

Sugar: Life or Death?

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Background

Saccharomyces cerevisiae is a fungus commonly known as Brewer's Yeast, which has been used for basic research pertaining to cell division and numerous biological processes. *S. cerevisiae* cells are a good model organism because they are eukaryotic (contain a nucleus), have linear chromosomes, divide by mitosis and meiosis, respond to signaling molecules, and respire. This makes yeast cells biologically similar to human cells, meaning our experiment can be directly tied to human cell division. Our study asked whether the concentration of glucose in the Yeast, Peptone, and Dextrose (YPD) media would affect the cell growth and proliferation of yeast cells.

Methods

To make the total 6 different concentrations of dextrose of YPD media, we used 6 separate Erlenmeyer flasks to combine different ratios of yeast extract, peptone, and dextrose (glucose). Every flask contained 500mL of DI water along with 5.0 g of yeast extract and 10.0 g of peptone was added to each flask. The baseline recipe (2%) called for 5.0 grams of dextrose, and we calculated the rest of the concentrations from there (1% having 2.50g, 4% having 10.0g, etc.). After inoculating each concentration with 300µL of yeast cells grown from an overnight culture to 10mL in each test tube respectively, we took the optical density of each concentration using the spectrophotometer set to 600 nm. Cell viability was observed using a methylene blue assay- 10µL of each concentration on a microscope slide with methylene blue. The optical density and cell viability was assessed periodically over a time frame of 6 hours, every hour. Between the 60 minute recorded intervals, each yeast concentration was placed in a shaking incubator at 30°C.

