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Research paper

Genetic diversity of *Candida albicans* isolates recovered from hospital environments and patients with severe acquired brain injuries

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ABSTRACT

Although the epidemiology of pathogenic *Candida* species causing invasive human diseases is changing, *Candida albicans* still remains the most common cause of bloodstream infections worldwide. The propensity of this pathogen to cause infections is undoubtedly the result of its unique genetic plasticity that allow it to adapt and respond quickly to a myriad of changing conditions both in the host and in the environment. For this reason, we decided to investigate the genetic diversity of this important fungal pathogen in a particular category of patients with severe neurological deficits including the hospital environments where they are hospitalized.

Genetic diversity of 21 *C. albicans* isolates recovered from blood, hands of healthcare workers and hospital environments was evaluated by using multilocus sequence typing (MLST) which revealed a high genetic heterogeneity with a set of 18 diploid sequence types (DSTs) recovered among 21 isolates investigated. Interestingly, 13 of these 18 MLST genotypes were completely new and added to the *C. albicans* MLST central database. Six eBURST clonal complexes (CC-1, CC-2, CC-6, CC-9, CC-27 and CC-42) and three singletons contained all DSTs found in this study. Among all the new DSTs identified, DST3388 was the most intriguing as this genotype was recovered from a typical *C. albicans* isolate clustering within the MLST-Clade 13, the most divergent evolutionary lineage within *C. albicans* population containing only isolates with unusual phenotypes originally known as *Candida africana*.

In conclusion, the results of this study expand our understanding of the molecular epidemiology and global population structure of *C. albicans* suggesting that further studies on different categories of patients and hospital environments are needed to better understand how the population of this species adapts and evolves in heterogeneous hosts and changing environments.

1. Introduction

Today, fungal infections represent a serious public health problem that needs urgent attention as evidenced by over 1.5 million of people supposed to die each year worldwide (Brown et al., 2012). This mortality rate currently exceeds that of other well-known infectious diseases, such as malaria (435.000 deaths in 2017), hepatitis B (887.000 deaths in 2015), or HIV/AIDS (940.000 deaths in 2017) and is rather similar to that of tuberculosis (1.6 million deaths in 2017) and other important human diseases including lung cancer (1.76 million deaths in 2018) and diabetes (1.6 million deaths in 2016) (www.who.int/mediacentre/factsheets/en). However, this incidence is far from being a realistic estimate, as the extent of the global burden of fungal infections appears to be considerably much wider (Bongomin et al., 2017; Denning, 2017). In fact, recent global estimates indicates that over 300 million people are afflicted with a serious fungal infection and 25 million are at particularly high risk of mortality or visual impairment (www.gaffi.org). In this scenario, invasive candidiasis poses an emerging public health concern as proved by over 750,000 cases reported every year worldwide (Bongomin et al., 2017). Undoubtedly, candidemia makes a substantial contribution to the global burden of invasive *Candida* infections with incidence rates reported to be between 2.1 and

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21 cases per 100,000 persons in several population-based epidemiological studies (Bongomin et al., 2017). Of note, in Italy the annual incidence rate of this disease has increased considerably reaching the unacceptable value of 21.8 cases per 100,000 people (Bassetti et al., 2018), one of the highest in the world (Bongomin et al., 2017; Bassetti et al., 2018).

The impact of invasive fungal infections on the economy of the countries and their health systems is also enormous, with billions of euros being spent each year because of the burden of healthcare-associated infections (HAIs). In European intensive care units (ICUs), about 8% of the bloodstream (BSI) and 11.7% of the urinary tract infections are caused by pathogenic *Candida* species acquired by patients during their stay in the hospital (ECDC, 2018). The most part of these HAIs have been linked to the use of invasive devices (e.g. vascular and urinary catheters) (ECDC, 2018) emphasizing the key role of hospital environments in the cross-transmissions of microbial pathogens (Rajendran et al., 2016). However, a recent epidemiological meta-analysis by Koehler et al. (2019) showed that the incidence and mortality of candidemia in Europe vary considerably between hospitals, regions and over time with an estimated global incidence rate of 3.88 cases per 100,000 persons and a 30-day mortality rate of 37% in the ICUs.

