Subject: Environmental Systems and Societies SL

Title: An investigation into the effect of different temperature conditions on the rate of growth of common wheat seeds.

Word count: 2245

Candidate number: hlx449

Identifying the context

Research question: To what extent does increased environmental temperature decrease the amount of germination and initial growth of *Triticum aestivum* (common wheat) seeds?

Environmental issue: Global warming due to human activities and the impact of this on food production

A serious environmental issue faced today is global warming. The Earth has been increasing in temperature since the 1800s. Ongoing research shows that since 1880, the Earth has warmed by 0.8˚C (GISTEMP, 2019). It is predicted to rise further by 2.5 - 4.5˚C before the end of 21st century (IPCC, 2007). Most temperature increase has occurred recently, with 9 out of 10 of the warmest years on record occurring since 2005 (NOAA, 2019). Scientists agree that this temperature increase is due to human activity (NASA, 2020). This is supported by trends in surface temperature anomalies since the early 1900s, when industrialisation spread globally. As seen in Figure 1, the surface temperature anomalies had increased from the reference value by almost 1.00˚C in 2016. The pattern suggests that global anomalies will continue to rise.

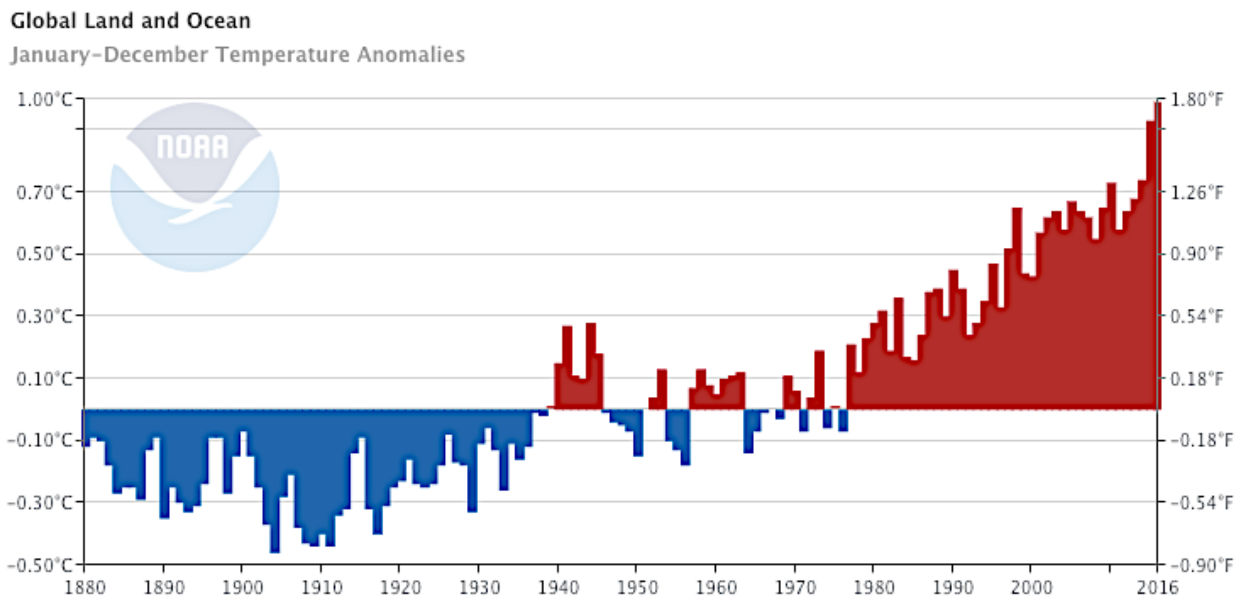


Figure 1 (NOAA, 2019) – a bar chart of annual temperature anomalies since 1880, showing a pattern of an uninterrupted increase in warming since the late 1970s.

Many species have an optimum temperature range in which they can grow (Miller & Stillman, 2012). Global temperature changes may mean that this thermal optimum no longer exists for certain species as abiotic factors within ecosystems change. Species such as wheat, rice and corn are key species in agriculture (Pimentel, 1993). If temperatures change due to global warming, then growing conditions, and even biome locations may shift and/or deteriorate. This could lead to fewer/poorer quality crops which could have detrimental implications for feeding Earth's ever-growing population.

