1 Summary

State the aims and objectives of the experiment.

Give a brief outline of the experiments you performed.

Summarise the most important results and major findings/trends, expressed quantitatively.

State the major conclusions which can be drawn from the results of the experiment

2 Introduction

The aim of the experiment is to establish a calibration curve against which we can track and compare the progress of our bioreactor. We do so by defining the appropriate frequency for best potential difference output on a phosphate buffer solution, as well as a 20 g/l yeast solution. Finally, we produce the calibration curve by analysing the potential difference at a range of yeast concentrations.

Background knowledge. Impedance spectroscopy is a widely used modality to establish cell composition, diagnosis of lymphatic edema and, most importantly for our purposes, quantify biological cell suspensions [1, 2]. Alternatively optical density can be used, which we in fact do use to verify our results.

Impedance (Z) is effectively resistance in an AC electrical circuit, which means that it follows Ohm's Law. Furthermore, we can represent a cell in extracellular liquid as capacitor in a electrical circuit, which allows us to use an important definition from AC theory; where V is potential difference and I is current, C is capacitance, f is the frequency, j is the imaginary unit:

$$V = ZI \qquad\qquad Z_c = \frac{-j}{2\pi f C} \tag{1}$$

In practice, we cannot determine C, but by combining both equations, we know the following relationship

$$V_{cell} \propto \frac{1}{f} \tag{2}$$

Which tells us that as we increase the function generator's frequency, the potential difference "lost" to the cells is reduced, and therefore, the potential that we measure in the extracellular liquid is higher.

$$V_{liquid} \propto f \tag{3}$$

3 Materials and Methods

3.1 Materials

- Combined amplifier, filter and driving circuit
- Function generator and oscilloscope. NI myDAQ used for both.
- Phosphate buffer solution (PBS)
- Activated yeast (*Saccharomyces cerevisiae*)

3.2 Methods

1.

Variables

- Control:
  - Output voltage on function generator: 10V.
  - Probes 180° from each other. Sense leads 30° from the probe and 180° from each other. See Figure 1.
  - Amplifier gain $\frac{15k}{150} = 100$.
  - Liquid volume 250 ml, 60mm height measured from inside the cup.
- Independent: Frequency.
- Dependent: Potential difference

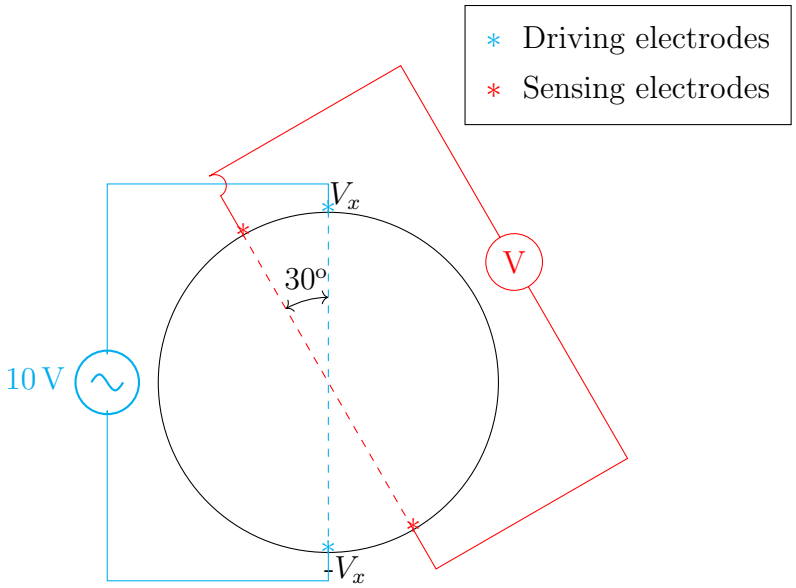


Figure 1: Orientation of electrodes

Considerations Due to a limited power input into the amplifier, a gain ratio of 1000 led to *gain saturation*, where we no longer observed the expected amplification. With gain of 10, the maxima appeared in different frequencies as to those of 100 and 1000, though we are unsure why this would happen.