Candida genus is a complex and important group of Ascomycetes yeast-like fungi that comprises over 300 species ubiquitously distributed in nature (Lachance et al., 2011). Some of these species are also commonly found as normal colonizers of the human body where can act as opportunistic pathogens if host conditions become compromised (Prieto et al., 2016). A typical example is *Candida albicans*, one of the most common cause of fungal infections in humans, especially in immunocompromised individuals (Quindós et al., 2018). This species, together with other important *Candida* pathogens (e.g. *Candida parapsilosis*, *Candida tropicalis*), is part of the so-called CTG clade in which the CUG codon is translated as serine rather than leucine (Turner and Butler, 2014). This flexibility of the genetic code, along with several other unique genetic proprieties such as changes in ploidy, loss of heterozygosity, isochromosome formation and the distinctive organization of chromosome hallmarks (Hirakawa et al., 2015), could explain the extraordinary adaptability of *C. albicans* to different niches in the host and a wide range of natural environments including those in the hospitals (Hirakawa et al., 2015; Hall, 2017; Legrand et al., 2019).

In this study we describe the population structure and genetic diversity of *C. albicans* isolates recovered from blood samples, hands of healthcare workers (HCWs) and hospital environments in one of the most important centre in Southern Italy for the treatment of patients with serious neurological disorders and/or severe acquired brain injuries.

2. Materials and methods

2.1. Clinical and environmental samples

This study was conducted in an ICU of the IRCCS Centro Neurolesi Bonino-Pulejo, Messina, Italy, and it is part of a surveillance programme funded by the Italian Ministry of Health for the prevention and control of healthcare-associated fungal infections (project GR-2011-02347606).

A total of 21 *C. albicans* isolates were examined in this study (Table 1).

Seven isolates were recovered from blood samples (5 isolates), a central venous catheter (CVC) (1 isolate) and a nasogastric tube (1 isolate) (Table 1) of patients with severe acquired brain injuries and phenotypically identified by the Vitek 2 system (bioMerièux, Italy) as part of the ordinary activity of the diagnostic microbiology laboratory of the hospital. Eleven isolates were collected from various surfaces using Sabouraud dextrose agar (SDA) contact plates (Ø 55 mm) or pre-moistened swabs, depending on the type of surface (flat or irregular) (Table 1). The remaining 3 isolates were recovered from the hands of healthcare workers working in the ICU using the broth bag method (Delfino et al., 2014). The study was approved by the ethical committee of the IRCCS Centro Neurolesi Bonino-Pulejo (study code: HCW_GR2011-02347606; protocol number: E12/17).

2.2. Phenotypic and molecular identification of C. albicans

Before molecular identification, all fungal isolates were subjected to phenotypic characterization using the rapid ID32 C system (bioMerièux, Italy) and conventional tests (germ-tube formation in serum at 37 °C for 2 h and chlamydospores production on corn meal agar at 25 °C for 7 days). The chromogenic medium CHROMagar *Candida* (BD diagnostics, Italy) was also used for presumptive identification of *C. albicans* and recognition of mixed cultures. Subsequently, all yeast isolates were subjected to molecular characterization by using a species-specific PCR-based method which allows to identify properly *C. albicans* by excluding the possible cryptic presence of the closely related yeasts *Candida dubliniensis* and/or *Candida africana* (Romeo and Criseo, 2008).

2.3. ABC genotyping

ABC-genotyping was performed using a simple PCR-based assay which can classify *C. albicans* strains into three different genotypes (A, B and C) on the basis of the presence or absence of a transposable group I intron in DNA sequences encoding 25S rRNA (McCullough et al., 1999).

In vitro amplifications were carried out using 2× Top Taq PCR Master Mix (Qiagen, Italy), DNA template (50 ng) and the following primers: CA-INT-L 5′-ATAAGGGAAGTCGGCAAAATAGATCCGTAA-3′ and CA-INT-R 5′-CCTTGGCTGTGGTTTCGCTAGATAGTAGAT-3′ (McCullough et al., 1999). PCR fragments were analyzed by 1.2% agarose gel electrophoresis and genotypes inferred according to the number and the size of the amplicons obtained: genotype A (~450 bp), genotype B (~840 bp) and genotype C (~450 and ~840 bp) (McCullough et al., 1999).

2.4. Multilocus sequence typing (MLST)

The MLST scheme employed for *C. albicans* genotyping was based on partial amplification and sequencing of seven protein-coding genes (*AAT1a*, *ACC1*, *ADP1*, *MPIb*, *SYA1*, *VPS13* and *ZWF1b*) according to the international reference MLST scheme established for this species (Bougnoux et al., 2003). Seven independent PCR amplifications were performed for each isolate by using the DreamTaq™ PCR master mix, 100 ng of genomic DNA template and 10 μM of each primer. The MLST primer set and the experimental conditions used in PCRs were the same as those reported in previous studies (Bougnoux et al., 2002; Tavanti et al., 2003). The amplified PCR products were examined by agarose gel electrophoresis to confirm the presence of the expected amplicons and then purified with a QIAquick PCR Purification Kit (Qiagen, Milan, Italy). Subsequently, DNA fragments were bi-directionally sequenced at the Eurofins Genomics, Ebersberg, Germany (www.eurofinsgenomics.eu) using the same MLST primers used for PCR.

