Writing Information

Instructions to Authors

Follow these instructions or your paper will be returned to you, and incur late penalties.

One of the learning objectives of your research project in the course is to develop your scientific writing skills. In science, writing is the most important means of communicating research findings. Major scientific findings are rarely kept secret. Instead, scientists share their ideas and results with other scientists, encouraging critical review and alternate interpretations from colleagues and the entire scientific community. In most cases, scientists report the results of their research activities in scientific journals in a standard written format. In this course, you will practice writing using this same standard scientific format and style.

4.0 TIP: Write like a scientist. Write your papers so that anyone who reads your manuscript could not tell it was from a student, but assumes it must have been from a scientist in a lab at MSU.

A scientific paper includes the following: a **TITLE** (statement of the question or problem), an **ABSTRACT** (short summary), an **INTRODUCTION** (background and significance of the problem), a **METHODS** section (report of exactly what you did), a **RESULTS** section (presentation of data), a **DISCUSSION** section (interpretation and discussion of your results), and **REFERENCES** (books and periodicals used). Data is also represented by **FIGURES** and **TABLES**.

Throughout the laboratory, you will practice scientific research and writing. Your papers will be reviewed by the course professor, TAs, and your peers in order to point out your areas of weakness and make suggestions for future improvements. By the time you have completed the course, you will have submitted the equivalent of two full scientific papers. If you are not certain about the level of independence and what constitutes plagiarism in this program, ask your instructor to clarify the class policy. ***Plagiarism will not be taken lightly and will be evaluated by instructors and software at turnitin.com. See syllabus for more info.***

Predictions: Science is not about explanation, but in fact it is about the ability to predict. All scientists must have models or hypotheses that can be used to then make predictions of what will occur. Thus prediction is a very important part of writing you papers. As a result in your early DRAFT1 and DRAFT2 in many cases your predictions may be all the data you have on a certain experiment and thus they should be well supported by papers from the literature. Use future tense when discussing Predictions. In general whether it's in the Abstract, Introduction, Results or Discussion, whenever you discuss something that you predict it should be in the format of:

"We predict..[what].. because..[rationale].. (citation of paper)."

**Note: A hypothesis is different than a prediction. The hypothesis is the model that explains how you believe things are working (e.g. we hypothesize electricity sparks gas in the cylinders of a car engine) while the prediction is what you think should happen during the experiment (e.g. if hydrogen gas is injected into the cylinder of a functioning engine we predict there should be a significant explosion).

Overview of Manuscript Sequence & Format
(This is what professional journals expect)

1. Page one is the Title Page (≤100 characters in title)
2. Page two will have ONLY the Abstract (≤250 words)
3. Page three will start with the Introduction. The Methods, Results, and Discussion sections can follow without starting a new page for each one (although you may start a new page if you are near the bottom of the current page). Clearly label each section with the section headings (ex. Introduction) and who authored it (Written by: Jill Sanders, Revised by: Bob Roberts).
4. The **Introduction** will provide the reader with the background information necessary to understand the rest of the paper.
5. **Methods** section will list materials used (Bought Vitamin C with Rose hips from the General Nutrition Store (GNC) 324 E. Grand River East Lansing MI) how stock solutions were prepared and explain exactly what you did in your research. After reading the Methods section, an incoming student should be able to repeat your work. Reference the original protocol.
6. The **Results** section follows Methods. This section will clearly and succinctly state what you observed upon performing each experiment.
7. The **Discussion** section follows Results. In this section you will discuss the significance of results and how your results relate with research performed by others.
8. The **References** section follows the Discussion. This is a list of the references cited within the paper.
9. Start a new page with the **Figures** section after the References. Figures will be sequentially numbered in the order that they were cited in the Results section (figures are most always cited ONLY in the Results section, not in Methods, not in Discussion). One figure per page with extensive figure legend paragraph ONLY at the bottom of the figure. The first sentence of a figure legend is its title. Follow the title with sentences explaining the figure as if someone did not have the Results section or in fact any other part of the paper available as a reference.
10. The **Tables** section will follow the Figures section. Tables get a title ONLY on the top with some explanation. Tables will be sequentially numbered in the order that they were cited in the Results section (Tables are most always cited ONLY in the Results). One table per page.
11. Figures and tables MUST be created on a computer unless otherwise instructed.
12. After the Tables section, a single white page will follow entitled, **Appendix**. Then append any laboratory notebook pages that indicate signed data for all members (photocopies from your notebook) and a photocopy of the first page of any articles cited and referenced in report.
13. Double space or 1.5 space typeface is required. Preferred font size is 12 point.

***Once returned, rejected papers (like late papers) lose one point in the first 24 hrs grace period but then the penalty becomes more severe: 10% off for 2 days late, 20% off for 3 days, and so on. After 5 days, you will receieve a "0". Unlike late papers, a rejected paper also loses 1 pt per rejection.**

A more detailed description of each section of a scientific paper follows also, review the published papers provided in the course packet for examples. As you write your paper, clearly label each section (except the title page), placing the title of the section on a separate line, centered, bold, but not underlined (like shown below).

Title Page and Title

The title page is the first page of the paper and includes the title of the paper, your name, the course title, your lab time, your lab instructors' names, the due date for the paper, and your groups' website address. The title should be as short as possible and as long as necessary to communicate to the reader the question being answered in the paper. Consider the following titles for a paper that describes the molecular mechanism of an antiviral drug.

1. "Inhibition of Mengovirus Replication by Dipyridamole"
2. "Antiviral Action of Dipyridamole"
3. "A Study Examining the Inhibitory Effects of the Drug Dipyridamole on Mengovirus Replication"

Title 1 is short and communicates the question being investigated. It conveys the mechanism of action (inhibition of replication), the name of the virus being inhibited (Mengovirus), and the name of the drug doing the inhibiting (Dipyridamole). Title 2 is short but too vague for the reader to know the subject matter of the paper. Title 3 is too long. The words "A Study Examining" are superfluous, and "Drug" and is redundant.

Place the title about 7 cm from the top of the title page. Place "by" and your name(s) in the center of the page, and place the course title, lab time, lab instructors' names, due date, and your groups' website address, each on a separate centered line, at the bottom of the page. Leave about 5 cm of white space below this information.

Abstract

The abstract is placed at the beginning of the second page of the paper, after the title page. The abstract summarizes the question being investigated in the paper, the methods used in the experiment, the results, and the conclusions drawn. The reader should be able to determine the major topics in the paper without reading the entire paper. As mentioned previously, predictions are an essential element of science and thus should appear in the Abstract of DRAFT1 and DRAFT2 and in the format: ***"We predict..[what].. because..[rationale].. (citation of paper)."***

Introduction

Start the introduction on page three. The introduction should generally be short, only 4-5 paragraphs in length and focus are background information of the following types:

1. Describe the question and hypothesis being investigated and background on the importance of the topic.
2. Review the background information that will allow the reader to understand the purpose and topics of the paper. There is usually a paragraph on the specimens studied, also one that provides evidence to support the hypothesis posed. A hypothesis is an educated guess; the Introduction should provide the "education." Include only information that directly prepares the reader to understand the question investigated. Most of this information should come

from outside sources, such as scientific journals or books dealing with the topic you are investigating.*

3. In a paragraph state background information on the methods chosen to investigate the hypothesis. Explain how these methods will address the question and describe the predicted outcomes. Why were they chosen?
4. In the last paragraph briefly state a hint of the results and conclusions of the investigations (or predictions). This generally comes only at the very end of the Introduction.

*All sources of information must be referenced and included in the References section of the paper, but the introduction must be in your own words. ***No "quotations" are permitted in any part of the paper.*** Refer to the references when appropriate. As you describe your investigation, include only the question and hypothesis that you actually investigated. It is a good idea to write down each item (question, hypothesis, supporting evidence, prediction) before you begin to write your introduction.

Write the introduction in past tense when referring to elements of your experimental investigation that are completed. When relating the background information, use present tense when referring to another investigator's published work. Use future tense when discussing Predictions.

Methods

The Methods section describes your experiment in such a way that it may be repeated exactly. Make the Methods professional just like in published papers, but target as your audience a student in LB-144. The majority of the information in this section comes from the Procedures or Protocols section of the Laboratory Guide and in your paper, this information should not be a list of steps. Write the Methods section in a paragraph format in past tense. Be sure to include levels of treatment, numbers of replications, and control of treatments. If you are working with living organisms, include the species and the sex of the research organism. Do not include failed attempts unless other investigators may wish to try the technique used. Do not try to justify your procedures in this section of the report.

If you describe an experiment from the lab guide, unless instructed otherwise, you may simply refer to the procedures listed in the guide (and page numbers). Under those circumstances, your Methods section should point out changes in procedure that are not indicated in the Lab Manual. When writing a full Methods section (with no reference to the Lab Guide), write these procedures concisely, but in paragraph form. The difficulty comes as you decide the level of detail to include in your paragraphs. You must determine which details are essential for the investigator to repeat the experiment. For example, if in your experiment you incubated potato pieces in different concentrations of sucrose solution, it would not be necessary to explain that the pieces were incubated in plastic cups labeled with a wax marking pencil. In this case, the molarity of the sucrose solutions, the size of the potato pieces and how they were obtained, and the amount of incubation solution are important items to include

Results

The Results section consists of two components: (1) one or more paragraphs that describe the results of each experiment/test and include the actual data with observations, specific numbers [and units] and math, (2) reference to figures (graphs, diagrams, pictures), and reference to tables.