Connections between issue and experiment: Common wheat (*Triticum aestivum*) is relied on by 4.5 billion people globally for daily calories (GCARD, 2012). It is the most widely grown crop globally, with demand expected to grow by 60% due to predicted global population of 9 billion people by 2050 (GCARD, 2012). It has an optimum germination temperature of 12-25˚C (Acevedo et al, n.d.). It is one of Australia's major crops, with 22 million tonnes produced annually (Agricultures Australia, 2019). Western Australia has a production area of 5 million hectares (Agricultures Australia, 2019). Wheat is planted in winter/spring for harvest late spring/mid-summer (Curtis, 2019). Western Australia's average spring-time (September, October, November) temperature is between 9.6°C–26.8°C (Curtin University, 2018), similar to wheat's optimum germination temperature of 12-25˚C (Acevedo et al, n.d.). Therefore predicted increases of up to 4.5˚C (IPCC, 2007) could impact Western Australia's wheat production if maximum spring temperatures increased to 31.3˚C: 6.3˚C warmer that wheat's uppermost optimum germination temperature.

This led to the research question:

To what extent does increased environmental temperature decrease the amount of germination and initial growth of *Triticum aestivum* (common wheat) seeds?

This will be investigated through an experiment on common wheat seeds to see if controlled temperature increase impacts their germination and initial growth. If temperature increase proves detrimental to *Triticum aestivum* germination, it may have negative implications as to the impact of global warming on wheat growth both in WA and globally.

Planning

Equipment List:

- 25 petri-dishes
- 250 common wheat seeds (*Triticum aestivum)*
- 4 Incubators set at 20˚C, 25˚C, 30˚C, 35˚C
- Distilled water
- Measuring cylinder
- 4 Thermometers(˚C)
- Max/min digital thermometer(˚C)
- 3% sodium hypochlorite solution
- Beaker
- Sieve
- Filter paper
- Weighing scales(mg)

Independent and dependent variables:

	What is it?	**How is it measured?**
Independent	The temperature of the *Triticum aestivum* seeds manipulated by incubators set at different temperatures.	Measured by thermometers in each of the incubators. Room temperature range measured with a max/min digital thermometer and the average found.
Dependent	A count of the number of seeds germinated.	The number of seeds in each trial counted by hand and recorded at each observation.
Dependent	The biomass of the seeds at the end of the experiment.	Final germinated seeds dried, and their dry biomass weighed.

Controlled variables:

Variable	How was it controlled?
Type of wheat seeds	All seeds were *Triticum aestivum* and came from same seed batch.
Number of wheat seeds per petri dish	Seeds were counted by hand as they were placed into dish.
Number of replicates	Every temperature trial had 5 replicates.
Sterilisation of seeds	Seeds were sterilised altogether prior to separation to ensure a consistent amount of sterilisation.
Time in incubators	All trials had the same amount of time in the incubators as they were all removed and measured at the same observation times, for the same duration.

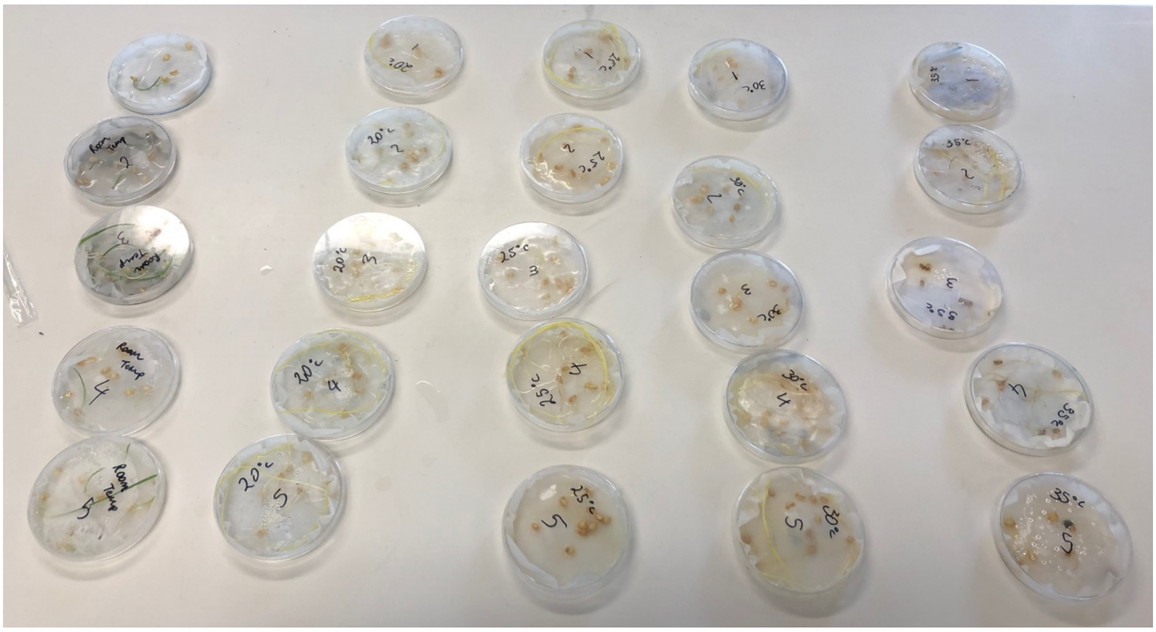
Method:

