



Strongarch Education

Student Notes on Writing Scientific Reports

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2024

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The Purpose of Scientific Reports: Audience and Conventions

Who is Reading the Report and Why?

The purpose of scientific reports varies, depending on the writer, the audience, and why the paper is being written.

Journal articles are written to create a public record of observations and conclusions made, and are read by a broad audience that assumes the authors are skilled and informed. Generally, it is assumed that journal articles are suitably peer reviewed prior to publication. Readers of journal articles are busy professionals, seeking clarity, brevity, accuracy and focus.

On the other hand, student reports including theses, are written for a very small audience of examiners. Examiners do not assume the student is skilled in the area, but instead read the report or thesis, hoping the student demonstrates understanding and skill.

Most scientific reports are written with intention to persuade. In the case of journal articles, authors seek to persuade readers that their data has been carefully obtained, sensibly related to the literature, supports the conclusions made, and adds meaningfully to human knowledge.

In the case of student reports and theses, the same objectives apply, but students must additionally persuade the examiner that the student really does know what they are talking about.

For this reason, it is very important for student reports to clearly spell out the logic of why and how experiments were performed, and also how data has been interpreted and seen in context of the literature. As such, student reports are usually more detailed and have more tightly justified arguments, compared with most published journal articles.

These notes relate most specifically to student reports and theses, but also to a lesser extent to journal articles of original research, where a more abbreviated approach is usually adopted.

Why the Conventions Matter

This document outlines widely accepted conventions for scientific writing, but it does seem important to briefly consider why authors, and particularly students, should stick to them. It is probably reasonable to think of the conventions as having evolved to make scientific reports easier to write, easier to read, and easier to understand. However, a persuasive case could also be made for defying any of the conventions this document describes. Why then, take such trouble to adhere so painfully, to what are in some ways, arbitrary rules?

As outlined above, additional to creating an appropriate scientific record, the author also seeks to persuade. Irrespective if the reader is: a reviewer for an article submitted for publication, somebody reading the paper after publication, or an examiner; the author always hopes to convince the reader of the scientific integrity and quality of the work, and this is especially important when the authors propose a model that challenges current thought.

Where the authors particularly challenge the reader with new ideas, it is especially important for readers to not be distracted by otherwise irrelevant excursions in style, so that adhering to the established conventions avoids unnecessarily irritating the reader.

Just as important, however, is for the author to build the reader's confidence in the author's professional abilities, and part of that, is for authors to demonstrate familiarity with the accepted conventions of the profession. Failure to write in a way that is widely accepted, reveals unfamiliarity with the professional norms, and reduces confidence in what the authors have written in a way that may be unfair, but nonetheless understandable.



It badly undermines the purpose of doing research, if it is written in a way that colleagues cannot take seriously. And so, the student is strongly encouraged to become familiar with the usual conventions of scientific writing, and to simply use them as helpful tools, for propelling their own work and scientific ambitions.

The Overall Structure of Scientific Reports

The Usual Structure of Scientific Reports

Most scientific reports comprise the following components:

- Abstract
- Introduction
- Materials and Methods
- Results
- Discussion
- References

Some Abstracts are themselves formally broken into '*Introduction*' or '*Background*', '*Materials and Methods*', '*Results*' and '*Discussion*'.

Some publications, however, have a deliberately abbreviated structure. *Nature* for example, combines *Results* and *Discussion*, while the *Materials and Methods* are either on-line only, or summarised in the figure legends.

For most journals, '*Figure Legends*' follow the '*References*' section, and these are followed by '*Figures*', and then '*Tables*'.

For essays, and student reports, it is usually convenient and acceptable for *Figures*, *Figure Legends* and *Tables* to be incorporated into the *Results* section itself.

Thesis Structure

For a Thesis, a convenient overall structure is:

- Title Page
- Acknowledgements
- Contents Pages
- Lists of: Abbreviations, Tables, Figures, and Publications of work described in thesis
- Declaration
- Summary of Thesis (only a few pages at most)
- General Introductory Chapter, outlining the scientific question addressed and the main relevant literature
- A series of experimental chapters, each one having the usual structure of a normal journal article, but without the *Abstract* or *References* sections
- A General Discussion, addressing findings of the thesis as a whole
- Appendices of any relevant additional data or information
- References for the entire thesis.

Idiosyncratic Formatting

Please note that some journals and academic institutions make idiosyncratic demands with regard to formatting, so that it becomes important to adapt the general advice given in this document, to the requirements of the specific organization for whom the document is prepared.



On Referencing

When References Are Needed

Any 'statement of fact' made in a scientific paper, should be referenced.

For example, the following statement: '*Most chairs have four legs*', should be supported by one or more specific references to peer reviewed studies. If, however, no such reference can be found, the statement should be qualified in such a way that it is clear that it is just an inference or opinion, eg. '*it seems that most chairs have four legs*'; or perhaps better still, '*it is assumed that most chairs have four legs*'.

In this way, the statements made clearly describe the evidence upon which assumptions were made. A reader can either back-track through the literature or consider carefully the statement made, to make their own assessment of the strength or otherwise of supporting assumptions upon which the current work is based.

Where References Belong

References are usually needed in the *Introduction*, *Materials and Methods*, and *Discussion* sections. They are only sometimes included in *Abstracts*.

