Anesthesia on Yeast: The Effect of Lidocaine Cream on the Viability of *Saccharomyces cerevisiae* Cells and Its Portrayal of the Impact of Anesthesia on Human Health

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Introduction

Background on Anesthesia

Anesthesia is a medical procedure used to intervene with an individual's ability to feel paid during medical treatments, such as surgery or dentistry (Medline Plus, 2018). There are four types of anesthesia: general anesthesia, monitored sedation, regional anesthesia, and local anesthesia. While anesthesia works by preventing nerves from passing signals to the brain, "scientists don't know exactly how all types of anesthetics work." (National Institute of General Medical Sciences, 2023).

Topical, or over-the-counter (OTC) anesthesia is a type of local anesthesia that can be used to temporarily numb pain without a medical prescription. This type of anesthesia is spread on the surface, and is not an injection, meaning it is only in contact with the skin. While each anesthetic serves a different purpose, in general, they provide symptom relief by reducing pain and itchiness in an area (Clinic, 2024).

In this lab, Aspercreme with lidocaine HCl 4% was used. The National Library of Medicine (2025) states that this is a topical anesthetic that can be used for temporary relief of minor pain. Islam et al. (2024) explain that lidocaine without epinephrine is expected to be active for about 1-2 hours. It is recommended to avoid in areas with poor circulation and open wounds.

Background on S. cerevisiae *(Yeast)*

Saccharaomyces cerevisiae (S. cerevisiae), commonly known as baker's yeast or brewer's yeast, is an organism utilized in biotechnical laboratories for studying ecology and evolution (Stewart, 2014; Bai et al., 2022). Similarities, like their growth and division methods and nearly identical metabolisms, have made yeast a very common replacement for human cells,

especially as *S. cerevisiae* cells are much more amenable (University of Colorado Anschutz, n.d.). Historically, due to modelled similarities with mammal organisms like humans, dogs, mice, cows, and rats, *S. cerevisiae* has been used in studying biopharmaceuticals, vaccines, virus-related investigations, biotherapeutic products, and other medical-related experiments (Karathia et al., 2011; Maneira et al., 2025).

Research

Therefore, the aim of this research was to understand how the length of exposure to lidocaine anesthetics correlated with the percentage of still viable *S. cerevisiae* cells and how this reflected the effect topical anesthetics like lidocaine may have on human cells.

Research question: How does the length of exposure (0 min, 30 min, 60 min, 90 min, 120 min) to diluted anesthesia cream with lidocaine (0.1 g of anesthesia and 0.5 mL of distilled water) affect the viability of *S. cerevisiae* cells?

Hypothesis: An increased length of exposure to the diluted anesthesia cream with lidocaine correlates with a decrease in percentage of still viable *S. cerevisiae* cells.

Materials & Methodology

Ethics & Safety

There were no ethical concerns to consider in conducting this investigation. However, since the investigation was conducted in a high school laboratory, all materials and procedures needed to be safe for direct exposure to individuals, including juveniles. Therefore, *S. cerevisiae* cells were used to represent human cells, and no humans were directly exposed to any anesthesia. Since *S. cerevisiae* are unicellular fungi lacking a nervous system, they are generally

used in daily lives, such as baking and brewing. Thus, they are safe to use in an open high school laboratory. The lidocaine cream, a topical and over-the-counter anesthesia available for the public in the state of Washington, was used for this investigation. As the anesthesia was not used on any humans and this type of anesthesia can be used without a prescription, it was safe to use in an open high school laboratory.

Preparation of Investigation

The independent variable of this investigation is the duration for which the *S. cerevisiae* was exposed to the anesthesia. In medical settings, lidocaine cream is typically applied about 30 minutes to 60 minutes prior to treatment or procedure, meaning that the anesthesia is most effective about 30 to 60 minutes after application (St. Jude Children's Research Hospital, 2025). Therefore, for this lab, the increments of duration of exposure to the anesthesia was determined to be in 30-minute increments, ranging from 0 minutes to 120 minutes of exposure.

