
Who, What, When, Where, How, and Why: The Ingredients in the Recipe for a Successful Methods Section

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In a prior article on abstracts I discussed the need, when writing a paper, to tell the story by answering questions. If I extended that concept by asking what question(s) the Methods section of a paper answers, the first response that comes to my mind is “How did I perform the study?” Yet “how” is just one of the main ingredients in the recipe for a successful Methods section. An informative Methods section also starts with 1 part *what*, 1 part *when*, 1 part *where*, 1 part *who*, and 1 part *why*. As with any recipe, the proportions of each can be modified to taste, depending on the study type and journal format, but each must be added lest someone notice that something seems to be missing from the final product.

The Methods section is also called the Materials and Methods, Patients and Methods, Study Design, or Experimental section. The goals of this section are to allow readers to (a) understand how and why the experiments were performed, (b) better understand the remainder of the paper and how the results and conclusions derived from the experiments, (c) be able to reproduce the study with an expectation of success, and (d) acknowledge that the results and conclusions are valid based on the strength of the methods and study design. Making sure to include the important details about *who*, *what*, *when*, *where*, *how*, and *why* in the study can help achieve these goals. I have listed in Table 1 some example questions that, depending on the study, might be important to answer for the reader. In the remainder of this article I discuss other important ingredients that can help you develop a winning recipe for your Methods sections.

Length and Detail

Although the Methods section should not read like a procedure manual or cookbook, it is the one part of a research paper for which length (word count) is a sec-

ondary consideration after clarity and adequate detail. As long as you help readers reach the goals listed above, your Methods section should be as long as necessary to describe the important experiments in your study.

The importance of each question in Table 1 and the amount of detail required can vary depending on the type of study and the target audience. For example, if you are comparing 2 analytical methods for quantifying serum human chorionic gonadotropin and need to identify specimens from healthy individuals, pregnant women, or patients with renal failure or cancer, you might rely on the existing medical record and the prior judgment of several clinicians. Detailing where the diagnosis was made (e.g., clinic vs hospital) or who made the diagnosis (e.g., attending physician vs resident) becomes less important to the reader than detailing what protocols were followed to compare the assays, where the analyses were performed, and what instruments were used.

By comparison, if you are doing a clinical study for which a diagnosis, histopathology interpretation, or response to treatment is the major outcome, then who made the diagnosis or what diagnostic criteria were used become key details compared with who in the central laboratory tested the patient's blood or what was the analytical principle behind the commercial method used. In the second example you still want to state that the testing was performed in the central laboratory on a specified analyzer, but no additional details are needed.

Errors of omission (insufficient detail) are common in Methods sections. Experimental conditions and details sometimes become self-obvious to authors and may be unintentionally left out. One way to avoid leaving out important detail is to treat the first draft as if it were a standard operating procedure used for training individuals about the analyses, diagnostic criteria, drug preparation, or even surgery used in the research study. If you consider what details would cause the experiment to fail if left out, you may decide that some details (e.g., wearing latex gloves, the brand of pipette, where reagents are stored in the laboratory, type of sutures) are relevant only to your facility and do not need to be included in the final paper. But you may discover that you forgot to include something as simple, yet crit-

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Table 1. Who, what, when, where, how, and why questions to consider when writing the Methods section.

Who
Who maintained the records? Who reviewed the data? Who collected the specimens? Who enrolled the study participants? Who supplied the reagents? Who made the primary diagnosis? Who did the statistical analyses? Who reviewed the protocol for ethics approval? Who provided the funding?
What
What reagents, methods, and instruments were used? What type of study was it? What were the inclusion and exclusion criteria for enrolling study participants? What protocol was followed? What treatments were given? What endpoints were measured? What data transformation was performed? What statistical software package was used? What was the cutoff for statistical significance? What control studies were performed? What validation experiments were performed?
When
When were specimens collected? When were the analyses performed? When was the study initiated? When was the study terminated? When were the diagnoses made?
Where
Where were the records kept? Where were the specimens analyzed? Where were the study participants enrolled? Where was the study performed?
How
How were samples collected, processed, and stored? How many replicates were performed? How was the data reported? How were the study participants selected? How were patients recruited? How was the sample size determined? How were study participants assigned to groups? How was response measured? How were endpoints measured? How were control and disease groups defined?
Why
Why was a species chosen (mice vs rats)? Why was a selected analytical method chosen? Why was a selected experiment performed? Why were experiments done in a certain order?

ical, as the pH of a buffer, the need to perform sample preparation in glass vs plastic, or an antibiotic drip during the surgery.