Each electropherogram was visually checked using the FinchTV v1.4 software (GeoSpiza Inc., Seattle, WA) to confirm base-calls and detect the potential occurrence of heterozygous sites. The sequence data at each locus were used to query sequences deposited into *C. albicans* MLST database (http://pubmlst.org/calbicans) for assigning the allele numbers (Tavanti et al., 2003).

The seven allele numbers were combined to create a diploid sequence type (DST) or a MLST genotype. New allelic combinations (new DSTs) were submitted to the central MLST database where new numbers were assigned by the curator.

Table 1
Origin of *C. albicans* isolates, ABC and MLST genotypes obtained in this study.

Patient	Isolate n°	ABC type	Origin	MLST loci							DST	CC
				AAT1a	*ACC1*	*ADP1*	*MPIb*	*SYA1*	*VPS13*	*ZWF1b*		
P1	IRX-C1	A	Blood	62	12	21	1	42	194	4	3376	S
P2	IRX-C2	B	Blood	2	7	8	4	7	10	8	3377	2
P3	IRX-C3	B	Blood	70	14	8	4	7	10	8	656	2
P4	IRX-C4a*	A	Blood	2	2	5	2	2	6	5	277	1
P4	IRX-C4b*	A	Central Venous Catheter	2	2	5	2	2	6	5	277	1
P4	IRX-C4c*	A	Nasogastric tube	2	2	5	2	2	6	5	277	1
P5	IRX-C5	A	Blood	2	3	5	2	2	6	20	3378	1
–	IRX-En1	A	Bathroom sink	3	3	21	1	34	30	4	3379	9
–	IRX-En2	C	Bedside table	70	14	21	4	7	10	8	3380	2
–	IRX-En3	B	Bedside table	60	13	21	1	7	11	12	3381	6
–	IRX-En4	B	Bed handle	70	14	8	4	42	3	22	3382	S
–	IRX-En5	C	Bed handle	33	22	6	18	64	53	15	3383	42
–	IRX-En6	B	Bed handle	62	13	21	1	7	11	12	3384	6
–	IRX-En7	A	PC keyboard	3	14	8	4	7	3	8	3385	2
–	IRX-En8	A	Patient bed	3	14	8	4	7	3	8	3385	2
–	IRX-En9	B	Patient bed	3	4	8	26	7	4	8	3386	S
–	IRX-En10	B	Interphone	3	14	8	4	7	10	8	3387	2
–	IRX-En11	A	Bed handle	96	7	32	26	2	61	48	3388	27
–	IRX-Ha1	A	Hands (speech therapist)	2	5	5	9	2	24	5	185	1
–	IRX-Ha2	A	Hands (nurse)	3	5	5	2	2	6	5	184	1
–	IRX-Ha3	A	Hands (nurse)	2	5	5	2	2	6	5	69	1

DST = Diploid sequence type assigned by *C. albicans* MLST database; CC = Clonal Complex; *same patient; S = Singleton

2.5. Phylogenetic and population structure analysis

Phylogenetic analysis, using the unweighted pair group method with arithmetic averages (UPGMA) algorithm with the p-distance model, were conducted based upon the concatenated sequences of the seven housekeeping genes. Nucleotide sequences were modified as described by Odds et al. (2007) to label homozygous and heterozygous sites in order to allow the analysis of diploid sequence data with the MEGA7 software (Kumar et al., 2016).

A total of 72 selected DTSs, representing the five most populous *C. albicans* clades (clades 1, 2, 3, 4 and 11) including *C. africana* clade 13 (Chowdhary et al., 2017), were downloaded from the MLST database and included in the phylogenetic analysis. MLST clonal complexes (CCs) were predicted by e-BURST analysis using the goeBURST algorithm implemented in the PHYLOViZ 2.0 software (Nascimento et al., 2017). A clonal complex (CC) was defined to contain at least two DSTs sharing any 6 of the 7 MLST alleles (single-locus variant (SLV) analysis).

BioNumerics software (Applied Maths, Belgium) was used to generate a minimum spanning tree for inferring evolutionary relationships among all known *C. albicans* MLST-genotypes and shape the global population structure of this species.

