.

In this context, considering the growing popularity of wheat beers all over the world as well as the importance of durum wheat cultivation in the Mediterranean region, this study was designed to evaluate the cultivable microflora, the presence of mycotoxins and the susceptibility to malting of the durum wheat variety 'Senatore Cappelli' grown in Sardinia in 2012 and 2013.

Materials and methods

Wheat samples and malt analysis

The durum wheat variety 'Senatore Cappelli' (*Triticum durum* Desf.) was grown in Tertenia (39°31′42″ N; 9°35′11″ E; 10.9 m a.s.l.; Sardinia, Italy) from November to July in 2012 and 2013. The local meteorological records were provided by the Sardinian Regional Agency for Environmental Protection, with the daily temperature and precipitation data from the local weather station (NU). After harvest, the grain was stored at 20°C for 30 days, and then it underwent chemical and microbiological characterisation.

The chemical analysis of the wheat grain was performed using a whole grain analyser (InfratecTM1241; Foss Italia, Padova, Italy) with near-infrared transmittance technology. Micro-malting of the wheat was performed by the Crisp malting group (Fakenam, Norfolk, UK), according to the standard protocols for barley malting and analysed according to the European Brewery Convention *(16,17)*.

Microbiological media and culture conditions

A 10 g aliquot of wheat grain was mixed with 90 mL sterile saline solution (0.9 g NaCl in 100 mL double distilled water) and homogenised (Stomacher laboratory blender 400; Seward Medical, London, UK).

To isolate the bacteria, serial dilutions of the homogenates were used to inoculate the following agars: plate count agar (Microbiol, Italy) with 0.01% cycloheximide (Sigma, St Louis, MO, USA) at 32°C for 48 h; M17 agar (Microbiol, Italy) at 30°C for 48 h; and De Man, Rogosa and Sharpe agar (Microbiol, Italy) at 37°C for 48 h under aerobic and anaerobic conditions in a gas-pack system (Oxoid AnaeroGen, Thermo Scientific, Basingstoke, UK). The isolates were stored in MRS with 20% glycerol at −80°C.

To isolate the yeast, serial dilutions of the homogenates were used to inoculate plates of yeast malt agar (Microbiol, Italy) with 0.01% chloroamphenicol, 0.01% dichlorotetracycline and 0.02% Triton X100 (all from Sigma, St Louis, MO, USA), and incubation at 25°C for 3–5 days. Wallerstein Laboratory nutrient agar (Oxoid, Hamphire, UK) was used to define the different yeast isolates according to profile, contour and colour *(18)*. The isolates obtained were stored in 1% yeast extract, 2% peptone, 2% dextrose (YEPD; Oxoid, Hamphire, UK) with 20% glycerol at −80°C.

To isolate the filamentous fungi, 100 grains were put directly onto potato dextrose agar (PDA), and 100 grains were surface disinfected with 2% sodium hypochlorite for 3 min, followed by 3 washes in sterile water, and then put onto PDA (Microbiol, Italy) with 0.01% chlorotetracycline and 0.01% streptomycin (both from Sigma, St Louis, MO, USA). These PDA plates were incubated at 25°C for 5–7 days. Monosporic cultures were then prepared following Burgess *et al. (19)*. After 14–21 days of incubation on Spezieller Nährstoffarmer agar and/or PDA at 22–25°C under a photoperiod of 12 h light, the fungi were subjected to macroscopic and microscopic examination. Morphological identification of the *Fusarium* spp. was performed following the specifications described by Leslie and Summerell *(20)*. The filamentous fungi isolated were stored on PDA with 50% glycerol at −80°C.