Unless special circumstances apply, references should never be in the *Results* section. When an author finds themselves inserting references in the *Results* section, it is usually a sign that what they are writing, belongs in either the *Introduction* or *Discussion*.

Time and Tense

There are reasonably strict conventions regarding 'Time Tense', which at first seem counterintuitive, but which are nonetheless very important.

When Authors Refer to the Work of Others

Observations by others reported elsewhere are supported by references, and usually described in the present tense. Examples are: '*changes in cytokine synthesis **are** dose dependent (Zimelweis et al. 2014)*', '*increased expression of protein P **is** associated with reduced expression of agent Y (Milstein and Werner, 1987)*'; '*there **is** strong correlation between use of disposable tissues and rhinitis, (Buggins and Baxter, 2002)*'.

When Authors Refer to Their Own Work

On the other hand, current work and observations made by the authors and reported in the paper being written, is always described in the past tense. Examples are: '*changes in cytokine synthesis **were** dose dependent*'; '*increased expression of protein P **was** associated with reduced expression of agent Y*'; and '*we **found** strong correlation between use of disposable tissues and rhinitis*'.

Please note, that if the authors refer to their own earlier published work, either the present or past tense can be used, depending on sentence construction. The past tense is appropriate when the words 'we' or 'I' are used. For example, '*we found strong correlation between use of disposable tissues and rhinitis (Buggins and Baxter, 2002)*'. When the words 'we' or 'I' are not used, the present tense is applied, for example: '*there **is** strong correlation between use of disposable tissues and rhinitis, (Buggins and Baxter, 2002)*'. In both cases, the relevant reference to the author's own work should be given.

On occasion, earlier observations by the authors may not have been published, but might nonetheless be important for the paper. In that case, the bracketed phrase '*(unpublished data)*' may be used. An alternative construction would be '*Our earlier unpublished work indicated that use of disposable tissues correlates with rhinitis*'.



Why Different Tenses Used

All of this is potentially confusing, because counterintuitively the past tense is mostly used for 'current' data, while the present tense is reserved for earlier published 'past' data. What may be helpful, however, is the thought that using the present tense for peer reviewed published work, indicates acceptance of the earlier work as 'fact'. On the other hand, use of the past tense to describe the authors' own new work, helps project an appropriately humble attitude. This use of the present and past tenses, creates the impression that the authors elevate the work of their predecessors to the status of 'fact', and now merely report what they have themselves seen.

Avoidance of Bullet Point Formatting: Write in Whole English Sentences

It is often tempting to use 'bullet points' or 'lists' in scientific writing, especially in the *Materials and Methods* sections. But while 'bullet point formats' and 'lists' are common and acceptable in laboratory notes and slides presentations, they do not have wide acceptance in the written literature, and are only rarely used in published journal articles.

Students are strongly discouraged from writing in anything other than whole sentences and paragraphs.

Despite this pointed advice, I have used a bullet point style for some aspects of the current document, not by way of example, but only to help make some very specific points more clear.

In general, avoid lists, avoid bullet points, and use complete English sentences.

Take Care With Headings, Sub-Headings and Paragraphs

It is dispiriting for anybody trying to understand a scientific report, to be confronted by a solid block of text. This applies equally to essays as it does to scientific reports and journal articles.

Instead, it is very helpful for text to be broken into paragraphs addressing single main issues, and for headings and sub-headings to indicate a clear hierarchy of content. Font styles are used to indicate the hierarchy of sub-headings, and care must be taken to ensure these are applied consistently throughout any document.

Sometimes, sections are numbered, and sub-sections have subsidiary numbering, so that once again, care must be taken for this to be consistent and complete throughout any document.

Notably, failure to use headings, subheadings and paragraphs in a consistent and sensible way, suggests disorder and a lack of clarity, again undermining confidence of the reader in what the author has written.

Reviewing Documents

Few writers are able to produce clean copy without significant re-writing and text-polishing. While this always improves the quality of writing, it is also true that the more an author works over the same text, the more difficult it becomes for the author to detect their own errors. This strange 'document blindness' can be partly overcome by allowing a few days, or preferably a week or more to pass, before final review, correction and submission.

It is also often helpful to print the document, because errors apparent on the printed page, are often missed on the computer screen.



Finally, thought should be given to the font used. Serifs are the little decorations adorning the ends of lines in some fonts, such as in Times New Roman, which differs from the sans serif Arial font used throughout this document. Please note that 'sans serif' is French, and is translated to English as 'without serif'.

There seems to be broad acceptance that serif fonts are easier to understand than sans serif fonts, when hard-copy documents are read. On the other hand, there is also some data suggesting sans serif fonts are more quickly interpreted, and that they may be preferable for text on computer screens and public signage. Irrespective which font is used, it is probably sensible to be at least conscious of the effect font and presentation style can have on comprehension.

The Abstract

The *Abstract* usually comprises from 150 to 250 words that summarise the main points of the paper, while the internal structure of the abstract usually mirrors that of the paper.

Reading the *Abstract* alone, should leave the reader with a clear understanding of what was done, why it was done, and what new insights have been gained. A thesis summary is essentially similar to an *Abstract* but longer, and usually spreads over from two to four thesis pages.

