Writing in the Sciences

Unit 5: The original manuscript

Recommended order for writing an original manuscript

1. Tables and Figures
2. Results
3. Methods
4. Introduction
5. Discussion
6. Abstract

Good references

- Clinical Chemistry Guide to Scientific Writing: http://www.aacc.org/publications/clin_chem/ccgsw/Pages/default.aspx#
- Mimi Zeiger. *Essentials of Writing Biomedical Research Papers*, McGraw Hill Professional

Writing in the Sciences

Unit 5.1: Tables and Figures

Tables and Figures are the foundation of your story!

Editors, reviewers, and readers may look first (and maybe only) at titles, abstracts, and tables and figures!

Figures and tables should stand alone and tell a complete story. The reader should not need to refer back to the main text.

Tables and Figures are the story!

"An article about computational science in a scientific publication isn't the scholarship itself, it's merely advertising of the scholarship. The actual scholarship is the complete software development environment and the complete set of instructions which generated the figures."—Jon Claerbout, Stanford

Tips on Tables and Figures

- Use the fewest figures and tables needed to tell the story.
- Do not present the same data in both a figure and a table.

Tables vs. Figures

- Figures
  - Visual impact
  - Show trends and patterns
  - Tell a quick story
  - Tell the whole story
  - Highlight a particular result
- Tables
  - Give precise values
  - Display many values/variables

Table Title

- Identify the specific topic or point of the table.
- Use the same key terms in the table title, the column headings, and the text of the paper
- Keep it brief!
- Example: “Descriptive characteristics of the two treatment groups, means ± SD or N (%)”

Table Footnotes

- **Use superscript symbols to identify footnotes, according to journal guidelines;**
  - A standard series is: *, †,‡,¶,#,**,††, etc.
- **Use footnotes to explain statistically significant differences**
  - E.g., *p<.01 vs. control by ANOVA
- **Use footnotes to explain experimental details or abbreviations**
  - E.g., EDI is the Eating Disorder Inventory (reference)
  - Amenorrhea was defined as 0-3 periods per year

Table Formats

Model your tables from already published tables! Don't re-invent the wheel!!

- **Most journals use three horizontal lines: one above the column headings, one below the column headings, and one below the data**
- **Follow journal guidelines RE:**
  - Roman or Arabic numbers
  - centered or flush left table number, title, column, headings, and data
  - capital letters and italics
  - the placement of footnotes
  - the type of footnote symbols

Example table:

Three horizontal lines

Table 1. Descriptive characteristics of the study groups, means ± SD or N (%).

Characteristic	Bad Witches	Good Witches
N	13	12
Age (yrs)	45 ± 5	36 ± 6*
Female	11 (85%)	10 (83%)
BMI (kg/m^2)	21 ± 6	23 ± 3
Systolic BP (mmHg)	140 ± 10	120 ± 9*
Exercise (min/day)	30 ± 20	60 ± 30*
Employment status		
Unemployed	4 (31%)	0 (0%)
Part time	3 (23%)	4 (33%)
Full time	6 (46%)	8 (66%)
Smoker (yes/no)	6 (50%)	0 (0%)*

*p<.05, ttest or Fisher's exact test, as appropriate.

Example table:

Table 1. Descriptive characteristics of the study groups, means ± SD or N (%).

Characteristic	Bad Witches	Good Witches
N	13	12
Age (yrs)	45 ± 5	36 ± 6*
Female	11 (85%)	10 (83%)
BMI (kg/m^2)	21 ± 6	23 ± 3
Systolic BP (mmHg)	140 ± 10	120 ± 9*
Exercise (min/day)	30 ± 20	60 ± 30*
Employment status		
Unemployed	4 (31%)	0 (0%)
Part time	3 (23%)	4 (33%)
Full time	6 (46%)	8 (66%)
Smoker (yes/no)	6 (50%)	0 (0%)*

*p<.05, ttest or Fisher's exact test, as appropriate.

What not to do!

Remove grid lines!

Table 1. Descriptive characteristics of the study groups, means ± SD or N (%).

