CELL SIGNALLING

Quorum sensing with pheromones

The vast majority of fungi reproduce sexually and use secreted pheromones to signal to each other. A study now shows that these signalling molecules in the fungal plant pathogen *Fusarium oxysporum* activate a density-dependent autocrine signal that controls asexual spore germination.

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From studies on unicellular fungi (yeasts), we understand the mechanism of pheromone signalling in great detail[1,2]. For instance, in *Saccharomyces cerevisiae*, α-type cells secrete α-pheromone, which is detected by the specific G-protein coupled receptor (GPCR) Ste2 in a-type cells. Conversely, a-cells secrete the a-pheromone, which is detected by Ste3 GPCR in α-cells. Each mating type expresses a single pheromone and the cognate receptor for the partner's pheromone (Fig. 1a). Not so in *Fusarium oxysporum*! This plant pathogen, which causes wilt diseases in crops, has no reported sexual cycle[3]. Nevertheless, the *F. oxysporum* genome encodes both pheromones and both pheromone receptors. Turra et al. now show that *F. oxysporum* cells express both receptors and respond to both signals (Fig. 1b)[4]. In an elegant co-labelling experiment, they demonstrate that both signals are perceived by the same cell, which co-internalizes both labelled α-factor (which binds with Ste2) and labelled Ste3 (which is bound by a-factor). Thus, pheromones elicit autocrine, rather than paracrine, signalling in *F. oxysporum*.

Interestingly, α- and a-pheromone are sensed by the cells in a competitive manner. Indeed, when faced with competing gradients of α- and a-pheromones in a directional growth assay, the cells display preferential growth towards a-factor. The reason for this is that α-factor is degraded by the secreted protease Bar1; in absence of Bar1, competing a- and α-factor gradients induce a stalemate. The interesting observation is that disrupting a-sensing promotes chemotropism towards α-factor and, conversely, disrupting α-sensing enhances chemotropism towards a-factor. As both pheromones elicit the same intracellular signalling, involving Rho GTPase and a mitogen-activated protein kinase (MAPK) cascade, this likely reflects competition for limited signalling molecules inside the cell. Of note, this MAPK cascade is distinct from the one controlling chemotropism in *S. cerevisiae* and is instead homologous to that monitoring cell wall integrity[5], indicating significant re-wiring of the signal transduction pathway during evolution (Fig. 1b).

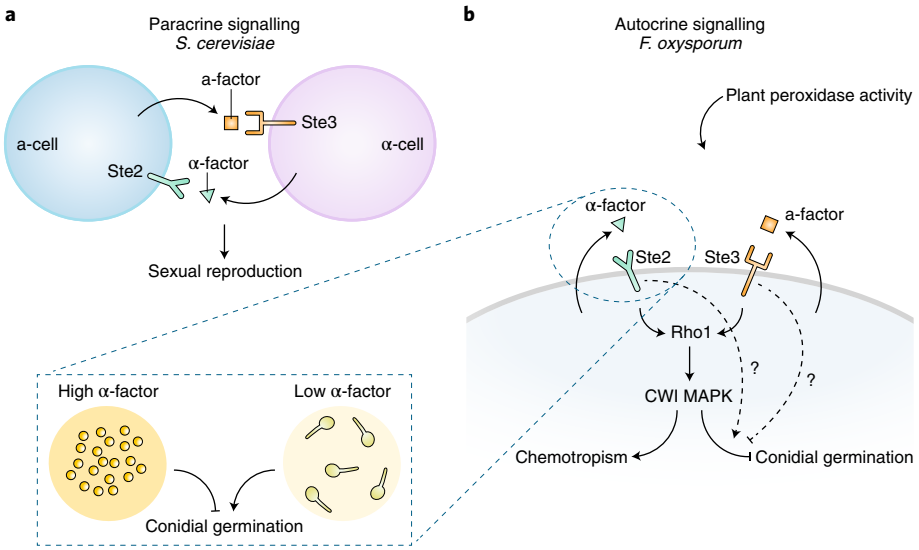

Fig. 1 | Paracrine versus autocrine pheromone signalling. a, *S. cerevisiae* α-type cells secrete α-pheromone, which is detected by Ste2 GPCR in a-type cells, while a-cells secrete the a-pheromone, which is detected by Ste3 GPCR in α-cells. Paracrine pheromone signalling will ultimately lead to yeast cell reproduction. **b**, *F. oxysporum* cells express both Ste2 and Ste3 receptors and respond to α- and a-pheromones. While α-pheromone signalling via Ste2 inhibits conidial germination at high cell densities, lower concentrations of α-pheromone at low cell densities promotes conidial germination (inset). Of note, Ste2 in *F. oxysporum* also responds to the activity of plant peroxidases to promote chemotropic growth. CWI, cell wall integrity.