The Introduction

What Should be in the Introduction

The *Introduction* is written to help the reader understand what is to come in the rest of the paper. By the time the *Introduction* has been read, the reader should have absolute clarity on the precise scientific question addressed in the paper, and why it is important.

Specific aspects that should be in the Introduction are:

- What is already known, supported by relevant references
- What is not known and that the study now investigates
- And thus why the study was performed

Also, the *Introduction* should contain background for any aspect of the materials and methods that might be otherwise confusing.

What Should Not be in the Introduction – Inappropriate References and Subheadings

The *Introduction* should not comprise an exhaustive review of the literature, because the literature is usually much too extensive to mention every possible relevant paper. Instead, reference should only be made to key papers substantiating statements made. Recent reviews are helpful, but references should otherwise be to the first instance a particular observation and or conclusion, was made.

For student reports, it is particularly important to demonstrate ability to distinguish important from less important papers. When students simply list every paper written on a subject, examiners conclude an inability to distinguish the critical from the trivial.

Please note that the same careful discrimination is required when selecting references in the *Materials and Methods*, and *Discussion* sections.

It is sometimes helpful for explanatory figures and or tables to be included in the *Introduction*, and presentation conventions for these are as detailed below for the '*Results Section*'.

Sub-headings are also not usually used in the Introduction, although this is sometimes relaxed in theses where the *Introduction* is usually more extensive.



The Materials and Methods Section

This section should be written in sufficient detail for scientific colleagues to reproduce results elsewhere. There are usually multiple sub-sections in the *Materials and Methods*.

Materials

It is usually convenient to have a discrete *Materials* sub-heading, where all materials used are listed, including the source from which these materials were obtained. The supplying companies, city, state and country of origin are usually indicated for all materials used, while for the sake of brevity, specific sources are usually mentioned only once.

Methods

Detail Appropriate to Authorship and Audience

Because the student has need to demonstrate a proper understanding of the methods used, the *Methods* sections of student reports are generally more detailed than in published papers. Each method should either have its own subheading, or be at least clearly grouped together with other related methods under an appropriate sub-heading.

Equipment and Software

Equipment is not listed under *Materials*, but is incorporated into relevant *Methods* sections. Also, only equipment used for data collection is usually specified, examples being plate readers, confocal microscopes, and spectrophotometers, which would be detailed respectively under relevant sub-headings such as '*Colorimetric assays*', '*Laser scanning confocal microscopy*', or '*Spectrophotometry*'. Details of software are provided in a similar way to equipment.

Equipment used for routine laboratory procedures such as cell culture, preparing buffers, or rotating samples, is not usually detailed in the report unless it has particular bearing on the results or interpretation.

The Results Section

General Aspects of the Results Section

The *Results* section should contain only what has been observed by the authors in the current study, and as a general rule has no references. *Results* are presented with the least possible interpretation, because interpretation of data is reserved for the *Discussion*. Also, *Results* are presented in careful order, such that the reader is brought stepwise towards whatever overall conclusion the authors believe their work leads.

Although saving interpretation for the *Discussion* section, it is nonetheless sometimes helpful to make brief statements as to the conclusions drawn from results, and this is most easily done by writing relevant sub-headings in the form of conclusions. Examples of such sub-headings, with regard to a fictional 'Protein Y' might be:

- *Protein Y Activity was Dependent on Protein Conformation*
- *Activity of Protein Y was Independent of the Purity of Protein Y Preparations*

This has the further advantage that by simply reading through the sub-headings, the reader is able to quickly see what the data shows.

Results are presented as text, with supporting *Tables* and *Figures*, which comprise graphs and or photographs. It is important that any *Figures* and *Tables* shown, are specifically mentioned in the text. However, text provided in *Figure Legends* should not repeat what is written in the body of the *Results* section. Instead, the two should work together to describe the data. For the novice author, it is thus helpful to make sure that all specific statements



made in the *Results* appear only once, in either the body of the text, or a *Figure Legend*, but never both.

Statistics

Wherever possible and appropriate, statements made in the *Results* section should be accompanied by supporting statistical evaluations. A 'p value' of 0.05 or less has become widely accepted as indicating 'statistical significance', representing a probability of 1/20 or less, that the specific observation described is a statistical accident that would not be recurrent were a larger sampling of data performed. It is important for any researcher to acquire at least a basic understanding of statistical ideas and procedures.

It is beyond the scope of this document to detail how and why particular statistical tests are performed, but it is perhaps helpful to be aware that different statistical tests are appropriate for different types of data, and that it is not sensible to just 'try different tests out' till one is found that yields a satisfyingly low p value.

For all data, the authors need to consider: if the data is at nominal, ordinal, interval or ratio level; if the data is normally distributed; or if the data are dependent. Once there is clarity on these points, the appropriate statistical test can be selected and applied.

Importantly, statistical tests do not quantitate 'how true' a statement is. Statistical tests only quantitate the uncertainty authors have with regard to the reproducibility of their reported data.

Conventions for Tables

Tables are prepared with a title sentence on the top line, separated from the rest of the table by a single horizontal line, and often enclosed above by a second line. There is also usually a table description at the bottom of the *Table*, which may contain several sentences, and is enclosed by two horizontal lines.

Headings for columns include units, as do headings for rows, while differences in font may be used to help distinguish headings from data.