In referencing figures, remember to number figures and tables consecutively in the order that they are mentioned in this section. Refer to figures and tables within the paragraph as you describe your results, using the word Figure or Table in parentheses, followed by its number, for example, "(Figure 1)." Avoid citing a figure with a full sentence or statement such as, "please see figure 1 for graphed data points" or even just "please see table 2." DO NOT place each figure or table at the end of each paragraph in which it is cited. Place figures and tables after the References section. If you have performed a statistical analysis of your data, such as chi-squared, include this data in the Results section. Explain what calculations you did and the result and direct the reader to the Methods section for more details. Then show an example of the results and refer the reader to a table with all the data.

The most common error a student makes is only making general comments and not including actual data (ie the results) in the Results. Results should include detailed observations (what did you see: odor, texture, etc.) and specific findings (what did you record: 15 grams, 20 degrees C, 20% increase, 32,000 cells etc.). Report your data as accurately as possible in the order that they happened. It is important to have separate paragraphs and topic sentences that introduce the results of each test but do not spend much time discussing the meaning of your findings, save that for the Discussion.

For "***Predicted Results***" - which will be the only focus of Draft 1 and will be included in subsequent drafts - these are what you predict or expect will happen, and these predictions must be based upon the primary literature you have gathered throughout your research on your topic/thesis. When including predictions follow this format: ***"We predict..[what].. because..[rationale].. (citation of a published paper or source)."***

Discussion

The Discussion section is where you will analyze and interpret the results of your experiments. The Discussion should show a possible relationship between observed facts—those observed by you and those observed by others and reported in published research papers. Write as clearly and succinctly as possible. A good Discussion will include the following:

1. Summarize the Introduction and restate the question and hypothesis being addressed.
2. Briefly summarize the results of the experiments. Do not include details regarding methods.
3. Interpret the results. Explain how the results answer the questions posed. State whether your results support or refute your hypothesis. Do not use the word "prove" in your conclusions. Your results will support, verify, or confirm your hypothesis. They also may negate, refute, or contradict your hypothesis. The word prove is not appropriate in scientific writing.
4. Discuss how your results and interpretations relate with previously published research. This will require you to cite outside references. Some may come from the Introduction, while you will also find new references that specifically relate to your findings. You can speculate and propose theoretical implications of your work.

5. Describe weaknesses in experimental design or technical difficulties that arose during the research. Explain how these problems specifically affected the outcome of the research. Any human errors (spills, etc) discussed should have been then corrected by certain steps.
6. Discuss experiments that would be performed if the research were to be continued. Explain how those experiments would contribute to answering the questions addressed by the research.

Figures

All figures should be computer generated. The format of the figure will depend on the type of data collected. Your figures will include mostly photographs and graphs. The photos and graphs must be done in a professional manner and include computer generated labels when appropriate, and always with only one figure per page. Under each photo or graph, there must be a legend paragraph. The legend paragraph will include the Figure number, a title sentence, and a description of what was done in the experiment and shown in different labeled parts of the figure. A reader must be able to understand the general concept of the experiment performed without reading the Methods section. In fact if a student from LB-144 picks up a single piece of paper with one of your figures on it, they should be able to explain to you what that experiment is about from the legend.

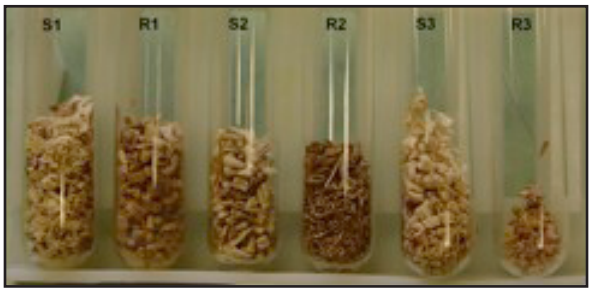

Figure 1. Preparation of corn root and stalk samples for sugar analysis. Samples were cut into units no greater than 4 mm3 using a surgical scalpel. Three stalk samples of 70 grams each and three root samples of 80 grams each were prepared. Root 1 (R1), Root 2 (R2), Root 3 (R3), Stalk 1 (S1), Stalk 2 (S2), and Stalk 3 (S3).

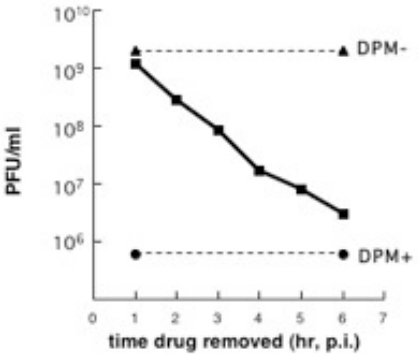

Figure 2. DPM and Virus Yield. HeLa cell monolayers were infected with Mengovirus at a multiplicity of 50 pfu/cell. DPM+ samples had 80 µM DPM (in ethanol) added to the media at the time of infection. DPM- samples were dosed with an equivalent volume of ethanol. Medium from DPM+ cultures was exchanged with drug-free medium at the indicated times. Virus was harvested at 8 hrs PI and the titer determined by plaque assay.

Tables

While Figures are often used in papers (graphs, photographs, gel images) tables are rare. Tables should only be used when all the data being presented cannot be reported in a simple and comprehensible manner in the Results section. The title appears at the top of the table; there is no legend. A footnote may be necessary to clarify an important point in the table.

Table 2. Mengovirus plaque phenotypes in the presence of DPM.

Concentration, μM DPM	Plaque Reduction (%)[a]	Relative Plaque Size[b]
80	100	N/A
60	98	minute
40	93	+
20	68	++
10	25	++
0	0	++++

[a] Values represent the average of two experiments each done in triplicate.

[b] Plaques in the absence of DPM averaged about 2mm in diameter.

Table 1. DNA glycosylases in human cell nuclei.

Enzyme	Size (amino acid residues)	Gene location at chromosome	Altered base removed from DNA
UNG	313	12q23–q24	U and 5-hydroxyuracil
TDG	410	12q24.1	U or T opposite G, ethenocytosine
hSMUG1	270	12q13.1–q14	U (preferentially from single-strand DNA)
MBD4	580	3q21	U or T opposite G at CpG sequences
hOGG1	345	3p25	8-oxo G opposite C, formamidopyrimidine
MYH	521	1p32.1–p34.3	A opposite 8-oxo G
hNTH1	312	16p13.2–p13.3	Thymine glycol, cytosine glycol, dihydrouracil, formamidopyrimidine
MPG	293	16p (near telomere)	3-MeA, ethenoadenine, hypoxanthine

Reference Citation Formatting

A References section lists only those references cited in the paper. You will cite all the references you used when you wrote your paper. In the text of the paper, cite the references using the author's name and publication year. If there are two authors you must state both of their names if there are greater than two authors, state the first authors name followed by et al.

For example: We predict that the 95% alcohol solution will kill all of the NIH-3T3 cells in our culture flask because Smith performed a similar experiment in his paper with another cell type (Smith et al, 1998). In previous research the p58 protein was associated with increased cell growth in C127 cells (Johnson, 2001; Benenson and Kortemeyer, 2003; Haenisch et al, 2006).

Types of Literature:

The vast collection of scientific literature can be generally divided into three categories based on how 'close' they are to the original experiments and descriptions of scientific phenomena. **1) Primary literature**: The bulk of scientific journal articles are primary, meaning that they report the findings of specific experiments or descriptive studies. **2) Secondary literature**: From time-to-time investigators write review articles or books that summarize what is and is not known about a particular topic. Rather than conducting new experiments, these authors rely heavily on the primary literature, therefore these review articles and books are considered a part of the secondary literature. **3) Tertiary literature**: More general texts that summarize what has been reported in review articles comprise the tertiary literature.

Most new research relies heavily on previous work reported in primary literature. However, review articles can be extremely helpful in understanding how your research project fits into the larger scope of scientific investigation, and can be used as a source to locate primary literature references for the topic of interest.

Note that websites were not included in the above description of scientific literature sources. This is because they are not refereed — that is, just about anyone can publish something on the web without some impartial reader reviewing it beforehand. Web pages are often wonderful sources of information, but they can just as often be replete with bad information. At this point, it is very difficult to determine the reliability of web sources and, in general, they should generally only be used as a starting point about a particular topic. ***Thus websites are only allowed as citations in DRAFT1 manuscripts.***

Examples of Proper Citation Formatting for the listings in your Reference section:

Journal articles:

Single Author:

Belsky, A. J. 1986. Does herbivory benefit plants? A review of the evidence. American Naturalist 127: 870–892.

