APP N15-2

Engineering 1282H Spring 2020

Team Y4

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This worksheet is a group assignment and is designed to help your team analyze the results of the Lab 03 and assess the procedure. This will require Excel **and** the fluid mechanics program you created. Questions 1-13 are about the data. Please submit both this worksheet and the spreadsheet to Carmen as a single PDF when complete.

1. Using the given spreadsheet, APP_N10_1_TABLE.xlsx, fill in the specific heights, change in volume, time of trial, number of cells after flow, percent of total cells and the experimental flow rate for each data point. These are all measured values or simple calculations. Place these columns from the spreadsheet in the box below.

		٦	Г able 1: Basic D	ata			
		Height (cm)	Volume (mL)	Time (min)	# of cells after flow	Percent of	Flow rate
Data Point 1	Height 1 (Low Pressure Flush)	25.4	6.46	3	675	1	2.15333
-	Height 2	30	13.91	2	273	0.404444	6.95
	Height 3	40	22.2	2	187	0.277037	11
	Height 4	50	31.52	2	180	0.266667	15.7
Data Point	Height 1 (Low Pressure Flush)	25.4	5.45	3	276	1	1.81666
2	Height 2	30	13.42	2	95	0.344203	6.
	Height 3	40	21.58	2	33	0.119565	10.
	Height 4	50	31.27	2	23	0.083333	15.6
Data Point 3	Height 1 (Low Pressure Flush)	25.4	6.93	3	215	1	2.3
3	Height 2	30	14.97	2	121	0.562791	7.4
	Height 3	40	23.04	2	63	0.293023	11.
	Height 4	50	32.76	2	27	0.125581	16.

2. Next, using your heights, calculate the pressure and shear stress in the channel for each data point. Enter these cells from the spreadsheet in the box below.

	Table 2: Pressu	re and Shear Stress	
Pressure using height	Shear Stress using Heigh	Shear stress using flow rate	Pressure using flow rate
24892	. 124.46	50.22	10044.704
29400	147	162.22	32443.149
39200	196	258.89	51778.426
49000	245	367.58	73516.035
24892 29400 39200 49000	147 196	156.50 251.66	8474.247 31300.292 50332.362 72932.945
24892 29400			10775.510 34915.452
39200			53737.609
			==:== :==

3. Using your above values, create a plot of Cell Percent vs. Shear Stress with trendlines and place it in the box below. Try different regressions to get a best fit for your data. Show the trendlines on the plot. (Please note that regressions should make physical sense as well as fit the data)

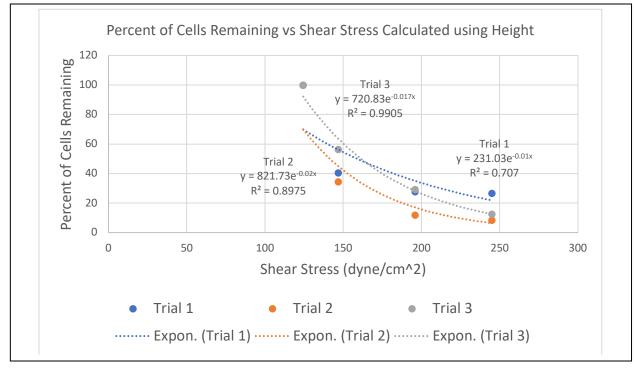


Figure 1: Plot of Cell Percent and Shear Stress

4. What did your group define as "Threshold Yeast Shearing"? How did you come to this decision?

This group defined the Threshold Yeast Shearing as the shear stress that allowed for less than 50% of cells to remain. The threshold yeast shearing that we determined was 175 dyne/cm². This allowed for less than 50% of the cells to remain in the channel.

5. Is this trendline method a valid way of determining threshold shear stress? If so, why? If not, how else could you collect data to get a better fitting trendline?

This method is an adequate form of data collection but could work better with more data points. Because there are only three points, one data point can skew the entire data set and make trendlines less representative of the data's characteristics. For all three sets, however, the r-squared value was above 0.7, providing ample assurance that there correlation between the data points and validity in using trendlines to analyze the data's trends.

6. Interpolate the threshold shear stress based on your team's defined threshold point and your trendlines. Fill the table with the results, and show a sample calculation

Table 3: Threshold shear stress calculated for each trial.