3. Results

During the three-year period 2015–2018, an important molecular surveillance program for fungal HAIs was implemented at the IRCCS Centro Neurolesi "Bonino-Pulejo" as a measure of quality of care in the ICUs in terms of infection control. Over the course of one of the environmental sampling (June 2017), we recovered a total of 14 *C. albicans* isolates from different hospital surfaces, including hands of healthcare personnel working in the same ICU where environmental sampling was conducted. Based on the laboratory records, 5 patients from this ICU were affected by candidemia due to *C. albicans*. All the patients (4 males and 1 female) showed severe acquired brain injuries caused by acute ischemic stroke (P1 and P3), anoxic encephalopathy (P2), severe respiratory failure (P4) and post-traumatic brain haemorrhage (P5). A total of 7 *C. albicans* isolates were recovered from blood cultures and implanted medical devices of these patients. One patient (P4) yielded multiple isolates from blood, CVC tip and a nasogastric tube respectively (Table 1).

The identity of all *C. albicans* isolates was confirmed by *HWP1* amplification which produced a DNA fragment typical for this species (~1000 bp) (Romeo and Criseo, 2008). By examining the proportion of the ABC genotypes obtained, we found that most part of the isolates belonged to genotype A (12/21; ~57%) followed by genotype B (7/21; ~33%) and C (2/21; ~10%) (Table 1). However most of the genotype B (5/7; ~71%), including the two unique type C isolates, were found in environmental samples recovered from hospital surfaces while type A isolates were isolated mainly from blood samples, medical devices and hands of HCWs (Table 1).

Sequencing of the internal gene fragments (range: 373–491 bp) (Table 2) of the multiple genetic loci included in the MLST scheme resulted in 2883 aligned nucleotides for each examined isolate. A total of 50 different alleles were found in the 21 genotyped *C. albicans* isolates (Table 2). The *VPS13* locus generated the most number of alleles ($n = 10$), while *ADP1* and *SYA1* loci produced the least ($n = 5$ each)

Table 2
Characteristics of the seven MLST housekeeping genes examined in this study.

Locus	Sequenced fragment size (bp)	N° of alleles	N° of polymorphic sites	Nucleotide position
AAT1a	373	7	9	7, 28, 40, 89, 124, 325, 352, 361, 373
ACC1	407	9	7	8, 29, 83, 211, 281, 317, 392
ADP1	443	5	12	35, 40, 46, 109, 125, 166, 205, 215, 225, 229, 232, 352
MPI b	375	6	16	21, 22, 27, 34, 36, 58, 66, 72, 88, 159, 193, 234, 237, 238, 276, 289
SYA1	391	5	7	1, 25, 61, 100, 142, 160, 351
VPS13	403	10	13	49, 53, 134, 212, 217, 241, 281, 282, 320, 322, 334, 370, 375
*ZWF1*b	491	8	9	23, 31, 43, 49, 55, 274, 337, 379, 482
Total	2883	50	73	–