The dependent variable is the percentage of *S. cerevisiae* cells that retained their viability, or were alive, after the designated exposure to the anesthesia. Since the number of cells present in each field of view for every trial cannot be controlled to be the same number, in order to accurately compare the viability of cells based on each length of exposure, the proportion of cells that are still viable from the total number of cells present in the field of view needs to be calculated. Therefore, by determining the percentage of *S. cerevisiae* cells that are still viable from each field of view, the difference in proportion of still alive to total number of cells can be used to understand how the length of exposure to anesthesia may affect the number of still viable cells.

Controlled variables:

- Type and dilution of of anesthesia
- Volume of methylene blue used
- Volume and concentrations of YPD and Agar powder used for petri dish bases
- Temperature at which *S. cerevisiae* cells were activated
- Type of cells

Materials:

- Distilled water (at least 600 mL)
- 30 g of YPD powder
- 3 g of agar powder
- 5 g of Red Star active dry baker's yeast (*S. cerevisiae*)
- 1 10 mL graduated cylinder
- 1 500 mL beaker
- 2 1000 mL Erlenmeyer flask
- 11 petri dishes
- 2 pipettes
- 2 sterile loops
- 3 stirring rods
- 5 weigh boats
- 1 thermometer (in Celsius)
- 1 refrigerator
- 1 microwave
- 1 mass balance
- 1 incubator
- 1 light microscope
- 1 clicker, for counting
- Aspercreme with lidocaine HCl 4% (about 1 g)
- Methylene blue (about 10 mL)
- Microscope slides (at least 30)
- Cover glass for microscope slides (at least 30)
- Oil immersion oil (about 10 mL)
- 1 1000x oil immersion lens
- Isopropyl alcohol (about 15 mL)
- Lens paper (at least 30)
- Paper towels
- 1 Sharpie marker

- 1 phone (to take photos and for timer)

Methodology

Part 1 - Creating the base formula for the petri dish plates to grow the *S. cerevisiae*

1. Measure 200 mL of distilled water in a clean 500 mL beaker and pour into a 1000 mL Erlenmeyer flask.
2. Using the weigh boat, measure 20 g of YPD powder on the mass balance and add to the Erlenmeyer flask.
3. Using the weigh boat, measure 3 g of agar powder on the mass balance and add to the Erlenmeyer flask.
4. Using a stirring rod, stir the Erlenmeyer flask until all of the powder has dissolved in the distilled water.
5. Place the Erlenmeyer flask in the microwave and heat the solution until it is a clear solution and no powder particles are visible. Keep watch of the flask while it is in the microwave and be careful that it does not overflow due to bubbling. If the solution begins to bubble, pause the microwave and let the solution settle before heating it again.
6. Remove the Erlenmeyer flask from the microwave and let it cool enough so that it will not melt the plastic of the petri dishes but that the solution is still liquid in the flask.
7. While waiting for the solution to cool down, use the Sharpie marker to label the ten petri dishes by number.
8. Pour a thin layer of the solution (up to about a third of the height of the petri dish) into each of the labeled petri dishes.

9. Wait until the solution in each plate has cooled and solidified, then cover each petri dish with a cover.
10. If not inoculating the petri dishes immediately, place them inverted in a refrigerator until further use.

Part 2 - Activating the *S. cerevisiae*

1. Measure 200 mL of distilled water and pour into a 1000 mL Erlenmeyer flask.
2. Using a microwave, heat the flask to 30 degrees Celsius.
3. Using the mass balance, measure 10 g of YPD powder on a weigh boat and add it to the Erlenmeyer flask.
4. Using the mass balance and a weigh boat, measure 5 g of Red Star active dry baker's yeast (*S. cerevisiae*) and add it to the Erlenmeyer flask.
5. Using a stirring rod, stir the solution gently, until the *S. cerevisiae* and YPD powder appear to be "dissolved" in the flask.
6. Cover the Erlenmeyer flask with a paper tower and leave it at room temperature for 12 hours.
7. After the 12 hours, use a sterile loop to inoculate the now activated *S. cerevisiae* onto petri dish 1 from part 1. Label this petri dish "control."
8. Invert the inoculated plate and place it in an incubator at 35 degrees Celsius for 24 hours.
9. After 24 hours, inoculate the other eight petri dishes with the *S. cerevisiae* now grown on the control plate.