Molecular methods

Bacterial DNA extraction was carried out according to Martín-Platero *et al. (21)*. The bacterial genotypic diversity was evaluated by randomly amplified polymorphic DNA polymerase chain reaction (RAPD-PCR) and repetitive element palindromic-polymerase chain reaction (rep-PCR), using the primers M13 (5′GAGGGTGGC GGTTCT-3′) and GTG5 (5′-GTGGTGGTGGTGGTG-3′), respectively *(22)*. Amplification and sequencing of the 16S rDNA were performed using the primers W001 (5′-AGAGTTTGATCMTGGCTC-3′) and W002 (5′-GNTACCTTGTTACGACTT-3′), as previously described *(23)*.

Yeast genomic DNA isolation was performed according to Burke *et al. (24)*. Identification of the yeast isolates was performed by amplification and sequencing of the ribosomal DNA Internal Transcribed Spacer (ITS) region *(25)*.

Amplicons were purified with PCR purification kits (QIAquick; Qiagen GmbH, Hilden, Germany), following the manufacturer's instructions, and then sequenced by BMR Genomics (Padova, Italy). The sequences obtained were compared with those in the GenBank database using the BLAST programme *(26)*, and with those in the Ribosomal Database project (http://rdp.cme.msu.edu/edu/index.jsp). Sequences with ≥97% identity were considered to represent the same species.

Quantification of mycotoxins

Quantitative analysis of the mycotoxins deoxynivalenol, T2-HT2, fumonisins, aflatoxins and ochratoxins was carried out using a 'rapid one-step assay' system (Charm Lateral Flow R.O.S.A.; Foss and Charm). The limits of detection for each of these micotoxin with this system were, respectively, 100, 10, 10, 250, 2 and 2 μg/L. Three replicates were performed for each sample.

Statistical analyses

Cluster analysis of the band profiles obtained from RADP and rep-PCR analysis was performed using the Infoquest software (version 4.5; Bio-Rad). A similarity matrix of bacterial banding profiles was calculated using Pearson's correlation similarity coefficients. Cluster analysis of the single and combined RAPD and rep-PCR band profiles was performed using the unweighted pair-group method with arithmetic averages. Different bacterial strains were distinguished as those with an arbitrary cluster cut-off of 85%.

Results and discussion

Chemical composition of 'Senatore Cappelli' grain

The chemical composition and technological properties of cereals are affected by the meteorological conditions during cultivation. In the growing area, rainfall in 2012 and 2013 was lower than the 10-year average, and 16% higher in 2013 than in 2012. In 2012, February and April saw the highest rainfall, while in 2013, this

was the case for November and December (Figure S1 in the Supporting Information). Less than 20 mm/month rain was recorded during the summers (June–August) in both years. The maximum temperatures were very similar for these two years, and they exceeded 30°C in June, July and August, while the minimum temperatures were steadily above the 0°C threshold. These rainfall and temperature conditions are typical of Mediterranean regions, where the moderate water deficit in spring results in moderate water stress during the anthesis stage, and in more severe constraint throughout the grain filling period *(8)*. In addition, the thermal conditions during the grain filling period significantly influence the amounts of nitrogenous substances accumulated in grain, as earlier sowing results in reduced protein content in durum wheat varieties *(27)*.

To determine the effects of the site of cultivation on the chemical composition, raw grains of 'Senatore Cappelli' cultivated in Tertenia were analysed (Table 1). The total protein and gluten contents of the 'Senatore Cappelli' grain were lower than those observed for this cultivar grown in a different geographical location *(28)*. A high protein content is detrimental in brewing, as it can cause fermentation problems *(6)*. In this respect, the 11.4% protein content of 'Senatore Cappelli' was very similar to that of the durum wheat variety 'Simeto', which was also proposed for brewing by Alfeo *et al. (7)*. Furthermore, the high germinability of the grain of 'Senatore Cappelli' is consistent with the economic sustainability of the malting process.