Characteristic	Bad Witches	Good Witches
N	13	12
Age (yrs)	45 ± 5	36 ± 6*
Female	11 (85%)	10 (83%)
BMI (kg/m^2)	21 ± 6	23 ± 3
Systolic BP (mmHg)	140 ± 10	120 ± 9*
Exercise (min/day)	30 ± 20	60 ± 30*
Employment status		
Unemployed	4 (31%)	0 (0%)
Part time	3 (23%)	4 (33%)
Full time	6 (46%)	8 (66%)
Smoker (yes/no)	6 (50%)	0 (0%)*

*p<.05, ttest or Fisher's exact test, as appropriate.

What not to do!

Make sure everything lines up and looks professional!

Table 1. Descriptive characteristics of the study groups, means ± SD or N (%).

Characteristic	Bad Witches	Good Witches
N	13	12
age (yrs)	45 ± 5	36 ± 6*
female	11 (85%)	10 (83%)
BMI (kg/m^2)	21 ± 6	23 ± 3
Systolic BP (mmHg)	140 ± 10	120 ± 9*
Exercise (min/day)	30 ± 20	60 ± 30*
Employment status		
Unemployed	4 (31%)	0 (0%)
Part time	3 (23%)	4 (33%)
Full time	6 (46%)	8 (66%)
Smoker (yes/no)	6 (50%)	0 (0%)*

*p<.05, ttest or Fisher's exact test, as appropriate.

What not to do!

Use a reasonable number of significant figures.

Table 1. Descriptive characteristics of the study groups, means ± SD or N (%).

Characteristic	Bad Witches	Good Witches
N	13	12
Age (yrs)	45.076 ± 5.032	36.007 ± 6.032*
Female	11 (85%)	10 (83%)
BMI (kg/m^2)	21.223 ± 6.332	23.331 ± 3.333
Systolic BP (mmHg)	140.23 ± 10.23	120.23 ± 9.23*
Exercise (min/day)	30.244 ± 20.345	60.123 ± 30.32*
Employment status		
Unemployed	4 (31%)	0 (0%)
Part time	3 (23%)	4 (33%)
Full time	6 (46%)	8 (66%)
Smoker (yes/no)	6 (50%)	0 (0%)*

*p<.05, ttest or Fisher's exact test, as appropriate.

What not to do!

Give units!

Table 1. Descriptive characteristics of the study groups, means ± SD or N (%).

Characteristic	Bad Witches	Good Witches
N	13	12
age	45 ± 5	36 ± 6*
female	11 (85%)	10 (83%)
BMI	21 ± 6	23 ± 3
Systolic BP	140 ± 10	120 ± 9*
Exercise	30 ± 20	60 ± 30*
Employment status		
Unemployed	4 (31%)	0 (0%)
Part time	3 (23%)	4 (33%)
Full time	6 (46%)	8 (66%)
Smoking	6 (50%)	0 (0%)*

*p<.05, ttest or Fisher's exact test, as appropriate.

What not to do!

Omit unnecessary columns!

Table 1. Descriptive characteristics overall and by gro (%), and p-values for the comparison between the groups.

Characteristic	Overall	Bad Witches	Good Witches	P-value
N	25	13	12	n/a
Age (yrs)	41 ± 6	45 ± 5	36 ± 6	0.0005
Female	21 (84%)	11 (85%)	10 (83%)	0.80
BMI (kg/m^2)	22 ± 5	21 ± 6	23 ± 3	0.31
Systolic BP (mmHg)	131 ± 12	140 ± 10	120 ± 9	0.0001
Exercise (min/d)	45 ± 40	30 ± 20	60 ± 30	0.0069
Employment status				
Unemployed	4 (16%)	4 (31%)	0 (0%)	0.17
Part time	7 (28%)	3 (23%)	4 (33%)	
Full time	14 (56%)	6 (46%)	8 (66%)	
Smoker (yes/no)	6 (24%)	6 (50%)	0 (0%)	0.01

Types of Figures

1. Primary evidence
    - electron micrographs, gels, photographs, pathology slides, X-rays, etc.
    - indicates data quality
    - “Seeing is believing”
2. Graphs
    - line graphs, bar graphs, scatter plots, histograms, boxplots, etc.
3. Drawings and diagrams
    - illustrate an experimental set-up or work-flow
    - indicate flow of participants
    - illustrate cause and effect relationships or cycles
    - give a hypothetical model
    - represent microscopic particles or microorganisms as cartoons

Figure Legends

**Allows the figure to stand alone.

May contain:

1. Brief title
2. Essential experimental details
3. Definitions of symbols or line/bar patterns
4. Explanation of panels (A,B,C,D, etc.)
5. Statistical information (tests used, p-values)