Two Authors:

Brown, J. H. and D. W. Davidson. 1977. Competition between seed-eating rodents and ants in desert ecosystems. Science 196: 880–882.

Multiple Authors:

Free, C. A., J. R. Beddington, and J. H. Lawton. 1977. On the inadequacy of simple models of mutual interference for parasitism and predation. Journal of Animal Ecology 46: 543–554.

If the source was published in an online journal do not cite the URL, treat it the same as printed:

Thomas, J. A., M. G. Telfer, D. B. Roy, C. D. Preston, J. J. D. Greenwood, J. Asher, R. Fox, R. T. Clarke, and J. H. Lawton. 2004. Comparative losses of British butterflies, birds, and plants and the global extinction crisis. Science 303: 1879-1881

Books:

Chapter within a book:

Goldberg, D. E. 1990. Components of resource competition in plant communities. Pp. 27–50 in J. B. Grace and D. Tilman, eds., Perspectives on Plant Competition. Academic Press, San Diego.

An entire book:

Hynes, H. B. N. 1970. The Ecology of Running Waters. University of Toronto Press, Toronto.

Theses:

Watson, D. 1987. Aspects of the population ecology of Senecio vulgaris L. Ph.D. thesis, University of Liverpool.

The textbook:

Campbell NA. and Reese JB. 2007. Biology – 8th ed., Chapter 13 "Mendel and the Gene". Benjamin Cummings, CA.

The Lab Maunal:

Haenisch, et al. 2009. LB145 Course Pack. MSU Printing Services, East Fee Hall. Michigan State University, East Lansing, MI

A Web Site: [only allowed in DRAFT1 manuscripts and follows a text citation (Author(s), Year published).]

Anonymous. 2002. Wisconson Fast Plants Web Site. http://www.fastplants.org/Introduction/Introduction.htm, last accessed 7/10/02

References (used in the creation of this appendix):

McMillian, V. E. 2001. Writing Papers in the Biological Sciences, 3rd ed. St. Martin's Press, Inc., New York.

Appendix

Note everything you place in your appendix will be discarded during grading.

An Appendix section includes only materials that are not actually required in the paper and could be thrown away with no effect to the paper. Often special extra information is included here. In LB-145 you are expected to place any laboratory notebook pages that indicated signed data for all members (photocopies from your notebook) and a photocopy of the first page of any articles cited and referenced in the report.

Reminders

Before writing your paper, refer to the following hints to make your paper stronger:

1. Write clearly in short, logical, but not choppy sentences.
2. Use past tense in the Abstract, Methods, and Results sections when discussing things that have been completed. Also uuse past tense in the Introduction and Discussion sections when referring to your experiment. Use future tense when making predictions about future experiments.
3. Write in grammatically correct English, but use METRIC UNITS.
4. When referring to the scientific name of an organism, the genus and species should be italicized, the first letter of the genus is capitalized, but the species is in lower case; for example *Drosophila melanogaster*.

Making a Website

Making a webpage is a useful skill to have and is not as hard as you might think. This primer is intended as a walkthrough for the basic beginner. Anyone looking for a more advanced explanation should enroll in a web design course already.

There are many "places" on the internet where you can load your webpage (each website name corresponds to a file on a computer somewhere in the world) but we'd like you to load your file onto MSU's computers through their AFS system. For the uninitiated, the basic idea of this system is that MSU has provided hard-drive space that users (students and faculty) can access and use from any computer connected to the internet. Most people use this to store and back-up files or run webpages. If you'd like more info on what this system is and how it works, go to the ATS helpdesk website at help.msu.edu.

The first step is to generate your file and save it as an html. There are many programs that can do this for you, but most versions of MS Word made in the last decade have a "save as webpage" or "save as html" function in the file menu. You should title your document "index" (you need at least one index file in the Web folder for MSU to be able to run your web page, so this might as well be it), unless you would like to make another index file that has hyperlinks to the document you are currently trying to upload. If you have images in your file (which you really should have) then Word will automatically make a separate folder titled "index_files" in which all of the images are stored. You will need to create a folder with the same name in your AFS space and upload all of these files into it.

Go to netfiles.msu.edu and log in with your MSU netID. Click on the "Web" folder which should be near the bottom of the file/folder list in the middle of the page. Select "Upload file(s)" from the options on the left. Click on the "Browse"" button, locate your index.htm file and click "Upload file(s)". To create your image folder, click on the "Create a new folder" option on the left side of the screen and enter the name EXACTLY the same as the one generated by Word (index_files). Click on the folder and repeat the file uploading process until of the folder"s contents are in your AFS space.

Now test your website by going to www.msu.edu/~yournetID. If you are having problems getting things to work, contact the ATS help-desk or a resident computer whiz.

Samples: Student Papers

Genotypic Identification of CF Patients with the R553X Mutation using IB3 and S9 Cell Lines and Allele Specific PCR

By: Sarah Bonczyk, Nathan Johns, Elizabeth LeMieux, and Mitch Wood

LB 145 Cell and Molecular Biology
Tuesday 7 PM
Ashley Coulter and Jason Mashni
4/24/2009

http://teamhouselb145.tripod.com/

(Title page written by: Nathan Johns
Revised by: Mitch Wood,
Finalized by: Liz LeMieux)

Abstract

Written by: Nathan Johns
Revised by: Mitch Wood
Finalized by: Liz LeMieux

The R553X mutation of the cystic fibrosis transmembrane conductance regulator (CFTR) accounts for 0.7% of all cystic fibrosis cases (Hull *et al,* 1993). The mutation involves a single base pair substitution in the 553rd amino acid from CGA (Arginine) to TGA, a stop codon, leading to a truncated protein (Bal *et al,* 1991). Allele specific polymerase chain reaction (ASPCR) was used to determine whether samples of DNA are wild-type, heterozygous, or homozygous for the R553X mutation in the CFTR gene. DNA was extracted from epithelial bronchial cells of known CF patients (Qiagen Inc, 2007). We hypothesized that by controlling annealing temperatures and salt concentrations in the PCR reaction, a single base pair mismatch can be used to determine the presence of the R553X mutation, based on previous PCR diagnostic testing (Chavanas *et al,* 1996). The resulting amplified DNA was then analyzed using agarose gel electrophoresis to determine the genotype of the DNA. We were able to determine whether samples were heterozygous, homozygous wild-type, or homozygous for the mutation by the presence or absence of bands 1,056 base pairs long and based on which forward primer used (Wu *et al*, 1989). Research surveys were distributed and analyzed in order to examine different public opinion on genetic testing between students of various studies at Michigan State University. Results showed no significant difference in opinions on genetic testing among various residential colleges. These tests are significant in helping doctors diagnose cystic fibrosis patients for specific mutations faster and more accurately than previous testing.

Introduction

Written by: Nathan Johns
Revised by: Mitch Wood
Finalized by: Liz LeMieux

Cystic fibrosis (CF) is an autosomal recessive disease caused by mutations in the gene coding for the cystic fibrosis transmembrane conductance regulator (CFTR) protein. The role of the CFTR protein is to serve as a chloride ion channel in epithelial cells (Rowe *et al*, 2008). Epithelial cells with a CFTR mutation, most often in the lungs, pancreas, and intestines, secrete large amounts of mucus, which builds up and creates complications in the affected tissues (Welsh and Smith, 1995). Mucus buildup in the respiratory tract often leads to pulmonary infection, the most common cause of death in CF patients (Golshahi *et al,* 2008). Current treatments attempt to remove this mucus in the lungs and avoid affection, often by using percussive therapy and antibiotics (Welsh and Smith, 1995).

Over one thousand mutations in the CFTR gene are identified to cause CF, with the ΔF508 mutation, a deletion of three base pairs at position 508, being the most common, and accounting for approximately 70% of all cases (Teem *et al*, 1993). The R553X mutation is the sixth most common, accounts for 0.7% of cases, being most prevalent in German communities (Hull *et al*, 1993). R553X is a nonsense mutation caused by a C to T substitution at the 553rd amino acid. This changes what would normally be arginine to a stop codon (Hull *et al,* 1993). A nonsense mutation is a mutation that causes the DNA sequence to result in a premature stop codon, or a nonsense codon in the mRNA which results in a truncated, incomplete and nonfunctioning protein. In the case of R553X, research has shown that the premature stop codon often results in exon skipping in RNA translation (Aznarez *et al*, 2007). The loss of the exon causes an unstable mRNA of the truncated protein and therefore does not undergo the process of translation (Aznarez *et al*, 2007). The R553X mutation is a class I mutation because of the unstable mRNA synthesized in the nucleus caused by nonsense alleles. (Gambardella *et al.* 2006)

Polymerase chain reaction (PCR) is a technique used to amplify a desired section of DNA (Saiki *et al*, 1988). In PCR, DNA is heated during a denaturing step in order to break the hydrogen bonds between nucleotide bases to separate complimentary 5' and 3' strands. Once