Table 1. Chemical characterisation of the durum wheat variety 'Senatore Cappelli'

Grain sample	1000 grain weight (g)	Moisture (%)	Protein content (%)	Dry gluten (%)	Moist gluten (%)	Yellow Index	Test weight (kg/hL)	Gluten index	Gluten (%)		Starch (%)	Germinability (%)	Ash content (%)
									Past	Residual			
2012	57.0	12.4	11.7	8.6	24.4	9.9	80.0	10.0	2.0	0.2	57.8	97.0	4.2
2013	56.5	12.1	11.1	8.3	24.2	10.8	80.0	7.0	2.0	0.1	56.6	94.0	4.0
Mean	**56.8**	**12.3**	**11.4**	**8.5**	**24.3**	**10.4**	**80.0**	**8.5**	**2.0**	**0.2**	**57.2**	**95.5**	**4.1**
SD	**0.4**	**0.2**	**0.4**	**0.2**	**0.1**	**0.6**	**0.0**	**2.1**	**0.0**	**0.1**	**0.8**	**2.1**	**0.1**

SD, Standard deviation.

Microflora of 'Senatore Cappelli' grain

The microflora of cereal grain from different geographical locations tends to form distinct groups, which indicates the great importance of the geographical region of cultivation *(29)*. Indeed, the soil and the environment are the main microbial reservoirs for contamination of wheat grain. Bacteria, fungi and yeast species identified for these 'Senatore Cappelli' grain through culture-dependent approaches are shown in Table 2.

Bacteria. The bacterial concentrations associated with the wheat grain varied over the two years: $4.42 \times 10^4 \pm 2.91 \times 10^3$ CFU/g in 2012, and $8.18 \times 10^6 \pm 4.05 \times 10^5$ CFU/g in 2013, probably because of the above differences in rainfall. These data are in agreement with Berghofer *et al. (30)*, who reported bacterial concentrations from 10^2 to 10^5 CFU/g in an extensive survey that investigated the microbiological quality of >600 wheat and flour samples. The analysis of the morphology of the bacterial colonies followed by genotyping with the combined banding profiles of RAPD-PCR and rep-PCR allowed the bacterial isolates to be grouped into 35 clusters (Figure 1). Thirty one strains represented single cluster strains. According to Carafa *et al. (31)*, the great number of singletons accounts for a high microbial biodiversity. Further, a similar number of singletons has been determined in cereals as well as in other food matrices *(23,32)*. The 16S rDNA sequencing of representative isolates from each cluster identified 40 strains at the genus level and 11 strains at the species level. In agreement with Minervini et al. *(33)*, *Staphylococcus* spp. and *Bacillus* spp. were dominant and constituted the core *Firmicutes* microbiome of durum wheat grain. *Bacillus* spp. are also dominant in the roots and flours of durum wheat *(33)*. Among the genera detected at lower concentrations, members of the genus *Kocuria* are commonly isolated from various natural habitats, such as the rhizosphere, soil, fermented food and marine sediments *(34)*. Also, the presence of *Rhodococcus* spp. in the wheat rhizosphere has

Table 2. Cultivable microflora isolated in raw grain of durum wheat variety 'Senatore Cappelli'