Because *F. oxysporum* does not exhibit sexual reproduction, these findings of competitive pheromone signalling inside single cells raise the question of the physiological role for pheromone sensing in this organism. Indeed, because pheromone signalling is autocrine, whether the cell uses pheromones as chemo-attractants is unclear. Importantly, *F. oxysporum* hyphae appear to not only respond to, but also produce, both α- and a-factors. Indeed, water-soluble, heat-resistant peptides from hyphal exudates elicit the same responses as synthetic pheromones. Interestingly, Turra et al. found that pheromones are physiologically important to regulate the germination of asexual spores (called conidia), an important initial event for plant invasion[3]. They first discovered that conidial germination was significantly inhibited at high, but not low, cell densities, indicative of quorum sensing. Quorum sensing is well defined in bacteria, but is also prevalent in fungi[6,7]. In these conditions, the pheromones were also upregulated. Remarkably, the inhibition of germination at high cell densities was partly relieved by blocking α-factor signalling. Conversely, germination at low cell densities was inhibited by addition of synthetic α-factor or by deletion of the α-factor protease Bar1. Thus, at high cell densities, high α-factor levels inhibit conidial germination, whereas at low cell

densities, lower α-factor levels are efficiently degraded by Bar1, thus permitting efficient germination (Fig. 1b, inset). These findings demonstrate that the α-pheromone is a quorum sensing molecule that adapts germination success to cell density.

Unexpectedly, a-factor signalling produces the opposite effect: addition of a-factor promoted germination at high cell densities; conversely, blocking a-factor signalling partly compromised germination at lower densities. Because a- and α-factor signalling occurs through a common MAPK cascade that leads to an identical output for chemotropism, this raises the question as to how the cell discriminates between the two signals during conidial germination. The authors speculate that α-factor–Ste2 signalling represses germination by activating not only the MAPK cascade, but also an additional pathway that synergizes with the MAPK signal at high cell densities. Alternatively, a-factor–Ste3 signalling may elicit a second pathway that antagonizes the inhibitory effect of the MAPK cascade on germination (Fig. 1b).

There are a number of other examples of shared signalling pathways that elicit distinct outcomes depending on input. For instance, the filamentous growth and pheromone pathways in *S. cerevisiae* are activated by distinct signals (nutrients and pheromones, respectively) that use essentially the same MAPK core to promote distinct transcriptional programmes and physiological responses[8]. GPCRs are also known to be able to differentiate between distinct activating ligands in yielding appropriate downstream signals[9]. Understanding how the Ste2 and Ste3 GPCRs transduce appropriate signals will be all the more interesting in *F. oxysporum*, as Ste2 not only responds to α-factor but also to plant-secreted peroxidases, which provide chemo-attractive signalling[5]. It will be fascinating to discover how the cell iteratively and differentially uses the same receptors during plant infection to discriminate quorum sensing during germination from plant perception during chemotropic growth. ❐

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References

1. Martin, S. G. *J. Cell. Sci.* **132**, jcs230706 (2019).
2. Alvaro, C. G. & Thorner, J. *J. Biol. Chem.* **291**, 7788–7795 (2016).
3. Gordon, T. R. *Annu. Rev. Phytopathol.* **55**, 23–39 (2017).
4. Vitale, S., Di Pietro, A. & Turra, D. *Nat. Microbiol.* https://doi.org/10.1038/s41564-019-0456-z (2019).
5. Turra, D., El Ghalid, M., Rossi, F. & Di Pietro, A. *Nature* **527**, 521–524 (2015).
6. Hogan, D. A. *Eukaryot. Cell* **5**, 613–619 (2006).
7. Mehmood, A. et al. *Molecules* **24**, 1950 (2019).
8. Saito, H. *Curr. Opin. Microbiol.* **13**, 677–683 (2010).
9. Stallaert, W., Christopoulos, A. & Bouvier, M. *Expert Opin. Drug Dis.* **6**, 811–825 (2011).

Competing interests

The author declares no competing interests.