Introduction
Molecules from nature have an excellent proven track record of providing lead compounds for pharmaceutical and agrochemical products. The research of Hypha Discovery is aimed at stimulating the production of novel bioactive molecules from tropical higher fungi (Figure 2), a new method that stimulates fermentations of fungi has been developed. Basidiomycetes, a spore producing group of fungi, are difficult to grow effectively under laboratory conditions and are notoriously unproductive. Using the new stimulatory technique, a vast promotion of chemical production obtained when using Hypha’s unique stimulatory conditions is shown in the chromatographic analysis (Figure 3).

Solid Phase Extraction of Biologically Active Compounds after Stimulatory Fermentation of Basidiomycetes

Figure 1: Production of novel biologically active molecules from temperate and tropical mushrooms and toadstools

Fractionated Solid Phase Extraction

The primary extracts generated from the proprietary process were processed through Solid Phase Extraction cartridges (Bond Elut Plexa®) to remove macromolecules known to cause interferences in target based screening (proteins / enzymes / antibiotics). Bond Elut Plexa is a new type of polymeric SPE product, designed for improved analytical performance and ease of use. The base material is a macroporous styrene divinylbenzene co-polymer. The monodisperse polymer particles are functionalized with hydrophilic substituents. These hydrophilic-containing moieties create a ‘water rich’ environment on the polymer particles at the hydrophobic core. Proteins and polysaccharides are removed, the compounds of interest can then be selectively eluted in a buffer-wash step. With the macromolecules thus washed away in a buffer-wash step. With the macromolecules thus removed, the compounds of interest can then be selectively eluted according to their polarities.

The Method

The fermented sample is loaded on to the 500 mg Bond Elut Plexa cartridge in the presence of buffer enabling retention of very polar N components from the broth sample.

The SPE on Bond Elut Plexa works using a combination of extraction and fractionation mechanisms. When compared to other polymeric materials we have used at Hypha, this material greatly increases the precision and speed of our research. The advanced design of the pore facilitates a highly efficient sample throughput by allowing us to quickly remove unwanted sources of assay interferences such as proteins and oligosaccharides.

The advanced pore architecture facilitates a highly efficient sample throughput by allowing quick removal of unwanted assay interference, in addition to offering compound fractionation typically not found in SPE methods (Figure 5).

Results

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Conclusion

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