1) The wheat seeds were sterilised by soaking in 3% bleach solution (sodium hypochlorite) for 15 mins, and then thoroughly washed with water.
2) Filter paper placed in labelled petri-dishes. Using tweezers, 10 sterilised seeds were placed in each and 20ml distilled water tipped into each dish.
3) The lids were placed on the dishes and 5 petri-dishes placed in each temperature zone (16 ˚C, 20˚C, 25˚C, 30 C, 35˚C).
4) The dishes were checked at 3, 5 & 10 days, the number of seeds that germinated in each counted and recorded. If any trays were found to be dry, 5-20mls of distilled water was added to adequately dampen them, depending on how dry they were.
5) After 10 days, all petri-dishes were placed in 35˚C incubators with their lids off for 5 days to dry them out.
6) The biomass of the dried seeds from each dish was then recorded by first weighing the seeds on the filter paper, then scraping the seeds off, weighing the paper alone and subtracting the first value from the second.

<u>*Justification of method:*</u>

The data collection in this experiment had 10 seeds in each petri-dish, with 5 replicates of each temperature (Image 1). This increases sample size and therefore improves the validity of the results, as a greater sample size is a more accurate representation of the population. The seeds were sterilised before use to prevent mould growth which could impact germination. The temperatures of the dishes were chosen to represent at regular intervals the possible increased temperatures in Western Australia due to global warming. 16˚C, 20˚C and 25˚C represent 'optimal' growth temperatures for *Triticum aestivum* seeds (Acevedo et al, n.d.) and 30˚C and 35˚C represent the upper temperatures that could be expected in the future (IPCC, 2007). Originally, 20ml of distilled water was added, as it was a sufficient amount to keep the seeds moist to allow germination between observations (they were being heated and so evaporation had to be considered to ensure the seeds did not dry out). Either 5ml, 10ml or 20ml of distilled water was then added to different petri-dishes if they became drier, as it was important to ensure the seeds were wet to allow the best opportunity for germination.

Image 1: The 25 labelled petri dishes with germinated seeds on the final day of the experiment.



Risk assessment:

Risk	Why is it a risk?	Mitigation
Handling glass	Glass is easily breakable, and broken glass is dangerous as it can cut skin.	Care taken when handling glass equipment. If any glass equipment is broken, gloves should be worn to pick it up and a teacher should be informed.
3% hypochlorite solution	Solution is corrosive, and so can cause skin irritation on contact such as itching, redness and pain.	Gloves, goggles and lab coat should be worn to protect skin. If any solution contacts skin, it should be rinsed thoroughly with water.
Water near electrically powered incubators.	Water can cause electrical equipment to short circuit. It can also electrocute people if the water is in contact with electricity.	Care must be taken not to spill and water and to keep wires and sockets as protected as possible.

Ethical considerations:

The ethical considerations of this experiment are minimal. The 3% hypochlorite solution is corrosive, however is diluted enough to be safely disposed of domestically with minimal harm to aquatic life. There are no other animal considerations in this experiment. Wheat is a commonly available, non-endangered species, meaning that its use in the experiment does not threaten the species and is ethically sound. The germinated seeds and filter paper are safe to be disposed of domestically with no damage to the environment.