Lines may be provided that separate column headings from the underlying data, and such lines are often used to indicate groupings of data. Vertical lines are usually not used, but can be provided if they are of particular value.

No further lines should be provided, other than those mentioned above separating title sentences, table descriptions, and headings.

Special symbols can be used to direct the reader's attention to particular points in the *Table*, or to indicate statistical significance of observations made.

An example illustrating these conventions is provided below.



Table 1. Relative percentage values for compression considering volume (Rel. % of Volume) and Biological Response Units (Rel. %BRU) during application of incisive or molar bite force in individual teeth.

| | The Relative Percentage of Volume or BRU for Compression in Individual Teeth Coronal Soft Tissue Follicle | | | | Apical Soft Tissue Follicle | | | |
|---|--|--------------|--------------|-----------|-----------------------------|--------------|--------------|-----------|
| | Canine | 1st Premolar | 2nd Premolar | 2nd Molar | Canine | 1st Premolar | 2nd Premolar | 2nd Molar |
| Left Teeth Under Incisive Bite Force | | | | | | | | |
| Rel. % Volume | 42.2 # | 69.3 | 74.1 | 12.6 # | 24.2 | 44.6 | 32.0 | 8.0 |
| Rel. % BRU | 57.9 | 79.1 | 73.8 | 9.6 & | 22.4 | 45.2 | 25.0 | 5.6 |
| Left Teeth Under Right Molar Bite Force | | | | | | | | |
| Rel. % Volume | 38.4 # | 78.5 | 75.4 | 6.5 # | 20.3 | 50.2 # | 31.0 | 15.0 |
| Rel. % BRU | 51.0 | 89.0 | 67.8 | 2.9 & | 14.5 | 46.7 | 24.4 | 26.8 |
| Right Teeth Under Incisive Bite Force | | | | | | | | |
| Rel. % Volume | 84.5 | 56.1 | 71.4 | 4.1 # | 16.4 | 61.0 # | 43.5 | 72.0 # |
| Rel. % BRU | 89.4 | 74.7 | 79.7 | 0.8 & | 16.8 | 51.7 &a | 54.0 &a | 46.6 |
| Right Teeth Under Right Molar Bite Force | | | | | | | | |
| Rel. % Volume | 56.0 | 37.9 # | 62.1 | 47.9 # | 12.7 | 52.8 # | 73.8 # | 76.4 # |
| Rel. % BRU | 64.5 | 39.8 & | 67.3 | 49.6 &a | 6.5 | 52.4 &a | 52.9 &a | 64.5 & |

With the exception of second molars, a general pattern of compression in coronal tissues and tension in apical tissues predominated. Considering the relative percentage of volume under compression in canines and premolars only, exceptions (#) were seen in 7 out of 24 instances. Further refinement by evaluation of BRU reduced exceptions to 5 out of 24 (&). 4 of these exceptions (&a) in BRU were within 4 percentage points of the 50% value marking consistency with the general rule, and 3 of these were in right sided apical tissues during right molar biting, likely representing localized asymmetrical effects of molar bite force.

Table From Sarrafpour et al. PLOS ONE, 2013, 8(3):e58803



Conventions for Figures

Where possible, *Figures* should be prepared in black and white, rather than in colour. This convention is slowly changing, but has nonetheless survived from the time before widespread electronic publication. This convention also reflects the ongoing high cost of colour printing. A single colour image in a well regarded International journal, can cost the authors several thousand dollars to publish.

All axes in graphs should be labelled and units shown, but it is usually not necessary to provide a *Figure Title* in the figure itself. When photomicrographs are shown, size bars should be included.

The style of *Figures* should be consistent across the paper, and should also be consistent with that widely accepted in the literature, that is: bold black axes; axis ticks only where appropriate and helpful; clear numbering and lettering large enough to survive size reduction during printing; and symbols of sufficient size and appearance to be clearly distinguished from one another.

As a general rule, the eye is drawn to solid black shapes and lines more than it is to open outlined shapes or dashed lines. For this reason, it is often helpful to represent 'test' data with solid black, or where necessary bold bright coloured shapes, and to show 'control' data with open outlined shapes and dashed lines.

Microsoft Excel is often used to prepare *Figures*, but defaults to a style that includes guide lines and inappropriate axis formats. These features are easily corrected in Excel, while saving figures in PDF format facilitates convenient opening for final formatting in image processing software such as Adobe Photoshop.

Where composite figures are prepared, care must be taken to use the maximum available space with minimum possible unused area, and to also arrange *Figures* in a way that is at once logically sensible, and aesthetically pleasing.

Figure legends

Figures should always be supported by *Figure Legends*. The *Figure Legend* comprises two parts, the first being a title sentence describing the *Figure*, and the second part being one or more explanatory sentences that walk the reader across the *Figure*, and help the reader see what the authors are trying to convey.

Nothing in the *Figures* or *Figure Legends* should be ambiguous, so that it is helpful to clearly label important features in the *Figure* that are then described in detail in the *Figure Legend*. This is particularly the case in photomicrographs of histological sections.

These points are illustrated in the three examples of *Figures* and *Figure Legends* provided below.