Microflora	2012 harvest	2013 harvest
Bacteria (CFU/g)		
Kocuria rhizofila	$<1.00 \times 10$	$5.28 \times 10^5 \pm 2.61 \times 10^4$
Microbacterium areolatum	$<1.00 \times 10$	$6.60 \times 10^5 \pm 3.27 \times 10^4$
Staphylococcus spp.	$3.81 \times 10^4 \pm 2.51 \times 10^3$	$5.66 \times 10^6 \pm 2.80 \times 10^5$
Rhodococcus wratislaviensis	$2.04 \times 10^3 \pm 1.34 \times 10^2$	$< 1.00 \times 10$
Bacillus pumilus	$4.08 \times 10^3 \pm 2.69 \times 10^2$	$1.33 \times 10^6 \pm 6.59 \times 10^4$
Yeast (CFU/g)		
Cryptococcus chernovii	$1.02 \times 10^3 \pm 1.02 \times 10^2$	$9.00 \times 10^2 \pm 5.08 \times 10^2$
Cryptococcus festucosus	$3.00 \times 10^2 \pm 3.52 \times 10$	$1.27 \times 10^4 \pm 6.36 \times 10^3$
Rhodotorula glutinis	$3.69 \times 10^3 \pm 4.12 \times 10$	$4.18 \times 10^3 \pm 2.02 \times 10^3$
Saccharomyces cerevisiae	$1.32 \times 10^3 \pm 1.27 \times 10^2$	$1.80 \times 10^2 \pm 9.45 \times 10$
Fungi (CFU/100 grains)		
Rhizopus spp.	$4.34 \times 10 \pm 0.14 \times 10$	$5.53 \times 10 \pm 0.23 \times 10$
Alternaria spp.	$3.39 \times 10 \pm 0.09 \times 10$	$3.64 \times 10 \pm 0.07 \times 10$
Fusarium spp.	$0.53 \times 10 \pm 0.01 \times 10$	$0.52 \times 10 \pm 0.01 \times 10$
Penicillium spp.	$1.25 \times 10 \pm 0.01 \times 10$	$0.43 \times 10 \pm 0.01 \times 10$
Aspergillus spp.	$0.59 \times 10 \pm .01 \times 10$	<1

already been described *(35)*. *Microbacterium aerolatum* has been isolated from air samples, from the rhizosphere of indoor plants, and more recently in barley malt *(29,36)*.

Owing to the nutritional and technological aspects, a limited number of Gram-positive species and a few Gram-negative bacteria can grow in and spoil beer. In unpasteurised beer, *Lactobacillus* spp. and *Pediococcus* spp. are the predominant Gram-positive beer spoilers, while *Pectinatus* spp. and *Megasphaera* spp. are regarded as the most important Gram-negative spoilage bacteria *(37,38)*. Other bacteria isolated from brewery environments and associated with wort spoilage belong to *Enterobacteriaceae*, *Citrobacter*, *Hafnia*, *Klebsiella* and *Obesumbacterium (38)*. Among the beer spoilage bacteria, LAB are particularly dangerous, as they contaminate cereals and are considered part of the endophytic microbiota of cereals *(33)*. However, the absence of LAB in raw grain of 'Senatore Cappelli' in the present study was not surprising, as other studies have shown that, even after enrichment procedures, LAB cannot be isolated from 10% of durum wheat samples *(39)*. This is in agreement with the observation that the number of operational taxonomic units belonging to *Lactobacillus* strongly decreases during milk development and physiological maturity of durum wheat *(33)*.

Yeast. The total yeast count of the durum wheat grain were $3.0 \times 10^3 \pm 3.28 \times 10^2$ CFU/g in 2012, and $1.8 \times 10^4 \pm 9.55 \times 10^3$ CFU/g in 2013. These levels are similar to those already described in the literature for wheat flours *(30)*. Following morphological characterisation of the yeast isolates on Wallerstein Laboratory nutrient agar, three colony types were described: flat colonies with wrinkled surface, opaque and creamy texture, and pale red colour; smooth, opaque colonies, with creamy consistency, and cream to green colour; and colonies with a knob-like, convex surface, smooth, with butyrous texture and red colour. According to Pallman *et al. (18)*, these last two types of colonies correspond to isolates belonging to *Saccharomyces* spp. and *Rhodotorula* spp., respectively. Following ITS sequencing for 15 of the 156 isolates, the following species were identified: *Cryptococcus chernovii*, *Cryptococcus festucosus*, *Rhodotorula glutinis* and *Saccharomyces cerevisiae* (Table 2). The basidiomycetes genera *Cryptococcus* and *Rhodotorula* have already been described in wheat grain and in the durum wheat 'Senatore Cappelli' *(28,40)*. The uncontrolled growth of these yeast can cause problems, such as discoloration of the grain or alterations to the grain physiology during the malting process *(41)*. Nevertheless, *Rhodoturula* species can also contribute positively to the brewing process, as they produce different enzymes, which include α-amylase, β-glucanase, cellulose and endo-xylanase. These extracellular hydrolytic enzymes contribute to the overall enzyme spectrum of the malt *(29,42)*. Moreover, strains of *Rhodotorula glutins* were shown to be effective for inhibition of virulence of different fungal pathogens, such as *Penicillium expansum* and *Aspergillus niger (43,44)*. *S. cerevisiae* has already been identified in wheat grain and flour *(28)*. *S. cerevisiae* is the primary microorganism in some of the oldest biotechnological processes and selected strains are commonly used as starter cultures in brewing *(45)*. In this respect, wild *S. cerevisiae* isolates on raw materials might compete with the starter strains during the fermentation process. Cocolin *et al. (46)* showed that the inoculated starter strains generally do not dominate the fermentation of craft beers, as only 18% of strains isolated were ascribed to the starter culture.