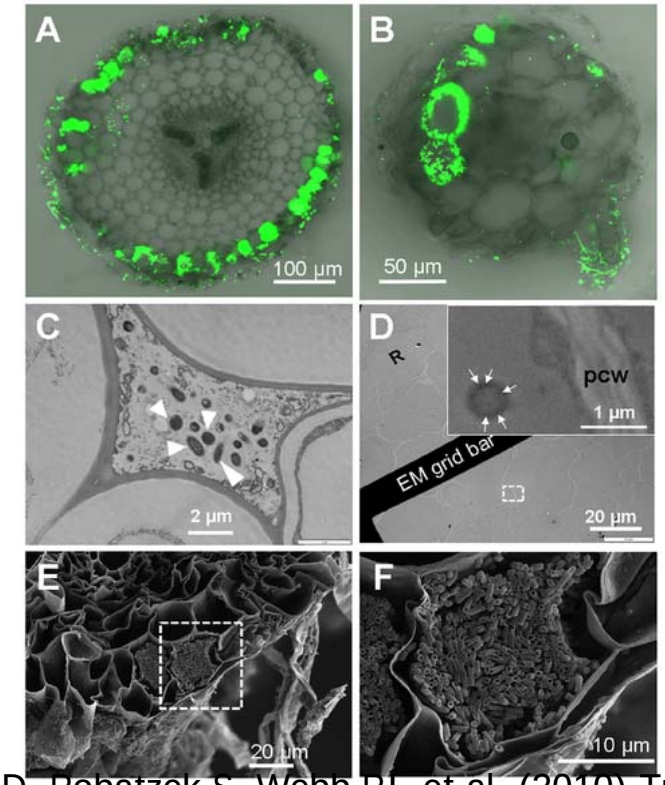
Example Legend

- Figure 2. Root transverse sections and electron micrographs of tomato and Arabidopsis show GFP E. coli in the apoplast and inside root cells. E. coli was detected inside tomato roots (A, C and D, E and F) and Arabidopsis roots (B). (A and B) Fluorescent images of transverse sectioned roots taken by CLSM. (C and D) Images taken by a transmission electron microscope. White triangles in (C) indicate E. coli cell present in apoplast. (D) Roots were probed with immunogold-labeled anti-GFP revealing E. coli in root cortex cells. Sub-image in (D) is a detail of dash-white square box. Gold labeling is marked with white arrows. Rhizodermis cell (R) and plant cell wall (pcw) is indicated. (F) is a detail image of (E) showing plant cells containing E. coli, and both images were taken by SEM.

- Paungfoo-Lonhienne C, Rentsch D, Robatzek S, Webb RI, et al. (2010) Turning the Table: Plants Consume Microbes as a Source of Nutrients. PLoS ONE 5(7): e11915. doi:10.1371/journal.pone.0011915
- http://www.plosone.org/article/info:doi/10.1371/journal.pone.0011915

