

Effect of salt stress and various physical extraction methods on the phycocyanin yield from *Spirulina* sp.

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ABSTRACT

Synthetic pigments were used as a colorant or dye in many industries for ages, but due to their hazardous impact on the human body (causes cancer, irritation in skin and eyes) and the environment (pollution), there was a shift to biologically derived pigments in the recent years. Additionally, phycocyanin, a natural blue-colored pigment predominantly found in *Spirulina* sp. has gained researchers' interest due to its enormous properties such as anti-oxidant, anti-microbial, anti-cancer, and also ease in culturing and maintenance. However, the lack of low-cost extraction strategies, without compromising the purity or quality of the product (*i.e.*, phycocyanin) is one of the major obstacles to scaling up the production processes for commercial purposes. Here in this study, phycocyanin was extracted from the *Spirulina* sp. (pre-treated with hexane for defatting and with ethanol for depigmentation) using three different physical cell disruption methods (ultrasonication, homogenization, and freeze-thaw cycles) and two different buffers (phosphate buffer, sodium acetate and sodium chloride buffer), and water (as control). Further, enhancement in phycocyanin yield from the *Spirulina* sp. under salt-stressed conditions was studied. This study focuses on both the upstream and downstream processing of the eco-friendly microalgal pigment production system, which can have a possible application in various industries.

Keywords: Phycocyanin, pigments, *Spirulina* sp., extraction, physical methods, microalgae

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Introduction

- Phycocyanin is an accessory photosynthetic blue pigment present in *Spirulina* sp. which is responsible to absorb around 50% of the sunlight. The structure of the pigment consists of a heterodimer of α (alpha) and β (beta) subunits of molecular weight 18 and 20 kDa respectively (Glazer, 1989; de Morais et al., 2018).
- There are a few drawbacks of phycocyanin that make it difficult to scale up and use on the industrial scale, which include low stability, high extraction and purification cost, and low yield (Pez Jaeschke et al., 2021).
- An extraction and processing method for the extraction of pigments from *Spirulina* sp. needs to be optimized which could increase the yield of the compound along with increasing the stability and shelf life of the product.