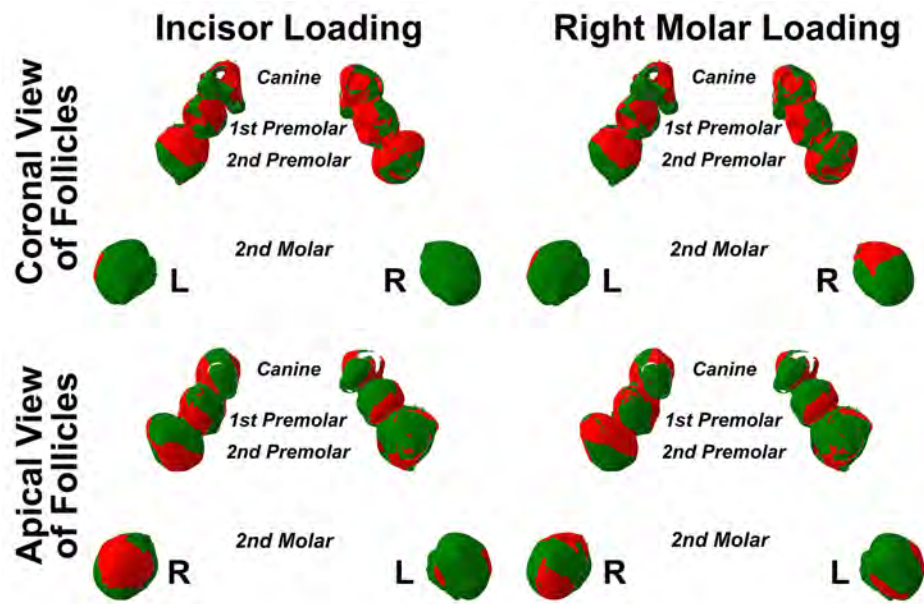


Figure 1. Dental follicle compression (red) and tension (green) during incisor or right molar bite force. The surface of dental follicles is seen from coronal or apical perspectives, while left (L) and right (R) sides are indicated. The upper surfaces of dental follicles for unerupted canines, first premolars and second premolars appeared subject to greater compression during both incisor and right molar loading, as compared with the lower surfaces of the same teeth which were in general subject to greater tension. This general pattern did not, however, appear to apply in the case of the unerupted second molars.