Although we defined probe orientation, there are two factors we are disregarding: the cup is not perfectly cylindrical, and the leads are not straight – so the distance between electrodes is smaller than what the experimental setup implies. Instrumental uncertainty becomes largely irrelevant, as we expect there to be some movement in the probes as we

Maximum amplitude for PBS around 3000hz, maximum amplitude for yeast at 5000hz. We pick 4000 Hz. There is no concern for cell disruption at the range of frequencies we are analysing (Yerworth, R., 2019 November 28)

4 Results and Discussion

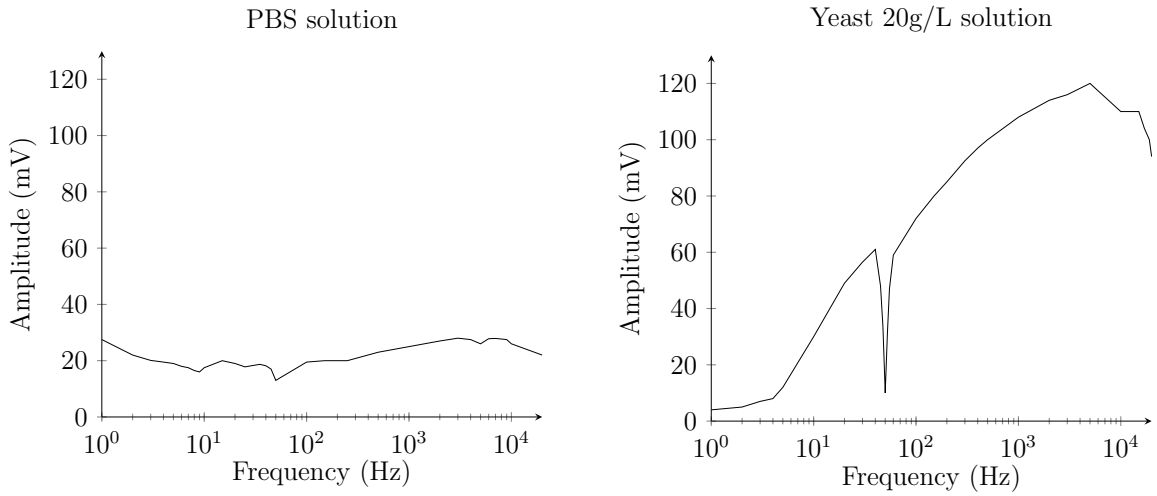
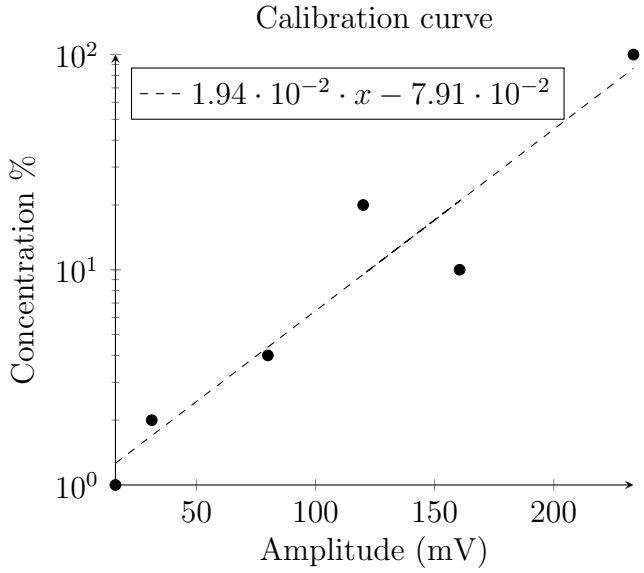


Figure 2: Amplitude based on frequency. Note the dip at 50 Hz due to the filter.



Limitations and improvements Originally it was expected that the impeller would not affect our results, but during the 24h run, we saw that the magnetism that drives the impeller had a very large effect on the numbers seen. This was not taken into consideration in our design, but it definitely should have been.

Assess performance of reactor prototype; cell density with final design

important comments on results, relate to theory and knowledge

5 Conclusion

References

[1] V. F. Lvovich. *Impedance spectroscopy : applications to electrochemical and dielectric phenomena / Vadim F. Lvovich.* 2012.

[2] A. K. Polat, U. Karabacak, V. Mutlu, L. Tomak, and A. Bilgici. Early diagnosis of lymphedema after breast cancer treatment: Bio-impedance spectroscopy. *The journal of breast health*, 13(2):83, 2017.