Trial:	Threshold Shear Stress (H, dyne/cm^2)
1	153.05
2	139.97
3	1156.96

Sample Calculation:

$$50 = 231.03e^{-0.01x}$$

$$0.216 = e^{-0.01x}$$

$$-1.53 = -0.01x$$

$$153.05 = x$$

7. Now, instead of using your height values, you will use the volumetric flow rate values to calculate the pressure. Calculate the shear stress using these pressures in the channel during each trial. Enter these cells from the spreadsheet in the box below.

	Height (cm)	Volume (mL)	Time (s)	Shear stress using flow rate
Height 1 (Low Pressure	25.4	C 45	100	50.22
Flush)	25.4			
Height 2	30	13.91		
Height 3 Height 4	40 50	22.2 31.52		
Height 1 (Low Pressure Flush) Height 2 Height 3 Height 4	25.4 30 40 50		120 120	156.50 251.66
Height 1 (Low Pressure Flush) Height 2 Height 3 Height 4	25.4 30 40 50	14.97 23.04	120 120	174.58 268.69

8. Now, using your shear stress values from the volumetric flow rates and your cell percentages, construct a plot similar to that in Step 3. Make sure to add the trendline equations.

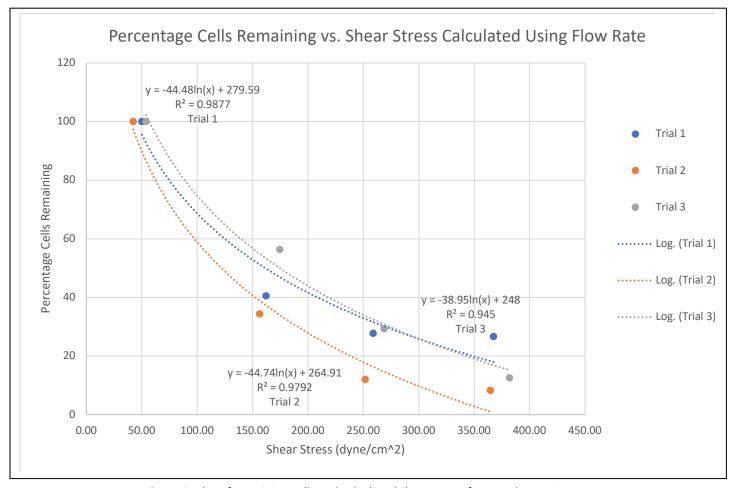


Figure 2: Plot of remaining cells and calculated shear stress from each experiment.

9. Calculate threshold shear stress based on your values and defined threshold level. Show the shear stresses in the table below as well as a sample calculation.

Table 6: Threshold shear stress calculated for each trial.

Trial:	Threshold Shear Stress (Q, dyne/cm^2)
1	174.45
2	121.94
3	161.33

Sample Calculation:

$$50 = -44.48 \ln(x) + 279.59$$
$$-229.59 = -44.48 \ln(x)$$
$$5.16 = \ln(x)$$

$$174.45 = x$$

10. Calculate the average of all values (excluding time) for each height. Now, calculate percent difference from average between each data point and its respective average. Do this for: volume, cell count, cell %, flow rate, pressure (Q), shear stress (Q), and threshold shear stress (H and Q). Place only the percent difference results in the table.

			Volume	Cell Count	Cell %	Flow Rate	Pressure using Height	Shear Stress using Height	Shear Stress using Flow Rate	Pressure using Flow Rate
Difference	Data point 1									
		Height 1 (Low Pressure Flush)	2.825745683	53.83892197	0	2.825736811	C	0	2.820314341	2.82574848
		Height 2	1.356658336	50.4587156	7.77137356	1.356370787	(0	1.355157353	1.35665825
		Height 3	0.329785639	65.87677725	18.60750886	0.329725777	(0	0.330353298	0.32978529
		Height 4	1.041502288	80.51948052	50.86599819	1.041375388	(0	1.041037068	1.04150233
	Data Point 2									
		Height 1 (Low Pressure Flush)	14.15174766	33.90170512	0	14.15133426	C	0	14.15308137	14.1517470
		Height 2	4.941860465	52.71317829	23.79042538	4.942158346			4.943913585	4.94185989
		Height 3	3.162055336	96.33507853	63.1352051	3.162180799			3.162519065	3.16205469
		Height 4	1.837769328	107.6923077	62.17948718	1.837897236			1.83855162	1.83776885
	Data Point 3									
		Height 1 (Low Pressure Flush)	9.841029523	57.53727223	0	9.840711823	C	0	9.847132518	9.84102641
		Height 2	5.985552116	29.57746479	25.13052731	5.985552116	(0	5.985998584	5.98555153
		Height 3	3.383845814	39.83050847	24.15309843	3.38390566	(0	3.384826942	3.38384488
		Height 4	2.816901408	95.81993569	23.1923602	2.816901408	0	0	2.817200495	2.81690099

11. Comparing τ_H and τ_Q , which is better to use when analyzing results? Support your answer by discussing both your interpolations from above and sources of error in the experiment.

