Writing/Speaking Support for 20.109



Atissa Banuazizi

12-111, 617-324-2172, atissa@mit.edu

Neal Lerner

14N-234, 617-452-2939, nlerner@mit.edu

Linda Sutliff

12-112, 617-324-3081, lsutfliff@mit.edu

Getting to know you: Two truths and one lie

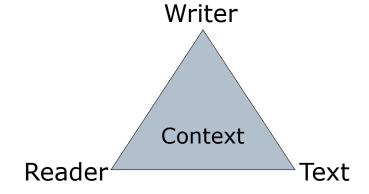
☐ Write three statements about yourself, two of them true and one a lie.



When you read a scientific research article, what do you expect to encounter?

Writing and Thinking Rhetorically about Science

Any writing act can be described in terms of a rhetorical triangle or set of relationships.



The rhetorical relationships for scientific writing can be complex and shifting.

Writer/Student/Professional/Scientist

Context: Classroom

Reader: Career Teacher Research

Research partner Scientific community **Text**: scientific data, the

"story" of your findings

Scientific writers need to control the rhetoric of scientific writing.



According to Aristotle, rhetoric is "the art of finding in any given case the available means of persuasion."

The goal of scientific writing is to court your audience.

Michael Halloran on Watson & Crick's 1953 "The Structure for DNA"

"The April 1953 paper, then, is really just the initial move in a rhetorical strategy aimed at gaining and holding the attention of an audience. As such, it presumes an understanding of science as a human community in which neither facts nor ideas speak for themselves, and the attention of the audience must be courted."



Research article scramble



☐ For the passages from a student's 20.109 laboratory report on homologous recombination: Which section (Introduction, Methods, Results, Discussion, Figure Captions) does each passage belongs to?

1.0 Introduction

By obtaining a more profound understanding of all aspects of DNA repair pathways, it may be easier for future breakthroughs in creating chemotherapeutic strategies that specifically and effectively attack cancers, and thus radically change modern cancer treatment. In order to contribute to this understanding of homologous recombination, we have created an assay that will enable us to determine when homologous recombination has taken place.

What features of this paragraph identify it as belong to the Introduction?

The introduction provides a framework for the story you are about to tell, and thus serves two main purposes. For one, you must provide sufficient background information for a reader to understand the forthcoming results. Just as importantly, you must motivate the audience to keep reading! How? Reveal the significance of the work through connections to both prior scientific accomplishments and interesting future applications.

From http://openwetware.org/wiki/20.109%28S11%29:Guidelines_for_writing_up_your_research

The Introduction establishes *context*, *focus*, and *justification*.

Context: Orient your reader to the published literature related to the topic and to essential background information

Focus: Define the research space, stake out territory. What questions are you addressing? What is your hypothesis?

Justification: Show how your work fits into and extends previous work. Argue for the importance of your work.

Swales (1990)

2.0 Methods

In order to perform bacterial transformation, 5 μ l of each purification ligation reaction was added to 50 μ l of competent bacterial cells, also a positive control was prepared with an uncut pCX-EGFP plasmid. These solutions were then heat shocked in a 42°C bath for 90 seconds so that the competent cells could uptake the DNA. 0.5 ml of LB media was then added to each reaction, and 200 μ l of each tube was plated onto separate LB + AMP plates using a sterile spreader. Each plate was then incubated at 37°C overnight.

What features of this paragraph identify it as belong to the Materials & Methods?

The methods section should allow an independent investigator to repeat any of your experiments. Use sub-section headings to allow readers to quickly identify experiments of interest to them (e.g., "Protein conjugation to hydrogels" or "Cell culture and fluorescent labeling"). When commercially available kits were used, it is sufficient to cite the name of the kit and say that it was used according to the manufacturer's protocol. The key to a good methods section is developing your judgment for what information is essential and what is extraneous.

From http://openwetware.org/wiki/20.109%28S11%29:Guidelines for writing up your research

Your research article should contain a Methods Section, not a Protocol.

A Protocol is . . .