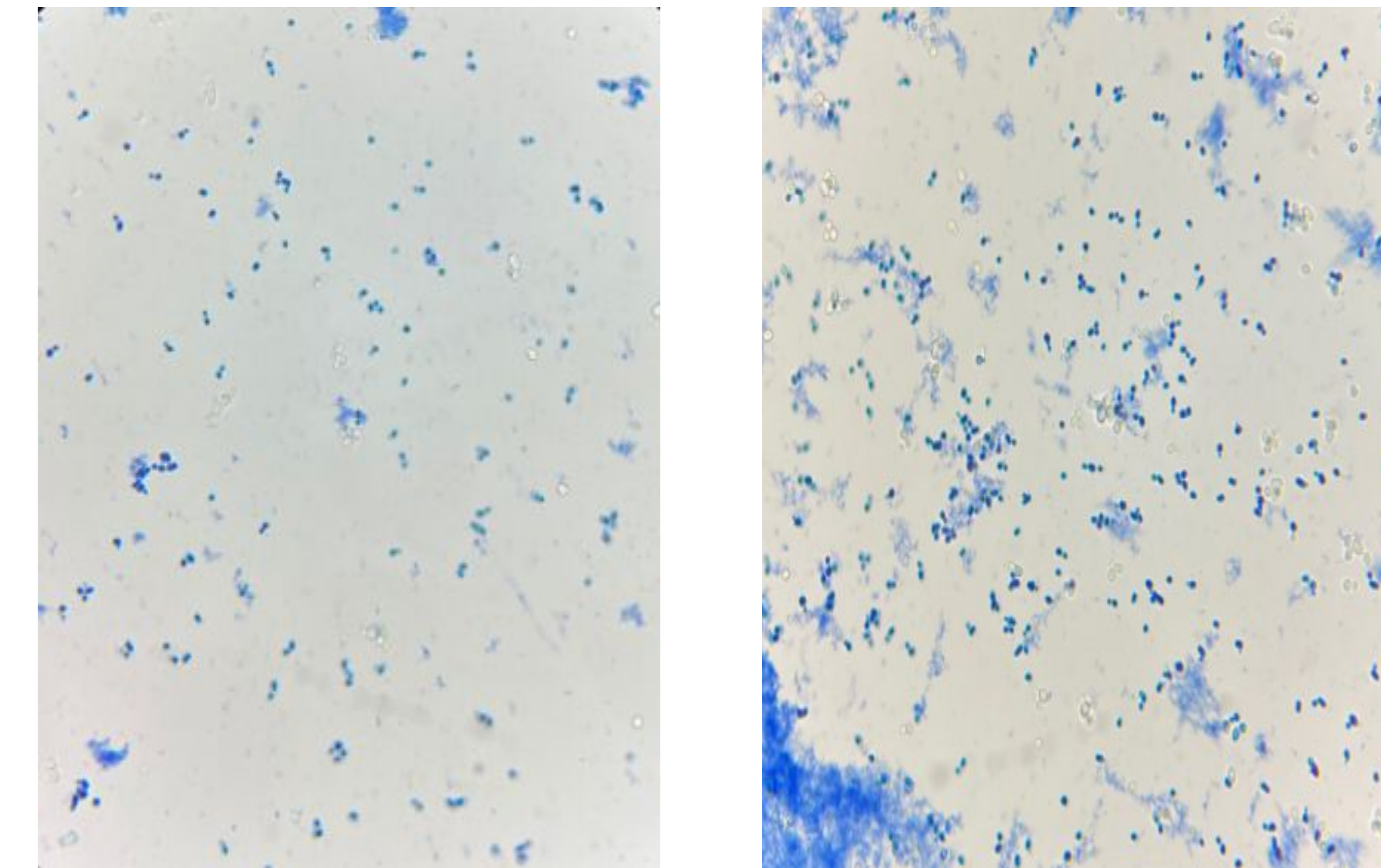