Results, Analysis and Conclusion

There were two sets of data collected from this investigation. The first was a count of the number of seeds germinated in each petri-dish at day 3, 5 and 10 days of the experiment. The second was total biomass of each petri-dish on the final day of the experiment. Each set of data had the mean, standard deviation and 95% confidence intervals calculated in order to provide further insight into the reliability of the results. Example calculations are shown below each dataset.

<u>Seed germination raw data:</u>

Table 1, 2 & 3: The number of seeds germinated in each trial at each temperature, as well as calculations for mean, standard deviation and 95% confidence intervals.

Monday 21st October, 3 days after planting		temperature of seeds (˚C)				
		16	20	25	30	35
Number of seeds germinated per trial	**1**	1	2	3	2	1
	2	0	2	2	1	3
	3	2	3	6	4	1
	4	1	3	1	1	3
	5	0	1	1	3	2
	Mean	0.8	2.2	2.6	2.2	2.0
	STDEV	0.8	0.8	2.1	1.3	1.0
	95% CI	0.7	0.7	1.8	1.1	0.9

Wednesday 23rd October, 5 days after planting		temperature of seeds (˚C)				
		16	20	25	30	35
Number of seeds germinated per trial	**1**	3	4	7	2	2
	2	1	4	3	2	4
	3	4	9	7	6	2
	4	1	4	4	3	6
	5	1	5	4	3	2
	Mean	2.0	5.2	5.0	3.2	3.2
	STDEV	1.4	2.2	1.9	1.6	1.8
	95% CI	1.2	1.9	1.6	1.4	1.6

Monday 28th October, 10 days after planting		temperature of seeds (˚C)				
		16	20	25	30	35
Number of seeds germinated per trial	**1**	4	6	8	4	3
	2	4	9	6	2	6
	3	9	10	7	7	2
	4	4	8	8	3	7
	5	9	7	5	4	2
	Mean	6.0	8.0	6.8	4.0	4.0
	STDEV	2.7	1.6	1.3	1.9	2.3
	95% CI	2.4	1.4	1.1	1.6	2.1

Calculations:

Example of finding the mean number of seeds germinated per trial.
Example – Table 1, column 1 (16˚C)

The sum of the *x* values (Σx) must be found and then divided by the number of *x* values (*n*):

$$\bar{x} = \frac{\Sigma x}{n}$$

$$\bar{x} = \frac{1 + 0 + 2 + 1 + 0}{5}$$

$$\bar{x} = \frac{4}{5}$$

$$\mathbf{\bar{x} = 0.8}$$

Example of finding the standard deviation of Table 1, column 1 (16˚C)

$$sx = \sqrt{\frac{\Sigma(x - \bar{x})^2}{n - 1}}$$

The mean of *x* values ($\bar{x}$) and the number of *x* values (*n*).

$\bar{x} = 0.8$

$n = 5$

A table was created to work out $\Sigma(x - \bar{x})^2$:

x	$x - \bar{x}$	$(x - \bar{x})^2$
0	-0.8	0.64
1	0.2	0.04
2	1.2	1.44
1	0.2	0.04
0	-0.8	0.64

$$\therefore \Sigma(x - \bar{x})^2 = 0.64 + 0.04 + 1.44 + 0.04 + 0.64$$

$$= 2.8$$

$$\therefore \quad sx = \sqrt{\frac{2.8}{n - 1}}$$

$$= \sqrt{\frac{2.8}{4}}$$

$$= 0.8367 \ldots$$

$$sx = 0.8$$

Confidence intervals were calculated using the mean and standard deviation values from previous calculations in Microsoft Excel.