Primary Evidence

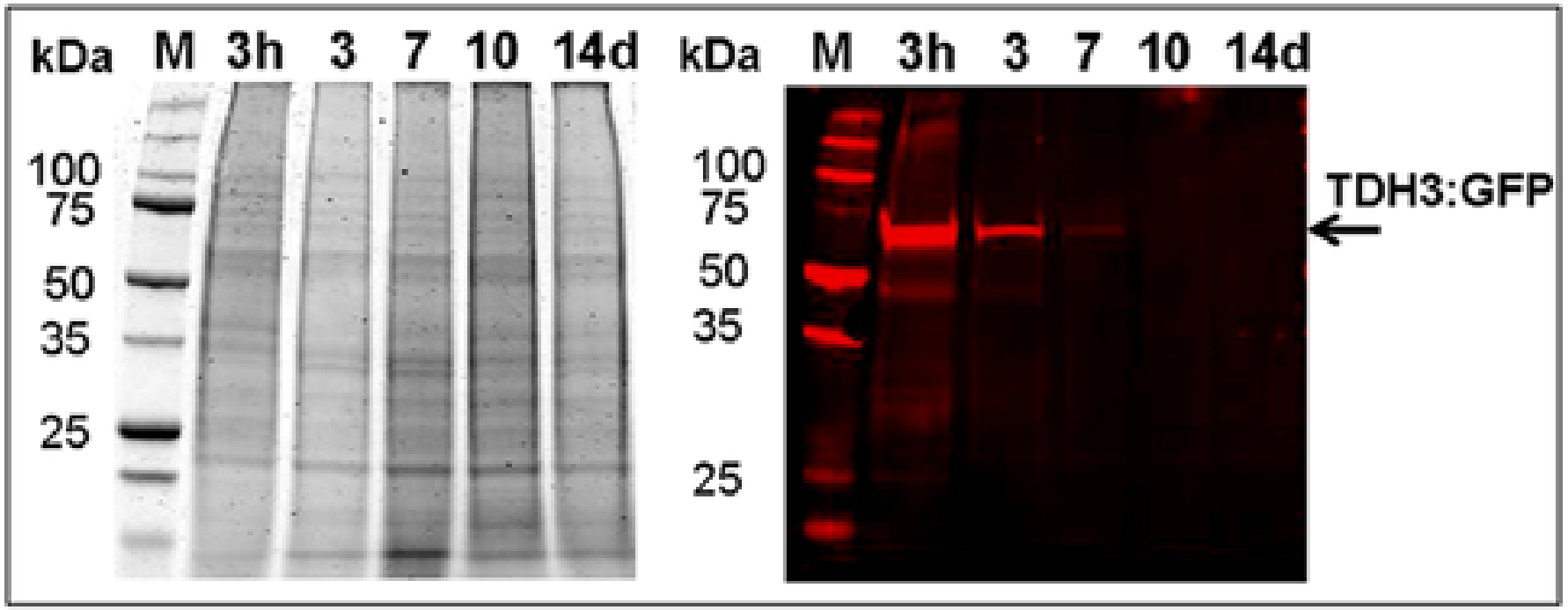
- **Figure 2. Root transverse sections and electron micrographs of tomato and Arabidopsis show GFPE. coli in the apoplast and inside root cells.**



Paungfoo-Lonhienne C, Rentsch D, Robatzek S, Webb RI, et al. (2010) Turning the Table: Plants Consume Microbes as a Source of Nutrients. PLoS ONE 5(7): e11915. doi:10.1371/journal.pone.0011915
http://www.plosone.org/article/info:doi/10.1371/journal.pone.0011915

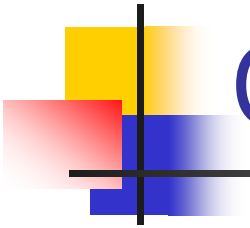
PLoS one

Primary Evidence



Paungfoo-Lonhienne C, Rentsch D, Robatzek S, Webb RI, et al. (2010) Turning the Table: Plants Consume Microbes as a Source of Nutrients. PLoS ONE 5(7): e11915. doi:10.1371/journal.pone.0011915
http://www.plosone.org/article/info:doi/10.1371/journal.pone.0011915

PLoS one



Graphs

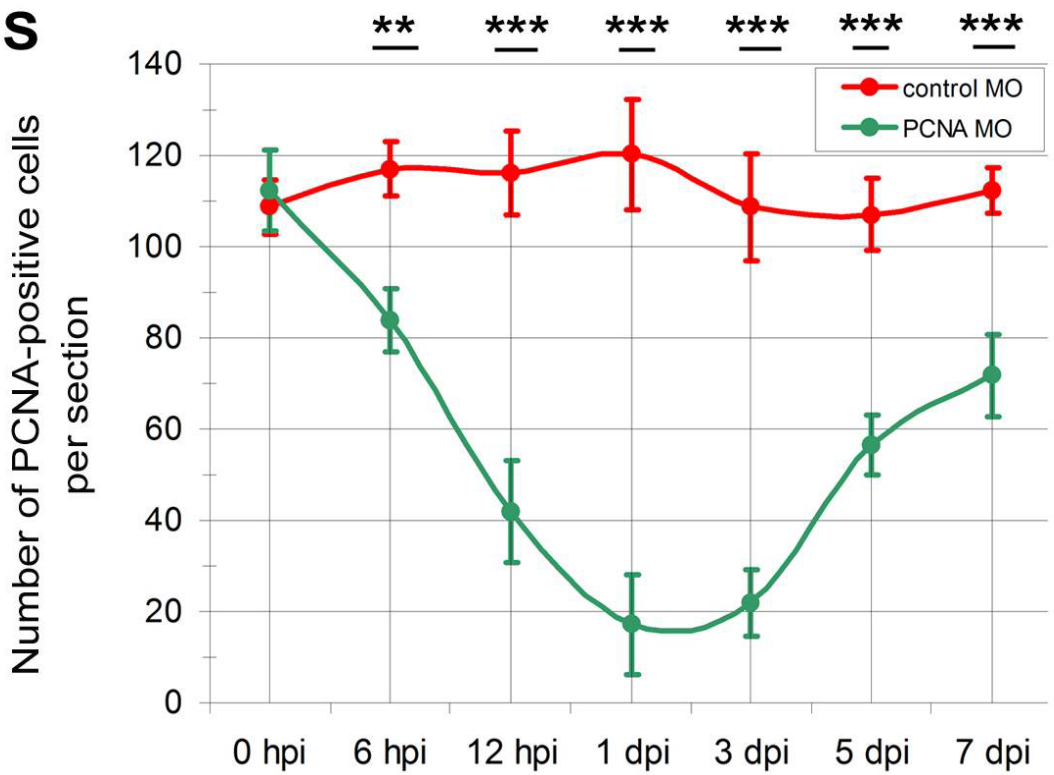
- line graphs
- scatter plots
- bar graphs
- individual-value bar graphs
- histograms
- box plots
- survival curves

