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Review Article

Quorum sensing in fungi – a review

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Quorum sensing (QS) is a mechanism of microbial communication dependent on cell density that can regulate several behaviors in bacteria such as secretion of virulence factors, biofilm formation, competence and bioluminescence. The existence of fungal QS systems was revealed ten years ago after the discovery that farnesol controls filamentation in the pathogenic polymorphic fungus *Candida albicans*. In the past decade, farnesol has been shown to play multiple roles in *C. albicans* physiology as a signaling molecule and inducing detrimental effects on host cells and other microbes. In addition to farnesol, the aromatic alcohol tyrosol was also found to be a *C. albicans* QS molecule (QSM) controlling growth, morphogenesis and biofilm formation. In *Saccharomyces cerevisiae*, two other aromatic alcohols, phenylethanol and tryptophol were found to be QSMs regulating morphogenesis during nitrogen starvation conditions. Additionally, population density-dependent behaviors that resemble QS have been described in several other fungal species. Although fungal QS research is still in its infancy, its discovery has changed our views about the fungal kingdom and could eventually lead to the development of new antifungal therapeutics.

Keywords: quorum sensing, pathogenic fungi, farnesol, tyrosol, aromatic alcohols, Candida albicans

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Introduction

The regulation of the expression of virulence genes is a crucial step in pathogenesis and in microorganism adaptation to host tissues [1,2]. One successful strategy of regulation of virulence developed by many pathogenic bacteria to overcome host defenses involves synchronizing the expression of virulence factors as a function of the population density in a process known as quorum sensing (QS) [3].

A major mechanism of microbial communication, QS occurs by the continuous release and monitoring of hormone-like molecules called auto-inducers or quorum-sensing molecules (QSM). The concentration of these molecules increases proportionally to the population and, after reaching a critical threshold, a regulatory response is triggered leading to the coordinated expression or repression of QS-dependent target genes in the whole bacterial group [4,5]. QS communication in bacteria was first observed during studies on genetic competence in *Streptococcus pneumoniae* and bioluminescence in marine *Vibrio* species in the 1960s and 1970s, respectively [6]. After that, QS was observed in many bacterial species regulating the most diverse processes including secretion of virulence factors, motility, biofilm formation, sporulation and antibiotic production [3]. In pathogenic microbes, the coordinated expression of virulence factors during infection of a host probably constitutes a significant survival advantage by enhancing the chances of establishing infection and escaping the immune response [7,8]. In addition, some QSMs can be considered virulence factors by themselves, since they are toxic to host cells and/or can modulate host immunity [9].

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Despite the burgeoning amount of information about bacterial QS in the last decades, QS in eukaryotic organisms was unknown until the discovery of farnesol as a QSM in the pathogenic fungus *Candida albicans* [10]. In the 10 years since this groundbreaking work was published, it inspired work that has led to almost 90 PubMed papers with the words '*Candida*' and 'farnesol' and to many others involving the effects of this molecule in other organisms. In addition to farnesol, the other known fungal QSMs are all alcohols derived from aromatic amino acids tyrosine (tyrosol), phenylalanine (phenylethanol) and tryptophan (tryptophol). Tyrosol was the second QSM described in *C. albicans* [11], whereas the other two were initially discovered as autoantibiotics inhibiting filamentation in *C. albicans* in the 1960s [12], and later were found to be *Saccharomyces cerevisiae* QSMs [13]. This review will initially focus on the known QSMs and their physiological effects, followed by other possible fungal QS systems in which the molecule has not been identified.

Discovery of QS in *C. albicans* – farnesol, farnesoic acid and MARS

C. albicans is a polymorphic fungus normally found in the human microbiota that can cause devastating infections in situations when its relationship with the human host is altered by immune suppression or compromise of epithelial barriers. Polymorphism between yeast, hyphae and pseudohyphae forms is critical for its virulence and corresponds to an adaptive response to environmental changes [14–17]. Many triggers for this morphological switch have been described, such as temperature, pH, CO_2 concentration, and the presence of mammalian serum [18].