Of course, errors of commission (irrelevant detail) can detract from the Methods section as well, and you must make sure to avoid adding information that can be cited and found elsewhere. For example, if you used a previously published method without modification, it is sufficient to reference the method and its principle (e.g., “We used the LC-MS/MS method of Anderson for quantifying testosterone.”). However, if you modified the published method in any way, then it is imperative that you include details of any modifications that were made (and why). Or if in a clinical study you compared continuous secondary endpoints with use of the Wilcoxon rank-sum test, there is no need

to provide a detailed description that this test is a nonparametric alternative to the 2-sample *t*-test for assessing whether 2 independent samples of observations come from the same distribution. Readers can access information about this statistical test elsewhere.

Style and Format

The Methods section should be divided into subsections with associated subheadings. The use of subheadings helps organize the material in the reader’s mind. When the materials used in the study are described, 3 formats are appropriate—as a listing under the subheading of reagents and supplies, as part of the description of an individual experiment, or both. Generic reagents, such as solvents, chemicals, and buffers, that are used throughout the study or in multiple places in the study protocol can be listed in a subheading labeled as Materials. However, if a reagent is specific to an individual experiment or method, such as a PCR experiment, then the reagents, enzymes, etc. used solely for the PCR should be listed in the paragraph detailing the PCR experiment, thereby helping the reader associate the importance of specific reagents with specific experiments. Be sure to include the source or vendor for all chemicals, reagents, animals, and instruments used in the study. Some journals also request that the location of the vendor be included the first time that the material is mentioned.

The Methods section should be written in the past tense because you are describing experiments and protocols that you did in the past:

- *The experiment was performed at room temperature.*
- *We quantified the drug by immunoassay.*
- *Assay correlation was determined by Spearman rank correlation.*
- *We performed a 2-way ANOVA.*
- *Study participants were recruited from the blood donor service.*
- *Human embryonic kidney cells were cultured in Dulbecco’s modified Eagle’s medium.*
- *Tissue release of C-reactive protein was monitored after blood flow restriction.*

The 2 exceptions to the use of the past tense are the presentation of summarized data and the introduction of figures and tables within the Methods section:

- *Our protocol is summarized in Figure 1.*
- *Figure 1 illustrates the steps in the procedure.*
- *The data are summarized as median and interquartile range.*
- *The results from these analyses are presented as relative risk ratios with 95% CIs.*

Table 2. All passive voice, all active voice, and combined use of both.
<p>All passive voice:</p> <p>Study participants were recruited from the blood donor clinic. Each participant was asked to fill out a questionnaire, which was then used to classify each individual. Heart rate, blood pressure, and temperature were evaluated by a nurse for each individual, at which time a 10-mL blood sample was obtained from each participant by a phlebotomist for routine chemistry testing. This information was compiled and used to create a database of patient characteristics vs laboratory results.</p>
<p>All active voice:</p> <p>We recruited study participants from the blood donor clinic. We asked each participant to fill out a questionnaire, which we then used to classify each individual. A nurse evaluated each individual for heart rate, blood pressure, and temperature, at which time a phlebotomist obtained a 10-mL blood sample from each participant for routine chemistry testing. We compiled this information and created a database of patient characteristics vs laboratory results.</p>
<p>Combined passive and active voice, irrelevant information removed:</p> <p>We recruited study participants from the blood donor clinic. Each participant was asked to fill out a questionnaire, which we then used to classify each individual. Heart rate, blood pressure, and temperature were evaluated, and a 10-mL blood sample obtained for routine chemistry testing. We compiled this information and created a database of patient characteristics vs laboratory results.</p>

Resources on writing differ in their preference for either the passive or the active voice for the Methods section. Either style is acceptable if used properly, but a combination of both provides the reader with some variation in presentation of the experiments. Whatever form you choose, be sure to avoid monotony in the presentation, as shown in Table 2. The first 2 examples in Table 2 also include potentially irrelevant information such as who measured heart rate and blood pressure, and who obtained the blood samples. Another way to avoid monotony when using predominantly the passive or active voice is to use transition phrases:

- *After mixing for 1 minute, we added 7 mL methylene chloride . . .*
- *To evaluate ion suppression, extracts were prepared . . .*
- *Because the cells did not adhere to polypropylene, fibroblasts were grown . . .*
- *Because of its signal-enhancing properties, cinnamic acid was added . . .*
- *Based on previous reports of calcitrol-induced inhibition, we added calcitrol . . .*

In addition to avoiding monotony and improving the flow of the text, transition phrases can be used to start a new paragraph, introduce a new experiment, or

describe for the reader why the experiment was performed.

