Article addendum

PHYSARUM ATTRACTION: WHY SLIME MOULD BEHAVES AS CATS DO?

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A plasmodium, the vegetative phase of the acellular slime mould *Physarum polycephalum*, has recently became a popular biological substrate for making experimental laboratory prototypes of living computing devices[1]. The Physarum-based computing devices are programmed using gradients fields generated by discrete configurations of chemo-attractants. Laboratory experiments shown that the plasmodium of *P. polycephalum* is attracted to glucose, maltose, mannose, galactose, many aminoacids (e.g. phenylalanine, leucine, serine, asparagine, theonine). Recently we found, see paper[2], that the plasmodium is strongly attracted to herbal calming/somnipherous tablets Nytol[3] and Kalms Sleep[4]. To select the principle chemo-attractant in the tablets we undertook laboratory experiments on the plasmodium's binary choice between samples of dried herbs/roots: *Valeriana officinalis*, *Humulus lupulus*, *Passiflora incarnate*, *Lactuca virosa*, *Gentiana lutea*, *Verbena officinalis* (Fig. 1). We constructed a hierarchy of chemo-attractive force (Fig. 2) and found that Valerian root was the strongest chemo-attractant for *P. polycephalum*[2]. A possible link between sedative activity of valerian and its chemo-attraction --- via relaxation of contractile activities --- is outlined in[2].

Valerian contains hundreds of identified, and possibly the same amount of not yet identified, components including alkaloids, volatile oils, valerinol, and actinidine (Fig. 3a)[5-8]. We can postulate that slime mould in the plasmodium stage may be attracted to a plant because the plant roots or stem harbour high level of food (bacteria), or the plant may provide protection for the slime mould from insect predators (e.g. fungus gnats, round fungus beetle, many plants including *Nepeta cataria* are known to have insecticidal secretions), the plant's volatile secretions “pheromones” may be chemically similar to “pheromones” of slime mould. This may be coincidence or there may be some beneficial symbiosis between certain plants and slime moulds (An example is the case of the recently discovered night flowering orchid which scientists believe mimics the plasmodial stage of a slime mould visually (chemically?) in order to attract pollinators.)

Actinidine is structurally close to the terpenoid nepetalactone (Fig. 3b), the active substance of catnip *Nepeta cataria. N*epetalactone and actinidine both have a similar bicyclic structural skeleton, and are classed as monoterpene derivatives. Actinidine as well as having the same dramatic attractive effect as Nepetalactone on cats, rats etc. is also a pheromone or allomone of many insect species (ant, stick insect). Boschniakine (Fig. 3h) also acts as a defence substance for stick insects[9], and shares significant chemical structure with actinidine. So we may expect Boschniakine to impart a chemoattractive effect on Slime moulds if the cyclic functionality is important for this action. It should be noted that in our experiments catnip exhibited a lesser slime mould attractive potential then valerian,[2] thus we can postulate that despite their structural similarities actinidine and nepetalactone act differently on the slime mould's metabolism. However, it is interesting that the two substances show such a range of activity across many species.

Isovaleric acid (Fig. 3f) and actinidine (Fig. 3a) are identified in the anal gland secretion of *Iridomyrmex nitidiceps* ant, and isovaleric acid is considered to be a distress indicator[10]. We can speculate that these components are also pheromones of *P. polycephalum* and can be considered in a framework of pheromones of cellular slime moulds[11-13] (indeed, there may be pitfalls in projecting physiology of cellular mould to their acellular counterparts).

The slime moulds are fairly primitive organisms therefore we could *argue that receptor bound by actinidine is a generic one and not developed to impart selectivity to one specific chemica*l.