Shear stress using flow rate would be better to use when analyzing results. As seen in the spreadsheet above, this metric changed with the experimental data points, and was not a "one size fits all" value like the ones calculated from height. These give us a better picture of the shear stress for our lab. Moreover, it also alerts us to potential errors in our work as we can check its difference from calculated shear stress as a canary for possible error.

12. Comment on these values. How repeatable is this experiment? How consistent are your results?

The percentage differences of many of the values varies. For example, cell count from height 4 exhibited 94 percent difference from the average in data point 3. Since data points two and three exhibited relatively similar values in cell count, but data point 1 had an extremely high count, it seems that the average was skewed. Unfortunately, much of the data from this lab followed the same pattern of unusual variation. The percentage difference between the cell counts was by far the highest, but the variation in shear stress was notable as well, ranging from 1 to 5 percent variation across the data points.

Outside of the cell count and percentage remaining, this lab did yield fairly consistent results across data points. However, due to the high variability exhibited in the cell count measurements, it cannot be seen as consistent. The rest of the lab seemed to have high repeatability and consistency, though, as the variability ranges from 1 to 14 percent difference across all other metrics and data points. Therefore, this lab was somewhat consistent but contained man anomalies and uneven data in the cell count and cell percentage metrics.

13. If you were unable to collect sufficient data, discuss what challenges prevented you from gathering data.

This lab became problematic at several different times. It started off well, with consistent results coming in from day 1, but soon became much less fruitful. After manufacturing our own chips, we found immense difficulty in achieving flow of any kind. The TAs confirmed this was an expected issue, as they believed the batch of PDMS chips we made were "faulty" and were expected to have poor results. However, our lab did not get any easier, even after trying to use new chips. Achieving a consistent flow became much more of a challenge and most of the final moments spent in the lab were used to clean up the sheer mess we had made from our numerous attempts to rectify the chips.

Questions 14 – 18 are about the experimental procedure.

14. Why might the use of a reverse flush step be useful prior to performing a shearing trial?

The use of a reverse flush step ensures that the remaining yeast cells are the ones only adhered to the walls of the chamber, allowing for an accurate count of the cells before and after the shearing. Using this method, it is easier to determine whether the test variable had an effect on the experiment.

15. What are the pros and cons of allowing the yeast to incubate for relatively longer periods of time before proceeding with the next steps?

Allowing the yeast to incubate for a longer period would help to ensure the maximal production of biofilm. It would also allow for the most cells to adhere to the walls of the channel. However, this would also mean that no cells would be sheared, so a data point would not be able to be collected.

16. What design features in your chip can help address issues with delivery, incubation, clogging, or clumping?

Making the entrance well bigger would help future implementations of this experiment as it would allow for more yeast to incubate in the channel. The exit port is smaller to allow for a more laminar flow. Additionally, keeping the channels more centered in the chip would help to evenly distribute pressure and would prevent clumping (because the yeast would flow and incubate in the channels evenly).

17. Do you think the flow rates in the channel with yeast cells should be the same or different from a channel with no yeast cells in it? Why or why not?

The flow rates of channels inoculated with yeast cells would be much less than those of normal, untouched channels, as the cells would take up most of the free space inside the channel and prevent efficient flow. There would be more drag along the walls of the yeast-lined channel as well, since the surfaces would be uneven and would interact with the liquid as it collided with each cell and broke portions of the wall off.

18. Could it be possible (could you) and practical (should you) to run multiple shearing trials on a single chip? How would you modify your procedure? Could you demonstrate that conditions are the same?

It is possible to run multiple shearing trials on a single chip since it can be cleaned out and reincubated with yeast cells. In order for all the trials to be tested under the same conditions, we should absolutely run several shearing trials on one chip. Since the chip would be effectively cleaned each time, we could show (using a microscope) that the layout of the channel remains the same across tests.