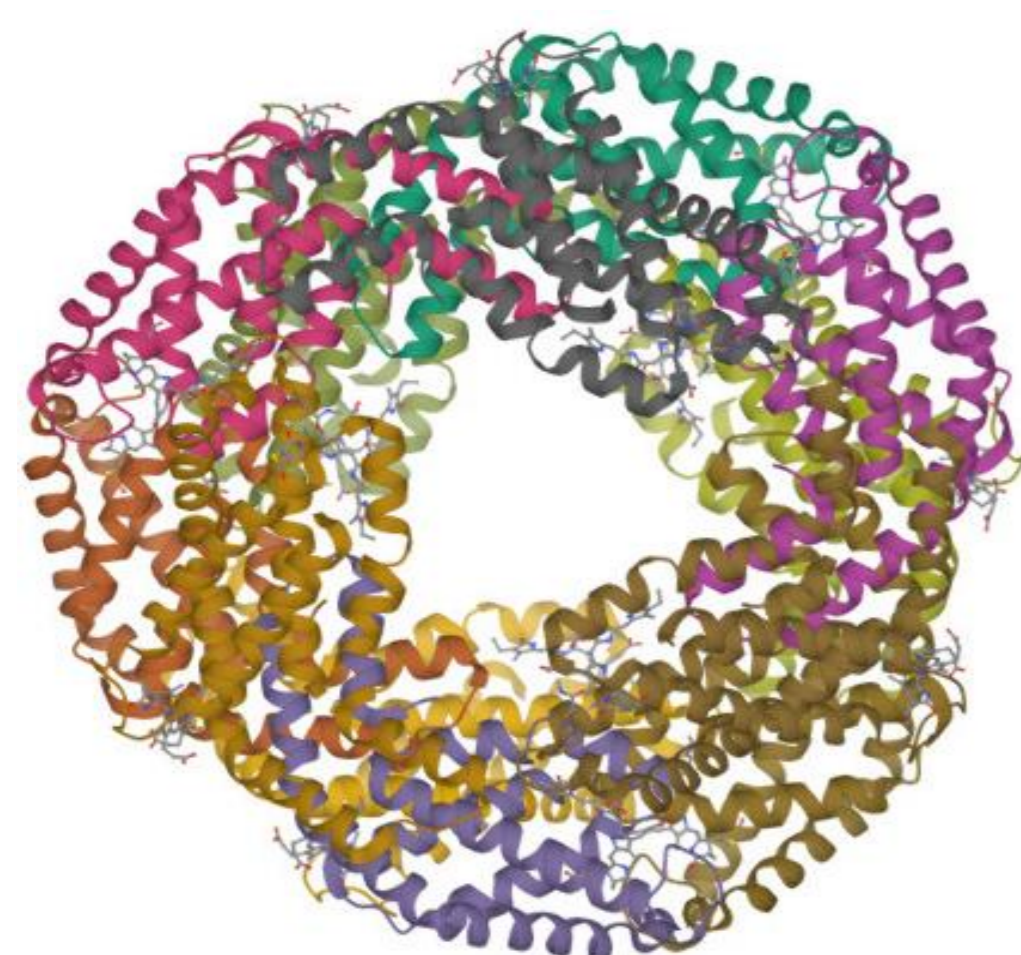


Fig 1: Crystal structure of phycocyanin from *Spirulina platensis* (Wang et al., 2001)

Objective

- To optimize a method for phycocyanin extraction from *Spirulina* sp.
- To enhance phycocyanin production in *Spirulina* sp. by subjecting it to salt stress using different concentrations of salt