- ☐ A series of steps to be carried out.
- ☐ Written in sequential or temporal order.
- ☐ Intended for the reader to achieve a final result.

A Methods Section is

- □ A series of steps already completed and is written in <u>past</u> <u>tense</u>.
- Written in logical order.
- ☐ Intended for the reader to replicate the experiment.

3.0 Results

As expected the digestion of plasmid backbone (Lane 2) displayed a band of about 4.8 kbp in length, as digesting with SalI would linearize the DNA. However, two other bands were seen in addition to the expected band, which could be due to poor enzyme efficiency. Lanes 3-5 in Figure 6 also confirm the projected length fragments of 3.7 kbp and 1.6 kbp (from Figure 5). This result indicates that the candidate clones were indeed the desired construct.

What features of this paragraph identify it as belong to the Results?

The purpose of the results section is to present your data in a relatively unbiased way, but with some guiding framework. Begin with a short description of the goal and strategy of your overall experiment, and then delve into specific sub-sections that describe each piece of the work. . . .

To write the results section, use the figures and tables as a guide. . . . Present the data as fully as possible, including stuff that does not quite make sense at first glance. Ultimately, each sub-section should begin with an overview sentence that introduces the present experiment and end with a sentence stating the primary conclusion reached from that experiment. . . . The overview and/or concluding sentences should also provide a transition to the previous/next piece of data when possible. Within a sub-section, be sure to stick to one topic per paragraph; sub-sections will generally require a few paragraphs each.

From http://openwetware.org/wiki/20.109%28S11%29:Guidelines_for_writing_up_your_research

What Differentiates Results from Discussion?

Results = Data Presentation

("Experiments showed that")

Discussion = Data Interpretation

("Experiments suggest that")

However, you still need to choose which data to present in your Results Section (an act of interpretation!).

Student results example: What makes this results opening effective?

In this study, a rational protein engineering approach was used to design two calcium sensors with different calcium sensitivity by mutating an existing sensor, IPC. First, SDM was used to incorporate mutations into the IPC plasmid, and the mutant DNA was amplified, isolated, and transformed into bacterial cells containing the *lac* operon protein expression system. Protein production was induced using IPTG, and after the overexpressed proteins were purified, a fluorescence assay and data analysis were used to characterize calcium sensitivity.

Construction and Amplification of Mutant Plasmids

SDM was used to create mutant plasmids from the template pRSET-IPC plasmid. Two mutants were made (Fig. 1), one in which the 124^{th} residue of CaM in IPC was mutated from methionine to serine (M124S), and one in which the BLAH residue was mutated from X to Z. For each mutant, a silent mutation creating a new restriction site was also incorporated [...]

Comment: Sets context well and concisely.

Student results example: Use of a subheading

Gel of digested IPC, M124S, and Q104R suggests successful mutagenesis reaction.

After performing a mutagenesis reaction and amplifying the plasmid, digestion of the plasmids was performed to assess the success of the mutagenesis and an agorose gel was run (Figure 2). The IPC (expected size of 4.17Kb) can be seen in the super coiled (\sim 2.5Kb in gel) configuration when uncut and in the linear (length of \sim 4Kb) configuration when cut by any of the enzymes. The M124S mutant was treated with *Accl* enzyme to produce two bands in the gel with lengths of about 3.2Kb and .8Kb, close to the expected lengths after digestion [...]

Comment: Great heading; effective intro sentence; efficient analysis.

Student example: Results in the context of class data

Other mutated variations of IPC were also analyzed by our colleagues. In parallel with us, they also examined WT-IPC and the M124S mutant. With outliers excluded, average WT-IPC K_d was 4.6e-7 M^4 with a standard deviation of 6.0e-8. The average M124S K_d was 8.22e-7 M^4 with a standard deviation of 1.38e-7.

For wt IPC, the data for 8 of 13 samples indicated a K_d within 10% of 0.43 μ M, while 11 of 13 samples had a Hill coefficient of >6.5. However, for M124S, 8 of 13 data sets indicated a K_d within 12% of 0.9 μ M, while 10 of 12 samples had a Hill coefficient of >2.