Methods

Written by: Liz LeMieux
Revised by: Sarah Bonczyk
Finalized by: Mitch Wood

Primers

Before the PCR tests were run, primers for the PCR tests were designed using the Cystic Fibrosis Mutation Database. Forward and reverse allele-specific primers that worked with both the wild type and with the mutation were needed. FPrimer1 is a forward primer ending in Guanine, complementary to the wild type amino acid where the mutation should be present. The second of the forward primers, Fprimer2, seeks the mutant type base sequence at the mutation site, base pair number 1789, which results in the primer ending in Adenine. The reverse primer, Rprimer, was designed to bind to the DNA strand 1,022 base pairs past the mutation site between base pairs 2828 and 2811 and was used in both tests. Fprimer1 is 16 base pairs long with the sequence of: GACTCACCTCCAGTT**G** and should properly bind to the wild type gene sequence of **C**AACTGGAGGTGAGTC. Fprimer2, the mutant seeking primer, is also 16 base pairs long with the sequence of: GACTCACCTCCAGTT**A**; the only difference from the previous forward primer is the last base pair, which should properly bind to the mutant R553X gene sequence of **T**AACTGGAGGTGAGTC. The reverse primer that will be used for both tests is 18 base pairs long with the sequence of: CATGAGAGAAGAGAC, which should bind to the gene sequence of TCTGTCTCTTCTCTCATG, which is the same in both mutant and wild type genes. All of the primers are written in 5' to 3'. These primers were ordered from the biological laboratory company Integrated DNA Technologies (IDT).

After the primers were designed, the annealing temperatures at which the primers would bind were determined by first calculating the theoretical melting temperatures of each primer using the following formula:

$$Tm=64.9^\circ C + 41^\circ C \times (\text{number of G's and C's in the primer} - 16.4)/N$$

where N is the length of the primer (Wright *et al,* 2009*)*. The calculated theoretical melting temperatures were: Fprimer1- 45.94°C, Fprimer2 – 43.36°C, and Rprimer – 45.77°C. Based on these calculations, an annealing temperature of 42°C was used in the PCR tests.

DNA Purification

DNA Purification was used to collect DNA from cultured cells of both mutant type and wild type samples to be used in PCR. Two different sources of Human DNA came from Human bronchial epithelial cells from a CF patient without the R553X mutation (IB3 stock cells) while another set of stock cells was found to obtain the R553X mutation, therefore were used as the mutant type test (S9 cells). Each of these stock cells were used to contrast the effectiveness of the allele specific primers. (Gambardella *et al*, 2006) Before beginning purification, all reaction vessels were kept on ice until used and frozen cultured cells were allowed to thaw in a water bath set to 37°C. Then, 200ul of the sample (either human bronchial epithelial cells without the mutation or cells from a CF patient with the mutation, depending on the PCR test being done) was added to a capture column and was incubated at room temperature for 30 minutes. Afterwards, 400ul of DNA Purification Solution 1 was added to the sample. The resulting mixture was incubated for 1 minute at room temperature. After incubation, the mixture was centrifuged for ten seconds at 2,000-12,000 times the force of gravity (xg). The capture column was then transferred into a new waste collection tube. Another 400ul of DNA Purification Solution 1 was added to the mixture and the solution was incubated at room temperature for 1 minute. The solution was then centrifuged again for ten seconds at 2,000-12,000 x g. Then, 200ul of DNA Elution Solution 2 was added and the mixture was centrifuged for ten seconds at 2,000-12,000 x g. The capture column was transferred to a clear DNA collection cube and 100ul of DNA Elution Solution 2 was added. The solution was incubated for ten minutes at 99°C and then centrifuged for 20 seconds at 2,000-12,000 x g. (Quigen Inc. 2007)

PCR

To analyze DNA obtained from the purification process, two different Polymerase Chain Reaction (PCR) tests were designed. Test #1used Fprimer1 and the Rprimer. Test #2 used Fprimer2 and the Rprimer. For each of the tests, the combination of 2.0ul of the target DNA template obtained through DNA purification, 5.0ul 10X PCR buffer, 0.2ul Taq polymerase, 2.0ul forward primer (100 uM), 2.0ul reverse primer (100 uM), 1.0ul 10mM deoxynucleotide building blocks (dNTP) of DNA and 40.4ul water was added to a test tube on ice. The combination was mixed and spun down in a centrifuge. Once the sample was placed in the Labnet thermocycler, the cocktail went through five steps: initial denaturation, denaturation, primer annealing, extension and final extension. In the initial denaturation stage the temperature was raised to 94°

Predicted Results

Authored by: Kevin Werner

In this experiment for the template used in PCR, genomic DNA will be extracted and purified from human cells. Cultured cells from Crohn's Disease patients with the R702W-CARD15 mutation, without the mutation, and heterozygous for the mutation will be obtained from CRISI Inc (Sacramento, CA). For our diagnostic assay, PCR will be used to amplify a DNA segment at a locus containing the R702W mutation on the CARD15 gene that causes a form of Crohn's Disease using a specialized approach for detecting SNPs developed by Hidenobu Yaku (Yaku et al, 2008). The resulting PCR products will be analyzed using agarose gel electrophoresis in either a traditional TBE or fast LB buffer system.

Primer Design

We predict PCR of the genomic DNA will amplify the anticipated products from the R702W locus on the CARD15 gene on chromosome 16 (Figure 1). This will occur because of carefully selected primer design, reactant concentrations, and PCR temperatures and times (Saiki et al, 1998). Thus primers R (annealing at 20,866 base pairs to 20,887 base pairs on the CARD15 gene) and CDF (annealing at 19,866 base pairs to 19,881 pairs on the CARD 15 gene) will amplify a 1021 base pair product, and primers R and WTF (annealing at 19,866 base pairs to 19,881 base pairs on the CARD15 gene) will amplify a 1021 base pair product (Wright, et al. 2010) (Figure 2). Also, the forward primers CDF (forward primer designed to anneal to mutant type template) or WTF (forward primer designed to anneal to wild type template) will not anneal when their

bases are not completly complimentary to the DNA template bases at the respective annealing loci of the primers, preventing amplification from occurring (Schochetman et al, 1988). The forward primers are known as discriminating primers because they are designed to be completely complimentary to either the mutant type or wild type genotype, but not both (Wittwer et al, 1993). Hence it is also predicted that heterozygous DNA will allow for some amplification with both of the discriminating primers. It then follows that amplification of DNA template from the CDF discriminating primer supports the presence of the R702W mutation, which could cause a maladaptive pro-inflammatory response in Paneth cells along with other genetic and environmental factors (Figure 3) (Lala et al, 2003).

We did a preliminary PCR amplification of the 1542 base pair long 16S rDNA locus of *Escherichia coli* to use as a control throughout our experiment (Haffar et al, 2010). The bands we produced during gel electrophoresis exhibited non-specific binding that we were able to reduce by raising the annealing temperature (Figure 4) (Livak et al, 2010). Also, the validity our concentrations of forward primer, reverse primer, dNTPs, buffer, and taq polymerase will supported by these experiments, thus supporting the validity of our Crohn's experiment.

Genomic Purification

In our preliminary research and assay design we found that DNA is extracted from samples in four steps: cell lysis, membrane disruption with a detergent, protein removal, and precipitation (Qiagen 2010) (Figure 5). We predict DNA yield of purified samples will be in the range of 3-8 μg, because this is the normal theoretical yield of

understanding of issues like genetic testing significantly more than any other surveyed group (Figure 6). Using the 1-10 scale for all questions pertaining to genetic testing, all surveyed groups on average responded above the median answer of 5, from which it can be interpreted that Michigan State University students tend to be supportive and knowledgeable about genetics (Table 2).

Discussion

Written by: Mitchell Wood
Revised by: Nathan Johns
Finalized by: Sarah Bonczyk

Experiment Summary

Cystic fibrosis, the most common autosomal recessive disease in Caucasians (Aznarez *et al*, 2007), has dramatic effects on multiple organs, including the lungs, pancreas, intestines, and liver (Welsh and Smith, 1995), due to mutations of the CFTR gene on chromosome seven, causing defects in sodium and chloride transport in epithelial cells (Aznarez *et al*, 2007). The R553X mutation is a specific variation of cystic fibrosis, involving a single base pair substitution at the 1789th base pair in the 553rd amino acid, from cytosine to thymine (Hull *et al*, 1993). The subsequent change from the amino acid arginine to a premature stop codon causes early truncation of the CFTR protein, thus altering the folding sequence (Gambardella *et al.* 2006). Although PCR has been proven effective for diagnosing genetic disorders such as cystic fibrosis (O'Leary *et al*, 1997), the question we are addressing is whether or not a PCR test can be designed to identify this specific mutation. We hypothesized that allele specific primers and a single base pair mismatch could be used to develop an accurate diagnostic test for patients with the R553X mutation using experimentally determined optimal conditions of PCR in terms of annealing temperature, primer concentration, and salt concentration.

