

Session 3: Coordinating your Figures and Tables with Results, Figure Legends and Materials and Methods

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Objectives

- Define the purpose and content of the Materials and Methods, Figure Legends and Results sections.
- Utilize the three sections to improve the presentation of the data.

What we are trying to avoid...



The “Big Picture”

- In order for other scientists (competitors, reviewers or colleagues) to understand the significance of your data/experiments, they must be able to:
 - Understand precisely what you did
 - See your data in a clear and simple way
 - Clearly know what you believe the data demonstrates
 - Determine if the data you present justifies your interpretation and conclusions

The “Big Picture”

- In order for other scientists (competitors, reviewers or colleagues) to understand the significance of your data/experiments, they must be able to:
 - Understand precisely what you did **(M & M)**
 - See your data in a clear and simple way **(Legend)**
 - Clearly know what you believe the data demonstrates **(Results)**
 - Determine if the data you present justifies your interpretation and conclusions

Key Points to Remember

- Understand your target audience/journal and describe your work at the appropriate level
- Avoid repetition but verify interdependence of the sections
 - Materials and Methods
 - Figure Legends
 - Results

Materials and Methods

- Definition
 - Provides enough experimental detail so a reader familiar with the field could replicate the exact experiments described in the manuscript.

Materials and Methods

- Provide short, focused subsections
- Critical details regarding compounds, reagents and concentrations
- Provide reagent sources or references to sources
- Organize according to the order in which the data are presented (figures/tables)
- Define commonly used terms and abbreviations
- References or citations to other papers should be limited to common techniques or procedures
 - PLEASE CONFIRM THAT THE DESCRIPTION IN YOUR CITATION IS IN FACT ACCURATE

Figure Legends

- Definition
 - Provides enough experimental detail so that a reader familiar with the topic would be able to understand how and why the experiment was done.
 - Define all terms and abbreviations that are essential for understanding the data (lane numbers, concentrations, statistical analysis etc.)

Figure Legends

- Start with a summary sentence similar to a subsection heading or title.
- Focus on the figure and describing what is being presented.
- Avoid conclusions or interpretations

Results

- Definition
 - A cogent, precise description of what new or important data/information the experiment provides.

Results

- Rationale for an experiment can be provided
- The data from an experiment can be related to other data
- Interpretation of the data should be very limited
- Subsections/headers can cover multiple figures if they are related to each other

Interdependence

- The sections should build and integrate with each other
 - M & M provides the details of how things were done and critical reagents/conditions
 - Figure legends allow for appropriate understanding of the experiment
 - Results state the how the data fits with other experiments and what the author feels is important and novel about the data

Example:

Manuscript on influenza viruses which escape inhibition by anti-M2 protein antibodies

- Target Journal/Audience
 - Specialized journal focused on virology or vaccinology
 - Interested audience will primarily be individuals interested in influenza virus and influenza vaccines

Example: Materials and Methods

Virus infections. MDCK cells were infected with influenza WSN wt or the antibody escape mutant WSN esc at various multiplicities of infection (MOI) as defined by 50% tissue culture infectious units (TCID50) units per cell.

Example: Materials and Methods

Western blotting. For analysis by Western blotting, the infected cells (MOI=5.0) and a mock-infected MDCK cell culture were lysed in 1% SDS in PBS (phosphate buffered saline, Sigma, Inc.) at 9 hours post infection (9hpi) The lysates were passed through a 20g needle 10 times then stored at -20C. Procedures for SDS-PAGE and Western blotting were previously described (12). Briefly, cell lysates were mixed with an equal volume of 2x SDS-PAGE sample buffer, placed in a 100C water bath for 10 minutes, then placed on ice for 5 minutes. Polypeptides from cell lysates were separated by SDS-PAGE on 17.5% polyacrylamide gels. The polypeptides were transferred to PVDF membranes (Immopilon-FL, Millipore). The anti-M2 protein monoclonal antibodies 14C2 (specific to the extracellular domain of M2) and 1F5 (specific to the cytoplasmic tail of M2) were utilized at a final concentration of 5 ug/ml while a monoclonal antibody to b-actin (Sigma, Inc.) was used at a final concentration of 1 ug/ml. Primary antibodies were detected using goat anti-mouse IgG conjugated to Alexa Fluor 647 (Invitrogen; final concentration 10 ug/ml). The blots were imaged with a FLA-5000 phosphorimager (Fujifilm).

Example: Figure Legend

Figure 3. Western blotting of the influenza M2 protein with domain-specific monoclonal antibodies. The indicated MDCK cell lysates were probed for the expression of the M2 protein using (A) the 14C2 or (B) the 1F5 antibody. An antibody specific for b-actin was used to demonstrate equivalent loading of cell lysates.

Example: Results

To determine if the M2 protein from the WSN esc virus was still recognized by the 14C2 antibody used to select the variant virus, virus-infected MDCK cell lysates were analyzed by Western blotting using the 14C2 antibody or the 1F5 antibody which recognizes the cytoplasmic tail of the M2 protein (Figure 3). The M2 protein from WSN wt and WSN esc infected cells was recognized by both antibodies, indicating the 14C2 epitope was not lost in the WSN esc virus. This data demonstrate that the WSN esc virus is not a classically defined antibody escape variant since it maintains reactivity to the selection antibody (Figure 3) while losing sensitivity to the inhibitory effects of the same antibody (Figure 2).

Are we ready?

References

- William Wells. Me write pretty one day: how to write a good scientific paper. J. Cell Biology 165(6):757-758, 2004.
- Ushma S. Neill. How to write a scientific masterpiece. J. Clinical Investigations 117:3599-3602, 2007.
- Robert A. Day. How to Write and Publish a Scientific Paper. 1998. 5th edition. Oryx Press