Additionally, it has long been observed that at densities lower than 10^6 cells/ml, *C. albicans* cells develop into filamentous forms, whereas at higher densities the fungus grows as budding yeasts [10]. Hornby and collaborators discovered that this behavior was controlled by a QSM, which they identified as being the isoprenoid farnesol [10]. In the same year, Oh and colleagues described in a different strain of *C. albicans* that farnesoic acid acts like an autoregulatory substance (ARS) also inhibiting filamentation [19]. Farnesoic acid was found only in a strain of *C. albicans* that does not produce farnesol and its effects are less intense [19,20]. Long before farnesol and farnesoic acid were isolated, a morphogenic autoregulatory substance (MARS) was isolated by Hazen and Cutler [21] that produced similar effects on filamentation of *C. albicans*. However, the chemical features of this molecule differed from farnesol and farnesoic acid and its identity has not been determined yet [20].

Physiological effects of farnesol

Inoculum size effects on filamentation

The first described effects of farnesol as a QSM were on the regulation of *C. albicans* filamentation. Hornby and collaborators showed that commercial and conditioned media (CM)-purified farnesol inhibited the yeast-to-mycelium conversion triggered by three different germ tube formation inducers: L-proline, N-acetylglucosamine, and serum [10]. On the other hand, the isoprenoid had no effects on fungal growth rate, something the authors deemed as an essential feature of a QS molecule [10]. However, despite its effects on yeast-to-mycelium conversion, farnesol showed no effects on cells already committed to mycelial development [22].

Biofilm formation

Biofilms are surface associated organized microbial communities embedded within an exopolymeric matrix [23]. Infections caused by biofilms are difficult to eradicate because these structures are particularly resistant to antimicrobial agents and host immune factors [24,25]. Bacterial biofilm organization is highly dependent on QS, and the interaction between these two processes is considered pivotal in bacterial pathogenesis [26]. Due to the importance of the different *C. albicans* morphological forms in biofilm structure Ramage *et al*., evaluated the effects of farnesol on biofilm development [27]. They observed that in addition to its role in regulating *C. albicans* morphology, farnesol inhibited biofilm formation. Additionally, they showed that the rate of inhibition was dependent on how much time the cells had to adhere before farnesol was added. Once the cells started to filament the addition of farnesol had no effect on the development of biofilm structure but cells on mature biofilms responded to the isoprenoid and this effect may influence biofilm dispersal [20,27]. Microarray analysis of biofilms exposed to farnesol revealed that genes related to drug resistance, cell wall maintenance, cell surface hydrophilicity, iron transport and heat shock proteins were influenced in addition to the genes associated with hyphae formation [24].

Oxidative stress

Westwater and collaborators showed that conditioned medium (CM) from *C. albicans* stationary-phase cultures conferred protection against oxidative stress induced by hydrogen peroxide and by the superoxide anion-generating agents menadione and plumbagin [28]. Purified farnesol partially recapitulated the effects of CM. Additional gene expression experiments showed no induction of an

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oxidative stress response in response to commercial farnesol (35 μM) in *C. albicans*, suggesting a role for farnesol as an antioxidant in addition to a signaling molecule. In another study, Shirtliff and collaborators showed that farnesol concentrations of 40 μM or 100 μM were able to induce upregulation of *C. albicans* proteins involved in protection against oxidative stress [29].

Modulation of drug efflux

Sharma *et al*. showed that farnesol can specifically modulate *C. albicans* drug efflux mediated by ABC multidrug transporters without affecting the multidrug extrusion pump protein CaMdr1p from the major facilitator super family (MFS)[30]. In addition farnesol potentiated the effects to azoles and polyenes by increasing reactive oxygen species (ROS) levels *C. albicans* [30].