Methods

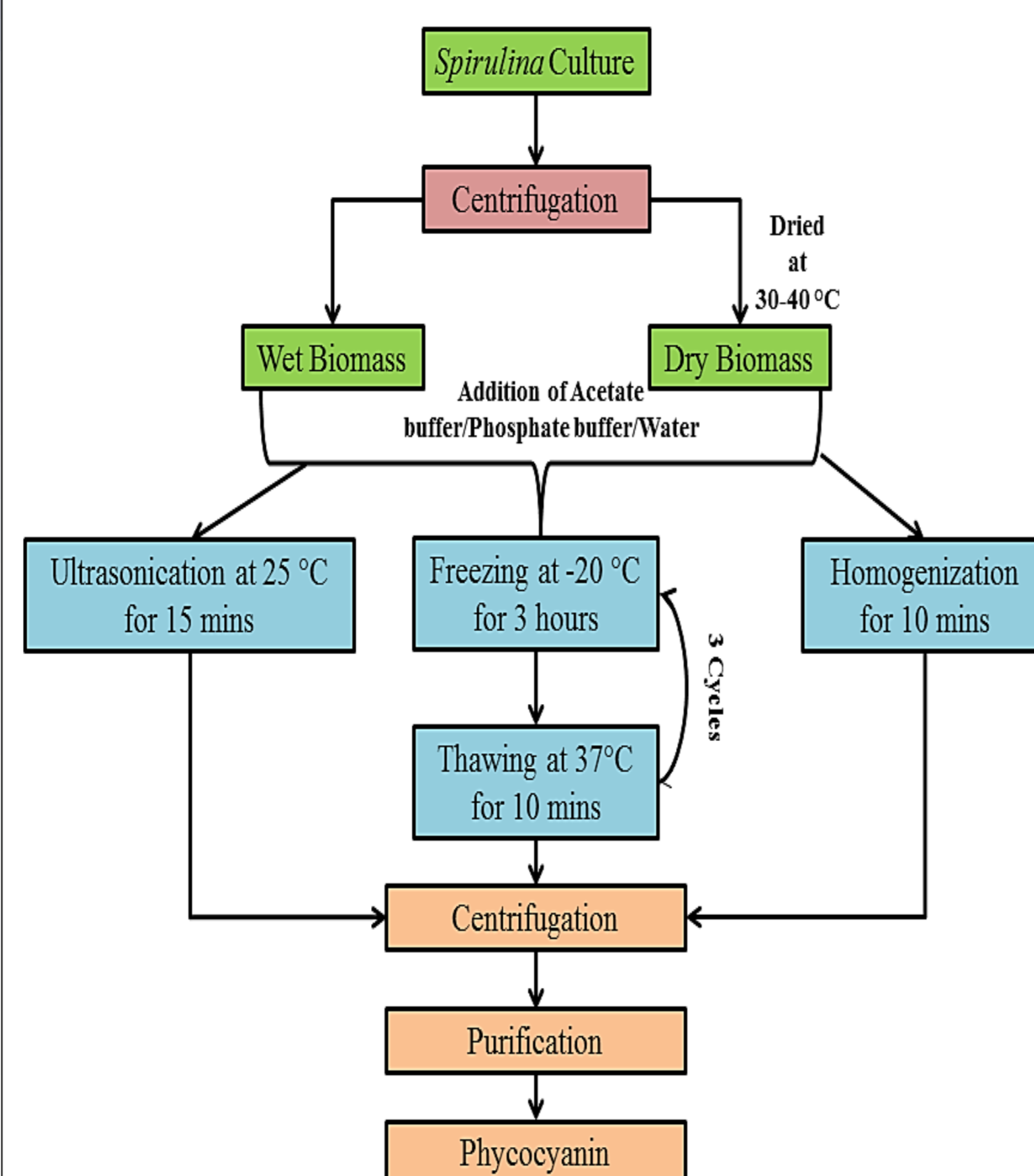


Fig 2: Workflow for the extraction of phycocyanin

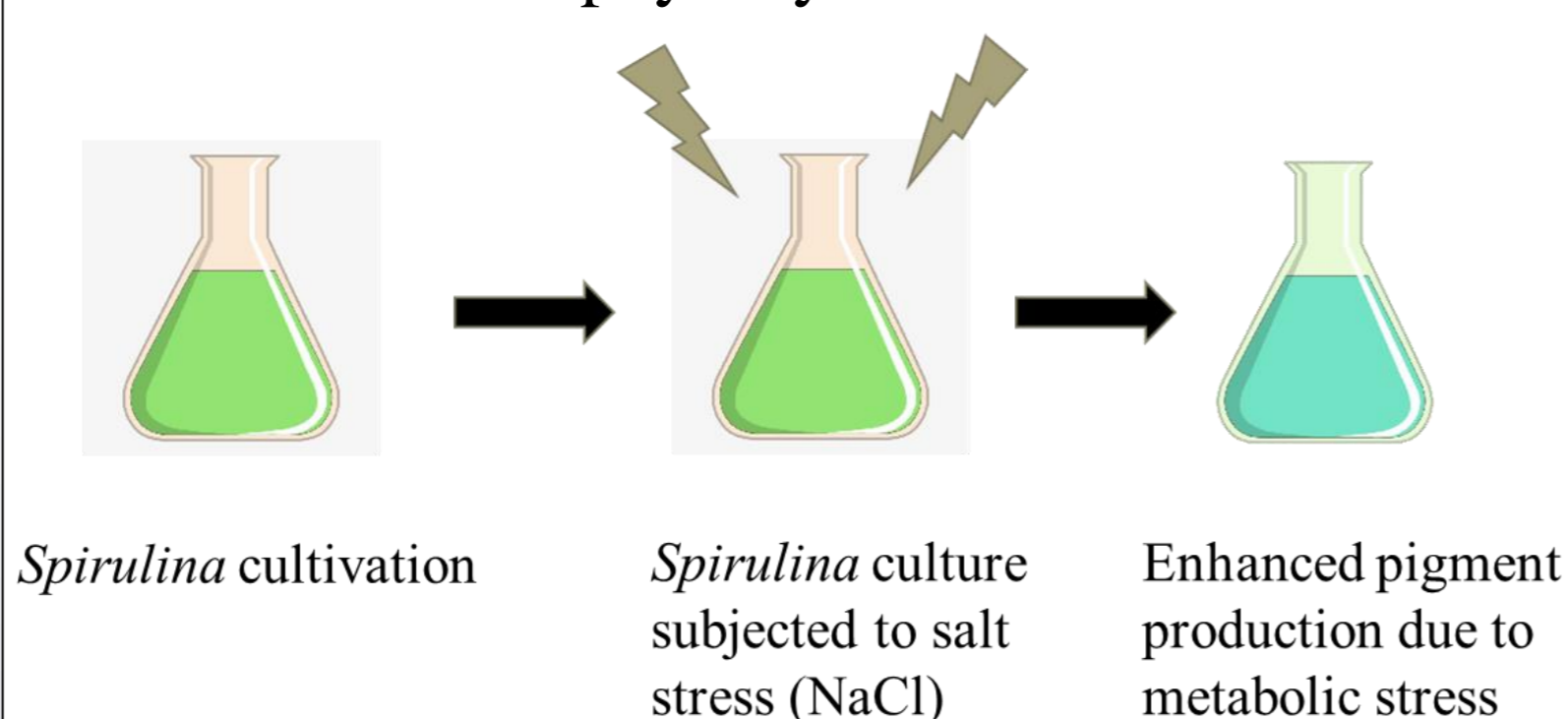


Fig 3: Enhancing phycocyanin production in *Spirulina* by increasing salt concentration (10-30%)

Results

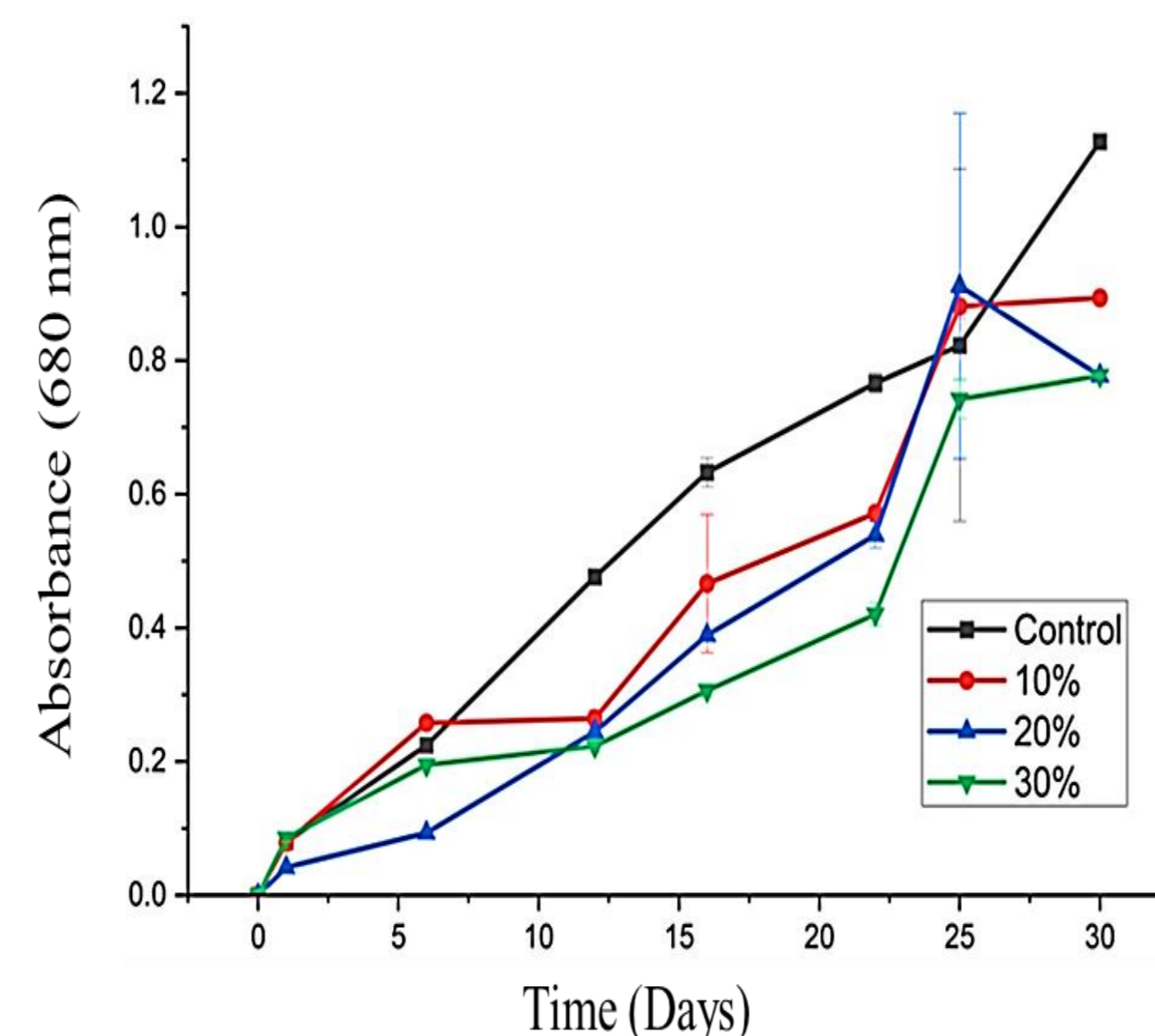


Fig 4: Growth curve of non-stressed (control) and the salt-stressed *Spirulina* culture