In addition to primer design, we bridged the gap between laboratory experiments and the sociology behind diagnosing genetic diseases by surveying student opinion on genetic screening and the effects of genetic diseases on the human race in the long run. In recent years modern medicine has extended the life expectancy of people with cystic fibrosis allowing those affected to live to childbearing age (Ratjen 2008), meaning CF genes are more likely to be passed down to future generations. Samples were taken from Lyman Briggs, James Madison, the College of

Natural Science, and general university students. It was hypothesized that Lyman Briggs students would be more supportive of genetic testing due in part to their background in required integrated studies and their greater understanding of how the frequency of a genetic disease could impact a gene pool (Singer *et al*, 2008).

Original Predictions

By amplifying DNA from IB3 human bronchial epithelial cells from a CF patient and S9 epithelial cells from a leukemia patient via allele specific PCR, the length of the amplified DNA was interpreted through gel electrophoresis to show the presence or absence of the R553X mutation. Two different forward primers, Fprimer1 and Fprimer2, were designed to discriminate between the wild-type and mutant CFTR genes through allele specificity based on a single base pair mismatch on the 3' end. The mismatch was positioned on the 3' end of the primers to more effectively reduce the amplified product by decreasing DNA polymerase and dNTP binding efficiency (Yaku *et al*, 2008). Successful annealing of the primers and the subsequent extension phase was hypothesized to result in a band of 1,056 base pairs, thus indicating a positive test. The lack of a band was hypothesized to indicate a disruption in the extension phase due to the single base pair mismatch (Chavanas *et al*, 1996). A homozygous wild-type genotype was expected to show a band of 1,056 base pairs when using Fprimer1 and show no band when using Fprimer2. In contrast, a homozygous mutant genotype was expected to show a band of 1,056 base pairs when using Fprimer2 and show no band when using Fprimer1. Lastly for heterozygous genotypes, faint bands 1,056 base pairs long were expected to appear in both tests, using either Fprimer1 or Fprimer2 due to the replication of both genotypes during PCR, causing neither set of forward primers to completely discriminate against the specific mutation site (Chavanas *et al*, 1996).

Results and Ultimate Findings

In order to determine optimal PCR conditions, multiple experimental trials were run with adjustments in DNA concentration and primer concentration. In addition, multiple experiments were run to establish the optimal annealing temperature, which can directly affect the annealing rates of designed primers (Elnifro *et al*, 2000) and magnesium chloride ($MgCl_2$) concentration to alter magnesium ion concentrations, which directly affect DNA polymerase activity in PCR (Ignatov *et al*, 2002). The optimal annealing temperature was determined to be 46ºC based off of the calculated primer melting temperatures (see Methods section) and which annealing

References

Written by: Mitchell Wood
Revised by: Nathan Johns
Finalized by: Sarah Bonczyk

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Predicted Figures:
Authored by: Kevin Werner

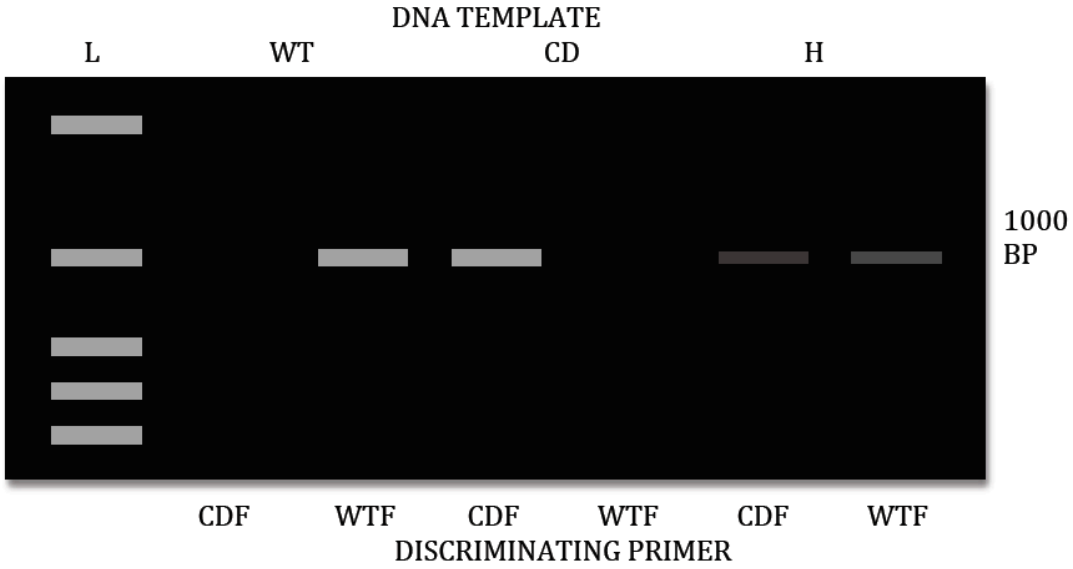

Figure 1 – Predicted results of from amplified products using gel electrophoresis. The non-discriminating oligonucleotide reverse primer, R, is 2% 3'-GTGGCTGCAGGGTTACAACTA-5'. The discriminating oligonucleotide forward primer CDF is 2% 5'-CGGGACGAGGCCGCG-3'. The discriminating oligonucleotide forward primer WTF is 2% 5'-GCGGGACGAGACCGCG-3'. The predicted binding site is on chromosome 16 on the CARD 15 gene from position 19,866 b.p. to 19,881 b.p. for the forward primer CDF, from position 19,866 b.p. to 19,881 b.p. for the forward primer WTF, and from position 20,866 b.p. to 20,887 b.p. for the reverse primer R with respect to the CARD 15 gene. It is predicted there will be a yield of 1.7 billion copies of the target DNA, along with a negligible amount of much larger DNA fragments (a by-product of PCR), and the original DNA template. The denaturing temperature will be set at 95°C, the annealing temperature at 55°C, and the extension temperature at 72°C. An initial denaturing time of two minutes will be used, then the times will be 30 seconds denaturing, 45 seconds annealing, one minute extending, and a final extension of 7 minutes for 30 cycles. The agarose gel contains 10% agarose. All bands are predicted to appear at 1021 base pairs beside the ladder. Bromphenol blue dye will be added to the gel to indicate when the gel electrophoresis is complete. L is the molecular marker or ladder, and the DNA templates WT, CD, and H are 2% DNA purified products from a wild type, mutant, and heterozygous genotype, respectively.

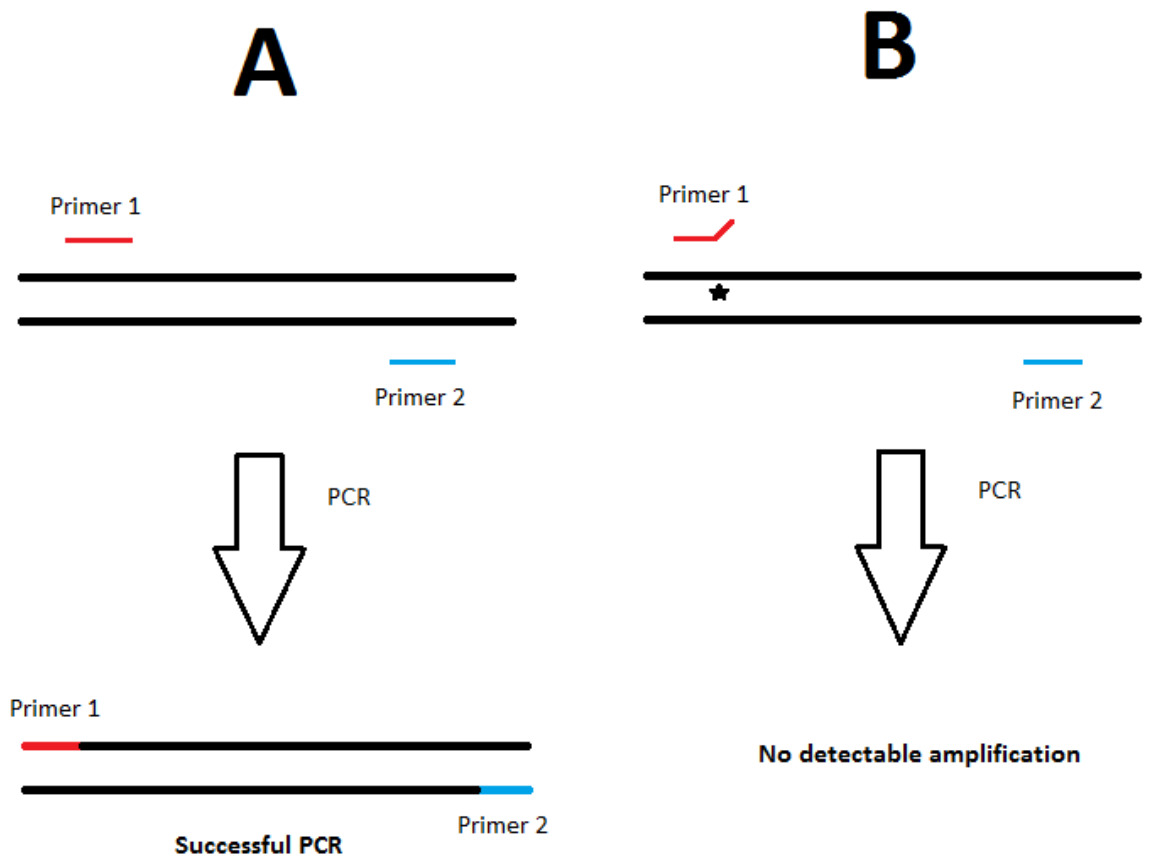

Figure 8. Representation of allele specific PCR. Column A represents wild-type DNA that was extracted using the "generation capture column kit". Column B represents mutant DNA. Primer 1 represents the forward wild-type primer and primer 2 represents the reverse primer. When primer 1 is used with wild-type DNA as seen in column A, PCR completes successfully. This is because the oligonucleotide bases match up with the DNA template and primers. However, when we use primer 1 with mutant DNA as shown in column B, the result is that there is no detectable amplification of DNA.