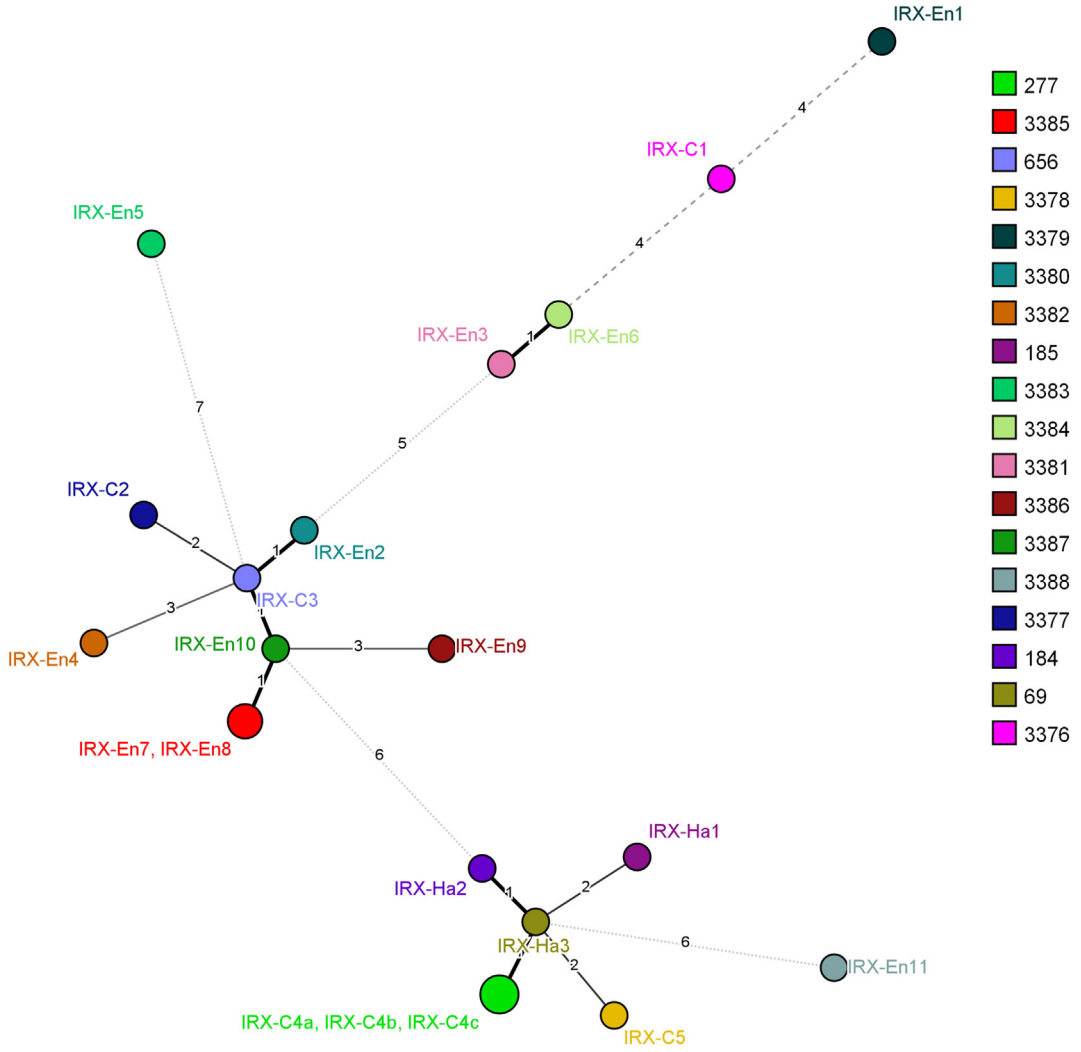

Fig. 1. Minimum spanning tree showing the genetic differences based on a categorical analysis between the MLST genotypes. Each colored circle represents a unique DST with on the side the number and code of isolates of that genotype. Numbers at the connecting lines correspond to the number of different alleles between DSTs. Similarities between MLST genotypes were visualized using BioNumerics version 7.6 treating the data as categorical information.

(Table 2). Overall 73 variable sites were detected among all sequenced loci (Table 2).

The *MPBIb* locus exhibited the highest number of polymorphic sites (n = 16), while *ACC1* and *SYA1* loci displayed the lowest (n = 7).

When all seven alleles numbers were combined, a total of 18 different DSTs were obtained from the panel of examined isolates (Table 1). Interestingly, among these DTSs, only 5 (DSTs: 69, 184, 185, 277 and 656) belonged to previously described MLST genotypes whereas 13 (~72%; DSTs: 3376–3388) (Table 1) were completely new and added to the *C. albicans* MLST central database.

One genotype (DST 277) was recovered from both blood and implanted medical devices (CVC and nasogastric tube) in a single patient (P4) (Table 1) but it was not found in hospital environments or hands of healthcare professionals, supporting the evidence of an endogenous acquisition of the clone rather than its possible horizontal transmission. However, the DST277 was genetically closely related to the DST69 genotype (IRX-Ha3 isolate) recovered from the right hand of a nurse (Fig. 1).

These two genotypes differ only at the ACC1 locus (Table 1) and inspection of the variant allele sequences reveals that they differ by 2 polymorphisms at nucleotide position 211 and 317.

A minimum spanning tree (MStree) showing the genetic differences between our 21 clinical, hands and environmental *C. albicans* isolates is shown in Fig. 1. Using the goeBURST algorithm, the allelic profiles of these isolates were further compared with those of 4391 isolates currently deposited in the *C. albicans* MLST database (as of 04 April 2018). The final dataset contained a total of 2983 confirmed DSTs that were grouped into 137 clonal complexes and 907 singletons. The two largest clusters (CC-1 and CC-2) generated by goeBURST analysis contained over 60% of our *C. albicans* isolates (Table 1; Fig. 2), whereas the remaining isolates were included within relatively less abundant clusters (CC-6, CC-9, CC-27 and CC-42) or were singletons (Table 1; Fig. 2). As showed in Fig. 2, one DST (DST3388), recovered from an isolate contaminating a bed handle in the ICU, was genetically highly related to a group of isolates clustering in the CC-27 containing the DST182 as the putative founding genotype and originally known with the name of *C. africana* (Odds, 2010).