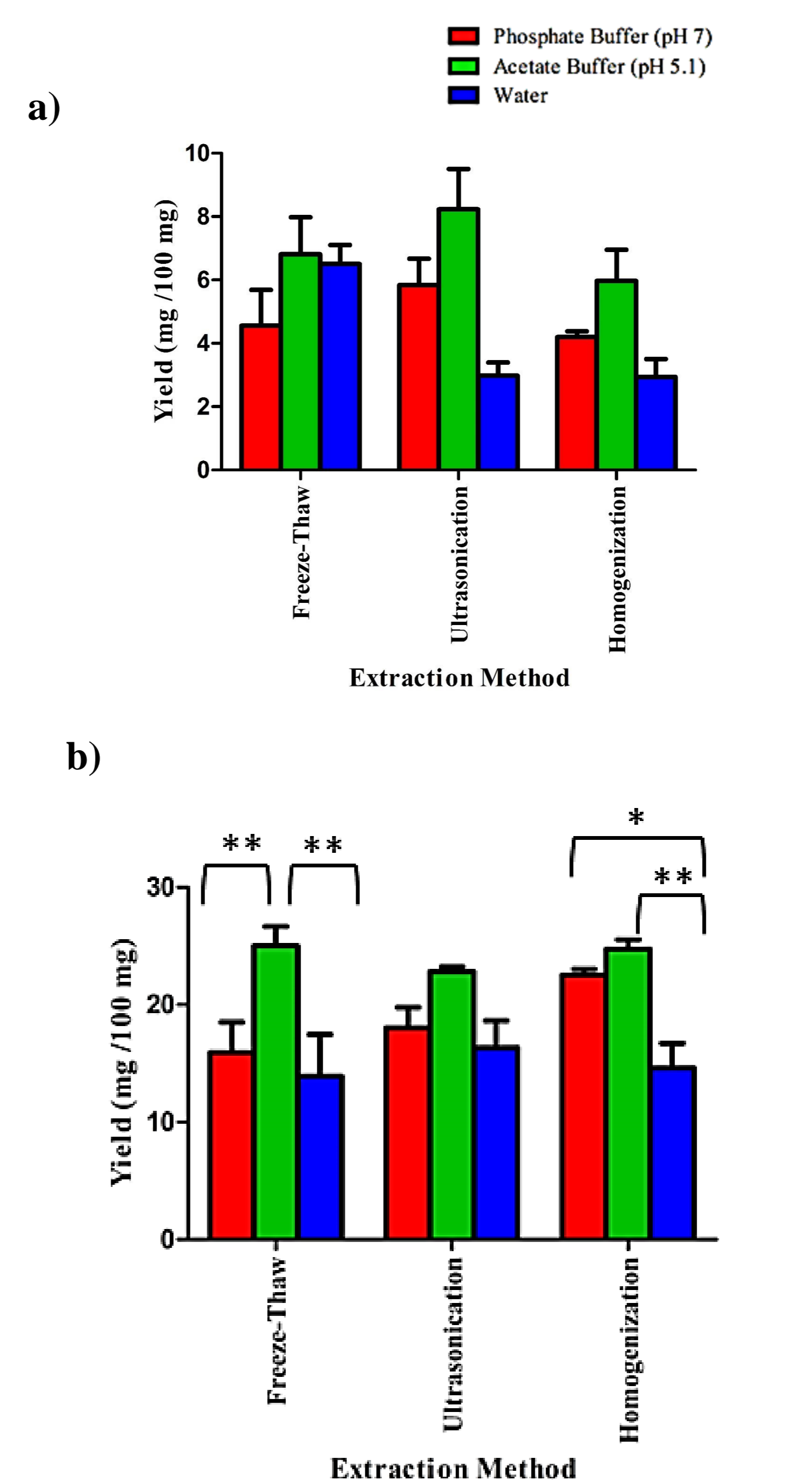


Fig 5: Comparison of yield between extraction methods using three different buffers in (a) wet biomass and (b) dry biomass (* $P < 0.05$, ** $P < 0.01$)