Line Graphs

*Used to show trends over time, age, or dose

(can display group means or individuals)

Line graph



Kizil C, Brand M (2011) Cerebroventricular Microinjection (CVMI) into Adult Zebrafish Brain Is an Efficient Misexpression Method for Forebrain Ventricular Cells. PLoS ONE 6(11): e27395. doi:10.1371/journal.pone.0027395
http://www.plosone.org/article/info:doi/10.1371/journal.pone.0027395

PLoS one

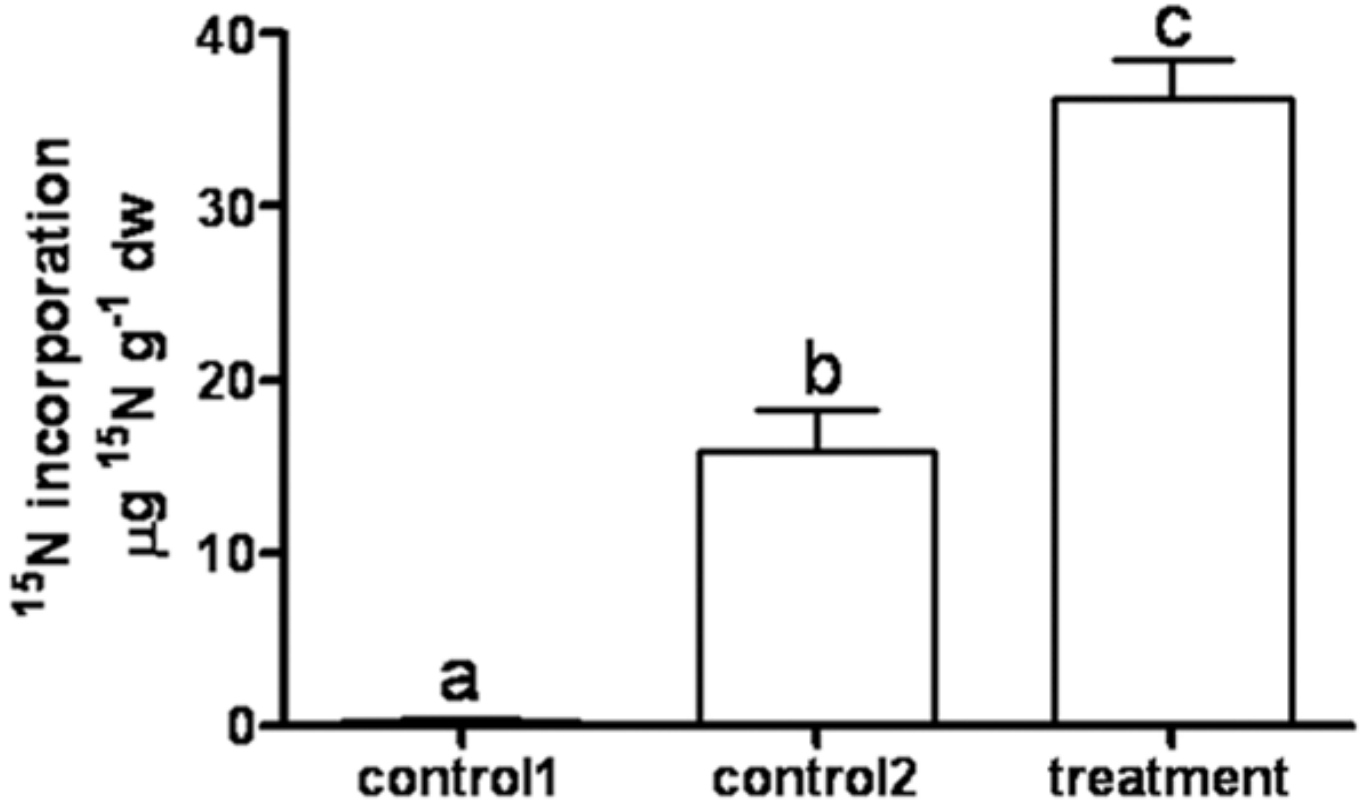
Bar Graphs

*Used to compare groups at one time point