The Methods section should present the experimental procedures in chronological order. An exception can be made to using this format if all of the experiments were performed independently of one another, with no clear ordering of their performance. In this situation the experiments can be ordered from most to least important, helping to ensure that the most important experiment is brought to the attention of, and retained by, the reader.

It is important that the details for specific experiments be presented chronologically as well. I remember one set of instructions for an instrument repair that stated something like, “Turn the luer lock counterclockwise to unlock and remove the valve. But first, bleed the gas to remove the backpressure so the valve does not fly outward and potentially cause injury.” No kidding! Similarly, chronology in sentence structure serves both the author and reader well. Instead of writing “the supernatant was transferred to another tube after centrifugation at 8800g for 10 minutes,” the actual sequence of events should be written as “after centrifugation at 8800g for 10 minutes, the supernatant was transferred to another tube.” Similarly, “we obtained a 3-mm punch biopsy sample after the patient gave informed consent” is better stated as “after the patient gave informed consent, we obtained a 3-mm punch biopsy sample.”

Tables and figures should be included in the Methods section only if they will save a large amount of text, and be of clear benefit in helping the reader understand the experiment being described. For example, in a methods paper you may have a large number of assay parameters that must be summarized (e.g., gradient conditions, mass transitions, voltage settings, detector settings, and programmed instrument changes). Describing these one after another in the text may result in a cumbersome paragraph of numbers and terms that would be more easily understood if summarized in a table. In another situation you may be describing a complex workflow protocol that is better understood as a schematic diagram. Nevertheless, the circumstances justifying a table or figure in the Methods section are few in number.

The decision of whether to put information in the Methods section vs the Results section can be confusing. The general rule is that anything known or planned at the beginning of the study goes in the Methods section, and anything that was not known or planned goes in the Results section. In some types of studies, however, the initial experiments described in the Methods section may yield data that lead to a change in subsequent experiments or to additional experiments. Because these later experiments are driven by data ob-

tained during the course of the study, the description of these experiments may make more sense if included in the Results section along with the corresponding results:

When we evaluated the data, we noted an apparent bimodal distribution related to sex. Because the original patient data set included 13 women and 47 men, we increased the number of samples obtained from women to 45 to confirm whether there was indeed a sex difference. Statistical analysis of the expanded data set (45 women and 47 men) confirmed a bimodal distribution [median (interquartile range) of 36 (14) mg/L for women and 61 (23) mg/L for men].

Lastly, make sure that the Methods section is consistent with all of the other sections in the final version of your paper. Is there an important method or experiment that is missing in the Abstract? Is there a method or experiment listed in the Abstract that is missing in the Methods section? Are there corresponding results in the Results section to match each method or experiment included in the Methods section? Is there an explanation, either in the Methods section or the Discussion, as to why each experiment was performed? As stated at the beginning of this article, you don't want a missing ingredient or the wrong ingredient to affect your final product.

Learning Exercise

Answer the following questions about the Methods section:

1. What are the questions that are answered in a Methods section?
2. Should the Methods section be written in the past, present, or future tense?
3. How does sentence structure differ between the passive and active voice?
4. In what ways are transition phrases helpful?
5. In what order are subsections organized in the text?
6. Are figures and tables allowed in the Methods section?

Final Thoughts

In Act II of William Shakespeare's *Hamlet*, Polonius states, "Though this be madness, yet there is method in

it." This statement has evolved into the modern phrase, "method to one's madness," meaning a rational plan that is hidden by a mysterious action, or a strange plan that manages to yield results. This strategy may have worked for Polonius, but will not work in a scientific paper. Poorly described experiments will trump the credibility of your results. If readers cannot understand how and why the experiments were performed, they will be hesitant to acknowledge the results and conclusions as valid. So make your Methods section work for you, not against you.

Resources and Additional Reading

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Answers to Learning Exercise

1. Who, what, when, where, how, and why.
2. Past tense, except for presenting summarized data and the introduction of figures and tables.
3. Active voice—the subject of the sentence performs the action (acts upon something). Example: Harold delivered the flowers.

Passive voice—the subject of the sentence receives the action (is acted upon). Example: The flowers were delivered by Harold.
4. Help the flow of the text, introduce a new experiment, or describe why an experiment was performed.
5. Chronological order or order of importance.
6. Yes, if they save a large amount of text and help the reader understand the experiment being described.