Comment: More robust to compare to class; explicitly justify model use.

4.0 Figure Caption

Results of gel electrophoresis on 1% agarose gel. Lane 1-4 contain the pCX-NNX backbone. In Lane 1 the vector is uncut. In Lane 2 the plasmid is cut with XbaI (? 4.8 kbp), while in Lane 3 it is cut with EcoRI (? 4.8 kbp). Lane 4 shows the backbone double digest with XbaI and EcoRI (?4.7kbp). Lane 5 is the 10Kb DNA Ladder. Lanes 6-7 contain the ?5-EGFP (PCR Product) insert. Lane 6 is the double digest (?0.66 kp), and Lane 7 shows the uncut insert. Lane 8 is the negative PCR-no template control. (Yellow Group W/F)

What features of this paragraph identify it as belong to a Figure caption?

Some readers begin by scanning the figures first. The figures, with the legends, should provide a self-explanatory overview of your data. Decide what the data show, then create figures which highlight the most important points of your paper.

Legends to the figures and tables explain the elements that appear in the illustration. Conclusions about the data are NOT included in the legends.

From http://openwetware.org/wiki/20.109%28S11%29:Guidelines_for_writing_up_your_research

Titles and captions allow figures and tables to stand on their own.

- ☐ **Guide** the reader to what is most important in the figure.
- □ Contextualize the data shown in terms of purpose and method.
- ☐ Focus attention on certain findings (e.g., relationship between values).
- Summarize the larger point.



Bonus tip!! Titles of tables go on TOP of the table while titles/captions of figures come BELOW the figure.

Connecting Results to Figures

From Kuroita, et al. "Structural mechanism for coordination of proofreading and polymerase activities in archael DNA polymerases." *JMB* 351, 2005, 291-298.

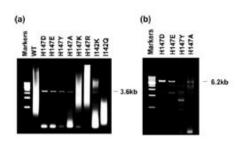


Figure 3. PCR with KOD polymerase mutants. (a) Agarose gel (1%) showing 3.6 kb PCR products. One unit of each mutant or WT enzyme was added to a mixture of 10 ng human genomic DNA and a primer pair designed to yield a 3.6 kb DNA fragment. (b) Long PCR with each mutant. One unit of each mutant was added to a mixture of 50 ng human genomic DNA and a primer pair designed to yield a 6.2 kb DNA fragment.

At first, a tragment of the human β-globin gene (3.6 kb) was amplified from different concentrations of human genomic DNA (final concentrations 2 ng/ μl and 0.2 ng/μl) by each mutated enzyme. Under the high template DNA condition (2 ng/μl), each mutant showed a distinct band at the expected position upon gel analysis (data not shown). The change in template concentration from 2 ng/μl to 0.2 ng/μl greatly increased the frequency of failed reactions. Only four mutants (i.e. H147D, H147E, H147Y and H147A) resulted in successful amplification. Although I142K also showed a faint band, conspicuous unexpected bands were amplified at the same time. The other mutants generated only indistinct non-specific bands (Figure 3(a)). This experiment indicates that the 3'-5'exonuclease activity is not the only cause of PCR failure, because some mutants exhibiting similar Exo/Pol ratios (e.g. H147E and I142Q) produced different results. From these experiments, it is concluded that the negative charge or hydrophobicity of the amino acid at position 147 plays an important role for the sensitivity of PCR.

Next, the mutants that showed successful amplification in the above experiments (H147D, H147E, H147Y and H147A) were applied to "long PCR". A DNA fragment of the myosin heavy chain (6.2 kb) was amplified from human genomic DNA (final concentration, 1 ng/µl). As shown in Figure 3(b), H147D and H147E successfully amplified 6.2 kb products. The yield with H147D was higher than that with H147E. The target was not amplified by H147Y and H147A. PCR with the other mutants and the WT enzyme also ended in failure (data not shown). These results indicate that a negative charge at residue 147 of KOD DNA

5.0 Discussion

With regards to the results obtained from flow cytometry, several unexpected results were observed. To begin with, all the negative controls had some cells that fell to the right of the diagonal line (greater FL1:FL2 ratio), suggesting that they expressed EGFP. This is likely due to the MES cells having background fluorescence or that there was contamination in the samples. However the most surprising result was the almost complete lack of homologous recombination in the ?3+75GgrAI samples. This was surprising as we hypothesized that an increase in distance of a double strand breaks would decrease HR; however, we still believed that it would be greater than having no double strand breaks.