*Tells a quick visual story

Bar graph

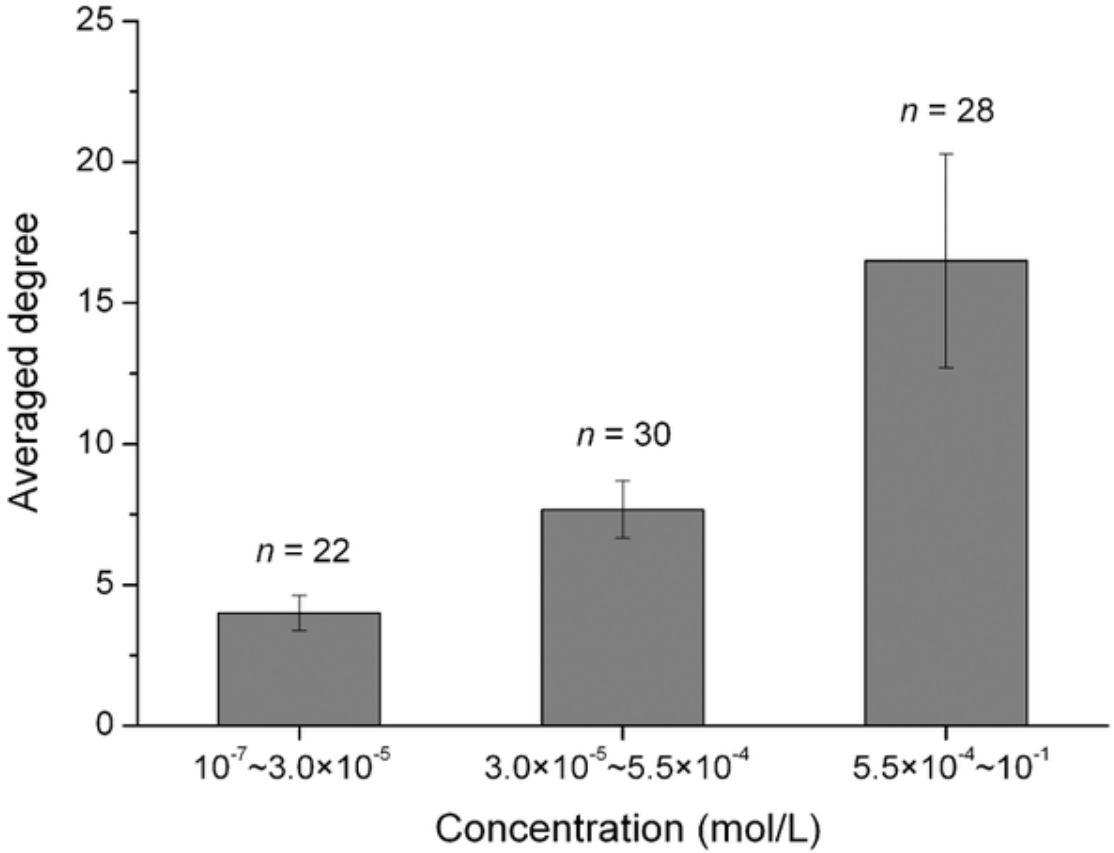
- **Figure 6. Incorporation of E. coli-derived 15N by leaves of tomato plants.**



Paungfoo-Lonhienne C, Rentsch D, Robatzek S, Webb RI, et al. (2010) Turning the Table: Plants Consume Microbes as a Source of Nutrients. PLoS ONE 5(7): e11915. doi:10.1371/journal.pone.0011915

Bar graph

Figure 3. Degree-concentration correlation for E. coli metabolites (P<.01, Kruskal-Wallis test).