Farnesol effects on other microbes

Farnesol was shown to have detrimental effects on many microbes including bacteria and other fungi, such as *Staphylococcus aureus*, *Saccharomyces cerevisiae*, *Aspergillus* species, *Paracoccidioides brasiliensis* and *Mycobacterium smegmatis* [31–38]. This section describes some of those effects.

Saccharomyces cerevisiae. Machida and collaborators reported that 25 μM of farnesol inhibited the growth of *Saccharomyces cerevisiae* without compromising cell viability. The inhibition was linked to G1 cell cycle arrest and a significant decrease in the intracellular diacylglycerol (DAG) levels. The reduced levels of DAG lead to inactivation of the protein kinase C (PKC) signaling pathway that regulates cell proliferation and growth in response to DAG [37]. The same group also showed that farnesol induced the formation of (ROS) by inhibiting the mitochondrial electron transport chain in *S. cerevisiae*, leading to the suggestion that ROS generation was an alternative signal leading to the cell arrest dependent on the PKC pathway [39].

Aspergillus nidulans. The addition of farnesol to cultures of *A. nidulans* had no effect on germ tube development, but triggered apoptosis in this fungus [31]. Farnesol-induced apoptosis was dependent on mitochondrial function and involved the production of (ROS), FadA heterotrimeric G proteins and a putative Poly(ADP-ribose) polymerase (PARP) which acts in response to DNA damage [40]. Similarly, co-cultivation of *C. albicans* and *A. nidulans* harmed the growth and development of the latter suggesting that farnesol might also function to reduce competition with other microbes [31]. In addition, Salvodi and collaborators found that *A. nidulans* farnesol-induced apoptosis is probably dependent on autophagy and protein kinase C (pkcA) function, since an *A. nidulans* mutant of the ATG8 gene, which is required for autophagosome formation was more susceptible to farnesol effects and a protein kinase c mutant (calC2) was more resistant [41]. They further confirmed the role of pkcA in *A. nidulans* farnesol response by showing that overexpression of pkcA enhances the cell death in response to farnesol [42].

Aspergillus niger. In contrast to its effects on *A. nidulans*, farnesol did not impair the growth of *A. niger* but produced noteworthy changes in the morphology of this fungus. Concentrations higher than 10 mM of farnesol completely suppressed conidiation and led to a 10-fold reduction of intracellular levels of cAMP [43].

Aspergillus fumigatus. Dichtl and collaborators showed that farnesol inhibited the growth of wild type cells of the human pathogen *A. fumigatus* [44], albeit to a lower extent of what was observed in *A. nidulans*. The toxicity was lower in osmotically stabilized medium and the addition of farnesol produced more profound inhibitory effects on cell wall mutants of *A. fumigatus*. The authors showed that farnesol acted as a cell wall stress agent, inhibiting the cell wall integrity signaling pathway and leading to misplacement of tip-localized Rho proteins that caused problems in hyphal morphology [44].

Fusarium graminearum. The addition of farnesol to cultures of this pathogen of wheat and other grains induced apoptosis and impaired macroconidia production, development, germination and viability. Due to the devastating economic impact of *F. graminearum* on cereal cultures the group also suggested that the isoprenoid may represent a potential new class of antifungal agent [45].

Candida dubliniensis. Farnesol had no inhibitory effects on *C. dubliniensis* growth, but prevented hypha and pseudohypha formation in this fungus similarly to its effects on *C. albicans* [46].

Candida parapsilosis. Exogenous farnesol causes transitory growth arrest of *C. parapsilosis* [47]. The growth arrest is not restricted to a specific stage in the cell cycle and is reduced in the presence of serum proteins. Additionally, about 20% of *C. parapsilosis* cells die after exposure to 50 μM of farnesol for 2 h, an effect that could be associated with overexpression of genes involved in cell aging

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[47]. Farnesol also inhibited the formation of *C. parapsilosis* biofilms, but because this fungus does not form true hyphae, the mechanisms of action are probably distinct from the inhibition produced on *C. albicans* biofilms. Also, in contrast with *C. albicans* gene expression studies, the main effects of farnesol exposure on *C. parapsilosis* cells were alterations in the expression of oxidoreductases and genes involved in sterol metabolism [47].