What features of this paragraph identify it as belong to the Discussion?

The purpose of the discussion section is to interpret and contextualize your data. You should begin by reiterating the purpose of your research and your major findings. Then you might do any or all of the following: connect your findings to other research (published or that of your peers); describe any ambiguities and sources of error in the data, and suggest future experiments to resolve uncertainties; explain where you expect your work may lead, and suggest specific experiments for extending your findings; describe any conceptual or technical limitations of the research. Finally, you should explain the significance of your findings to basic science and to engineering applications. Like the previous sections, the discussion should have a clear organization and narrative flow, whether or not you use sub-sections.

From http://openwetware.org/wiki/20.109%28S11%29:Guidelines_for_writing_up_your_research

Student discussion example:

The purpose of this experiment was to alter the sensitivity of inverse pericam (IPC) to Ca^{2+} concentration by creating unique constructs with a range of Ca^{2+} dissociation constants and degrees of cooperativity. We created two constructs with mutations in the CaM portion of IPC responsible for binding to the M13 portion of IPC; one mutation (M124S) showed a higher K_d value and less coopertivity compared to wild-type IPC, while the other (BLAH) showed little change in K_d value but an increase in coopertivity.

Discussion opens with a framing of the "problem" and offers key results

What are the Pitfalls of a Discussion Section?

- □ Not enough of a controlled analytical narrative.
- ☐ Failure to follow arguments set up in the introduction.
- ☐ Failure to focus on the current results.
- ☐ Speculating too much or not enough.
- ☐ Improper tense (Discussion largely in present tense).
- ☐ Hedging excessively.



Excessive Hedging

"The cause of the degenerative changes is unknown but *possibly* one cause *may* be infection by a *presumed* parasite."

Rule of thumb: One hedge word per sentence!



Don't forget abstracts!

The abstract serves as a condensed version (not >250 words) of your report, from motivational background to key results (and how they were found) to implications for the future. By convention, it should be single-spaced and not include citations.

The importance of a good abstract cannot be overstated since computers generally index the words in a paper's title and abstract, and thus these may be the only parts that many people read. The abstract may also be the way a journal's editor decides whether to send your paper out for peer review or reject it as uninteresting and not generally relevant.

From http://openwetware.org/wiki/20.109%28S11%29:Guidelines_for_writing_up_your_research

Example student abstract

Genetically encoded calcium sensors bind calcium with specific dissociation values and degrees of cooperativity; thus, they are useful only for specific ranges of calcium. Inverse pericam (IPC) is one such sensor consisting of a calcium binding protein, calmodulin (CaM), a circularly permutated yellow fluorescent protein (cpYFP), and M13, CaM's target peptide. When bound to Ca²+, IPC shows a decrease in fluorescence. We created two mutant constructs of IPC by altering a hydrophobic and a negatively charged residue (M124S and BLAH) on CaM necessary for M13 binding. The M124S mutant showed a decrease in calcium affinity and cooperativity indicating potential use at higher calcium concentrations over a broader range than wild-type IPC. The BLAHmutant showed a relatively small decrease in affinity but a large increase in cooperativity, which would be useful for monitoring binary calcium fluctuations.

Comments:

Frames and responds to a problem; mini-report, from intro to discussion.

Resources for Writing in 20.109

☐ Guidelines for writing up your research:

http://openwetware.org/wiki/20.109%28S11%29:Guidelines for writing up your research

- ☐ Assignment Descriptions:
- RNA Engineering Report:
 http://openwetware.org/wiki/20.109%28S11%29: RNA engineering report

System Engineering Research Article:

http://openwetware.org/wiki/20.109%28S11%29: System engineering report





