

## How protists crack the walls of algae

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Feeding, life history, and de novo transcriptome assembly of Orciraptor agilis (A) Orciraptor agilis extracting the chloroplast of Mougeotia sp. after perforating the algal cell wall (differential interference contrast). Scale bar, 5  $\mu$ m. (B) Annular dissolution of the algal cell wall resulting from an attempted attack (phase contrast). Scale bar, 5  $\mu$ m. (C) Distribution of F-actin (green: fluorescent phalloidin) reveals the lysopodium in Orciraptor agilis formed during



attack on Mougeotia sp. (overlay of differential interference contrast and fluorescence channels). The increased blue fluorescence (Calcofluor white) at the contact sites indicates lysis of the algal cell wall. Scale bar, 5 µm. (D) Scanning electron micrograph of a perforation by Orciraptor agilis reveals the degradation of both main structural components of Mougeotia's cell wall, gellike biopolymers (potentially pectins; indicated by "gel") and cellulose microfibrils (indicated by "fib"). Scale bars, 2 µm and 200 nm (inset). (E) Life history stages of Orciraptor agilis from which the samples were generated. (F) Benchmarked universal single-copy orthologs (BUSCOs) assessment of the assembled transcriptome. The analysis was performed with the "Eukaryota" dataset and the "Alveolata" dataset (sister group of Rhizaria). (G) Upset plot showing the number and overlap of ORFs annotated by the indicated annotation tools and databases. Only intersection sizes > 100 are shown. (H) Principal component analysis (PCA) based on the expression level of all transcripts for each replicate included in the experiment. Credit: Current Biology (2022). DOI: 10.1016/j.cub.2022.05.049

A team of researchers led by Dr. Sebastian Hess from the University of Cologne's Institute of Zoology has studied the expression of carbohydrate-active enzymes in the unicellular organism Orciraptor agilis by RNA sequencing. Orciraptor is a so-called "protoplast feeder" and lives exclusively from the cell contents of dead algae. To do this, it has to penetrate the cellulosic cell wall of the prey. In collaboration with colleagues at the Centre for Comparative Genomics and Evolutionary Bioinformatics (CGEB) in Halifax, Canada, the UoC researchers were able to identify a possible key enzyme for the highly specialized feeding act of the protist.

Upon contact with the algal cells, Orciraptor upregulates an <u>enzyme</u> that should be able to cleave plant cellulose, based on its gene sequence and predicted 3D structure. This enzyme could help the protist to dissolve algal cell walls. Until now, the <u>molecular basis</u> of how protoplast feeders



interact with their prey has been completely unclear. The article "Comparative transcriptomics reveals the molecular toolkit used by an algivorous protist for cell wall perforation" in the journal *Current Biology* now sheds some light on this phenomenon.

Furthermore, Orciraptor contains a number of unexpected proteins such as chitin-binding proteins, a chitin synthase and several chitinases. The potential function of chitin or similar biopolymers in the naked flagellate is still unclear. However, the enzymes suggest an important physiological role of chitin in the life history of Orciraptor. Enzymes that decompose recalcitrant biopolymers such as cellulose and <u>chitin</u> are also of great technological and industrial importance. Currently, <u>industrial</u> applications mainly utilize enzymes from bacteria and fungi—the traditional organisms in microbial biotechnology. In the published paper, Dr. Hess and colleagues point to the as yet untapped biotechnological potential of non-fungal microeukaryotes such as Orciraptor.

Orciraptor was discovered about ten years ago in nutrient-poor moorlands and described by Dr. Hess during his doctoral studies at the University of Cologne's Institute of Botany. However, there are many other <u>unicellular organisms</u> that show similar feeding strategies, but are not directly related to Orciraptor. Currently, dozens of such organisms are cultivated and genetically characterized at the Institute of Zoology. All of this is made possible by recent technological advances in the field of high-throughput sequencing. However, working with protoplast feeders also requires special knowledge in handling exotic microorganisms. The scientist are convinced that it is time for modern biology to turn back to the diversity of non-model organisms again. "The data from our study on Orciraptor highlight how fruitful future molecular analyses of little-known protists will be," Dr. Hess said.

**More information:** Jennifer V. Gerbracht et al, Comparative transcriptomics reveals the molecular toolkit used by an algivorous



## protist for cell wall perforation, *Current Biology* (2022). DOI: 10.1016/j.cub.2022.05.049

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