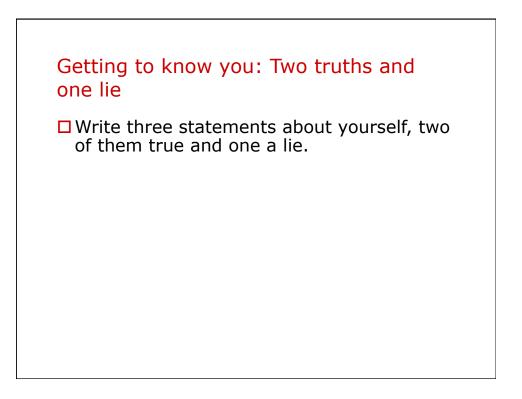
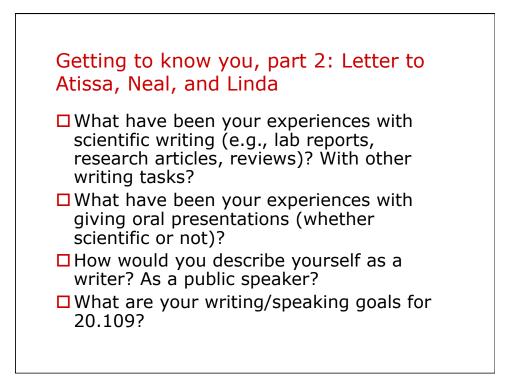
# Writing/Speaking Support for 20.109

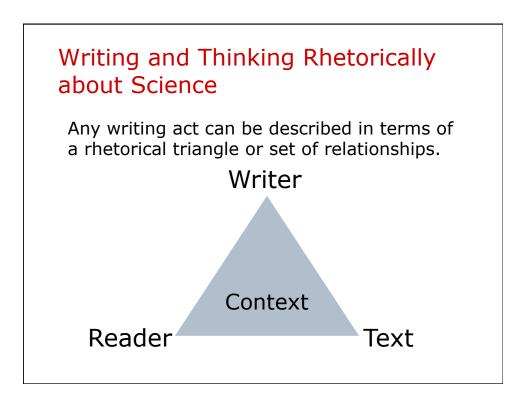
Atissa Banuazizi

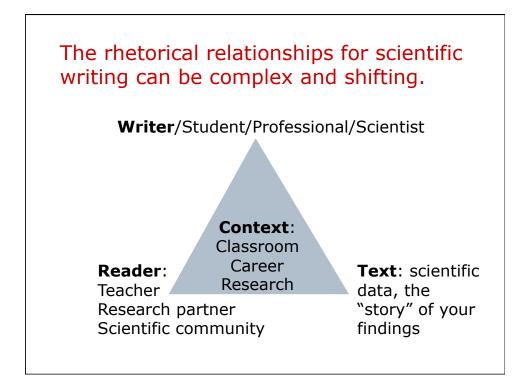
Neal Lerner

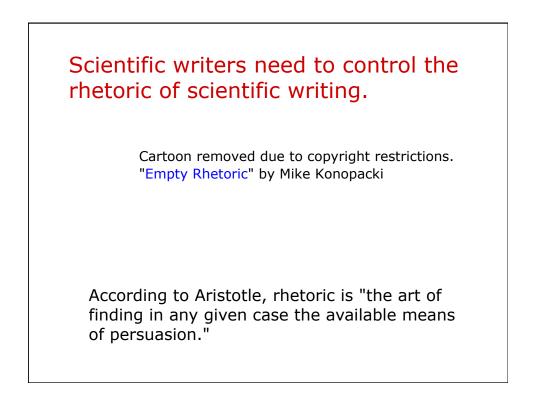
Linda Sutliff

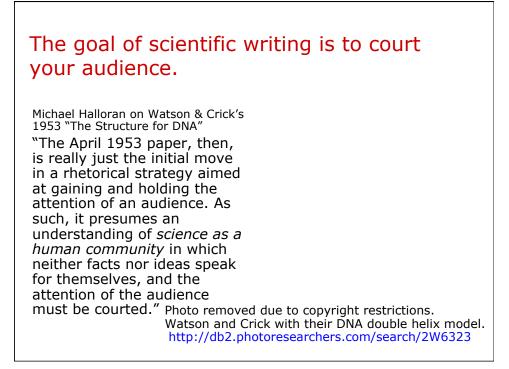














### **1.0 Introduction**

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 future applications.