However, unlike *C. africana* strains described so far, the DST3388 isolate (Table 1) was able to produce chlamydospores and assimilate *N*-acetylglucosamine. Nevertheless, UPGMA analysis confirmed the very strong genetic relationship between DST3388 and *C. africana* MLST genotypes deposited in the database by grouping all isolates in a single well-supported clade (bootstrap: 100%) (Fig. 3) previously referred to as MLST-Clade 13, the most divergent evolutionary lineage within *C. albicans* population (Odds, 2010; Chowdhary et al., 2017). Moreover, UPGMA analysis also revealed that more than 85% (18/21) of our isolates fall within the three most populous *C. albicans* MLST-clades: clade 1, clade 4 and clade 11 (Fig. 3) (Odds et al., 2007; McManus and

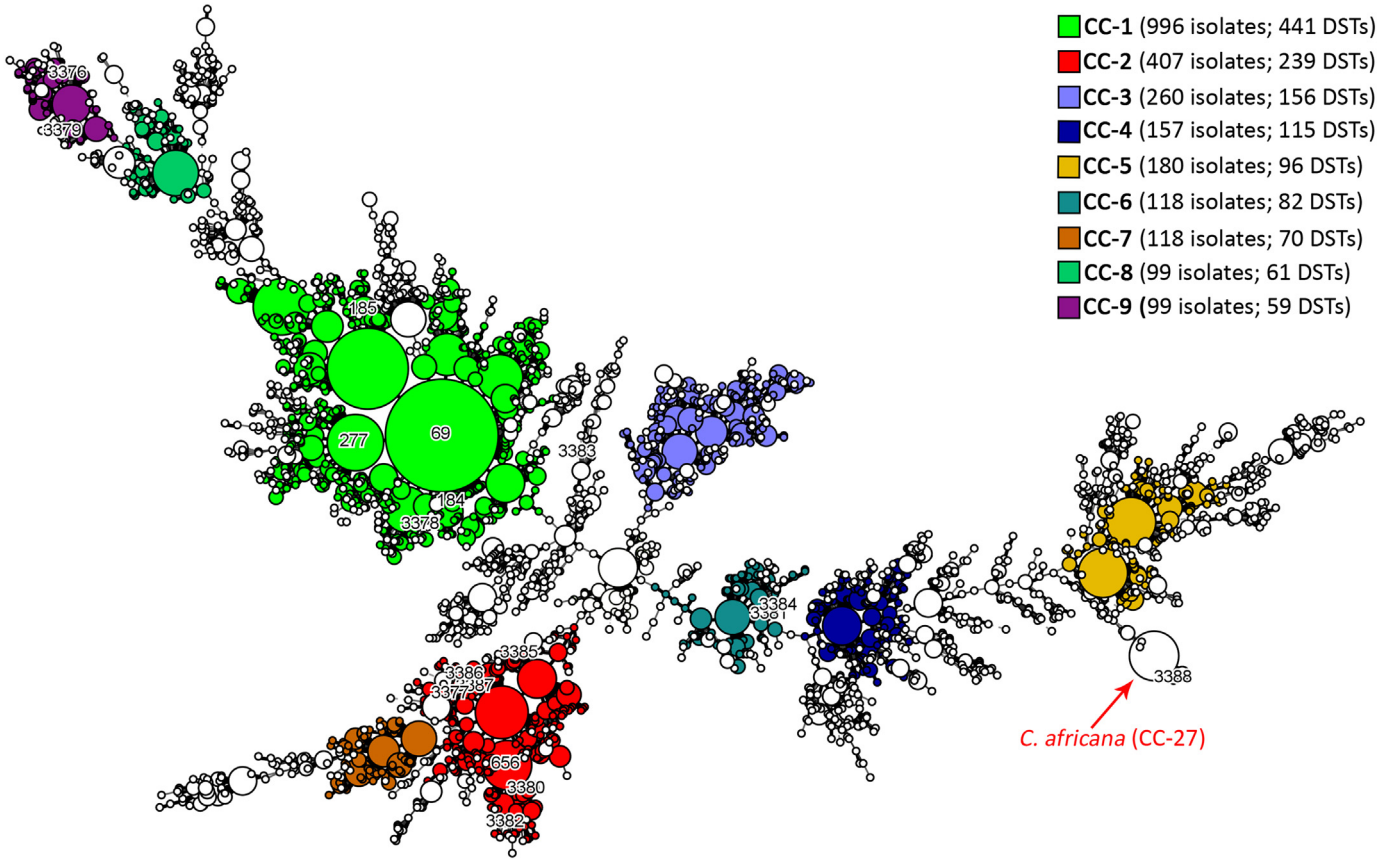

Fig. 2. Minimum spanning tree showing the population structure of *C. albicans* generated using our MLST data including those from 4391 isolates currently deposited in the MLST database. The position of the nine major clonal complexes (CCs) predicted by the goeBURST analysis is indicated, together with the CC-27 containing all *C. africana* MLST genotypes described so far. All the DSTs recovered in this study are also shown above the tree.

Coleman, 2014).