Part 3 - Counting the number of viable and total number of cells under the microscope

1. Using a pipette, place one drop of distilled water onto a microscope slide.

2. Using a sterile loop, pick up a small amount of *S. cerevisiae* and dilute it in the distilled water on the microscope slide.
3. Using a sterile pipette, place half a drop of methylene blue onto the diluted *S. cerevisiae*.
4. Using a sterile loop, gently mix the methylene blue, distilled water, and *S. cerevisiae* on the microscope slide.
5. Place a cover glass over the solution on the microscope slide.
6. Place a single drop of oil immersion oil on top of the cover glass.
7. Place a 1000x oil immersion lens onto the microscope.
8. Using the microscope and oil immersion methods, focus the microscope until individual cells, both dyed and not dyed by the methylene blue, are visible. Take a picture of the field of view for future data analysis.
9. Clean the microscope slide with isopropyl alcohol and distilled water. Clean the 1000x oil immersion lens with isopropyl alcohol and lens paper.
10. Repeat steps 1 through 9 of Part 3 four more times, for a total of 5 trials with no exposure to the anesthesia.
11. Take a photo of the field of view, and using the microscope pointer in the field of view for reference, divide the field of view into 8 equal sections.
12. Count the total number viewable *S. cerevisiae* cells in the clearest of the 8 sections and record it in the raw data table.
13. Count the number of still viable *S. cerevisiae* cells in the clearest of the 8 sections and record it in the raw data table.

Part 4 - Exposing the *S. cerevisiae* to the anesthesia

1. Using a weigh boat and the mass balance, measure out about 0.1 g of Aspercreme with lidocaine HCl 4% and place it onto the clean petri dish.
2. Using a 10 mL graduated cylinder, measure 0.5 mL of distilled water pour it over the Aspercreme with lidocaine in the petri dish.
3. Using a stirring rod, thoroughly mix the solution until all of the Aspercreme with lidocaine has dissolved in the distilled water.
4. Of the eight petri dishes, select a plate with an abundance of *S. cerevisiae* growth.
5. Using the Sharpie marker on the underside of the petri dish, outline sections of the petri dish where there is at least about 1 mm of *S. cerevisiae* grown. Number each of these sections.
6. Using a sterile transfer loop, transfer the diluted anesthesia onto one of the marked sections of *S. cerevisiae* in the selected petri dish. Gently spread the anesthesia over the cultured section until it appears to be evenly spread out.
7. Set a 30-minute timer and let the *S. cerevisiae* be exposed to the anesthesia for the set amount of time.
8. Follow the procedure steps 1 through 9 in Part 3 to determine the total number of cells in the field of view and the number of still viable cells in the field of view. Depending on how much *S. cerevisiae* grew on each petri dish, switch plates and mark new sections on each plate, as needed.
9. Using one of the other marked sections of cultured *S. cerevisiae,* repeat steps 1 through 8 of Part 4, 4 more times, for a total of 5 trials.

10. Repeat steps 8 and 9 of Part 4, increasing the length of exposure by 30 minutes until each increment of time between the range of 0 minutes to 120 minutes has been trials.