Figures

Written by: Sarah Bonczyk
Revised by: Liz LeMieux
Finalized by: Nathan Johns

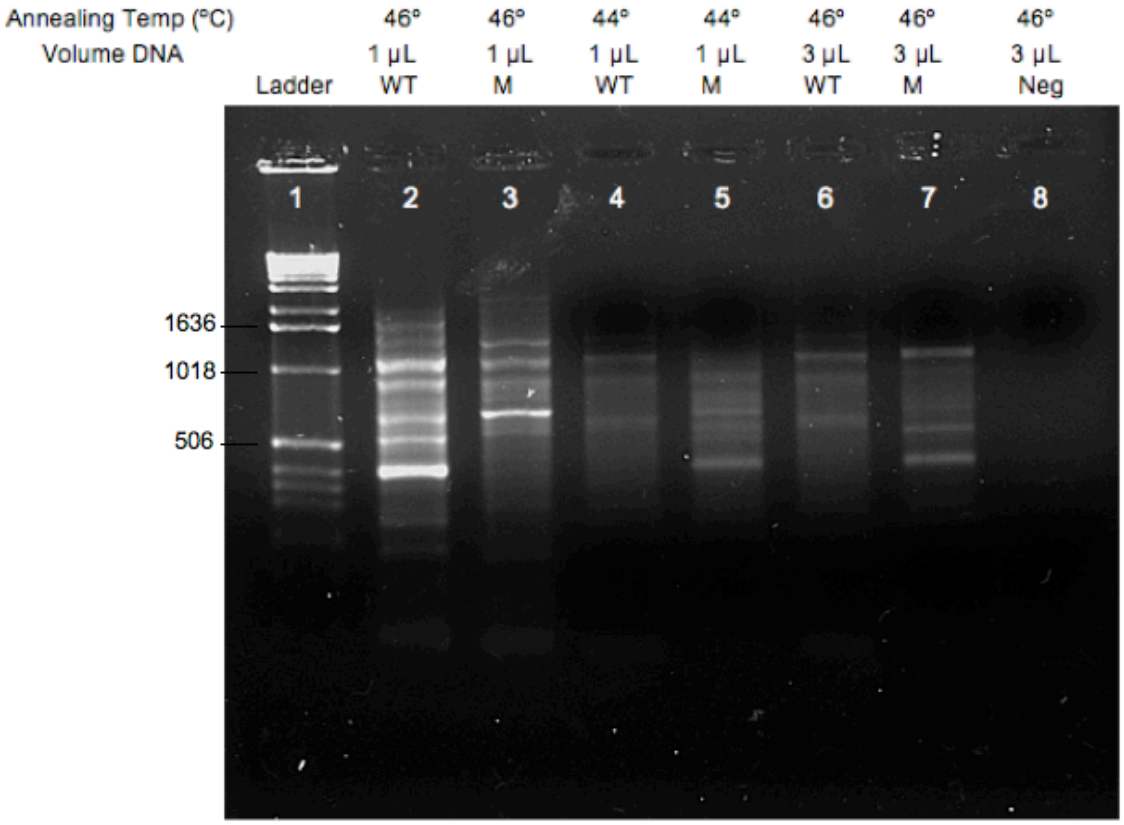

Figure 1: Amplification of DNA segment containing R553X mutation site while varying annealing temperatures and initial S9 DNA volumes. Thermocycling conditions included a 5-minute denaturation at 94 ºC with 30 cycles of 30 s at 94 ºC, 30 s at 46 or 44 ºC, and 60 s at 72 ºC, with a final elongation phase at 72 ºC for 7 minutes. All lanes show non-specific binding. Lanes 2,3,4,6, and 7 show bands near 1018. The targeted region of DNA is 1056 base pairs in length. Lanes using an annealing temperature of 46 ºC and lower initial DNA volumes of 1µL show higher intensity bands than other lanes. From this test it was determined that lower initial concentrations of DNA and an annealing temperature of 46 ºC are optimal for our designed primers.

Table 1. Chi-square test on adverse reactions to CF symptoms in public locations.

Location	Total People Observed	Expected Frequency*	People Observed with Reaction	People Expected with Reaction	$(O-E)^2/E$
Elevator	40	.717	27	28.68	.0984
Cafeteria	25	.717	17	17.93	.0482
Study Lounge	15	.717	9	10.75	.284
Restaurant	10	.717	6	7.17	.191
Total	**90**	**.717**	**59**	**64.53**	X^2=.6216
				n=4	df=3 p value associated with X^2=.90

*Values representative of the results of question one of Figure 5.

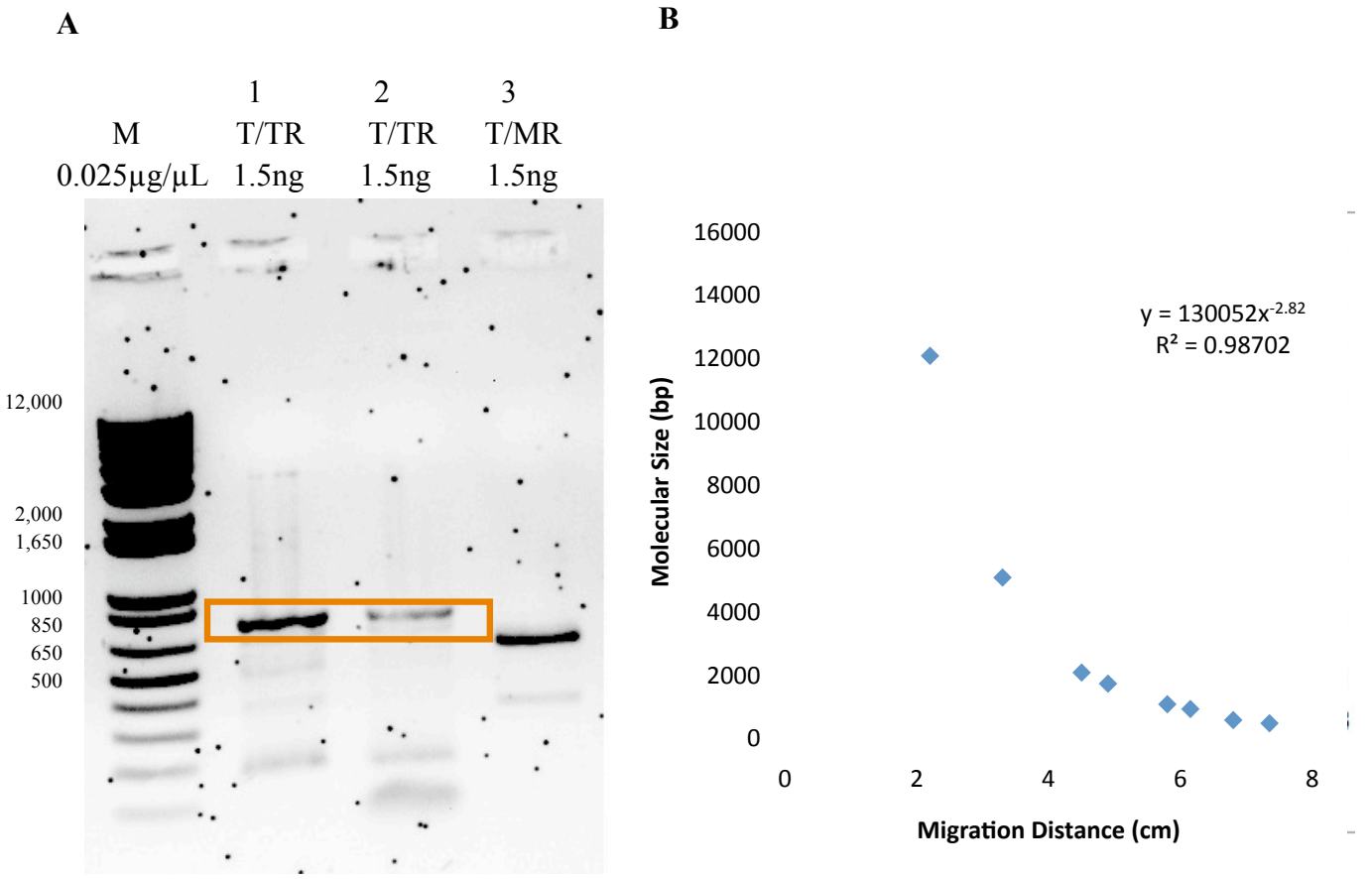

Figure 2. PCR amplification of the DMD gene from human DNA at an annealing temperature of 48ºC. A. After PCR amplification, gel electrophoresis was conducted in a 0.8% TBE gel run at 115V for 30 minutes to detect amplified regions of DNA. M is the molecular marker (1.25ng) 1-Kb Plus Ladder. Lanes 1 and 2 correspond to wild-type DNA samples tested with the wild-type forward/reverse primer set (T/TR). 10µL (1.5ng) of wild-type DNA was added to each of these lanes, and the expected amplification of a 765bp long fragment was observed. Lane 3 corresponds to a wild-type DNA sample amplified with the mutant forward/reverse primer set (T/MR). 10µL (1.5ng) of DNA was added to this lane and a PCR product of 589bp long was observed. For each one of the lanes, non-specific binding was also seen but the expected band was quite distinct.

