Towards the exploitation of cellular mechanosensitive nanosensors for bioprocess optimization.

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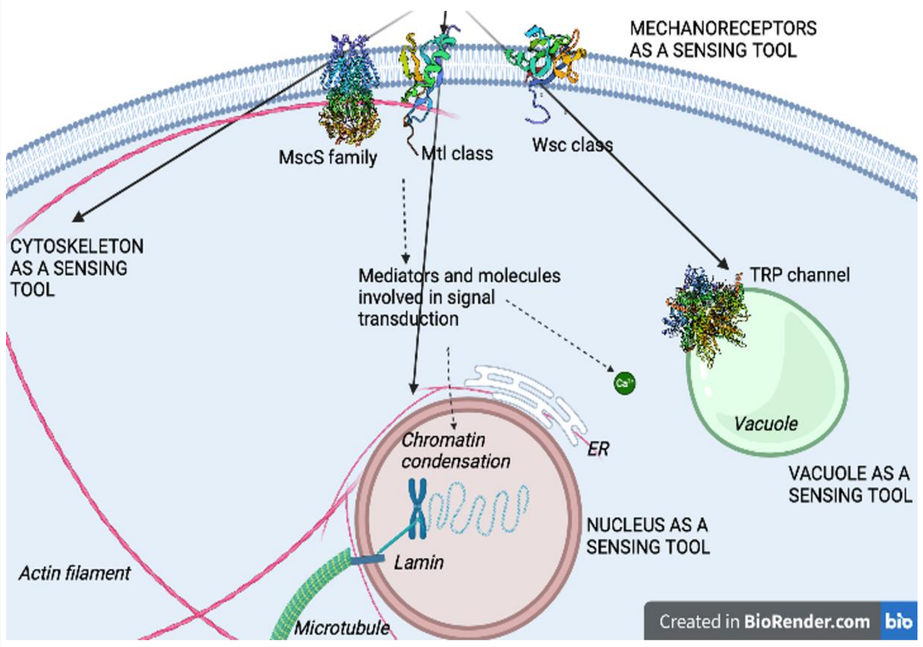
Introduction

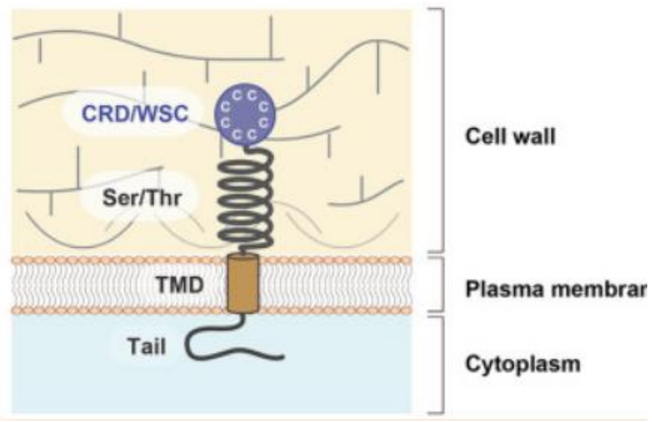
Ultrasound Wave stimulation(UWS), affecting living cells through the propagation of pressure waves in the culturing medium, gathered research interest for its effects on microbial growth and productivity. UWS was demonstrated to reduce fermentation length but few studies have focused on the characterization of the cellular components involved in the perception of mechanical stimulus signals and the correlated signal transduction pathways. To move forward in this direction we designed and developed a novel sonobioreactor (SNBR) prototype to precisely generate and transduce UW at fixed frequency and intensity. Currently we are focusing on the study of conserved cellular mechanoreceptors, nanoscaled sensors evolved to perceive pressure variation and activate downstream signalling. This will pave the way to understand cellular response to UW and may contribute to the sustainability of industrial bioproduction.



Mechanical stimules are renowned for activating mechanosensors. In microorganisms, mechanoreceptors are involved in cell wall re-shaping, modeling it in response to surrounding microenvironment (i.e. osmolarity variation, grazors presence) to the aim of cell wall integrity and homeostasis maintenance,which are MscS, Mtl and Wsc families.