Results

Table 1. Raw Data: Number of viable *S. cerevisiae* cells counted per ⅛ field of view

Duration of Exposure to Lidocaine Cream (min)	**Trial**	**Dilution/Concentration of Lidocaine Cream (± 0.1 g, ± 0.05 mL)**	**Number of Viable *S. cerevisiae* Cells per 1/8 of Field of View**	**Total Number of *S. cerevisiae* Cells per 1/8 of Field of View**
0	1	N/A	42	48
	2	N/A	57	60
	3	N/A	63	66
	4	N/A	59	65
	5	N/A	45	53
30	1	0.1 g lidocaine with 0.50 mL	36	45
	2	0.1 g lidocaine with 0.50 mL	32	56
	3	0.1 g lidocaine with 0.50 mL	27	48
	4	0.1 g lidocaine with 0.50 mL	34	40
	5	0.1 g lidocaine with 0.50 mL	38	57
60	1	0.1 g lidocaine with 0.50 mL	29	64
	2	0.1 g lidocaine with 0.50 mL	24	40
	3	0.1 g lidocaine with 0.50 mL	40	54
	4	0.1 g lidocaine with 0.50 mL	47	56
	5	0.1 g lidocaine with 0.50 mL	35	43
90	1	0.1 g lidocaine with 0.50 mL	10	34
	2	0.1 g lidocaine with 0.50 mL	22	50
	3	0.1 g lidocaine with 0.50 mL	16	43
	4	0.1 g lidocaine with 0.50 mL	18	38
	5	0.1 g lidocaine with 0.50 mL	21	44
120	1	0.1 g lidocaine with 0.50 mL	6	34
	2	0.1 g lidocaine with 0.50 mL	11	45
	3	0.1 g lidocaine with 0.50 mL	7	32
	4	0.1 g lidocaine with 0.50 mL	12	42
	5	0.1 g lidocaine with 0.50 mL	8	39

Table 2. Processed Data: Percentage of still living *S. cerevisiae* cells after exposure to the anesthesia in the ⅛ field of view counted.

Duration of Exposure to Lidocaine Cream (min)	Trial	Calculated Percentage of Viable *S. cerevisiae* Cells in ⅛ Field of View	Average Number of Viable *S. cerevisiae* Cells on ⅛ Field of View	Standard Deviation	2 Standard Deviation
0	1	87.5	90.7	4.62	9.24
	2	95.0			
	3	95.5			
	4	90.8			
	5	84.9			
30	1	80.0	69.0	13.1	26.2
	2	57.1			
	3	56.3			
	4	85.0			
	5	66.7			
60	1	45.3	68.9	16.2	32.3
	2	60.0			
	3	74.1			
	4	83.9			
	5	81.4			
90	1	29.4	41.1	7.81	15.6
	2	44.0			
	3	37.2			
	4	47.4			
	5	47.7			
120	1	17.6	17.4	4.16	8.32
	2	24.4			
	3	21.9			
	4	28.6			
	5	20.5			

The following formula was used to determine the percentage of viable *S. cerevisiae* cells in each ⅛ field of view.