B. Semi-log plot for 1.25ng of 1-Kb Plus ladder. Each point of the graph depicts the specific distance traveled by each band of the 1-Kb Plus ladder with respect to its well. An R^2 value of 0.98702 ($p<0.05$) was obtained for the logarithmic trend line of the plotted traveled distances. The equation obtained from the trend line was used to calculate the traveled distances of the bands from lanes 1, 2, and 3 to get more accurate band size values. Values of 757.83 ± 8.75bp, 775.33 ± 8.75bp and 589.03 ± 5.21bp were obtained for lanes 1, 2, and 3 respectively.

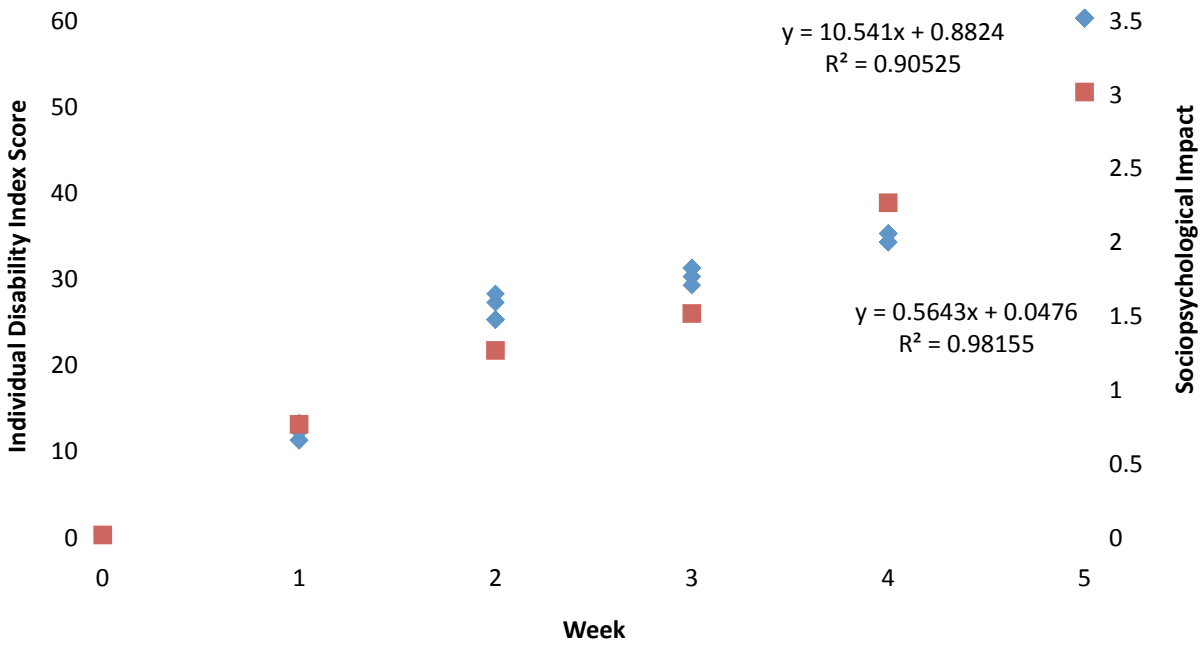

Figure 5. Correlation between the degenerative nature of Duchenne muscular dystrophy and the deteriorating socio-psychological state associated with the disease. Duchenne muscular dystrophy's symptoms were progressively added to each one of the researchers' lifestyles every week for a five-week period. At the end of each week, each researcher completed the Disability Index of Stanford University's Health Assessment Questionnaire (HAQ) in order to gauge the severity of disability that the researchers were living with. The HAQ gives scores between 0 and 60, with zero pertaining to no disability at all and sixty symbolizing complete disability and immobility. In addition to the HAQ, each researcher completed a survey that measured, on a scale of 0 to 3, the socio-psychological impact on the researcher as each symptom was added. A linear regression for the individual Disability Index scores is shown in blue while the linear regression for the Socio-psychological impact test scores is shown in red. An R^2 value of 0.90525 ($p<0.05$) was obtained for the Disability Index score and an R^2 value of 0.98155 ($p<0.05$) was obtained for the socio-psychological impact test.

Group Inquiry Research Paper DRAFT 1 (30 points)
Due at the start of Lab (week 3)

****REJECT and HAND BACK IMMEDIATELY if…***
__ The individual authorship of sections is unclear.
__ "Group responsibility" signature page is not completed
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ORIGINALITY/CREATIVITY/PROFESSIONALISM: (WORTH 2 POINTS *EC*) ____ ec pts?!

__ **Appendix: (WORTH 2 POINTS)** Do they have copies of signed data *and* the first page of all references?

Title: (WORTH 3 POINTS)
__ Paper gets 2 pts for having a title that says something about **methods** and their **specimen**.
__ Paper gets 1 points if their title predicts anything about their expected **findings**.

Abstract: (WORTH 5 POINTS)
__ Paper gets 3 points for having an abstract that explains well what they **will do and why.**
Including: ___purpose ___hypothesis ___experimental design ___significance in science
__ 2 points if abstract ***predicts*** anything about their expected **findings/results** *with* support from citations.

Introduction: (WORTH 5 POINTS)
__ Paper gets 4 points for having an introduction that tells you some **background** (several paragraphs) about what they plan to do and (one/final paragraph) what they **expect**/predict to see.
__ Paper gets the final 1 point if their introduction talks about **anyone else's research** on this topic or other related reference information from a book or paper etc.
__ Paper gets 1 BONUS point for having an Introduction that makes you **excited** and want to read on.

Methods: (WORTH 5 POINTS)
__ Paper gets 5 points for having a detailed methods section that clearly tells what they will do in a few pages. If you were an LB-144 student and ONLY had the Methods section, **could you repeat** the work?
Including: ___source of materials ___procedures and data analysis ___narrative & in past tense

"Predicted" Results and Figures: (WORTH 5 POINTS)
__ Paper gets 2 points if "Predicted" Results section discusses expected/predicted data that will be collected and supports assertions with rationale and citation of paper. Is each Figure cited properly, does it make sense?
Including: ___outcomes of experiments predicted with support ("We predict ... because... citation/paper")
__ Paper gets 1 point for each "prototype" figure that seems highly appropriate for their investigation.
Including: ___do Figures address research question ___ have a professional appearance ___have long legends with title and extensive description (so a 144 student could explain it with only that page).

Discussion: (WORTH 5 POINTS)
Try to develop a hypothesis that might be tested in your research. Present **why** you have predicted the results you did? The Discussion should be based on **evidence** found in your citations: books, magazines, literature.
__ 1. has an appropriate and interesting TOPIC for the circumstances;
__ 2. has a clear, and preferably original, specific POINT (also known as a THESIS);
__ 3. provides adequate SUPPORT (REASONING and EVIDENCE) for that point; *requires citations*
__ 4. is well ORGANIZED so that the audience can follow the points and examples;
__ 5. employs CLEAR, PRECISE LANGUAGE;
__ 6. is factually ACCURATE and also FAIR, including recognizing objections; *requires citations*
__ 7. is presented in a way that is ENGAGING to the audience;
__ 8. is of a REASONABLE SIZE for the circumstances (not too short or too long);
__ 9. clearly CREDITS OTHERS when their ideas and words are used; *requires citations*
__ 10. Indicates original PREDICTIONS and ultimate findings

"Group Responsibility" Signature Page (DRAFT 1)

Attach this sheet to the front of any written work that is to be turned in for a grade.

I, the undersigned, have read, edited and approved of the full manuscript my group is now submitting for grading. I can explain any part of it to Dr. Luckie and I am willing and excited to discuss the research on a moment's notice. In addition to the above, I am verifying that as a member of the Lyman Briggs community, I have held myself and my peers to the highest measures of honesty and integrity. My group has neither given nor received any unauthorized assistance in completing this work and we submitted our manuscript to http://turnitin.com/ for screening.

