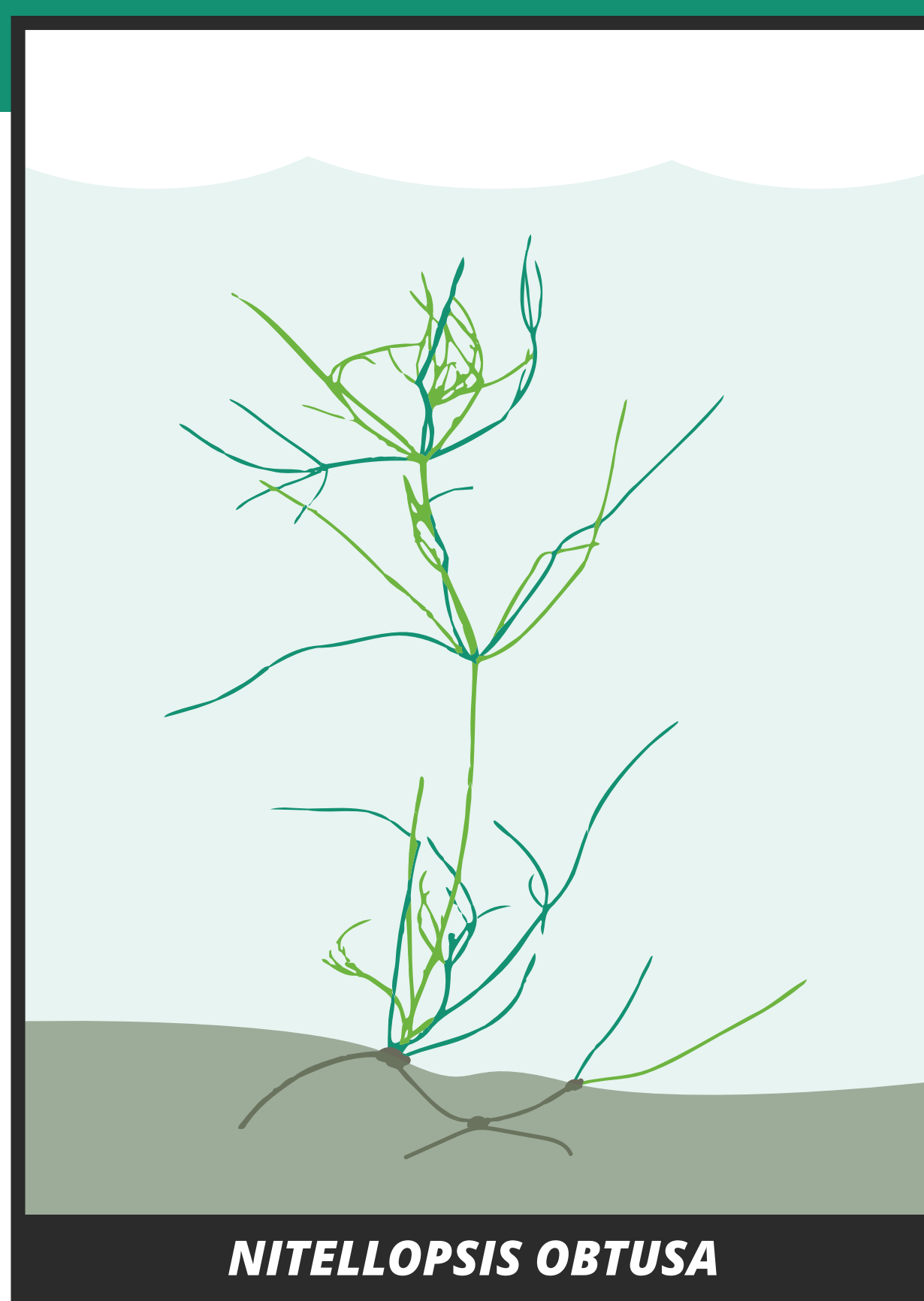


# Biomarker identification of isolated compartments of the cell wall, cytoplasm and vacuole from the internodal cell of characean *Nitellopsis obtusa*

## BACKGROUND

*Nitellopsis obtusa*, commonly known as starry stonewort, is a benthic alga and bioindicator of clean fresh or brackish water bodies.

Cells of characean algae such as *N. obtusa* are excellent for plant research because of their large size and close relation to higher plant cells.



NITELLOPSIS OBTUSA

## STUDY OBJECTIVES

- 1 **RE-EXAMINE**  
single-cell fractionation procedure in *N. obtusa*
- 2 **EVALUATE**  
obtained fraction purity using enzymatic markers
- 3 **RE-ESTIMATE**  
accumulation of copper oxide nanoparticles

## METHODS

Macrophytic algae *N. obtusa* was collected from Lake Obelija in south-east Lithuania.