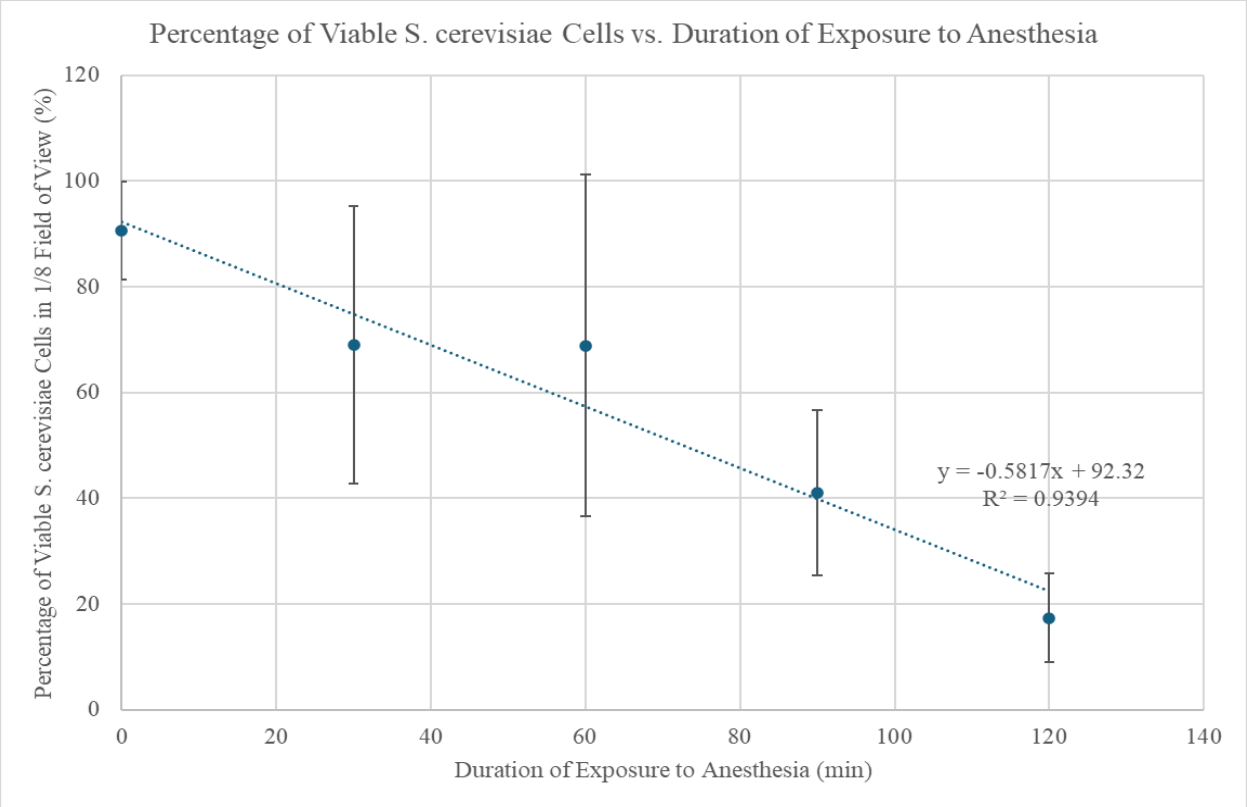
$$\left(\frac{number\ of\ viable\ S.\ cerevisiae\ cells}{total\ number\ of\ S.\ cerevisiae\ cells}\right)(100\%) = percentage\ of\ viable\ S.\ cerevisiae\ cells$$

The average percentage of viable *S. cerevisiae* cells in each ⅛ field of view for each duration of exposure to the anesthesia was calculated to represent the result of all five trials per manipulation of the independent variable. The standard deviation was calculated using the "=STDEV" function on Microsoft (MS) Excel. This standard deviation was then doubled to determine the margins of error of the data.

Justification of Uncertainties

The uncertainties of the average percentage of viable *S. cerevisiae* cells in ⅛ field of view for each duration of exposure was calculated to be two times the standard deviation, as shown in Table 2. A single standard deviation shows the possible variation from the best-fit trend line. By doubling this standard deviation, possible errors, including miscountings and variation in concentration of the diluted anesthesia are also considered (additional information about margins of error in the Evaluation, Weaknesses section).

Graph 1. Correlation between the percentage of viable *S. cerevisiae* cells and duration of exposure to the anesthesia



The data points on Graph 1 are based on the average percentage of viable *S. cerevisiae* cells in ⅛ of field of view per length of exposure (Table 2). The error bars are the two standard deviations that can also be found in Table 2.

Data Analysis

Based on Graph 1, it appears that there is an overall linear, negative correlation between the duration of exposure to the diluted anesthesia and percentage of still living *S. cerevisiae* cells. This means that an increase in length of exposure to anesthesia corresponds with a decrease in percentage of still viable *S. cerevisiae* cells. This supports the hypothesis.

MS Excel calculate that the equation of the trendline of the graph is

$$y = -0.5817x + 92.32$$

The slope -0.5817 percentage of cells per minute shows that for every one-minute that the *S. cerevisiae* was exposed to the anesthesia, the number of viable cells decreased by 0.5%.