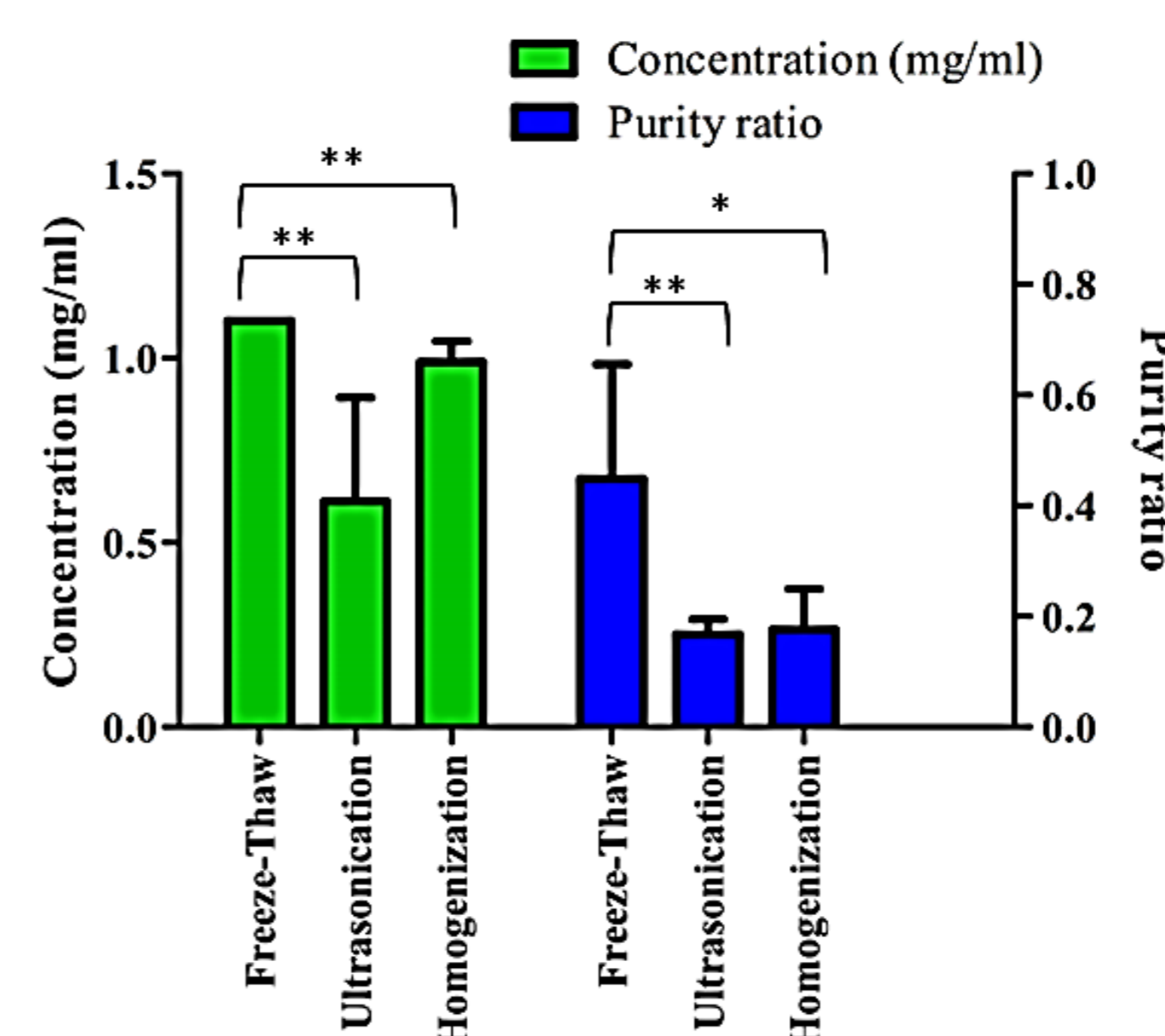


Fig 6: Comparison of the concentration (mg/mL) and purity ratio with the different extraction methods in dry biomass (* $P < 0.05$, ** $P < 0.01$)