Single-cell fractionation of internodal cells was completed and the purity of vacuolar fraction was examined using **vacuolar and cytoplasmic enzymatic markers**:

**$\alpha$ -mannosidase & malate dehydrogenase**

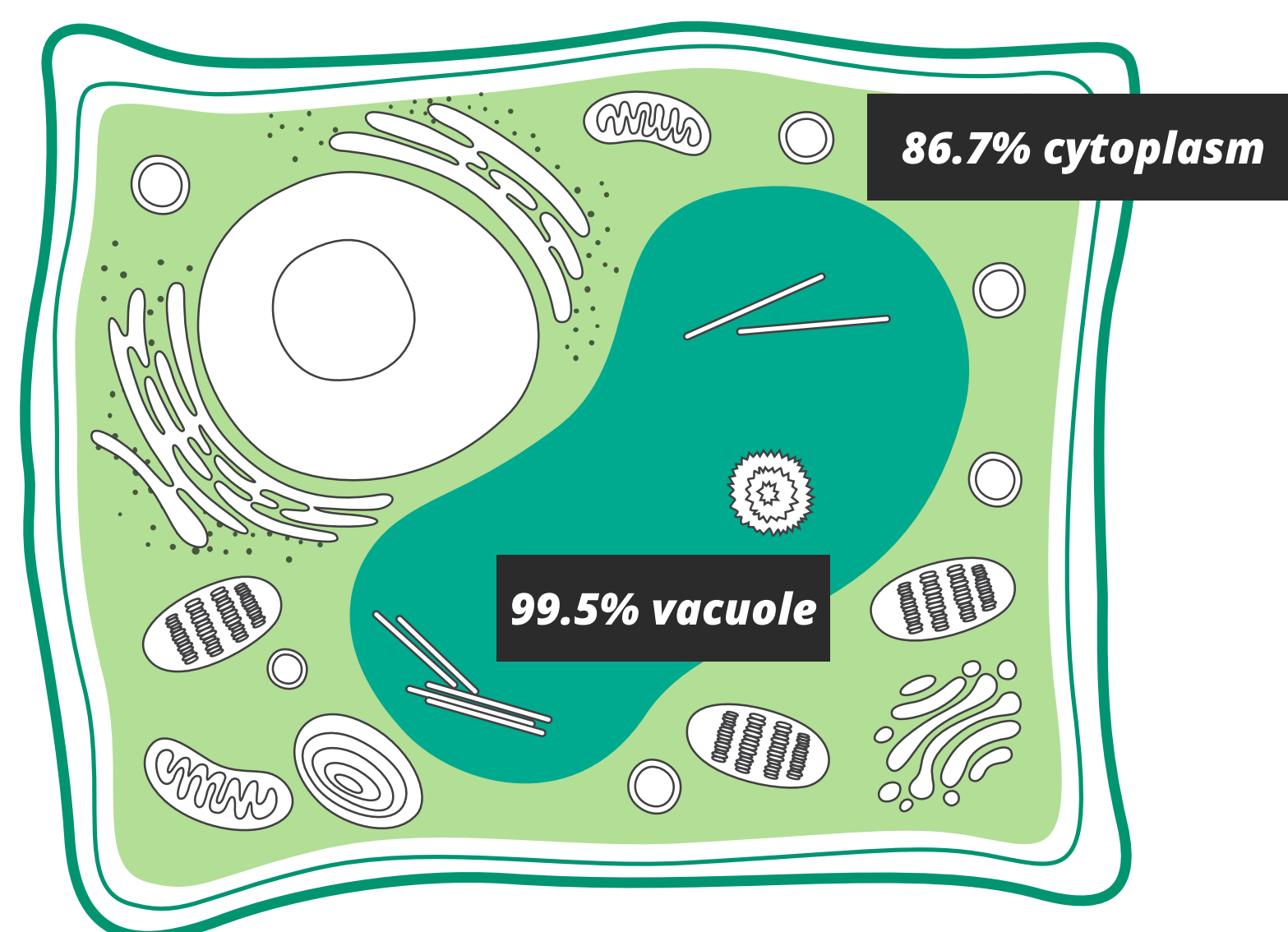


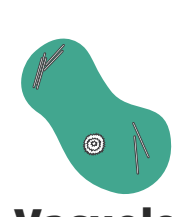
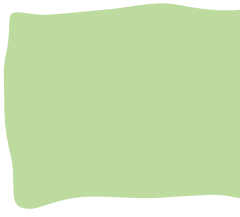

Protein content was measured and the content of photosynthetic pigments was analyzed. Additionally, copper oxide (CuO) nanoparticles were suspended and a statistical analysis was conducted by calculating the mean of three replicate experiments.

## RESULTS

*N. obtusa* cell results indicated **99.5% and 86.7% purity from vacuolar and cytoplasmic compartments**.

We observed slight cell wall compartment contamination by cytoplasmic (1.96%) and vacuolar (12.3%) material.



| COMPARTMENT  | 0.5 HR     | 3 HR       |
|--|------------|------------|
| <br>Vacuole   | 0.13 mg/L  | 0.18 mg/L  |
| <br>Cytoplasm | 2.39 mg/L  | 2.98 mg/L  |
| <br>Cell Wall | 0.93 mg/DW | 1.22 mg/DW |

## CuO NANOPARTICLE ACCUMULATION

When treated by 100 mg/L nCuO for 3 hours, compartments of *N. obtusa* cells **showed internalization of copper oxide (CuO) nanoparticles at around 30 minutes**.

## CONCLUSION

**Mechanical manipulation is a reliable method for isolating the main compartments of characean algae cells;** the application of enzymatic markers confirmed high purity of these obtained fractions. **Cell wall fraction was contaminated slightly** by vacuole and cytoplasm residues. Contamination from the vacuolar component of the cell wall fraction **could have been caused by the vacuoles from the numerous small cells in the nodes**.

**Estimating the purity of mechanically-separated cell fractions enabled the re-evaluation of copper oxide (CuO)-related concentrations in charophyte cell compartments.**