Paracoccidioides brasiliensis. Farnesol concentrations of 25–300 μM had a strong inhibitory effect on *P. brasiliensis* growth whereas at lower concentrations the isoprenoid inhibited the morphological transition from yeast to hyphae in this pathogenic dimorphic fungus [34].

Mycobacterium smegmatis. Farnesol inhibited mycobacterial multidrug efflux pumps, decreasing the minimum inhibitory concentrations of ethidium bromide and rifampicin against *M. smegmatis* [36].

Pseudomonas aeruginosa. C. albicans and *Pseudomonas aeruginosa* are commonly found in mixed opportunistic infections and both microbes engage in complex interactions involving their QS systems [48,49]. Hogan and collaborators found that a homoseryl lactone produced by *P. aeruginosa* inhibited filamentation in *C. albicans* without compromising fungal growth [50]. Additionally, *P. aeruginosa* is able to form biofilms on and kill filamentous *C. albicans*, but not *C. albicans* yeast cells. This suggests that the morphological switch triggered when *C. albicans* senses the presence of *P. aeruginosa* (by means of the bacterial QSM) could represent a survival strategy developed by the fungus. On the other hand, addition of farnesol to *P. aeruginosa* cultures inhibited the biosynthesis of *Pseudomonas* quinolone signal (PQS) one of the QS systems used by this bacteria, which consequently decreased production of the PQS-regulated virulence factor pyocyanin [49]. However, in mixed biofilms, where the concentrations of both PQS and farnesol were high, the overall effect was an increased production of phenazines. Similarly, farnesol was also shown to rescue phenazine production in *P. aeruginosa* mutants lacking the quinolone signaling master regulator (LasR) [51]. The group suggests that farnesol effects are due to the induction of ROS, since the effects are decreased in the presence of antioxidants and are similar to the *P. aeruginosa* response to hydrogen peroxide addition [51].

Tyrosol

Tyrosol acts as another QSM in *C. albicans*, decreasing the length of the lag phase of growth, and stimulating filamentation and biofilm formation. These effects are suppressed in the presence of farnesol, suggesting a fine QS-mediated control [11,52]. Additionally, tyrosol was shown to have an inhibitory activity against neutrophils possibly by interfering with the oxidative burst of these phagocytes [53].

Growth effects

Chen and collaborators [11] studied the lag phase time for diluted cultures of *C. albicans* to resume exponential growth, a phenomenon that increases with greater dilutions. The authors showed that the addition of CM shortened this lag phase and that the active molecule in CM was the aromatic alcohol tyrosol, a molecule continuously secreted during *C. albicans* growth. In addition to its effects on growth, tyrosol also stimulates filamentation in opposition to farnesol. However, tyrosol only promoted germ tube formation in conditions that normally induced these structures [11].

Biofilm formation

Tyrosol concentration correlates with the increase of biomass of both planktonic cells and biofilms of *C. albicans* and the addition of tyrosol at early stages of biofilm formation stimulated hyphal growth [52]. However, tyrosol production by dry weight is 50% higher in biofilms. When biofilms were treated with 50 μM of farnesol, higher concentrations of tyrosol (0.1–1 mM) abolished the filamentation-inhibiting effects of the first. In biofilms treated with higher concentrations of farnesol, the addition of tyrosol resulted in biofilms containing mostly yeast cells, suggesting that tyrosol could not counteract the effects of higher concentrations of farnesol. Additionally, experiments using biofilm conditioned medium showed that tyrosol effects exceeds those of farnesol after 14 h of biofilm formation, suggesting that tyrosol acts primarily in the early and intermediate stages of biofilm development [52]. On the other hand, in mature biofilms farnesol activity and concentration surpass tyrosol and possibly have a critical role on the release of yeast cells for biofilm dispersal as suggested before [20,27].