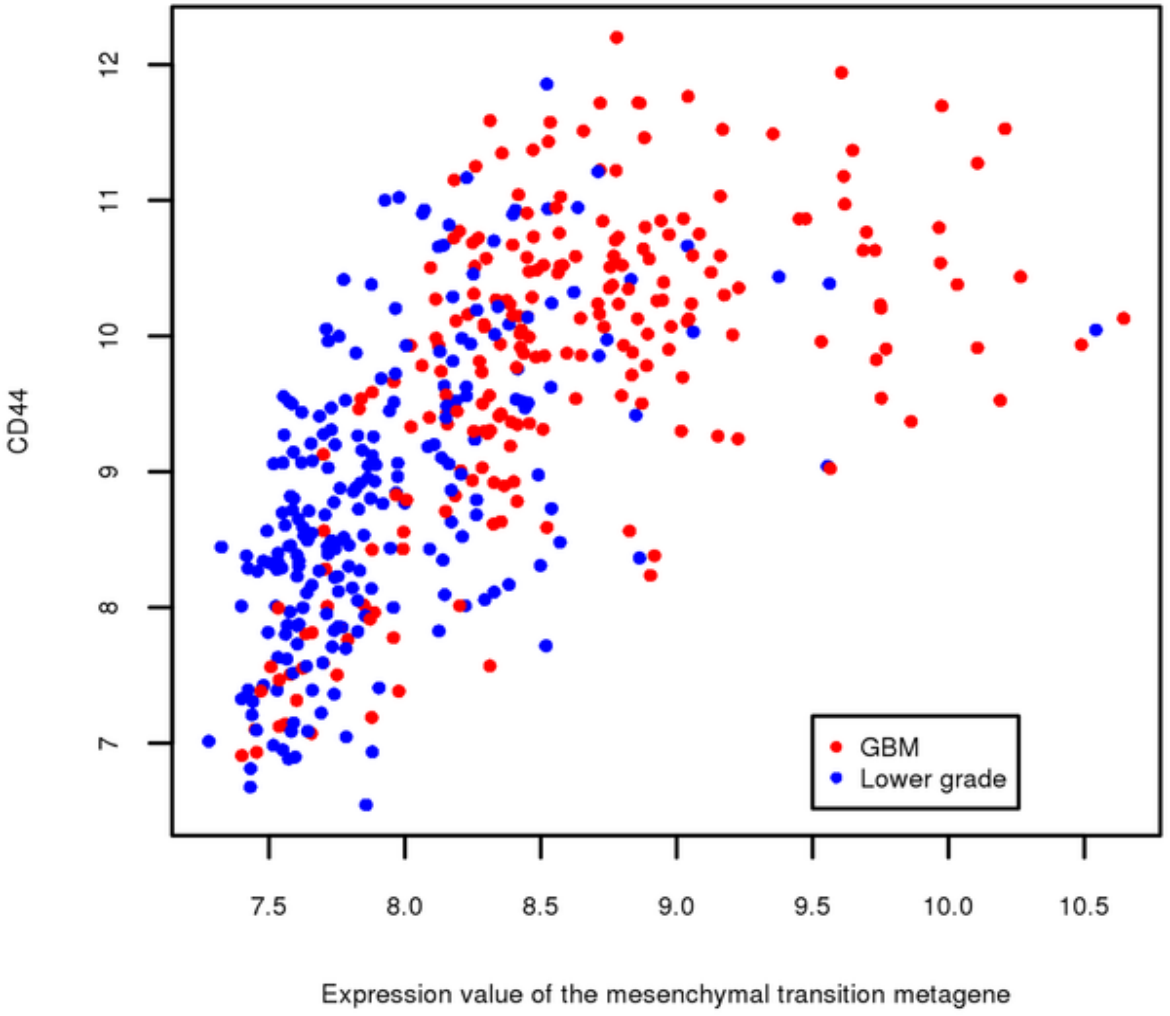
Zhu Q, Qin T, Jiang Y-Y, Ji C, et al. (2011) Chemical Basis of Metabolic Network Organization. PLoS Comput Biol 7(10): e1002214. doi:10.1371/journal.pcbi.1002214
http://www.ploscompbiol.org/article/info:doi/10.1371/journal.pcbi.1002214