Figure From Sarrafpour et al. PLOS ONE, 2013, 8(3):e58803

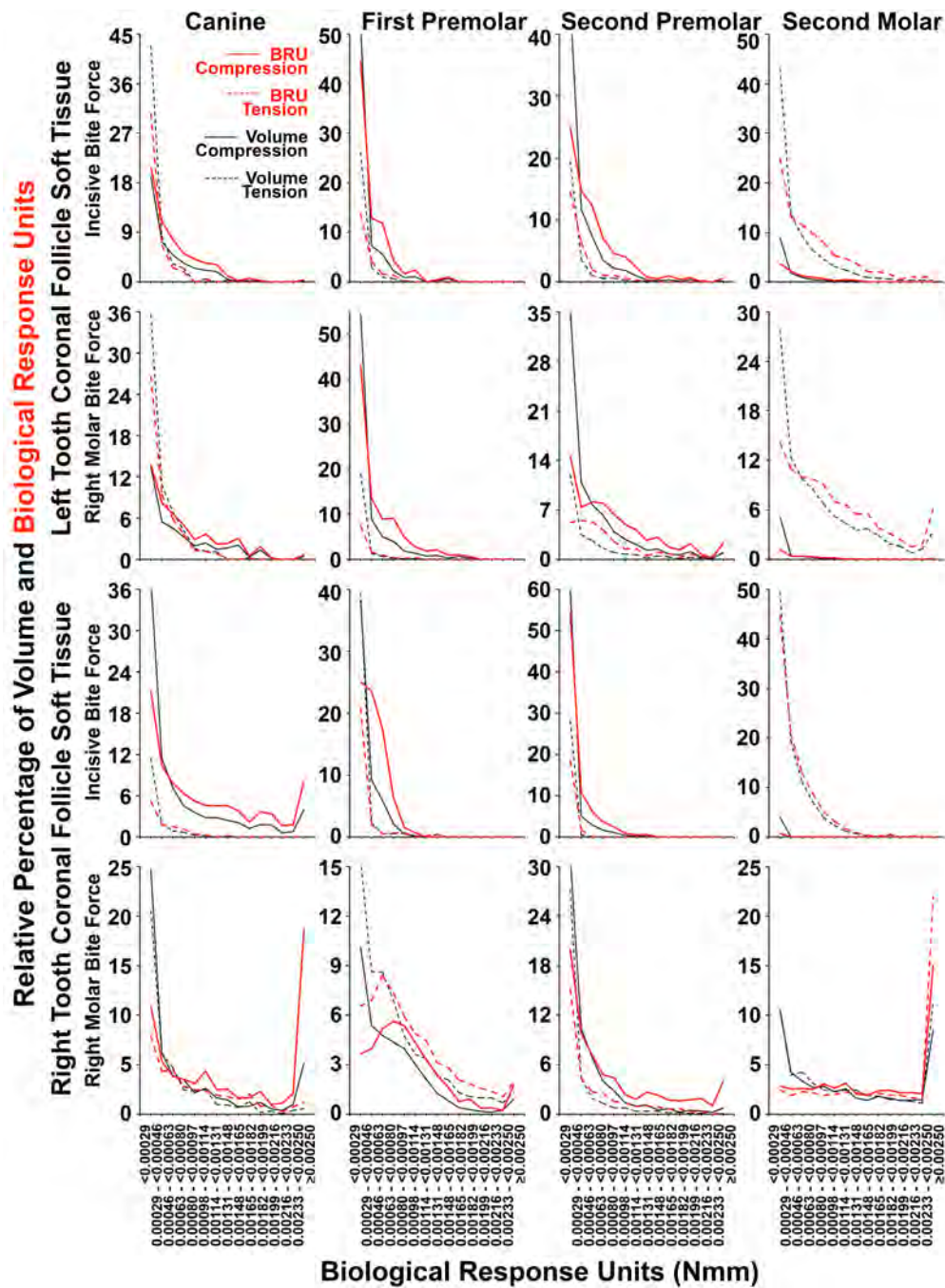


Figure 2. Percentage distribution of coronal follicle volume and summated BRU. As summarized in Table 1, considering tissue volumes alone (black lines), there was a strong tendency for compression (solid black lines) to dominate over tension (dashed black lines) across most BRU ranges. This was more pronounced when summated BRU was considered (red lines), such that there was a general right shift in BRU curves for compression (red solid lines) and sometimes a corresponding left shift in BRU curves for tension (dashed red lines). Exceptions for both volume and summated BRU were seen in the second molars, as well as in the right first premolar during right molar bite force application, while there were further exceptions considering volume alone in the left canine during both incisive and right molar loading.

Figure From Sarrafpour et al. PLOS ONE, 2013, 8(3):e58803

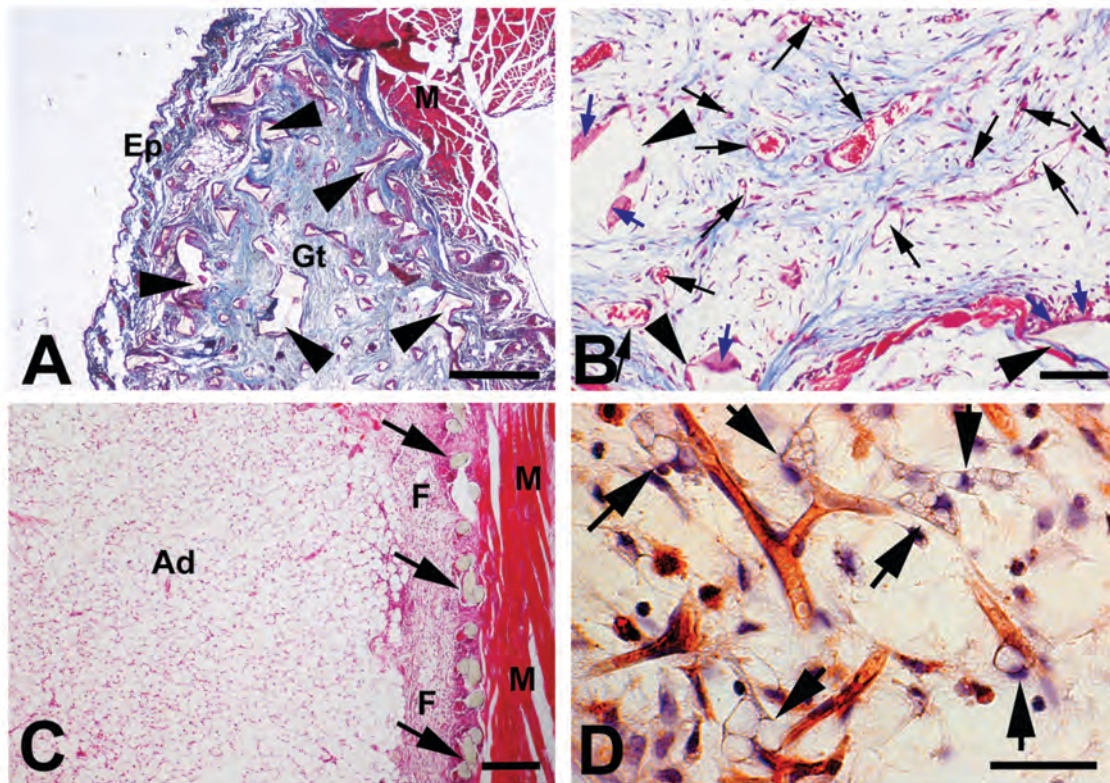


Figure 3. Photomicrographs of paraffin sections showing wound healing in mouse subcutaneous sponge implants at weeks 8 (A) and 3 (B), and mouse intramuscular adipogenic implants at weeks 6 (C) and 2 (D). (A) The epithelium (Ep) and underlying muscle (M) were separated by a mass of maturing granulation tissue (GT) penetrating into subcutaneously implanted sponge material (arrow heads), while collagen fibres stained blue using the Gomori trichrome method. (B) Blood vessels (black arrows) were readily identified in Gomori trichrome stained sections of subcutaneous implants. Clefts in tissues occupied by sponge material (arrow heads) were surrounded by multinucleated foreign body giant cells (blue arrows). (C) Granulation tissue matured to adipose tissue (Ad) in intramuscular implants, while the implant mesh material (arrows) was associated with fibrosis (F) and separated the newly formed adipose tissue (Ad) from surrounding muscle (M). (D) Multivacuolated lipoblasts (arrows) were prominent in week 2 granulation tissues of adipogenic lesions, and preceded the appearance of mature adipose tissue. These adipocyte precursor cells were often closely associated with vessels, which stained brown in *Bandeira simplicifolia* lectin histochemistry. (A & B, Gomori trichrome stain; C, H&E; D, *Bandeira simplicifolia* lectin histochemistry with Hematoxylin counterstain; Bars = 300 µm for A, 100 µm for B, 500 µm for C, 50 µm for D).