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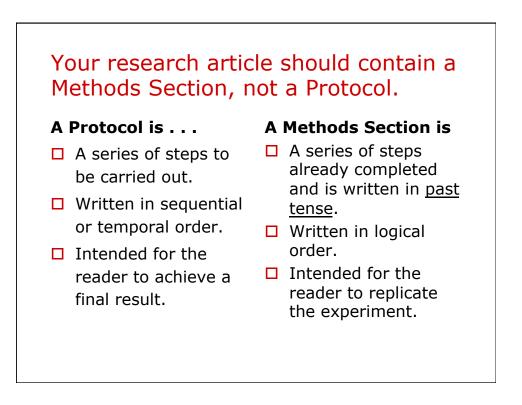
The Introduction establishes <i>context</i> , <i>focus</i> , and <i>justification</i> .		
<b>Context</b> : Orient your reader to the published literature related to the topic and to essential background information		
	Focus: Define the research space, stake out territory. What questions are you addressing? What is your hypothesis?	
Swales (1990)		<b>Justification</b> : Show how your work fits into and extends previous work. Argue for the importance of your work.

### 2.0 Methods

In order to perform bacterial transformation, 5  $\mu$ l of each purification ligation reaction was added to 50  $\mu$ 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  $\mu$ 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" and "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. Note that the methods section should be written in the past tense, since your experiments are completed at the time you are writing your paper. This section should also be written in complete sentences and paragraphs, not in bullet point form. From http://openwetware.org/wiki/20.109%28S10%29:Guidelines\_for\_writing\_up\_your\_research



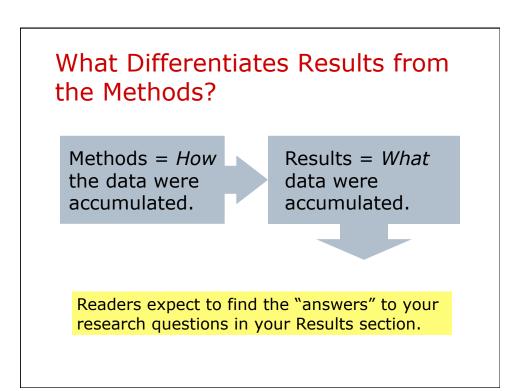
### 3.0 Results

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. (Sub-section headings and figure caption titles can also emphasize said conclusion.) The overview and/or concluding sentences should also provide a transition to the previous/next piece of data when possible. . . . Note that verbs in the results section are usually in the past tense. Only established scientific knowledge is written about in the present tense, "the world is round," for example. You cannot presume that your own data are part of the body of established scientific knowledge, and so when you describe your own results, use the past tense, "a band of 1.3 kb was seen," for example. There are, however, exceptions to this general rule. It is acceptable to say, "Table 3 shows the sizes of the DNA fragments in our preparation." It is also acceptable to say, "In a 1991 paper, Ebright and coworkers used PCR to mutagenize DNA."

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# 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!).

### 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?

Legends to the figures and tables explain the elements that appear in the illustration. Conclusions about the data are NOT included in the legends. As you write your first draft, you might state in a short simple sentence what the point of the figure or table is. In later drafts, make sure each element of the figure or table is explained. Your figure legends should be written in the present tense since you are explaining elements that still exist at the time that you are writing the paper.

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## 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.



Image: public domain (NASA: Earth as seen from Mars)

**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.

© Elsevier. All rights reserved. This content is excluded from our Creative Commons license. For more information, see http://ocw.mit.edu/fairuse. 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'exonucleasesome mutants exhibiting similar Exo/Pol ratios(e.g. H147E and I142Q) produced different results.From these experiments, it is concluded that thenegative charge or hydrophobicity of the aminoacid at position 147 plays an important role for thesensitivity 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+?5SgrAI samples. This was surprising as we hypothesized that an increase in distance of a double strand break 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.

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### Good MIT Resources

The Mayfield Guide On-Line http://www.mhhe.com/mayfieldpub/tsw/home.htm

The MIT Writing and Communication Center http://web.mit.edu/writing/





20.109 Laboratory Fundamentals in Biological Engineering Spring 2010

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