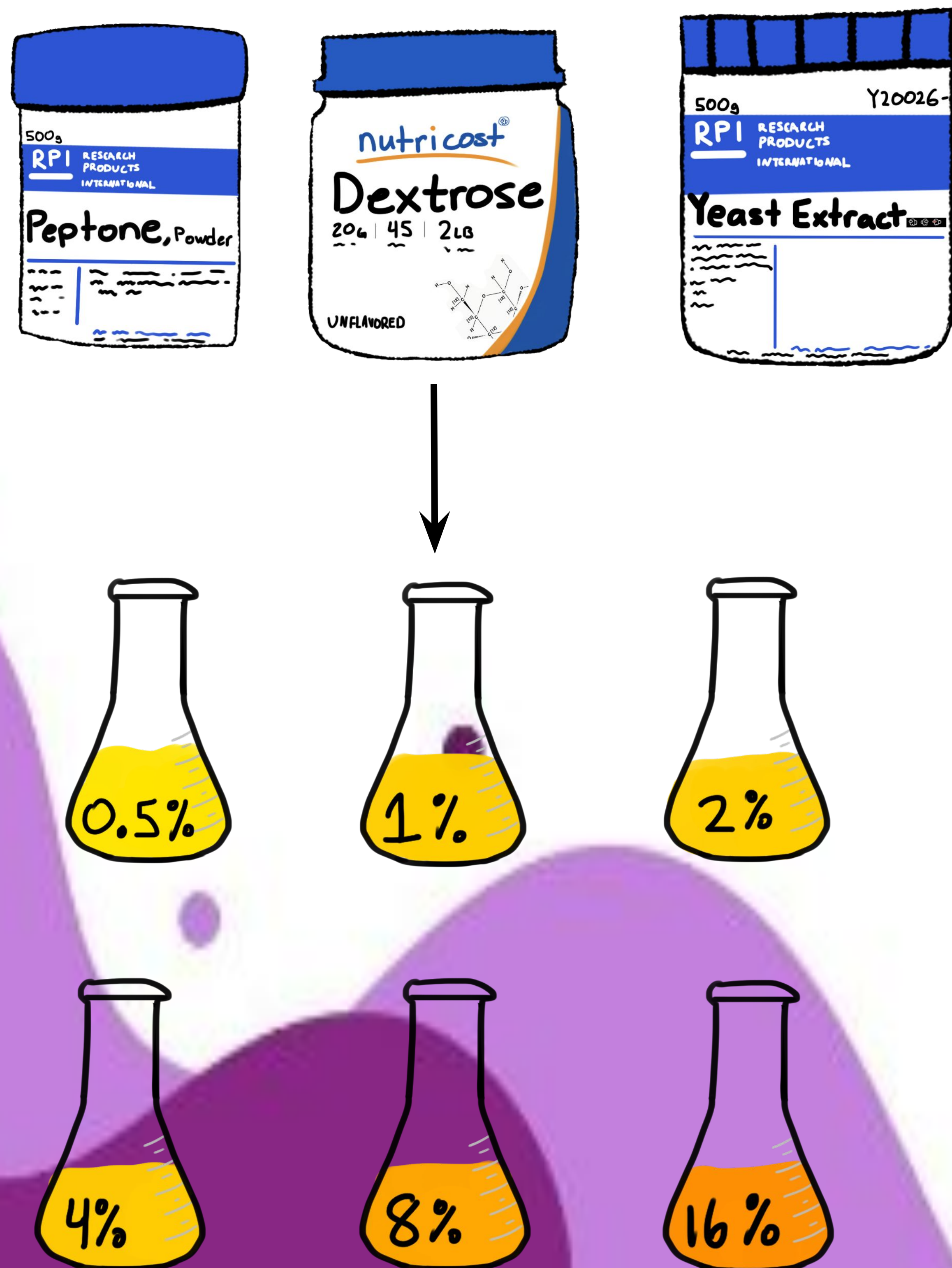