Oxidative stress response

Cremer and collaborators reported that tyrosol from *C. albicans*-conditioned medium inhibited the respiratory burst of human neutrophils [53]. In contrast, Westwater and collaborators did not observe any protective effect of exogenous tyrosol in *C. albicans* cells treated with hydrogen peroxide [28].

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Other aromatic alcohols

Chen and Fink reported that *Saccharomyces cerevisiae* secreted aromatic alcohols as QSMs that control morphogenetic changes in response to nitrogen starvation [13]. The switch from budding yeast to an invasive filamentous form requires the PKA signaling pathway and leads to the expression of Flo11, which is essential for filamentation of *S. cerevisiae*. Aromatic alcohol production in this yeast is tightly regulated by cell density and is repressed by ammonium, suggesting that nutrient-sensing and QS are interconnected as has been reported in some bacterial species [13].

In addition to tyrosol, *C. albicans* also secretes phenylethanol and tryptophol, Aromatic alcohol production in *C. albicans* depends on growth conditions, being stimulated by anaerobiose, alkaline pH and availability of amino acids [54]. However, it is still to be established if those two molecules could act as QSMs in this fungus.

QS signaling pathways and receptors

The signaling cascades controlling the expression of genes under QS regulation in *C. albicans* remain poorly understood. In 2009, Langford and collaborators published a review about signaling pathways possibly implicated in farnesol-mediated QS in *C. albicans* [55]. There is evidence that pathways which were previously implicated in the regulation of filamentation in *C. albicans*, such the Ras-cAMP-PKA pathway and the general repressor TUP1, are involved in the response to farnesol [56–58]. Davis-Hanna and collaborators demonstrated that farnesol inhibits the activity of the Ras-cAMP-Efg1 signaling cascade involved in hyphal formation, and that this hyphal growth can be restored by addition of exogenous dibutyril-cAMP [59]. The same group also showed that oxidative stress induced by farnesol is dependent on Ras1-adenylate cyclase signaling pathway [60]. More recently, Hall and collaborators confirmed the effects of farnesol on cAMP signaling by showing that the isoprenoid directly inhibited the activity of *C. albicans* adenylyl cyclase [61]. Studies analyzing the transcriptional profile of *C. albicans* in response to farnesol have also found upregulation of genes that are exclusively regulated by the MAPK pathway [58]. Additionally, Kruppa and collaborators found that farnesol was not able to inhibit filamentation or biofilm formation in *C. albicans* Chk1p histidine kinase mutants suggesting that this two-component signaling protein could be involved in QS [62]. However, a link between this protein and the cAMP signaling pathway has not been established yet.

These results suggested that farnesol can act through different regulatory pathways in *C. albicans* that are known to be also involved in several other physiological processes [56]. This fact, combined with the knowledge that farnesol is not simply a cell–cell communication molecule indicates great complexity and highlights the difficulty in studying QS using mutants of general signaling pathways or receptors, since some mutations can have overall effects not necessarily dependent on QS.

QS systems in other fungi

In addition to the *C. albicans* and *S. cerevisiae* QSMs, QS activities were also described in *Histoplasma capsulatum* [63], *Ceratocystis ulmi* [64], *Neurospora crassa* [65] and several other fungi. However, the molecules responsible for such activities have not been purified so far. Some of these activities are detailed below.

Histoplasma capsulatum

In *H. capsulatum*, a pathogenic thermo-dimorphic fungus, the regulation of α-[1,3]-glucan synthesis in the cell wall was shown to occur in response to cell density [63]. The α-[1,3]-glucan is absent from the *H. capsulatum* saprophytic mycelial form and its absence on the parasitic yeast cells lead to a loss of virulence by the fungus [66].