Fungi. The fungi genera isolated from the durum wheat grain are reported in Table 2. *Alternaria* spp. have already been identified among the prevalent fungal species in durum wheat samples *(47)*. The levels of *Fusarium* contamination for both the years reported were particularly low. The other potentially toxigenic species were *Aspergillus* spp. and *Penicillium* spp. Species that belonged to the *Fusarium* genus were further identified, as these can have an impact on wheat quality, with the production of mycotoxins, hydrophobins and enzymes that reduce the brewing wort β-glucan levels and viscosity, increase the wort soluble nitrogen content and change the wort colour. In 2012, only *Fusarium avenaceum* (5%) was isolated, while in 2013, *F. graminearum* (1%), *F. culmorum* (3%) and *F. solani* (1%) were also identified. These data are in agreement with Vujanovic *et al. (47)*, who reported that *F. avenaceum* was abundant in all tissues of durum wheat plants.

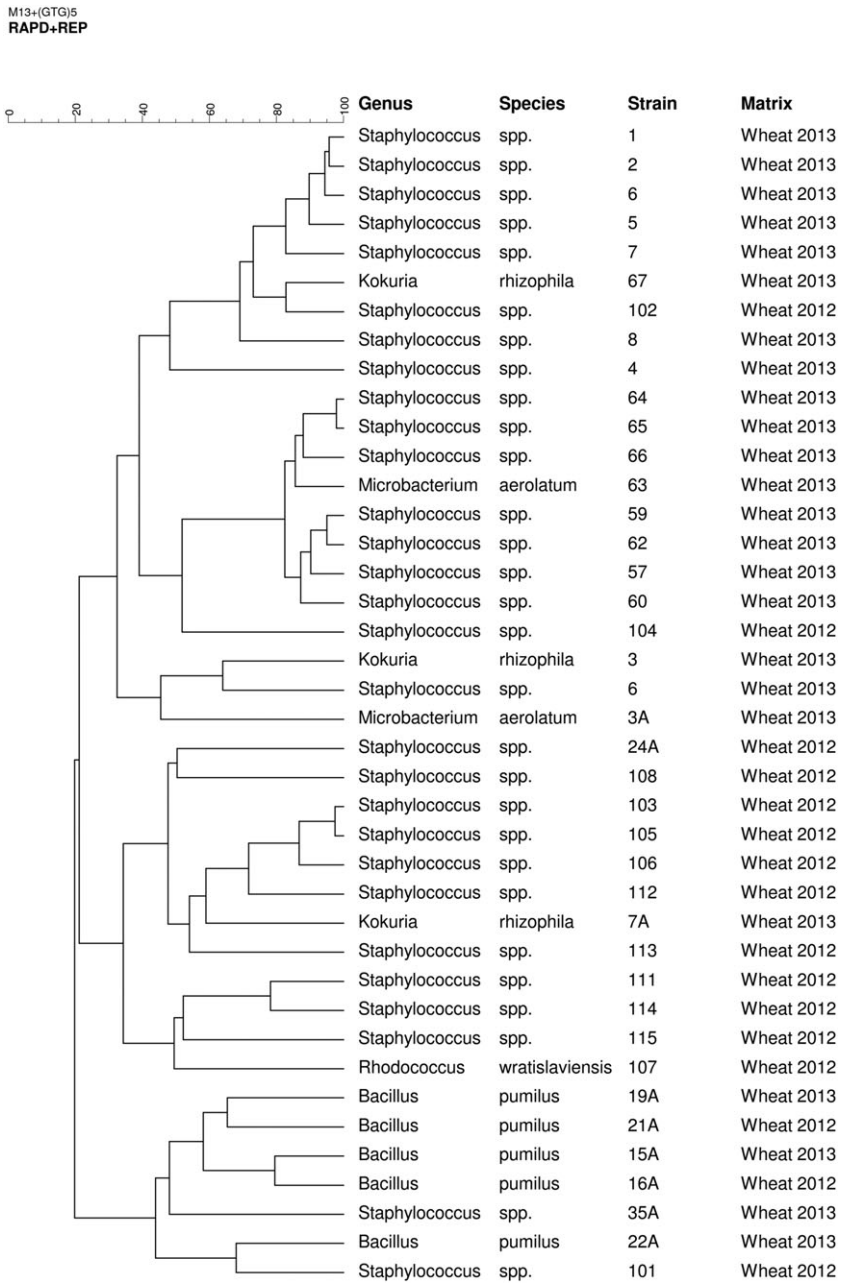

Figure 1. Genotyping of the isolates from the grain of durum wheat variety 'Senatore Cappelli' determined by RAPD-PCR and rep-PCR. The unweighted-pair-group method with arithmetic averages dendrogram is based on Pearson correlation coefficients of the combined M13 and GTG5 banding profiles.

It is well known that environmental conditions around the time of anthesis, including temperature and rainfall, are important factors in *Fusarium* infections and the composition of these species *(48)*. In particular, the lower temperatures in February and March 2012 were more suited for the growth of *F. avenaceum* rather than *F. culmorum* and *F. graminearum*. This infection might have occurred in April during anthesis, with lower rainfall during emergence (March) and greater rainfall in the spring at the resumption of vegetal growth (i.e. April–May). However, data regarding the prevalent fungal genera identified for durum wheat grain are due to their contamination during storage. Quantitative analysis of the mycotoxins identified contamination by T2-HT2 and aflatoxins here (Table 3). These results showed that mycotoxins were at levels below the fixed thresholds defined by European law: 1750 μg/L for deoxynivalenol, 200 μg/L for T2-HT2, 4000 μg/L for fumonisins, 4 μg/L for aflatoxins and 5 μg/L for ochratoxins (Community Regulation no. 856/2005). The aflatoxins were the only mycotoxins close to the threshold limits. In comparison, Peters *et al. (49)* reported that in craft beers, deoxynivalenol concentrations were frequently above the tolerable daily intake.

Malting quality

Despite the growing interest in wheat beers, very little attention has been paid to malting of wheat in comparison with malting of barley and other cereals. Recently, Alfeo *et al. (7)* evaluated the suitability for brewing of the malt obtained from durum wheat varieties 'Simeto' and 'Vivant'. In the same way, the malting quality

Table 3. Mycotoxin contamination in the raw grain of durum wheat variety 'Senatore Cappelli'

	Mycotoxin (μg/L)				
Grain sample	**Deoxynivalenol**	**T2-HT2**	**Aflatoxins**	**Ocratoxins**	**Fumonisines**
2012	<LOD	3.3	3.7	<LOD	<LOD
2013	<LOD	3.0	3.0	<LOD	<LOD

<LOD, Under the limit of detection.