Fig. 1. 1% glucose increased cell death after 5 hours (300 minutes) observed using methylene blue assay

S. cerevisiae exposed to variable glucose concentrations does not show much of a change in cell viability over time. All of the concentrations started with high cell viability, at some time stamps there was varying results that showed a drastic unpredicted change. This could be due to the assessment of the viability using the methylene blue assay, as it can be inaccurate for the whole concentrations rather than the small sample that is collected to be observed under the microscope.

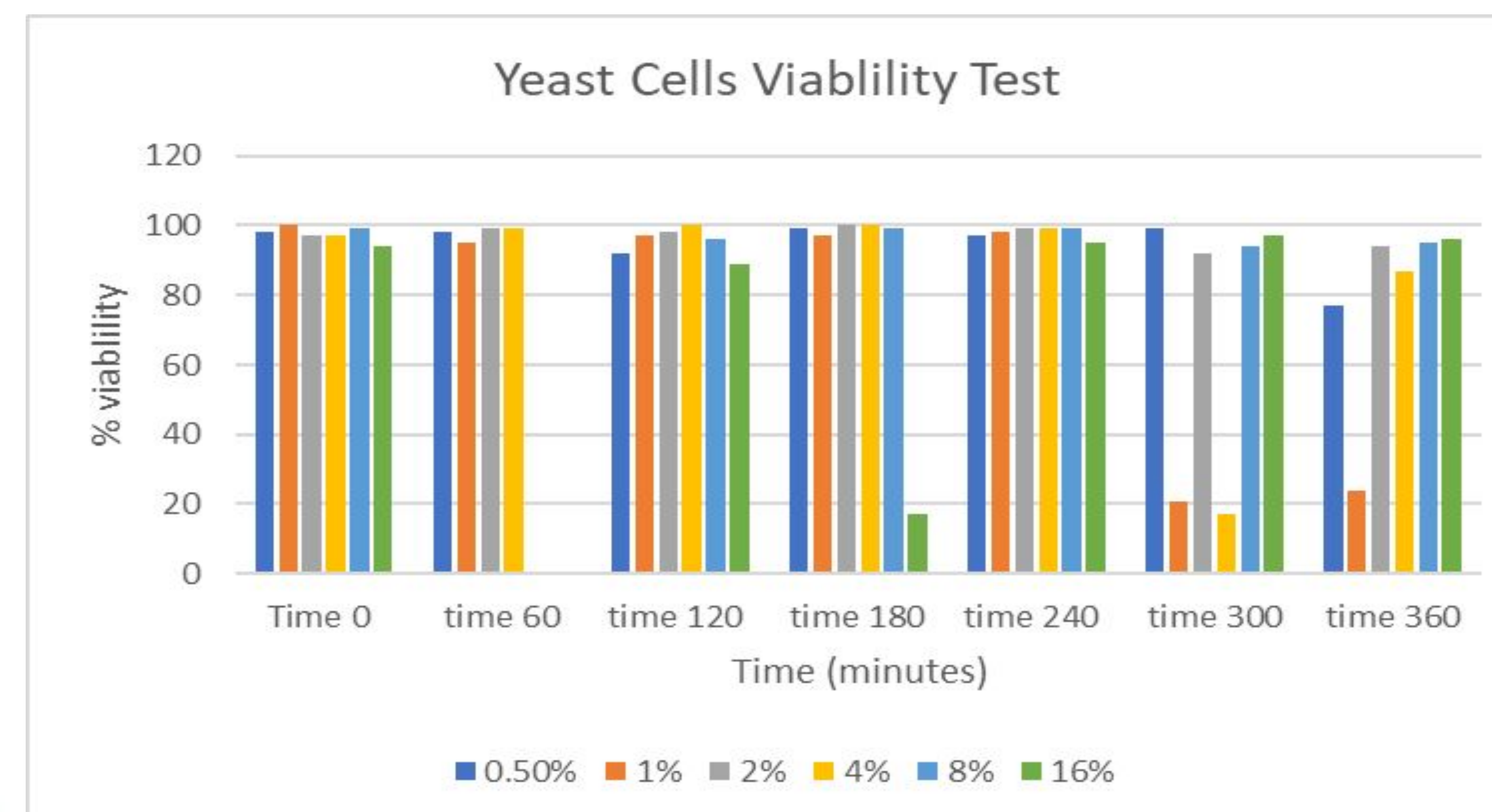


Fig. 2. *S. cerevisiae* exposed to various glucose concentrations shows varying cell viability observed via methylene blue assay.

Conclusion

We observed a trend that higher concentrations of glucose were less effective in aiding cell division over 6 hour time frame. *S. cerevisiae* exposed to high glucose concentrations shows slower growth, the differences between measurements of optical densities was less compared to the lower concentrations of glucose. The 16% concentration started off with the highest optical density and then it ended with the lowest optical density at 6 hours. The 0.5% concentration was the lowest at the start but it had more cell growth than the 8% and 16% after 6 hours. I think that yeast cells in high sugar concentration are having trouble with osmoregulation (holding on to water) and transportation of materials via the cellular membrane. In a study done by Souza et al., colon cancer cells exposed to higher glucose concentration in media affected the cell barrier and resulted in a change in transportation of materials entering and exiting the cell (2003). We saw a similar result with our yeast cells, the higher concentrated glucose medias slowed the rate in which our cells divided meaning this also could have resulted in an alteration of the physical cell barrier as well. As mentioned in the study conducted by Meijer et al., glucose plays an important role in *Saccharomyces cerevisiae* as a carbon source/ overall regulatory responses, and found that the glucose flux (change in glucose internally) is higher in lower concentrations of extracellular glucose media (1998). This coincides with the effects on the cell membrane in Souza et al., transportation of materials, and what we saw in our trend of decreased cell division under the conditions of high glucose concentrations.