The y-intercept (0, 92.32) shows that even when the *S. cerevisiae* was not exposed to any anesthesia, the percentage of still living cells was only about 92%. Ideally, the y-intercept should have been 100%, so this difference may have been due to a lack of nutrition in the petri dish or exposure to unwanted particles. This also may have influenced the following rates of dead cells, in addition to the increase in length of exposure to the anesthesia.

The coefficient of determination (R^2) value, as given by MS Excel, is 0.9394, meaning that there is a strong correlation between the best-fit linear line and the actual data points.

The following steps were taken to find the prospective x-intercept of the graph.

$$\text{Let } y = 0$$

$$0 = -0.5817x + 92.32$$

$$x = 159$$

This means that based on the best-fit trendline, after about 159 minutes of exposure to the diluted anesthesia, 0% of the *S. cerevisiae* cells in the field of view would still be living. After 159 minutes, there would no longer be any cells to reproduce, meaning that no more *S. cerevisiae* will be present in the field of view.

The error bars are relatively large, showing that there is possibility for a wide range of variation in the calculated data points. All of the errors bars overlap with the best-fit line on the graph, meaning that the actual data points are likely relatively accurate. However, the overlapping of the error bars with each other shows that the data points on the graph are not as

independent of each other. This implies that while within the range of data collected for this experiment, a linear, negative correlation may appear to be the best representation, if a wider range of data was collected, this correlation may be different.

Conclusion & Evaluation

Conclusion

Based on the results and data analysis of this investigation, it can be concluded that the longer the exposure to anesthetic processes such as lidocaine cream, the lower the survival rate of *S. cerevisiae* cells. As the negative linear representation of the graph shows, it can be assumed that there will be a point in the extended duration of exposure during which all *S. cerevisiae* cells will have died. While this investigation was conducted on *S. cerevisiae*, this correlation between duration of exposure to anesthetic processes and viability of cells should also be considered when applied to humans. Whether it is minor anesthesia like lidocaine cream to numb a small area, or a major anesthesia across the entire body in preparation for a surgery, it is important to consider and recognize how such applications may impact the overall health and survival of vital parts of the body, beyond simply reducing neural signals.

There have been other professional studies conducted on the effect lidocaine anesthesia and other types of topical anesthesia has on the viability of human cells. Jacobs et al. (2011) found that “local anesthetics containing lidocaine are significantly more toxic to mature human articular chondrocytes than a saline 0.9% control group,” recommending cautious clinical practices. Kim et al. (2020) state that local anesthetics, such as lidocaine and bupivacaine, “affect not only the viability but also the differentiation capacity of adult stem/progenitor cells from various anatomical sites.” However, these studies were all conducted in the context of being used

during a medical procedure. If these are just the possible effects of using lidocaine-like anesthetics under the supervision of a medical professional, it is even more important to understand the possible results of using over-the-counter anesthetics.

Weaknesses & Extensions

One major weakness of the experiment was the lack of precision in time of exposure to the anesthesia. The measured and recorded time of exposure was only while the *S. cerevisiae* was still in the petri dish. However, while preparing the microscope slide to view, transferring the sample of *S. cerevisiae* onto the slide, and focusing the microscope to view the cells, some time lapsed during which the cells were still exposed to the anesthesia. Therefore, the time measurements recorded do not accurately represent the exact length of exposure to the anesthesia. Since the aim of this experiment was to understand how an increased time of exposure impacts the viability of *S. cerevisiae* cells, while the recorded time lengths may not be precise, the increments should not influence the overall analysis of the data. However, if this experiment is conducted again, including the time it took to actually view and take a picture of the *S. cerevisiae* cells underneath the microscope would be helpful in more accurate data collection.

Another weakness is the inaccuracy that may have occurred because the field of views were divided into eight sections. While the field of view was divided into eight sections for practicality, the percentage of viable cells in other sections of the field of view may have been different. Thus, if the photo of the entire field of view had been considered, the analyzed data could also be different. Since it would not be efficient to count all the cells in the field of view, however, if this lab is conducted again, it may be more accurate to divide the field of view into a

grid pattern. Then, using a random number generator, about three of the grids can be selected for data collection. Averaging this collected data may provide a more accurate representation of the percentage of still living *S. cerevisiae* cells in each field of view after a set length of exposure.