Figure From Bolitho et al. *Wound Repair and Regeneration*, 2010, 18:211-222



The Discussion Section

Overall Purpose of the Discussion

While data is presented in the *Results* section, it is in the *Discussion* that results are finally interpreted to inform understanding. It is here that mere data, is converted to actual insight. Unfortunately, this very important part of any scientific paper is often neglected, especially by students.

A common error, is for the authors to simply restate their *Results* in the *Discussion*. This incorrectly shifts responsibility for interpretation from the authors to the readers. Because authors are more informed about their own data than readers can be, simply restating the results in the *Discussion*, fails to complete interpretation and produces an inadequate scientific record.

While there are no strict rules as to how the *Discussion* should be structured, the below approach seems effective and reasonably complete, especially for students wishing to demonstrate a mature and methodical approach to interpreting their own work.

Discussion of the Experimental Approach Used

There are often different ways in which any given study could be performed, using different techniques, different source materials, or different interpretative strategies. It is helpful for the authors to briefly explore different approaches that might have been used, and to recognize the advantages and disadvantages of each of these alternative approaches.

It is also important to discuss the advantages and disadvantages of the experimental approach that was actually used in the current study.

Having methodically discussed the alternative and actual approaches used, a reasonable case can then be made as to why, on balance, the approach finally decided upon was a reasonable choice, and was most appropriate to address the scientific question under study.

Interpretation of the Results

Having demonstrated that the experimental approach was reasonable, there can now be reasonable confidence in the fidelity of the results. This increases confidence for both the reader and investigator, that the results can be sensibly interpreted.

As mentioned above, results should not be restated, but instead interpreted in this section. For example, **if the Results contained the following observations regarding a fictional protein – ‘Protein Y’:**

- *Protein Y was inactivated when denatured by reduction.*
- *Denaturing reduction of Protein Y significantly increased Tryptophan and Bis-ANS fluorescence*
- *Protein Y was inactivated when fragmented by CNBr, and fragmentation was confirmed by SDS PAGE.*
- *Different commercial sources of Native Protein Y, with varying levels of purity as demonstrated by SDS PAGE, had identical dose response to recombinant Protein Y produced in yeast.*
- *Sequence identity for all Protein Y preparations was confirmed by trypsin digestions and MALDI-TOF MS.*

The Discussion might contain the following interpretation:

Because Protein Y activity was dependent on protein conformation, and identical regardless of source, we conclude that the activity under study was not a non-specific protein effect, but was due to an as yet unidentified active protein domain in Protein Y.

Although restating results in the *Discussion* is to be avoided, it is still sometimes helpful to briefly remind the reader, which specific results lead to the conclusions made. This is



especially so when papers have large amounts of data, and readers can become easily lost amidst the many different results provided. Were that the case with regard to the results outlined above, the *Discussion* might be written in a slightly longer way as follows:

Fluorimetry and protein structural analysis confirmed conformational modification of Protein Y by reduction and fragmentation, and this correlated with loss of Protein Y activity. Separately, the dose response for Protein Y was independent of Protein Y purity, including for recombinant protein. These observations support our conclusion that the activity under study, was not a non-specific protein effect, but was due to an as yet unidentified protein domain in Protein Y.

Please note that details of the specific fluorometric and structural analyses, or of the conformational changes made, or of the different sources of Protein Y, are not provided in the *Discussion*. These, together with any relevant statistical statements, belong in the *Results* and *Materials and Methods* sections.

Alternative Interpretations of the Results with Reference to Limitations of the Methods Used

Sometimes, results can be interpreted in different ways, and this may occasionally be due to limitations of the methods used. It is important for all researchers to consider alternative explanations for their data, but it is especially important for students to demonstrate this in their own research reports, because this is one aspect examiners specifically look for.

Notwithstanding demonstration of an open minded approach to the data, it remains important for the authors to indicate their own preferred interpretation, and for this preference to be clearly explained.

Using the results provided above as an example, the *Discussion* could include the following:

The possibility remains, that the activity under study is not due to a specific Protein Y protein domain, but that it is due instead to a contaminant bound to Protein Y in a strict stoichiometric relationship, and that this contaminant is lost following conformational modification of the protein. Such a contaminant could have a low molecular mass, and thus be released and not detected during preparation and analysis of Protein Y by MALDI-TOF MS. This seems unlikely, however, because any such contaminant would have to be equally lost from Protein Y following all conformational modifications studied, and would also be equally present in all native Protein Y preparations. Furthermore, yeast recombinant Protein Y is prepared in the absence of any native mammalian contaminants, so that the identical dose response of yeast recombinant and native Protein Y, would require yeast to produce the same putative mammalian contaminant in comparable quantity to that seen in mammals. Alternatively, yeast could produce a putative contaminant chemically different to the putative mammalian contaminant, but were that the case, it would seem an extraordinary coincidence for the dose response of bioactivity of different yeast and human contaminants to be identical, in addition to having identical Protein Y binding capacities. For these reasons, while we acknowledge the formal possibility that a contaminant of Protein Y may be responsible for the activity under study, we feel this is very unlikely and our preferred interpretation remains that Protein Y contains a specific protein domain mediating activity.