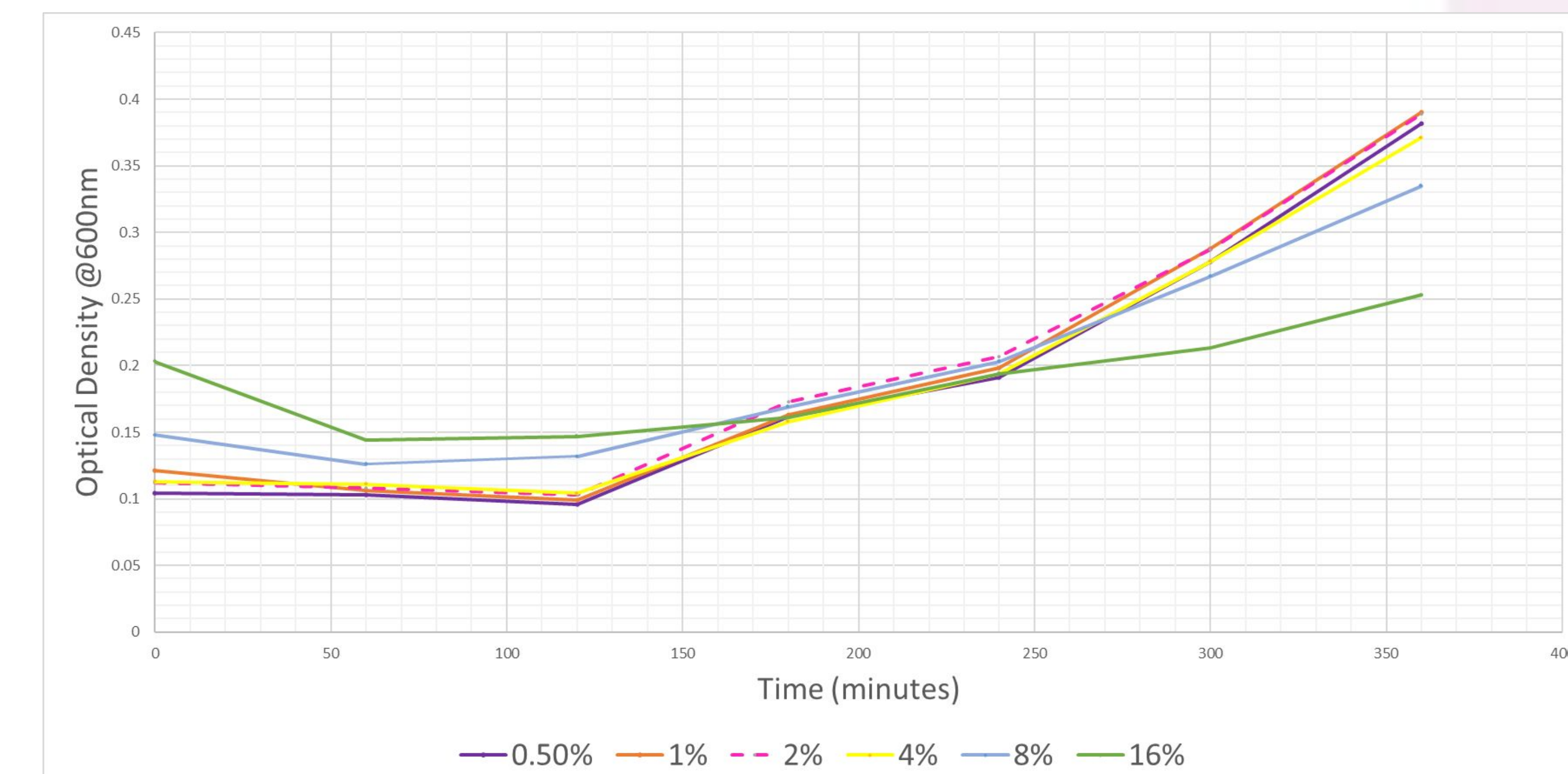


Fig. 3. *S. cerevisiae* varying glucose concentrations optical density measured in the spectrophotometer at 600 nm. Higher glucose concentrations 8% and 16% gradually changed less over time compared to lower glucose concentrations 0.5%

References

- Ashe, Mark P., et al. "Glucose Depletion Rapidly Inhibits Translation Initiation in Yeast." *Molecular Biology of the Cell*, vol. 11, no. 3, Mar. 2000, pp. 833–848.,
- Cullen, Paul J., and George F. Sprague. "Glucose Depletion Causes Haploid Invasive Growth in Yeast." *Proceedings of the National Academy of Sciences*, vol. 97, no. 25, 2000, pp. 13619–13624.,
- Meijer, Michelle M.C., et al. "Glucose Repression in *Saccharomyces Cerevisiae* Is Related to the Glucose Concentration Rather than the Glucose Flux." *Journal of Biological Chemistry*, vol. 273, no. 37, 11 Sept. 1998, pp. 24102–24107.,
- Saitoh, Shigeaki, and Mitsuhiro Yanagida. "Does a Shift to Limited Glucose Activate Checkpoint Control in Fission Yeast?" *FEBS Letters*, vol. 588, no. 15, 2014, pp. 2373–2378.,
- Souza, Vanessa M., et al. "High Glucose Concentration in Isotonic Media Alters Caco-2 Cell Permeability." *AAPS PharmSci*, vol. 5, no. 3, 14 Aug. 2003, pp. 17–25.,