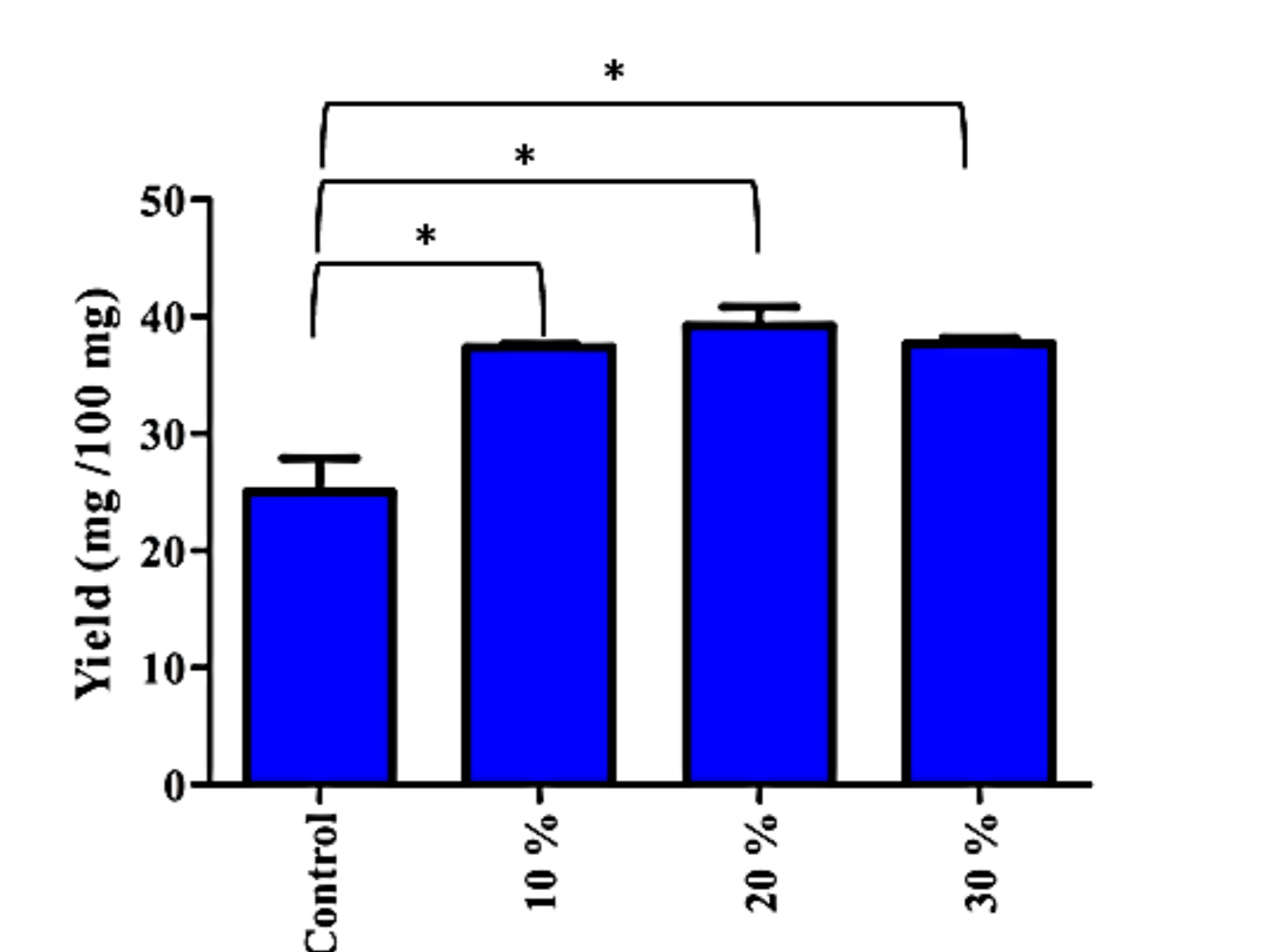


Fig 7: Comparison of yield between stressed and unstressed *Spirulina* culture (* $P < 0.05$)