4. Discussion

Recent epidemiological estimates have undoubtedly increased our perception of the role of pathogenic fungi in human infections and shaped an impressive global picture that poses significant future challenges in terms of research, management and treatment of fungal diseases (Bongomin et al., 2017; Denning, 2017; McCarthy et al., 2017). In this context *Candida* species represent a major cause of hospital BSIs (Lamoth et al., 2018), especially in Europe, where approximately 79 cases of candidemia are expected to occur every day of which 29 might have fatal outcome at 30th day (Koehler et al., 2019).

Among the different *Candida* species causing candidemia, *C. albicans* still remains the predominant pathogen (Lamoth et al., 2018; Koehler et al., 2019), probably, due to its innate ability to rapidly evolve strains with increased virulence and antifungal resistance (Popp et al., 2019). This its striking propensity to change genetically is further confirmed by our data which highlight an extraordinary genetic diversity among *C. albicans* isolates recovered from patients with severe neurological disorders including the hospital environments where they are routinely monitored and treated. In fact, in this study, we recovered 18 different MLST genotypes from 21 *C. albicans* isolates and found that 72% of the DSTs could not be assigned to any of the existing *C. albicans* genotypes in the MLST database (http://pubmlst.org/calbicans). Nevertheless, most of the clinical and hands isolates showed well-known MLST genotypes (Odds et al., 2007; McManus and Coleman, 2014) and were grouped in two clonal complexes only (CC-1 and CC-2), while the environmental ones were highly heterogeneous and included in five distinct genetic clusters (CCs: 2, 6, 9, 27 and 42) and two singletons. Interestingly, ABC typing data also indicated a higher genetic differentiation among environmental isolates supporting the hypothesis that in this kind of hospital environment, *C. albicans* is exposed to a range of stressful conditions which promote a more rapid evolution of this fungus (Forche et al., 2011). Nevertheless, based on our genotyping data, there were no evidence of intrahospital transmission of *C. albicans* infection and this is in agreement with recent data reported from Kuwaiti and German hospitals (Huyke et al., 2015; Asadzadeh et al., 2017). However, it should be noted that three isolates from the same patient (P4; Table 1) showed an identical genotype (DST277) which was not found in any other clinical or environmental sample, suggesting that the total genetic diversity observed for *C. albicans* blood isolates is consistent with the endogenous origin of the infections, as reported by others (Escribano et al., 2013; Huyke et al., 2015).

Among the different new genotypes reported in this study, DST3388 seems to be the most interesting as it was genetically closely related to a group of particular MLST genotypes specifically restricted to some *C. albicans* isolates originally known as *C. africana* (Figs. 2 and 3) (Romeo and Criseo, 2011; Chowdhary et al., 2017). Until now, all MLST-genotyped *C. africana* isolates have been grouped in a highly divergent lineage (MLST-clade 13) well-separated from other *C. albicans* clades (Odds, 2010; Chowdhary et al., 2017). Clade 13 isolates display a highly clonal nature and a number of genetic and phenotypic defects including the inability to produce chlamydospores and assimilate *N*-acetylglucosamine (Felice et al., 2016; Ropars et al., 2018). Conversely, our isolate (IRX-En11; DST3388 genotype) (Table 1), was not recognized as *C. africana* either by using molecular nor phenotypic methods of identification. In fact, the IRX-En11 isolate showed all the characteristics, including chlamydospores production and assimilation of the *N*-acetylglucosamine, usually seen in typical *C. albicans* strains