Ceratocystis ulmi

The phytopathogen *C. ulmi* also displays an inoculum size effect [64]. Hornby and collaborators reported that this process was mediated by an unknown molecule present in *C. ulmi* supernatants. The active molecule seems to be specific to *C. ulmi*, since the fungus only responded to its own CM and farnesol did not reproduce its effects.

Neurospora crassa

Roca and colleagues showed that the formation of specialized hyphae called conidial anastomosis tubes (CATs) in *N. crassa* was dependent on cell density [65]. CATs are structurally and functionally different from germ tubes and are associated with hyphal fusion. They found that the extent of CAT formation was reduced at low conidial concentrations ($\leq 10^5$ macroconidia/ml) and that this behavior was not dependent on cAMP but that MAP kinase signaling and a putative transmembrane protein are involved in the process [65].

Saccharomyces cerevisiae

Another interesting potential role of QS in *S. cerevisiae* was proposed by Severin and collaborators [67]. They speculated about the causes of apoptosis in unicellular

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organisms and suggested that the decision to undergo apoptosis is proportional to the degree of genetic damage, whose critical value is dependent of population cell density, since cellular suicide only makes evolutionary sense in a population context. They proposed that pheromones and ammonia can act as communication molecules in *S. cerevisiae*. Their affirmation was based on the fact that high concentrations of α mating factor induces apoptosis of yeast cells [68] and that an ammonia concentration gradient is formed during colony aging and this gradient induces the death of old cells close to the centre of the colony while allowing those in the periphery to grow to expand the colony [69]. Additionally, they found that phenylethanol can have a role in such communication systems because it suppressed the survival of yeast cells of the same mating type in stationary cultures kept for a prolonged time, possibly intensifying the signaling for apoptosis.

Cryptococcus neoformans

A QS-like behavior was described for mutants of the general repressor TUP1 of *C. neoformans* serotype D [70]. The mutant did not grow when inoculated at cell densities lower than 10^3 cells/ml. Growth was rescued by the addition of CM from high density cultures of the mutant which contained an oligopeptide responsible for its activity. This peptide had no activity on wild type cells, which were able to grow well in mycological media independently of inoculum size. Additionally, the same group later reported that the TUP1 QS-like effect was not general since it was not observed in TUP1 mutants from strains of *C. neoformans* serotype A, the serotype to which most *C. neoformans* clinical isolates belong [71].

Our group has also been working in *C. neoformans* QS. We observed that growth, melanization and secretion of GXM by *C. neoformans* cells are induced in the presence of CM from stationary phase cultures [72], although we have not yet identified the molecule responsible for such behaviors.

Effects of fungal QSMs in mammalian hosts

As mentioned before, tyrosol impaired neutrophilic killing of *C. albicans in vitro* by inhibiting the respiratory burst [53]. However, it is not known whether tyrosol is produced during infection and whether the concentrations produced would have the same effects.

Using a mouse model of systemic candidiasis, Navarathna and collaborators have shown that endogenous and exogenous farnesol have effects on *C. albicans* virulence [73]. They created a mutant that secreted six times less farnesol by knocking out the DPP3 gene encoding a phosphatase that converts farnesyl pyrophosphate to farnesol. Toxicity of exogenous farnesol was evaluated by oral or intraperitoneal administration in mice previously infected with *C. albicans*. They observed an enhanced mortality in the *C. albicans*-infected group that also received farnesol in comparison with the *C. albicans*-infected group, whereas farnesol alone had no detrimental effects on mice. Based on these results, they concluded that farnesol may represent a new virulence factor in *C. albicans* [73]. In another study, administration of farnesol to mice before intravenous infection with *C. albicans* reduced the levels of Th1 cytokines IFN-γ and IL-12 while inducing the expression of the Th2 cytokine IL-5, suggesting that farnesol could also inhibit proper immune responses to systemic infection with *C. albicans* [74].