of 'Senatore Cappelli' was evaluated (Table 4). According to Faltermaier *et al. (15)*, most of the characteristics of the malt obtained from 'Senatore Cappelli' were in the range suggested for good quality wheat malt, in terms of moisture, total protein, viscosity and extract. Levels of α-amylases and diastatic power were reported to be relatively high, in agreement with the observations of Briggs *(50)* and Alfeo *et al. (7)*, who reported that the contents of these enzymes were normally in excess in wheat malt, and were twice as high as in durum wheat when compared with common wheat. Viscosity, extract and nitrogen compounds (i.e. total protein, soluble nitrogen) all have important roles in the evaluation of malt *(51)*. Also, α-amylase and β-amylase activities are usually mentioned in the literature in terms of quality parameters *(50)*, where α-amylases degrade amylose and amylopectin to dextrins, β-amylases degrade the residual side chains and are the main contributors to the diastatic power of the malted grain. Thus, the activities of these enzymes are directly correlated to the levels of fermentable sugars during the mashing process.

On the negative side, the malt obtained from 'Senatore Cappelli' had a very high Kolbach index (KI). The KI is the ratio between soluble and total nitrogen (expressed as a percentage), and it measures the degree of degradation of the kernel protein. Titze *et al. (51)* suggested that the KI should be between 37 and 40% for a good wheat malt, with a maximum of acceptability at 45%. Also, Faltermaier *et al. (15)* set the malt quality range as a KI from 37 to 40%. Finally, Alfeo *et al. (7)* reported recently that the values of KI of malt from durum wheat varieties 'Simeto' and 'Vivant' were 40.3 and 38.0%, respectively. High values of KI are an indication of an excessive respiration rate, and consequently high malting losses *(52)*.

Table 4. Technological characteristics of the malt from the durum wheat variety 'Senatore Cappelli', analysed according to the European Brewery Convention *(17)*

Characteristic	**Unit**	**Analysis**
Moisture	%	4.7 ± 0.3
Extract 0.2 mm, as is basis	EBC	81.5 ± 3.3
Extract 0.2 mm, dry basis	EBC	85.5 ± 6.0
Visual colour	EBC	5.2 ± 0.1
Total nitrogen, dry basis	%	2.03 ± 0.1
Total protein, dry basis	%	12.7 ± 0.6
Total soluble nitrogen, dry basis	EBC	1.02 ± 0.0
Kolbach index	(%)	50.5 ± 2.4
Friability	%	60 ± 1.2
Homogeneity	%	98 ± 4.3
Partly unmodified grain	%	3 ± 0.1
Whole glassy corns	%	0.4 ± 0.0
α-Amylase, as is basis	DU	72 ± 2.4
α-Amylase, dry basis	DU	75 ± 3.8
Diastatic power, as is basis	°L	181 ± 843
Diastatic power, dry basis	°L	190 ± 10.8
Wort viscosity	EBC	1.64 ± 0.1
Wort pH	EBC	5.93 ± 0.3

DU, Dextrinising unit.

Conclusions

The data from this study shows that the locally grown grain of durum wheat variety 'Senatore Cappelli' has chemical and microbiological qualities that make this cultivar well suited as an adjunct in beer production. None of the bacteria and yeast isolated in the raw grain were of particular relevance to beer spoilage. On the other hand, some of the strains isolated in this study could represent a reservoir of strains of biotechnological interest. Indeed, some strains of *Bacillus* are known as plant growth promoting bacteria, and *Rhodococci* are recognised as being very versatile metabolically, and being active in biodegradation *(53)*. In particular, preliminary observations have shown that a strain of *Rhodotorula glutinis* isolated in the present study showed antagonistic activity against *Aspergillus ochraceus* and *Aspergillus flavus* (Angela Bianco, personal communication, 6 May 2018). In addition, very limited growth of filamentous fungi, and consequently little mycotoxin contamination, was measured for these locally grown grains; indeed, being locally grown, they will also be less subject to long storage times and transportation, and consequently to microbial contamination. However, and finally, the malt obtained from 'Senatore Cappelli' was not suitable for brewing, and further studies are needed to determine the optimal malting conditions to improve the KI of this durum wheat variety.

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Supporting information

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