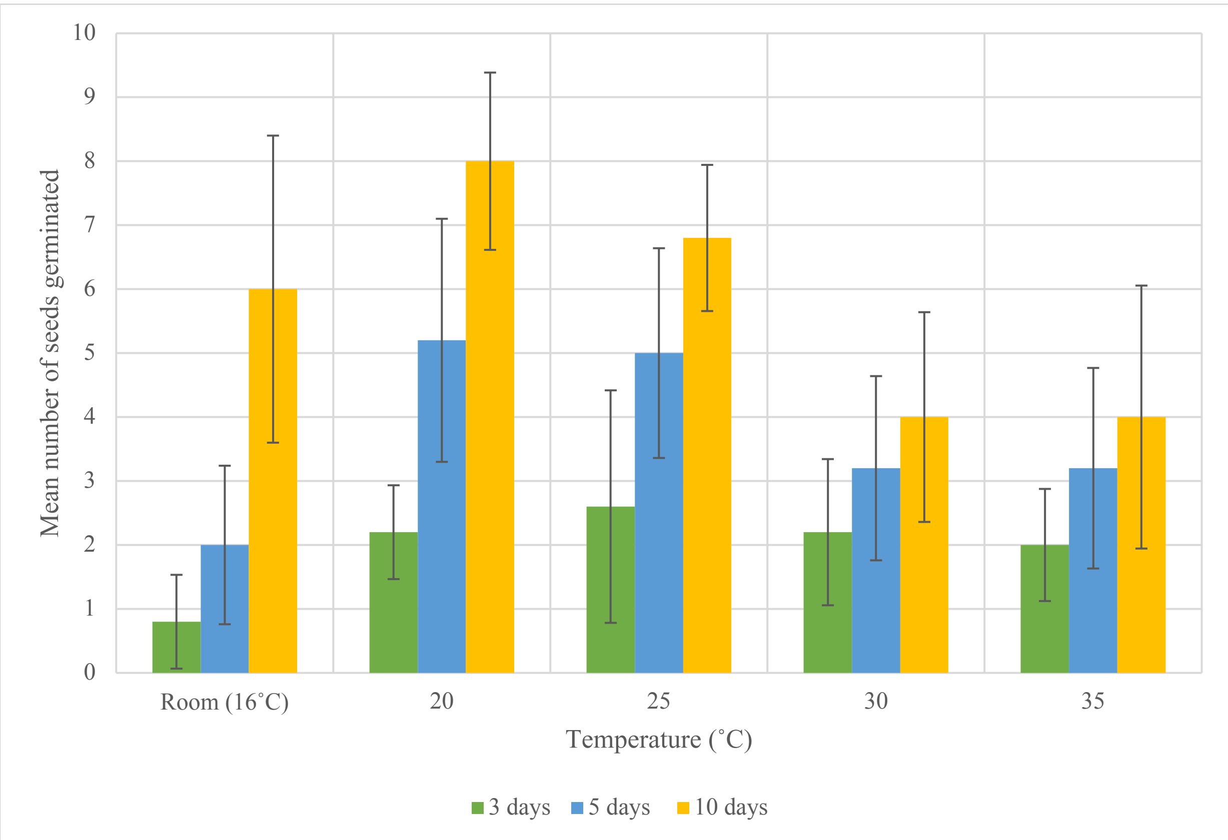
Qualitative observations:

The seeds germinated well in each trial. Dish 3 and 4 of 35˚C were found to be dry on day 3 and 5. By day 10, the seeds in the 16˚C environment had green shoots on them.

Summary of trends:

After 3 days, the most seeds germinated in the 25˚C trials (13/50 seeds). By day 5, the most seeds had germinated in 20˚C (26/50 seeds), and again after 10 days (40/50 seeds). The germination of the seeds in 30˚C and 35˚C remained similar to each other throughout the experiment, with fewer germinations than the lower temperatures. Overall the size of the standard deviations was proportionately small relative to the mean, indicating moderate reliability. The confidence intervals show this on Graph 1.

Graph 1: A bar chart that shows the mean number of seeds germinated at each temperature at each number of days after the experiment began. The error bars show the 95% confidence interval of each trial on each day.