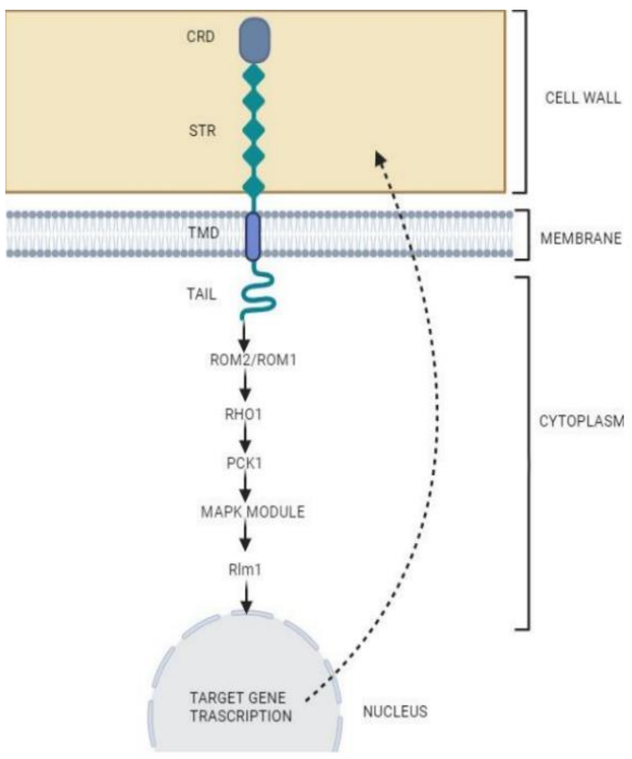
The role of mechanoreceptors

Wsc protein structure



Structural model and architecture of Wsc family sensors. Model showing the topology and proposed functional domains of WSC domain sensors, including the N-terminal cysteine-rich domain (CRD) or WSC domain, the glycosylated serine/threonine-rich domain (Ser/Thr), the transmembrane domain (TMD), and the C-terminal cytoplasmic tail.

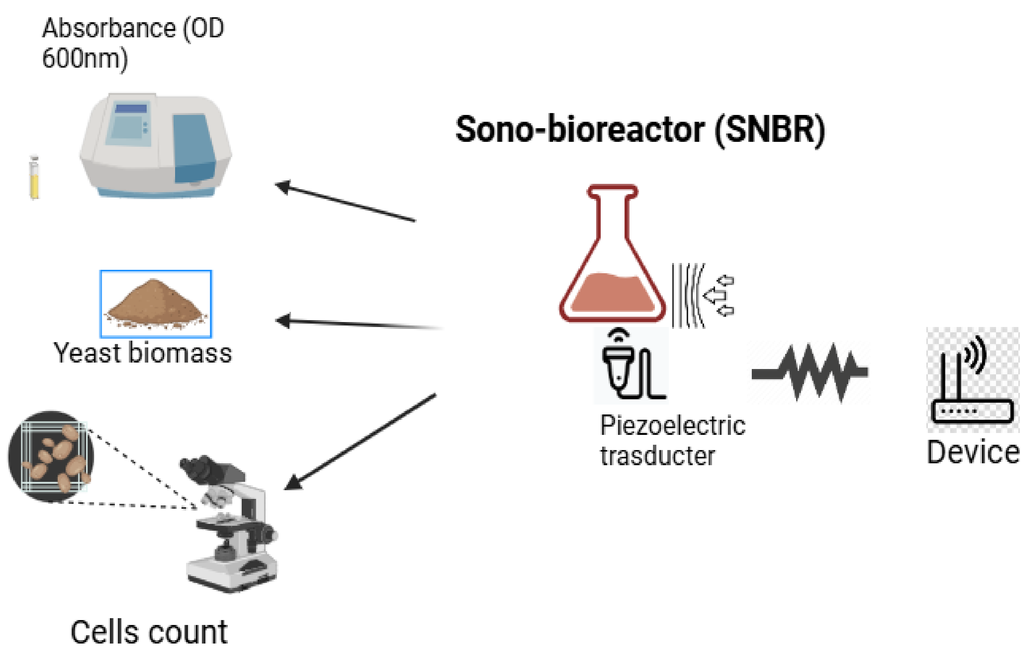
Wsc pathway



One of the receptors we are studying is Slg1 (former Wsc1), which is activated by mechanical stress. After activation, a cascade pathway begins, culminating with the expression of genes involved in the repair of cell walls, while others implicated in cell proliferation.