Additionally, manually counting the total number of cells and still viable cells likely resulted in inaccuracy. Thus, if this lab was conducted again, having multiple people count the cells in order to verify the number of cells present could be helpful in making the data more accurate. Ultimately, since the percentage of still viable cells was used for data analysis, the exact number of alive and total number of cells likely did not have a large impact in the final conclusion of the lab. However, taking further steps like checking the counted number of cells could be helpful in collecting more accurate data.

References

Bai, F.-Y., Han, D.-Y., Duan, S.-F., & Wang, Q.-M. (2022). The Ecology and Evolution of the Baker's Yeast Saccharomyces cerevisiae. *Genes*, *13*(2), 230. https://doi.org/10.3390/genes13020230

Clinic, C. (2024, March 27). *What Is a Topical Anesthetic? Uses, How They Work*. Cleveland Clinic. https://my.clevelandclinic.org/health/articles/topical-anesthetic

Islam, R. K., Tong, V. T., Robicheaux, C., Tageant, H., Haas, C. J., Kline, R. J., & Islam, K. N. (2024). The Impact of Anesthesia on Dermatological Outcomes: A Narrative Review. *Cureus*. https://doi.org/10.7759/cureus.72321

Jacobs, T., Vansintjan, P. S., Roels, N., Herregods, S. S., Verbruggen, G., Luc Herregods, & Almqvist, K. (2011). The effect of Lidocaine on the viability of cultivated mature human cartilage cells: an in vitro study. *Knee Surgery, Sports Traumatology, Arthroscopy*, *19*(7), 1206–1213. https://doi.org/10.1007/s00167-011-1420-5

Karathia, H., Vilaprinyo, E., Sorribas, A., & Alves, R. (2011). Saccharomyces cerevisiae as a Model Organism: A Comparative Study. *PLoS ONE*, *6*(2), e16015. https://doi.org/10.1371/journal.pone.0016015

Kim, Y. H., Park, G. Y., Rabinovitch, N., Solaiman Tarafder, & Lee, C. H. (2020). Effect of local anesthetics on viability and differentiation of various adult stem/progenitor cells. *Stem Cell Research & Therapy*, *11*(1). https://doi.org/10.1186/s13287-020-01905-2

Maneira, C., Chamas, A., & Lackner, G. (2025). Engineering Saccharomyces cerevisiae for medical applications. *Microbial Cell Factories*, *24*(1). https://doi.org/10.1186/s12934-024-02625-5

Medline Plus. (2018). *Anesthesia*. Medlineplus.gov; National Library of Medicine. https://medlineplus.gov/anesthesia.html

National Institute of General Medical Sciences. (2023). *National Institute of General Medical Sciences*. National Institute of General Medical Sciences (NIGMS). https://www.nigms.nih.gov/education/fact-sheets/Pages/anesthesia

National Library of Medicine. (2025). *Welcome To Zscaler Directory Authentication*. Nih.gov. https://dailymed.nlm.nih.gov/dailymed/drugInfo.cfm?setid=d5da67c3-c2ec-4e3e-abe9-cc862dcfc8bd

St. Jude Children's Research Hospital. (2025). *Using Lidocaine Cream for Needle Pain*. St. Jude Together. https://together.stjude.org/en-us/medical-care/medication-management/using-lidocaine-cream-for-less-needle-pain.html

Stewart, G. G. (2014). *Saccharomyces cerevisiae - an overview | ScienceDirect Topics*. Sciencedirect.com. https://www.sciencedirect.com/topics/neuroscience/saccharomyces-cerevisiae

University of Colorado Anschutz. (n.d.). *Yeast Molecular Genetics and Cell Biology*. Medschool.cuanschutz.edu. https://medschool.cuanschutz.edu/biochemistry/research/yeast-molecular-genetics-and-cell-biology