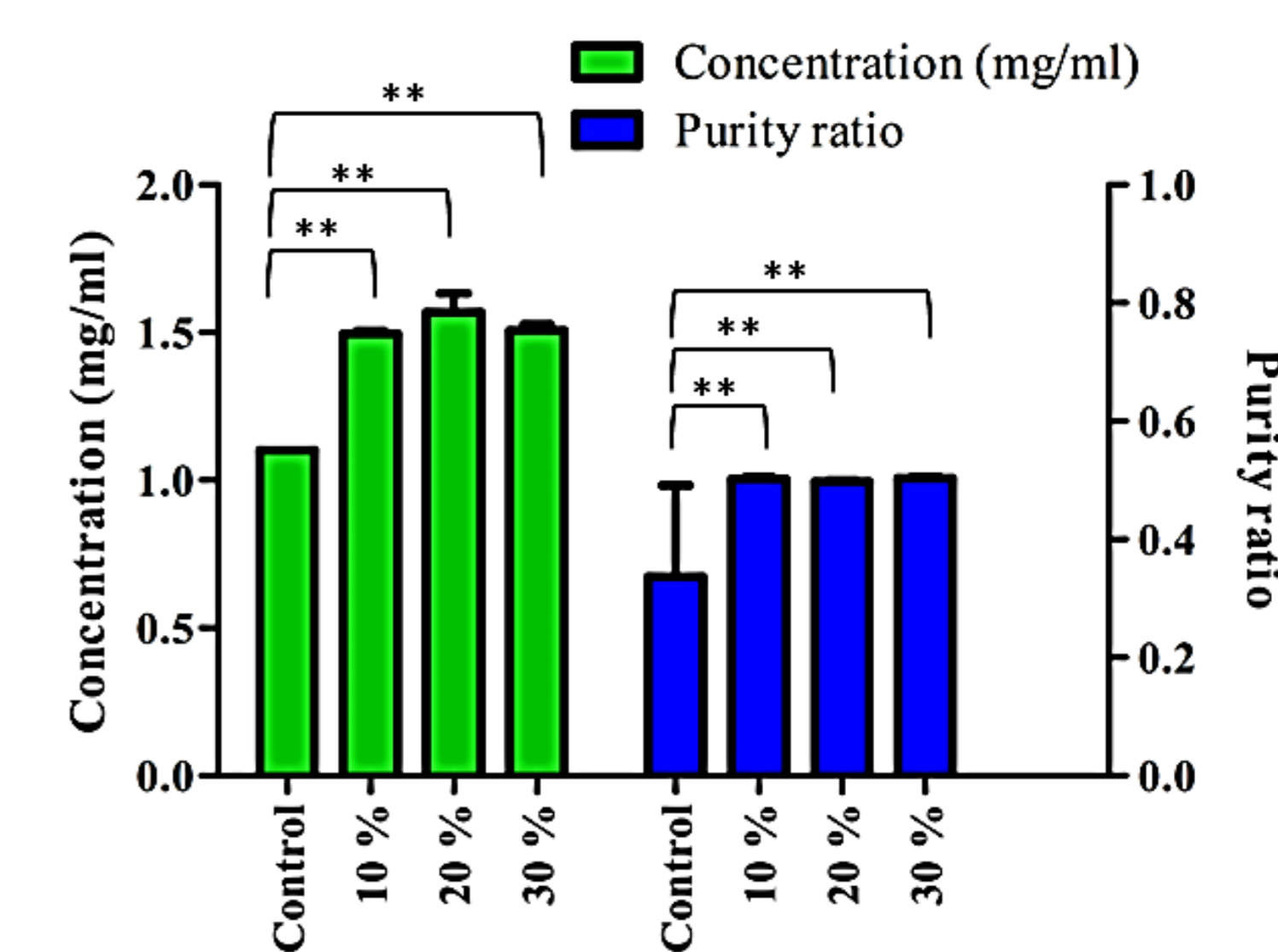


Fig 8: Comparison of concentration and purity of stressed and unstressed *Spirulina* culture (** $P < 0.01$)

Conclusion

- Optimized conditions for extraction of phycocyanin: (a) Freeze-thaw method, (b) Acetate buffer, and (c) dried biomass
- Salt stress increased phycocyanin yield by 1.48 fold

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