The Relationship of Observations and Conclusions Made to the Literature

Having made conclusions, it is now important to relate these conclusions, and the data upon which they have been made, to those of earlier workers. Notably, 'earlier workers' may include the authors themselves, in which case it is reasonable to alert readers to the possibility of some interpretive bias, by using expressions such as 'we earlier reported', or 'our previous study showed'.

The relationship of current work to previous literature has different forms, and it is helpful to be systematic in this part of the analysis.



How The Current Observations and Conclusions Add to Understanding

The way in which the current work adds meaningfully to knowledge must be explicitly spelt out. Without this, the actual value of the work remains unclear.

There may be some slight overlap with what is written in the Introduction, where what is currently known and the purpose of the current study is outlined. However, this should be minimized as much as possible.

With reference to the example given above, the following statement might be made:

Although earlier studies identified the Protein Y bioactivity (Milstein and Wiffle, 2006, Zimmel 2008), the current work demonstrates specificity in this response, and provides a basis for further structural analysis to define the precise Protein Y subdomain responsible.

Aspects of the Literature Consistent with Observations and Conclusions

Literature that supports the observations and conclusions should be cited. This is usually best achieved with a brief sentence such as:

Our study is consistent with the earlier reports of Protein Y bioactivity (Milstein and Wiffle, 2006, Zimmel 2008), as well as separate structural studies characterizing the specific bioactivity of other proteins (Matthews et al., 1988, Walker and Whitlock 2001, Xie et al. 2014).

Literature Inconsistent with Observations and Conclusions: Recognition and Reconciliation

There is often literature that is in conflict with the observations and or conclusions made, and it is very important to properly identify these conflicts and seek to reconcile them. It is never a good thing, to simply dismiss the work of our predecessors. Not only is it a terrible professional discourtesy, but it is also poor scientific practice to deny the work of others for the sake of convenience. Instead, conflicts with the literature should be carefully identified, and efforts made to understand and explain why these conflicts have arisen.

There are many reasons why others may have reported observations that conflict with current data. Differences in materials and methods or analytical approaches, may account for differences between current and earlier work.

Also, all interpretation occurs in context of whatever the current paradigms might be. Occasionally, a superficial reading of earlier studies may suggest inconsistency with current work, while a more careful examination of the earlier published data may reveal that the earlier observations had been interpreted in context of a now discarded paradigm, but are now consistent with current work when viewed from a more current perspective.

Again, with reference to the example given with our fictional 'Protein Y', the following might appear in a Discussion:

Contrasting with current results, are two earlier studies reporting activity in Protein P as opposed to Protein Y (Miller 1970, Miller and Weintraub 1972). This may be due to the use of rabbit serum as a source for both Proteins P and Y in these early studies (Miller 1970, Miller and Weintraub 1972), as opposed to our use of human Protein Y in the current work. Also, we have used a more sensitive cell line for our bioassay than was available at the time the earlier work was performed (Miller 1970, Miller and Weintraub 1972, Glick 2002). It is interesting that there is high sequence identity Between Proteins Y and P in rabbits, which is not seen in other mammalian species (Schmitz et al 2012), while no activity is reported for human Protein P (Bagroot 2010). This suggests the activity under study has been transferred from Protein Y to Protein P during the evolution of rabbits, but not in humans, accounting for the difference between our observations, and those of earlier workers (Miller 1970, Miller and Weintraub 1972).



Future Work

Journal publications rarely include discussion of future work, but examiners do require students to indicate how they see work progressing, based on their current observations.

Any discussion of future work should commence by addressing any limitations in the current study, as identified and discussed earlier in the *Discussion*. With reference to the examples given above, the following might appear in the *Discussion*:

It would be of interest to use a Protein Y affinity binding strategy, to explore the formal possibility that yeast produce a contaminant with the activity, and which also binds Protein Y. Also, in light of the reported difference between rabbits and other mammals with regard to Protein Y and Protein P activity (Miller 1970, Miller and Weintraub 1972, Schmitz et al. 2012, Bagroot 2010), it would be interesting to repeat the earlier reported rabbit studies (Miller 1970, Miller and Weintraub 1972), using the more recently developed bioassay described in the current investigation (Glick 2002).

Having shown awareness of how the current work could have been improved by further experimentation, it is then important to demonstrate a sense of how work could proceed to further explore the model proposed in the current paper. For example:

Future work will include fragmentation of Protein Y, with separation of fragments and identification of protein domains with activity. This will be coupled with structural analysis of CNBr cleavage sites, to help identify likely active protein domains. Further fragmentation of active isolated Protein Y fragments will identify the smallest active protein domain, which will then be expressed in either yeast or E Coli for site directed mutagenesis analysis. In addition, synthesised active fragments will be used for receptor binding studies, as well as to identify the relevant receptor. Biological activity and in-vivo relevance of the active Protein Y protein domain will be confirmed in animal experiments.