Additionally, farnesol augmented the response to *C. albicans* cell wall pathogen-associated molecular patterns (PAMPs), increasing the expression of inflammatory and regulatory cytokines in murine macrophage cell lines *in vitro* [75]. In the absence of cell wall PAMPs farnesol inhibited the production of IL-6, a key cytokine in resistance to mucosal and systemic candidiasis [75,76].

In oral candidiasis models, farnesol treatment inhibited the formation of tongue lesions and decreased the loss of weight in infected mice [77]. Farnesol also inhibited murine macrophage antifungal activity against *C. albicans* by inducing oxidative stress and apoptosis of macrophages [33]. Finally, farnesol induced apoptosis and necrosis of human spermatozoa suggesting a possible link between microbes and host infertility [35].Thus, farnesol seems to play multiple roles during infection both as a QSM and as a virulence factor per se.

Conclusions and perspective

Although cell density-dependent phenomena in fungi had been described for a long time, their characterization as QS and the identification of QSMs resulted in a remarkable change in how we view this group of organisms. However, only farnesol, farnesoic acid and three aromatic alcohols have been established as fungal QSMs. Many behaviors that are dependent on cell density and thus potentially regulated by QS have been described in fungi, but the molecules and pathways involved in those processes remain unknown at this time. It is hard to conceive that a kingdom so diverse as the fungi will rely only on a few QSMs and we believe that future work in this field will uncover many exciting new QS systems.

An important issue in the field is the absence of a consensus regarding the definition of fungal QSM. Based on two different sets of criteria previously suggested for bacterial QSMs [78,79], we propose that a fungal QSM should: (1) accumulate in the extracellular environment during fungal growth; (2) accumulate in a concentration that is proportional to the population cell density with its effects

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restricted to a specific stage of growth; (3) induce a coordinated response in the entire population that is not simply an adaptation meant to metabolize or detoxify the molecule itself after a threshold concentration is reached; (4) reproduce the QS phenotype when added to the culture exogenously; (5) not be solely a byproduct of fungal catabolism. Given that only a few fungal QSM have been isolated thus far and that fungi could have developed different solutions for intercellular communication during the immense evolutionary gap that separates them from bacteria, these criteria might need to be refined in the future.

An example of the problem that this lack of consensus in the terminology of microbial communication is that many researchers think that a QSM could not act to stimulate microbial growth as tyrosol does in *C. albicans*. Consequently, tyrosol is considered by many investigators as a minor QSM in *C. albicans*, and it has been less studied than farnesol [20,80]. However, a situation that has not been explored is the growth of *C. albicans* in anaerobic conditions, despite the fact that this fungus is part of the gastrointestinal microbiota. Interestingly, *C. albicans* does not produce and does not respond to farnesol under anaerobiosis [81]. On the other hand, the production of tyrosol and other aromatic alcohols in *C. albicans* is enhanced in anaerobic conditions [54]. However, as far as we know, the role of tyrosol as a QSM in *C. albicans* under low oxygen concentrations has not been investigated. Additionally, the idea that QS can only be involved in repression of growth was challenged with the discovery that QS can have either positive or negative effects on growth rates of *Vibrio harveyi*, a classical model of QS studies [82]. A very similar activity is reported by the so-called resuscitation factors of *Micrococcus luteus* and *M. smegmatis* which are responsible for recovering growth in viable non-replicating cells of these microbes after a prolonged stationary phase [83,84].

So far, the most studied fungal QS molecule is farnesol. Much has been learned about its roles on controlling *C. albicans* morphology, and interactions with other microbes and host cells. Furthermore, the studies on tyrosol reinforce the importance of QS for *C. albicans*, but also leave room for the speculation that other QSMs might exist in *C. albicans*. Given their importance in virulence, bacterial QS systems are being studied for the development of new therapeutic strategies [85,86]. The same could happen in fungi as new QS molecules and pathways are revealed in these organisms, potentially leading to more effective and less toxic treatment strategies for fungal infections.

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