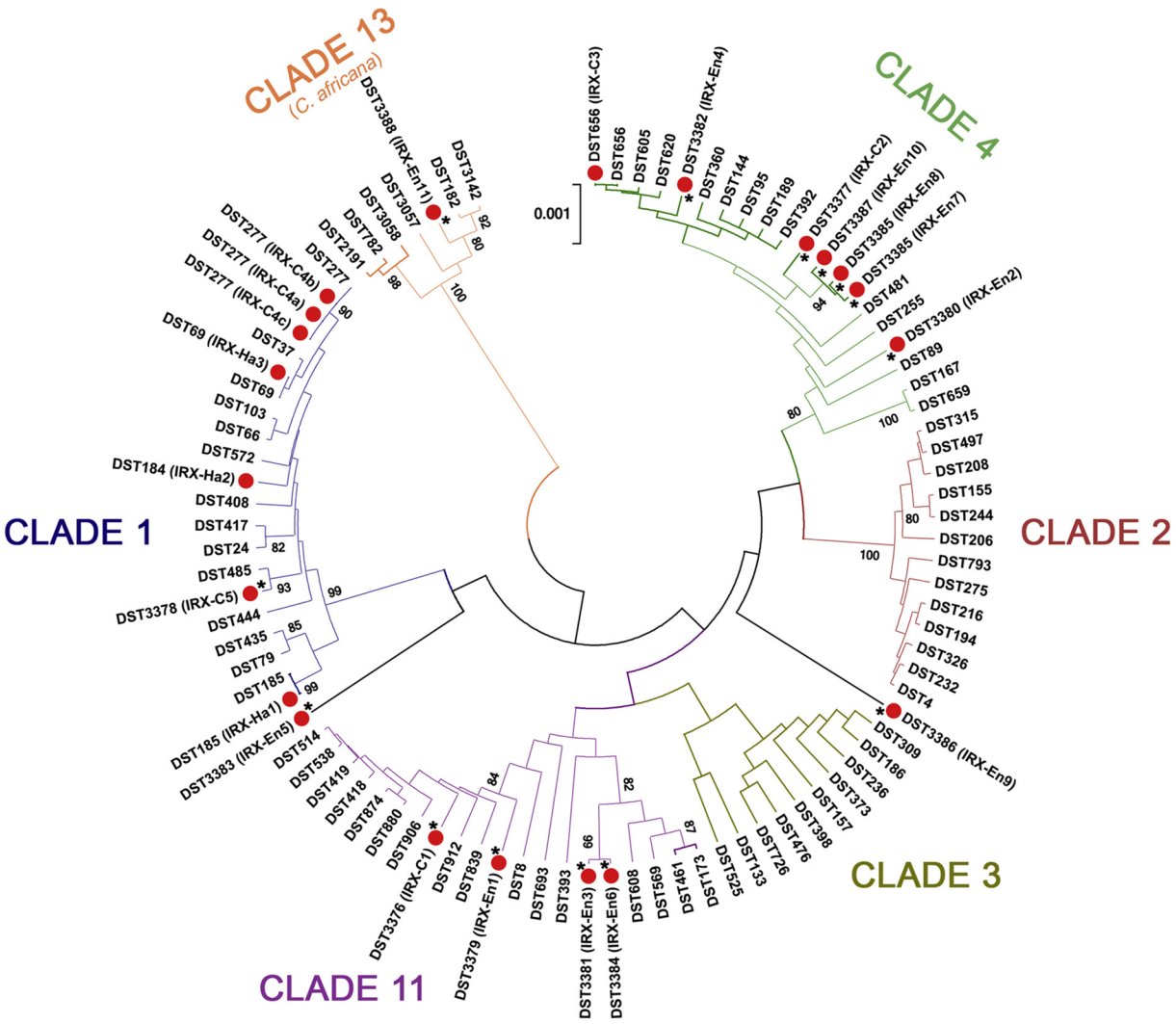

Fig. 3. Evolutionary relationships of 21 *C. albicans* isolates recovered in this study with isolates representing the major MLST clades (1, 2, 3, 4, 11 and 13). The evolutionary history was inferred using the UPGMA method. The evolutionary distances were computed using the p-distance method and are in the units of the number of base differences per site. The analysis involved 93 nucleotide sequences. All positions containing gaps and missing data were eliminated. There were a total of 5766 positions in the final dataset. All the DSTs recovered in this study are marked by a red dot at the end of the tree branches; *Asterisks indicates all new DSTs identified. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

and it was also identified as *C. albicans* by using the partial amplification of the *HWP1* gene (Romeo and Criseo, 2008). Noteworthy, the isolation of the IRX-En11 isolate supports the existence of additional evolutionarily divergent subpopulations of *C. albicans* and is in line with the high dynamism and plasticity of its genome (Ropars et al., 2018; Legrand et al., 2019) which results subject to a persistent diversification through the occurrence of gene flow in the population, most likely, through para(sexuality) (Ropars et al., 2018). In addition, this typical *C. albicans* isolate, clustering in the MLST-clade 13, led us to question the specific association, previously hypothesized in a large-scale MLST study (Chowdhary et al., 2017), between *C. africana* clade 13 isolates and some unusual phenotypes (i.e. lack of chlamydospores production and *N*-acetylglucosamine assimilation) and some genetic polymorphisms (i.e. size of the *HWP1* gene).

In conclusion, this study highlights that the environmental conditions encountered by *C. albicans* in our hospital could play a crucial role in increasing the genetic variability of this species, which in turn shapes the evolution of a wide range of closely related genotypes particularly prone to contaminate hospital surfaces rather than causing outbreak of infections. This is, in part, also confirmed by our internal epidemiological data which rank *C. albicans* as the third most common cause of BSIs after *C. tropicalis* and *C. parapsilosis* (Scordino et al., 2018), two species frequently isolated from our ICUs environments which could exert some antagonistic effects on *C. albicans* by limiting its virulence and its ability to cause invasive infections as recently demonstrated (de Barros et al., 2018).

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Declaration of Competing Interest

None.

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