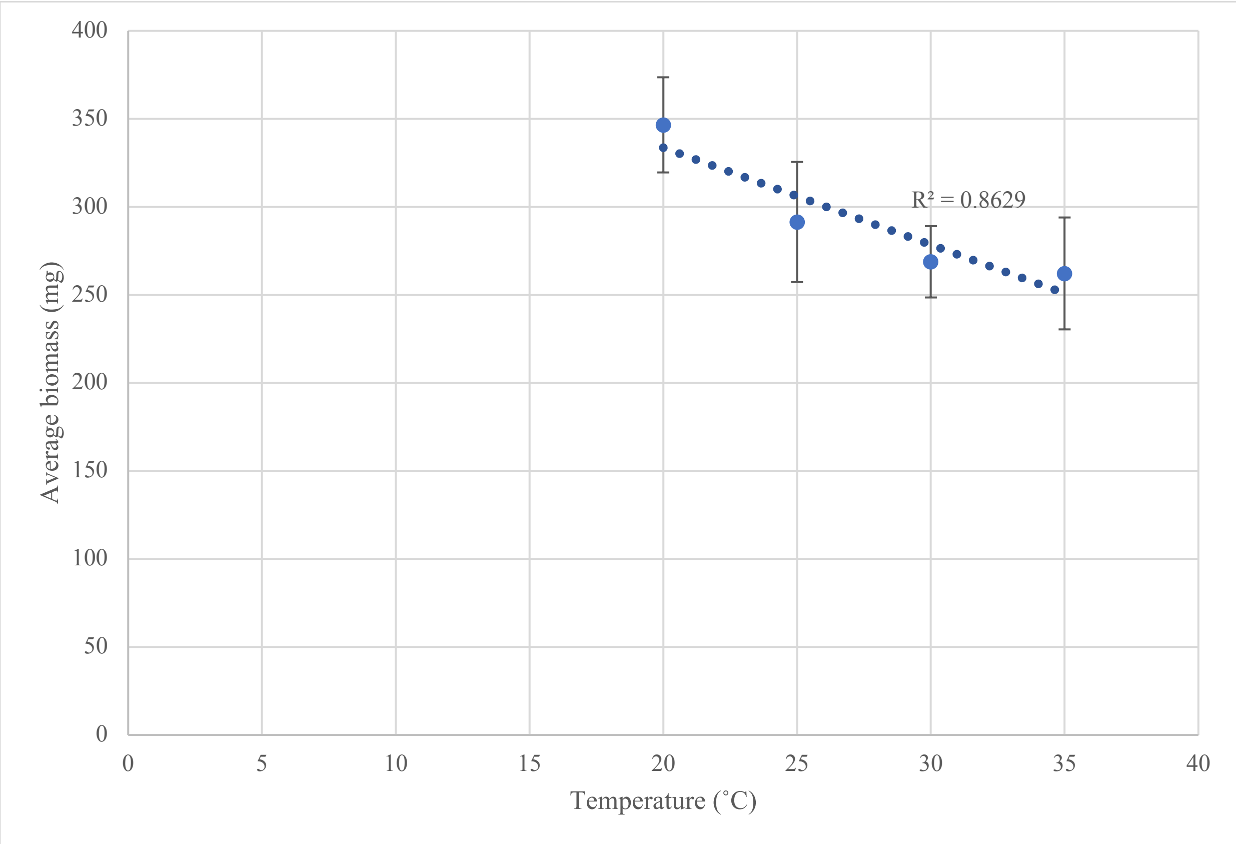
Graph 1 shows that the most germination occurred at 20˚C and 25˚C, with a final mean number of seeds of 8 and 6.8 respectively. Germination declined if temperatures were higher or lower than this. This is consistent for each set of results collected over the 10 days. The error bars are the smallest on day 10 of 20˚C and 25˚C, indicating that results would likely be similar if the experiment were repeated. The error bars on day 10 of 16˚C, 30˚C and 35˚C are comparatively larger, indicating less reliability in the results. Most error bars overlap, which further indicates weak trend across the data, though the error bars at 10 days for both 20˚C and 25˚C do not overlap with the day 10 error bars for 30˚C and 35˚C, suggesting there is a statistically significant difference, and that seeds at 20˚C and 25˚C are more likely to germinate than seeds at 30˚C and 35˚C.

<u>*Biomass data*</u>

Table 4: A table of the impact of temperature on biomass.

	biomass of seeds (mg)				
	temperature of seeds (˚C)				
trial number	**16**	**20**	**25**	**30**	**35**
1	378	338	290	287	210
2	344	398	273	278	210
3	333	313	266	233	278
4	410	375	306	315	322
5	380	309	322	231	291
AVG	369	347	291	269	262
STDEV	30.8	38.9	23.1	36.3	50.3
95% CI	27.0	34.1	20.3	31.8	44.1

Graph 2: graph of average final biomass of the seeds, with 16 ˚C data excluded as an outlier due to photosynthesis impacting biomass. The error bars show 95% confidence interval.



Graph 2 shows a strong negative correlation between average biomass and temperature, because the R^2 value is -0.86. This suggests that as temperature increases, the amount of germination and initial growth of wheat decreases. The error bars for each temperature do have a significant overlap with the bars either side of them, which implies the trend may not be as strong as the R^2 value suggests. However, the error bar for 20˚C does not overlap with the error bar for 35˚C, which implies a statistically supported overall decline in germination and growth as temperature increases.

