

Sterols used in chemical defence of the freshwater heterotrich ciliate Stentor polymorphus .

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Many species of ciliates belonging to the order Heterotrichida, are able to release toxic secondary metabolites for chemical defence against unicellular or multicellular predators. The chemical diversity of these toxic compounds is quite relevant and, sometimes, ciliate species on the same genus discharge very different kinds of molecules. In this study we analysed the chemical defence strategy of the freshwater heterotrich ciliate *Stentor polymorphus* against the predatory ciliate *Coleps hirtus*, and the microturbellarian flatworm *Stenostomum sphagnetorum*. The results showed that *S. polymorphus* defends itself discharging a mix of four sterols, ergosterol, 7-dehydroporiferasterol and their two peroxidised analogues. Sterols were isolated *in vivo* and characterized by liquid chromatography-mass spectrometry and nuclear magnetic resonance. Toxicity tests against other predators and competitors, indicated that at least, ergosterol and ergosterol peroxide, are essential for the efficiency of chemical defence in *S. polymorphus*. To the best of our knowledge, this is the first report about the use of sterols in predator-prey interaction among ciliated protozoa, highlighting the huge chemical diversity in the ciliate secondary metabolites.

<u>SESSIONE 3</u> BIODIVERSITA' E SCIENZE 'OMICHE NEI PROTISTI

Thinking outside the nucleus. A mitogenomic approach to ciliate phylogeny.

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In recent years, the usage of mitochondrial genes, or even entire mitochondrial genomes, to investigate phylogenetic relationships between organisms had seen a bloom in popularity. These kinds of analyses had been used, for example, to successfully depict cryptic species in protist, annelids and arthropods, or to discriminate between subspecies of Apis mellifera. In this regard, ciliates possess very peculiar mitochondrial genomes, being among the first to be identified as linear, showing several split rRNA genes and protein genes and being relatively long (~ 40 Kbp) in respect of other eukaryotes. Nevertheless, the low amount of available mitochondrial genomes of ciliated organisms in online databases, as well as the absence of representatives of many classes of this phylum, didn't allow to perform extensive and in-depth analyses using mitochondrial genome. For at least two representatives of each missing class in online databases, the whole DNA material was extracted, and the mitochondrial genome was sequenced and bioinformatically assembled. A selection of 17 protein coding genes was then used to perform the phylogenetic analysis, among 81 representatives of the phylum. Lastly some considerations about pros and cons about using mitochondrial genomes for phylogenetic analyses will be presented, as well as some consideration about synteny and genome content in ciliate mitochondria.