In 1979 Kincaid and Mansour[14] found that inhibitors of the enzyme cyclic 3’,5’-AMP-phosphodiesterase act as strong or moderate chemoattractants in *P. polycephalum*. Amongst

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substances tested strongest effect was observed with 4-(-3-butoxy-4-methoxybenzyl)-2-imidazolidinone (Fig. 3d) and moderate effects from theophylline (Fig. 3g) and other xanthine derivatives (interestingly they observed negative chemotaxis at high concentrations). Theopylline (Fig. 3g) is quite similar to caffeine and has a similar chemical structure to actinidine (Fig. 3a). They are bicyclic alkaloid/terpenoid molecules although the functionaliszation is distinct. Conversely, Nepetalactone does (Fig. 3b) not share the same structural similarities to the xanthine derivatives as actinidine. This may be the reason for the observed lesser effect as a chemoattractant for the slime mould *P. polycephalum*.

Acrasins (like cAMP), e.g. glorin[15] (Fig. 3c), which are implicated in the aggregation of slime moulds (not specifically physarum) also have certain structural similarities to compounds found in valerian. Limonene (Fig. 3e) and other terpenes have been found to bind to A_{2A} adenosine receptors[16]. Other antagonists are caffeine, theophylline (Fig.3g), istradefylline. So molecules with very limited structural similarity can bind to major receptors and impart a range of metabolic effects on various species.

In conclusion even though — the chemical structures of actinidine and nepetalacton are quite different — they induce the same behaviour in cats, rats and act as strong or moderate attractants for slime moulds. Thus we can postulate that the receptors involved are very non-specific and may have shared structure between primitive organisms and higher organisms. Therefore, there is significant “crosstalk” between pheromone like molecules and mimics — it appears especially when molecules have cyclic structure.

The original paper although searching for chemoattractants for applied research highlights the need for fundamental research into pheromones and chemo-attracts of primitive organisms such as slime moulds. The results show that significant information could be gained about the action of compounds on higher organisms.

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FIGURES

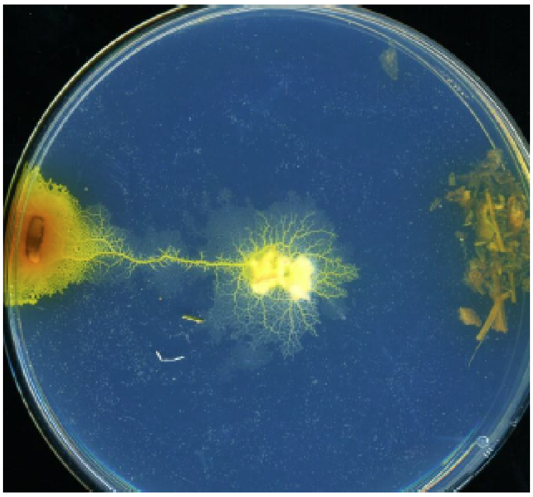


Figure 1. An exemplar experimental setup. The plasmodium is inoculate in the centre of a Petri dish and two portions of substances (Valerian root on the left and catnip on the right) and are placed at the end of diameter segment. See details in [1].

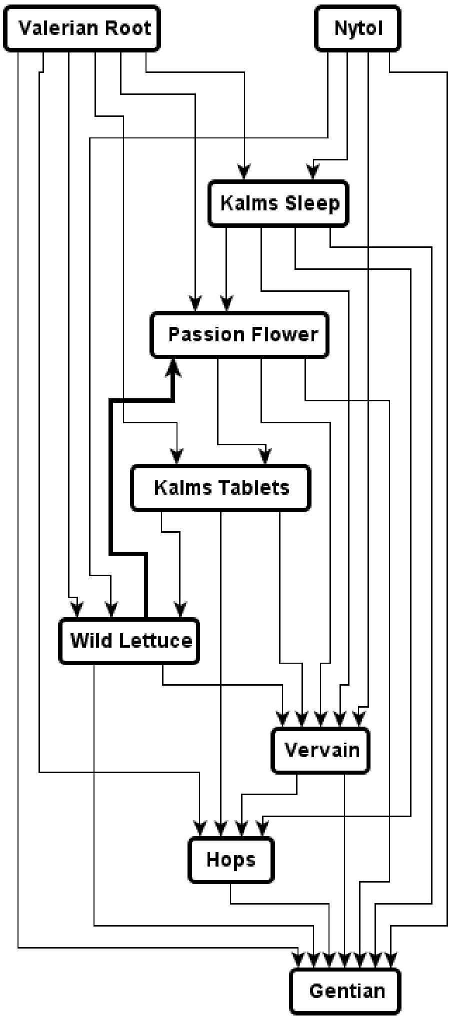
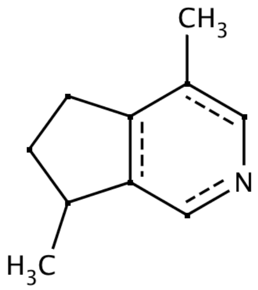
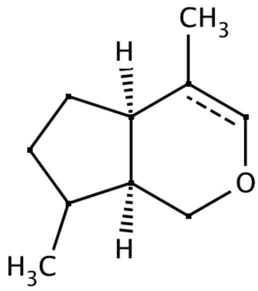