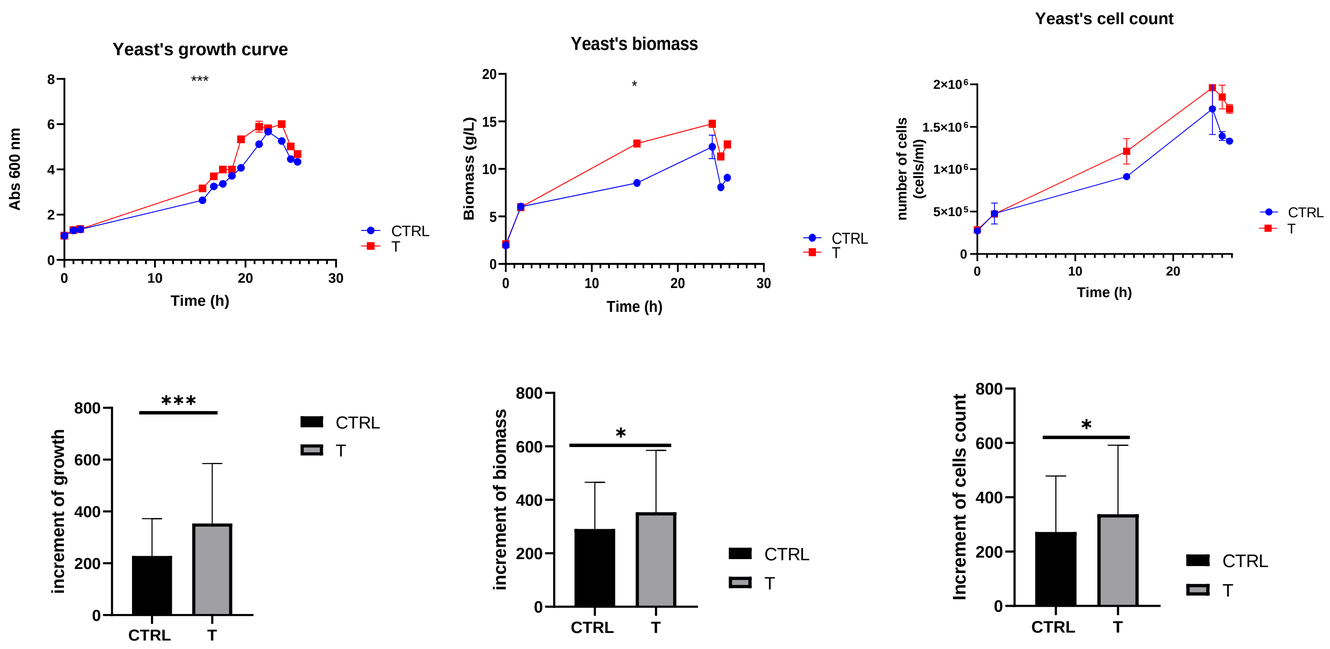
Hypothesis: can US influence yeast growth via mechanoreceptors?

Experimental design:



The experimental set up implemented in the growth tests conducted for yeasts. quantification studies. The treatment was applied 8 h in logaritmic phase and biomass was quantified during treatment analysing optical density (OD), biomass as cell dry weight (CDW) and cell count as parameters

Results:



The efficacy of ultrasound on yeast growth was evaluated through the utilisation of a SNBR. This involved the monitoring of absorbance; biomass production and cell count throughout the experimental period.The results showed, through the use of the US an increase in growth , for the absorbance of 15% compared to the control, for the increase of 20% compared to control, The number of cells increased by 18% compared to control.
SD, n=3. t-student , *** P value <0.001.

Conclusions:

Overall, our system provides a controlled platform to study the effect of US stimulation of microbial growth and the effects we measure on different organisms seem very promising toward the optimization of biotechnological industrial applications. This result led us to consider the hypothesis that ultrasound may activate various receptors involved in the expression of different genes. Our data confirm that this technology could be an excellent alternative in industrial processes to reduce the duration.

References:

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