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> ONE MONTH, ONE PROTEIN <

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give and take

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Survival is the essence of life. This may sound like an abysmal platitude but, in the living world, the act of survival implies an awful lot. To survive, many animals eat other animals, which they must first kill. Birds swallow seeds thus depriving them of a chance to grow into plants. Fungi destroy crops as they use them for their own reproduction. So life, or survival, is also strongly associated with death. A lot is going on at the molecular level too, where myriads of pathways are set into action as a response to nutrition, to infection or to a predator's attack. Plants are intriguing in that their survival cannot depend on mobility: they are unable to flee predators or infection, and quite unfitted to run after prey. Their survival depends on how their stems and leaves develop and move to catch sunlight, for instance, as well as on their means to fight off pathogens. For this, they may even benefit from the help of another species. One example has been described between poplar trees and fungi, in particular the fungus *Trichoderma asperellum*. *T.asperellum*, like all fungi, expresses small proteins known as hydrophobins which have a role in fungal growth and defence. One of these hydrophobins, HFB2-6, can prompt poplar signalling pathways that are crucial for the tree's own growth and defence.

Paysage à Marcillac

by Félix Vallotton (1865-1925)

Trichoderma asperellum is a widespread fungus which thrives in all sorts of environments: from fields to forests, in soil, decaying organic matter and plant roots. It frequently acts as a beneficial symbiont for plants by shielding the plant's roots from infection by other fungi. This is true for poplar plants, for instance. *T.asperellum* is known to colonize poplar roots. Colonization – which, in other instances could be called infection – is tolerated by the plant because the fungus may well supply nutrients while also offering protection against certain phytopathogens. Naturally, the fungus doesn't offer pathogen protection out of pure altruism but it's a sure way of keeping its source of nutrients – in other words the host plant – alive.

Poplar trees have been of great interest to humans for a long time and for a variety of reasons. Rapid growth is one. Belonging to the *Populus* genus, of which there are about 30 species, they are fast-growing trees and can thus be used to cover land with forests that do not take centuries to grow. In some countries, poplars are grown to line the edges of fields to protect their crops from wind erosion. Today, without feeling too guilty, poplars can be used commercially and on a relatively wide scale, in the knowledge that the trees that have been felled will be rapidly replaced. Poplars not only grow fast but they also spread rapidly, as a broken branch easily take root again. This is how colonies have literally sprung from just one original plant. The Pando Forest in the USA is a stunning example. Estimated to be about one thousand years old, almost 50'000 quaking aspen (*Populus tremuloides*) stems – all of which are genetically identical – are connected by one massive root system that stretches over an area as big as 75 football fields.

T.asperellum will undoubtedly have been involved in such a massive spread and growth of trees. We know, for instance, that the fungal hydrophobin HFB2-6 is involved in colonizing plant roots. We also know that it has a role in protecting plants against phytopathogens. Both roles would have helped to promote the expansion of poplar forests. How though, does HFB2-6 do this? HFB2-6 is a hydrophobin. Hydrophobins are small globular proteins that are found exclusively in filamentous fungi such as *T.asperellum*. They are secreted onto the surface of fungal hyphae where they self-assemble into rodlets to form a protective sheath. This sheath interacts with the environment to promote fungal growth, development and dispersal, but also host colonization – or indeed infection. Hydrophobins all sport eight cysteine residues which form four disulfide bridges. The smallness of the protein (*circa* 100-200 amino acids) and the existence of four disulfide bridges packs the protein sequence into a small bulbous shape that is particularly stable.

Besides forming a protective fungal sheath and their involvement in colonizing plant roots, hydrophobins are also highly expressed when *T.asperellum* is suffering from biotic or abiotic stress. This is echoed in the promoter region of HFB2-6's gene which harbours a series of what are known as 'stress response elements'. These are short stretches of DNA that respond to environmental stress – such as dehydration, low temperatures, starvation, drought and pathogens – by boosting HFB2-6 synthesis. The wonderful part is that poplar trees seem to benefit from this HFB2-6 boost by literally acquiring protection from phytopathogens (namely the very common fungus *Alternaria alternata*) while also being stimulated to grow.

How can this occur? How can a fungal hydrophobin influence the immunity and growth of its host? A plant to boot. There must be some kind of direct communication between *T.asperellum*'s protective sheath and the roots of the poplar plant. There is, although the molecular details are still very scant. It seems that, one way or another, HFB2-6 is capable of triggering off three crucial plant signalling pathways: the jasmonic acid signalling pathway, the salicylic acid signalling pathway and the auxin signalling pathway – all of which are equally set off naturally by plants as a response to environmental stress. The jasmonic and salicylic acid pathways induce host systemic resistance (ISR), and systemic acquired resistance (SAR), giving poplars broad-spectrum resistance to pathogens, while the auxin pathway promotes tree growth. Highly intertwined, they communicate with one another to form complex and dynamic networks whose ultimate aim is to balance the plant's growth with its response to environmental stress. In a way, HFB2-6 acts as a vaccine against phytopathogens for the poplar tree. Consequently, when the plant is actually attacked by a phytopathogen such as *Alternaria alternata*, it already has all it needs to protect itself.

This is a wonderful example of cross-talk between species to create an atmosphere of mutual assistance where each protagonist benefits from the other's presence. Understanding how hydrophobins, such as HFB2-6, cross-talk with other poplar trees may help farmers who grow poplar trees commercially. Indeed, besides afforestation, poplar has been used by humans for centuries and for so many different things. Its wood is particularly flexible, and is used to surround cheeses such as French Camembert or Swiss Vacherin, and to make the bodies of musical instruments like guitars. The widely-used 'plywood' is usually made with poplar wood, as are cheap wooden pallets. Meanwhile, poplar pulpwood is used to manufacture paper. Of great interest, too, is the faculty of poplar trees to clean up contaminated environments by absorbing, degrading and neutralising pollutants such as heavy metals, pesticides and hydrocarbons… All excellent reasons for us to understand better how hydrophobins talk to plants.

Cross-references to UniProt

Class II hydrophobin 6, *Trichoderma asperellum* (Filamentous fungus): I7AQG9

References

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