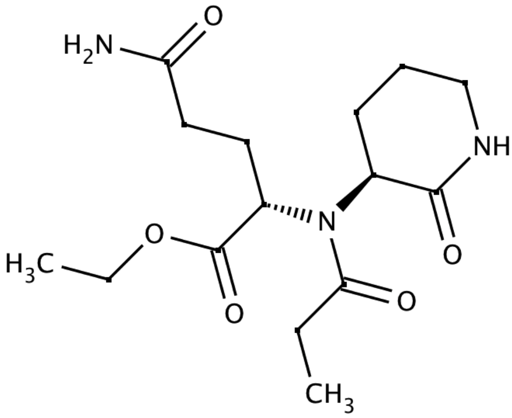
Figure 2. Hierarchy of Physarum preferences. The higher is a substance positioned in the hierarchy the more strongly the substance attracts *P. polycephalum*. From [1].



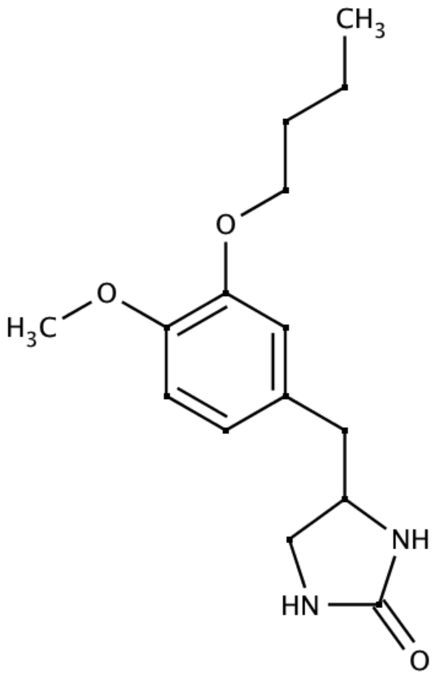
(a) Actinidine



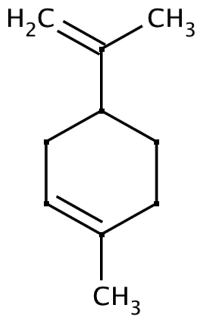
(b) Nepetalacton



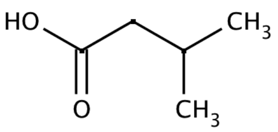
(c) Glorin



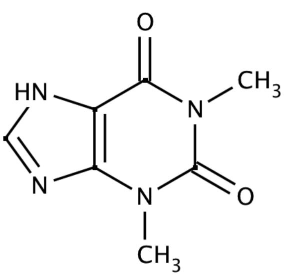
(d) 4-(3-Butoxy-4-methoxybenzyl)-2-imidazolidinone



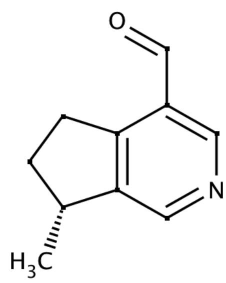
(e) Limonene



(f) Isovaleric acid



(g) Theophylline



(h) Boschniakine

Figure 3. Chemical structures of substances attracting slime moulds.**

** We used the ChemSpider (`http://www.chemspider.com`) chemical database for sourcing the structures and Marvin Sketch (`http://www.chemaxon.com`) for displaying and manipulating them.