Group Name _____ Date _____

Primary Investigator: Name _____ Signature _____

I developed/revised these sections of the paper: _____

Since the Proposal, I performed these duties inside and outside of lab towards the completion of this part of our group research project (e.g. assays, meetings, growing/buying plants, etc)

_____

Protocol Expert: Name _____ Signature _____

I developed/revised these sections of the paper: _____

Since the Proposal, I performed these duties inside and outside of lab towards the completion of this part of our group research project (e.g. assays, meetings, growing/buying plants, etc)

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Data Recorder: Name _____ Signature _____

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Since the Proposal, I performed these duties inside and outside of lab towards the completion of this part of our group research project (e.g. assays, meetings, growing/buying plants, etc)

_____

Laboratory Technician: Name _____ Signature _____

I developed/revised these sections of the paper: _____

Since the Proposal, I performed these duties inside and outside of lab towards the completion of this part of our group research project (e.g. assays, meetings, growing/buying plants, etc)

_____

Group Inquiry Research Paper DRAFT 2 (40 points)
Due at the start of Lab (week 6)

****REJECT and HAND BACK IMMEDIATELY if…***
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__ "Group responsibility" signature page is not completed
__ The individual responsible for each section is unclear (ie "revised by Jim Smith").
__ Submitted manuscript format doesn't follow "Instructions to Authors"
__ Submitted manuscript has not yet been uploaded to http://turnitin.com/ (have receipt?)

ORIGINALITY/CREATIVITY/PROFESSIONALISM: (WORTH UP TO 5 POINTS **EC)**
__ Paper gets 1-5 pts for having experimental design that is very creative and unique -or- extremely professional ie if they take previous research, cite it well, and extend it in an impressive scientific style.

APPENDIX
__ Do they have copies of signed data *and* the 1st page of articles they've cited in References?

Title: (WORTH 3 POINTS)
__ Paper gets 2 pts for having a title that says something about **methods** and their **specimen**.
__ Paper gets 1 points if their title predicts anything about their **findings**.

Abstract: (WORTH 3 POINTS)
__ Paper gets 3 points for having an abstract that explains well what they **will or did do and why.**
Including: ___purpose ___hypothesis ___exp design ___outcomes/data ___significance in science
__ Paper gets 2 points if the abstract discusses *both* actual *and* predicted **findings/results**.
__ Subtract points if authors make claims or assertions without any evidence to support those predictions.

Introduction: (WORTH 7 POINTS)
__ Paper gets 2 points for having an introduction that tells you some **background** (several paragraphs) about what they plan to do.

Paper gets 0.5 point for each of these present in the Introduction:
__ 1. has an appropriate and interesting TOPIC for the circumstances;
__ 2. has a clear, and preferably original, specific POINT (also known as a THESIS);
__ 3. provides adequate SUPPORT (REASONING and EVIDENCE) for that point; *requires citations*
__ 4. is well ORGANIZED so that the audience can follow the points and examples;
__ 5. employs CLEAR, PRECISE LANGUAGE;
__ 6. is factually ACCURATE and also FAIR, including recognizing objections; *requires citations*
__ 7. is presented in a way that is ENGAGING to the audience;
__ 8. is of a REASONABLE SIZE for the circumstances (not too short or too long);
__ 9. clearly CREDITS OTHERS when their ideas and words are used; *requires citations*
__ 10. Indicates original PREDICTIONS and some hint of findings

Methods: (WORTH 5 POINTS) The *Methods* section must appear complete for ALL experiments and ready for publication.
__ Are there all the protocols necessary to repeat their experiments exactly for their completed research?
__ Are there all the recipes and protocols clarified for their research during the upcoming weeks?
__ If an LB-144 student ONLY had the Methods section and a course pack, **could they repeat** the work?
Including: ___source of materials ___procedures ___data analysis ___narrative & in past tense

Results: (WORTH 4 POINTS)
__ Paper gets 4 points if Results section discusses actual findings alongside the expected/predicted data that was collected. Read the Results section and look at each cited Figure or Tables, is it cited properly?
Including: ___outcomes of experiments stated? ___has actual #'s from data? ___cite figures/tables properly?
__ Subtract points if authors make claims or assertions without any evidence to support those predictions.

Figures/Tables: (WORTH 5 POINTS)
__ Paper gets 1 point for each actual (not predicted) figure that seems highly appropriate and professional for investigation. At week 6, should have completed/actual DIY research figures and those from Genomic Prep.
Including: ___do F/T address research question? ___ have a professional appearance? ___have long legends with title and extensive description (so a 144 student could explain it with only that page)?
__ If they have NO photographs of research results from any experiment, subtract points (no evidence).
__ Paper gets up to 2 **BONUS** points if Figures completely ROCK!

Discussion: (WORTH 10 POINTS)
Try to develop a hypothesis that might be tested in your research. Present **why** you believe you have found those results or predicted the results you did? The Discussion should be based on **evidence** found in your experiments and citations: books, magazines, literature.
__ 1. has an appropriate and interesting TOPIC for the circumstances;
__ 2. has a clear, and preferably original, specific POINT (also known as a THESIS);
__ 3. provides adequate SUPPORT (REASONING and EVIDENCE) for that point; *requires citations*
__ 4. is well ORGANIZED so that the audience can follow the points and examples;
__ 5. employs CLEAR, PRECISE LANGUAGE;
__ 6. is factually ACCURATE and also FAIR, including recognizing objections; *requires citations*
__ 7. is presented in a way that is ENGAGING to the audience;
__ 8. is of a REASONABLE SIZE for the circumstances (not too short or too long);
__ 9. clearly CREDITS OTHERS when their ideas and words are used; *requires citations*
__ 10. Indicates original PREDICTIONS and ultimate findings

Completed experiments: (3 POINTS) Paper must have *final draft* profession quality for completed work from previous weeks. At week 6 DIY research should be mostly complete, genomic prep also complete.
__ Does the Introduction, Results and Discussion sections have a professional complete quality for all completed experiments in the research?
__ Do the Figures (photographs, graphs, drawings etc) and Tables for all completed research appear complete and ready for publication?

"Group Responsibility" Signature Page (DRAFT 2)

Attach this and the rubric sheet to the front of any written work that is to be turned in for a grade.
I, the undersigned, have read, edited and approved of the full manuscript my group is now submitting for grading. I can explain any part of it to Dr. Luckie and I am willing and excited to discuss the research on a moment's notice. In addition to the above, I am verifying that as a member of the Lyman Briggs community, I have held myself and my peers to the highest measures of honesty and integrity. My group has neither given nor received any unauthorized assistance in completing this work and we submitted our manuscript to http://turnitin.com/ for screening.

Group Name _____ Date _____

Primary Investigator: Name _____ Signature _____

I was responsible for these sections of DRAFT2: _____

Since DRAFT1, I performed these duties inside and outside of lab towards the completion of this part of our group research project (e.g. assays, meetings, growing/buying plants, etc)

_____

Protocol Expert: Name _____ Signature _____

I was responsible for these sections of DRAFT2: _____

Since DRAFT1, I performed these duties inside and outside of lab towards the completion of this part of our group research project (e.g. assays, meetings, growing/buying plants, etc)

_____

Data Recorder: Name _____ Signature _____

I was responsible for these sections of DRAFT2: _____

Since DRAFT1, I performed these duties inside and outside of lab towards the completion of this part of our group research project (e.g. assays, meetings, growing/buying plants, etc)

_____

Laboratory Technician: Name _____ Signature _____

I was responsible for these sections of DRAFT2: _____

Since DRAFT1, I performed these duties inside and outside of lab towards the completion of this part of our group research project (e.g. assays, meetings, growing/buying plants, etc)

_____

Characteristics of a 4.0 Final Manuscript

1. Reads like a scientist wrote it. The author is clearly well-read on the topics they studied, understands their goals and methodologies and communicates that well in a concise, smart, composition.
2. References section contains scientific research papers, recent books and news articles.
3. Figures looks professional with clear and detailed legends.
4. Introduction and Discussion sections have numerous citations and they are used regularly every few sentences. All sections are clearly broken up into sub sections, with sub titles, that focus on each step of the project (such as: Quantitation of DNA Found, DNA purification, PCR analysis).
5. Each step in the research project is addressed individually and thoroughly with context "why and how it was done" as well as evidence "photographs," data sets, mathematical analysis.
6. Error analysis is scientific and thoughtful; citing difficulties found by other researchers in publications, limitations or possibilities that explain problems in data collection.

Characteristics of a 0.0 Final Manuscript

1. Reads like a high school student wrote it. The author is clearly has not read about the topics they studied (even material presented in the course pack), and does NOT understand their project or communicate well in a composition.
2. References section contains anonymous websites and citations are not complete.
3. Figures looks incomplete with short confusing legends.
4. Introduction and Discussion sections have very few citations and all sections are just long paragraphs with no topic sentences and no sub-headings.
5. Each step in the research project is NOT addressed individually and the author seems only focused on quickly being done.
6. Error analysis is trivial where all the human error items listed could easily have been fixed by using controls or doing replications (or even coming to lab more than once a week and paying attention).