<u>*Conclusion:*</u>

The data collected on germination and biomass agree with each-other, with strong/moderate correlations that suggest past a certain point (25˚C), increased temperature decreases the amount of germination and initial growth of Triticum aestivum seeds. Graph 1 shows that optimum germination occurs at 20˚C-25˚C, decreasing as temperatures change either side of this. Graph 2 shows that biomass decreases as temperature increases, however, whether biomass decreases below 20˚C is not able to be shown from these results due to removal of 16˚C as an outlier. Overall, this data seems to support a reliable conclusion that as temperature increases the germination and initial growth of Triticum aestivum seeds decreases.

Discussion and Evaluation

Discussion of Conclusion in Context of Environmental Issue:

This conclusion of the seeds germinating less as the temperature increases supports the optimum germination temperature of Triticum aestivum being 12˚C-25˚C (Acevedo et al, n.d.). This conclusion also supports another experiment done on wheat which found that "high temperature reduced crop growth" (Chakrabarti et al, 2013). The reason for this similarity may be the due to the controlled lab conditions that were similar across experiments, however the results overall strengthen the conclusion that global warming could decrease agricultural productivity as growing conditions may change and no longer support optimum germination temperatures. This is potentially problematic both now and in the future, when the high projected population which will rely on crops like wheat to sustain it. Western Australia may remain somewhat within the optimum temperature range as temperatures increase, with a new range of 14.1˚C-31.3 ˚C, however on a global scale there could be a decrease in wheat production in current wheat growing areas.

Evaluation of Strengths. Weaknesses, and Limitations of Method:

A significant strength of this experiment was its large sample size. With 5 different treatments and 50 seeds in each treatment, there were 250 Triticum aestivum seeds in the experiment. This decreased the chance for error and genetic variability within the data, and increases the validity of the experiment and its conclusions due to it being a more accurate representation of the population.

A weakness of this experiment is that on day 10 it was found that dish 3 and 4 of the 35˚C treatment had cracks in them, explaining why water had to be added to these dishes more frequently than any other dish. Whilst this should not have significantly impacted the experiment, as water was still added regularly and so the seeds remained fairly damp, it lessens the experiment's validity because the dampness of the seeds was not effectively controlled. Another variable not controlled effectively was that light was received by the 16˚C dishes. This allowed them to photosynthesise, and whilst probably not impacting germination, since seeds germinate underground and therefore do not require light, it impacted the growth of the seedlings once germinated, evident in the biomass results, where the 16˚C seeds had an average final biomass of 369mg, which was larger than the other trial averages and an outlier in the findings of this investigation and the investigation of Chakrabarti et al. This was a significant flaw in the experiment, and thus it would not be accurate to compare the biomass of the 16˚C petri-dishes to the other dishes that were light controlled, hence the data was excluded in Graph 2. Having controlled data for the germination of wheat seeds in 16˚C would strengthen the validity of the conclusion. Another of the method's limitations was that it was unable to account for other factors that might impact germination in real life, such as soil conditions or rainfall. Therefore, a modification of this experiment would be to ensure all seeds were in the same light conditions at all times by setting up an incubator at 16˚C, which would be dark as it was for the other temperatures, preventing photosynthesis and the subsequent increase in biomass.

Applications

Potential Solution to Environmental Issue:

A potential solution to the issue of wheat production is to genetically modify the wheat seeds to have a higher optimum germination temperature. This would allow the continuation of wheat growth in areas that may be impacted by global warming. This technocentric approach to food production in a warming world has already occurred. A 2014 experiment (Honghong Hu, Lizhong Xiong) created genetically modified corn designed to withstand drought conditions. This has yet to be applied to wheat, however offers a potential solution to its production in an increasing global temperature.

Evaluation of the Solution:

A strength of this solution is that it would allow continued growth in agricultural areas that may be affected by global warming, which would maintain the industry and livelihood of wheat-growers in that area and be an effective way of providing food for the increasing population. A weakness of this solution is that it relies both on technological advances and a populations' willingness to consume genetically modified food, which is often a contentious issue - as seen in the debate surrounding GM soy, and fears that GM food could provoke allergic reactions (World Health Organisation, 2014).

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