Scatter Plots

*Used to show relationships between two variables (particularly linear correlation)

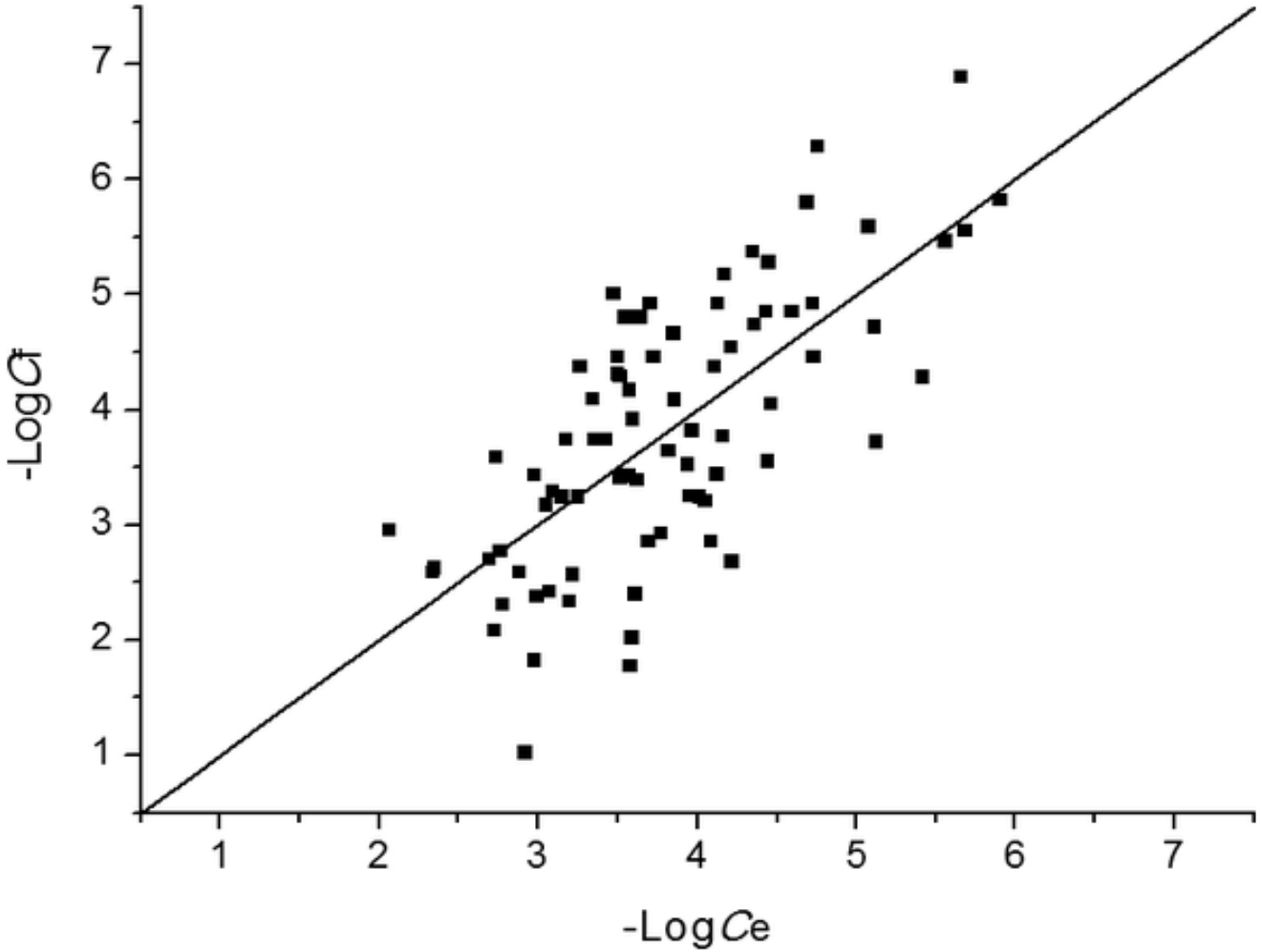
*Allows reader to see individual data points=more information!

Figure 4. Scatter plot for the expression levels of CD44 vs. the mesenchymal transition metagene.



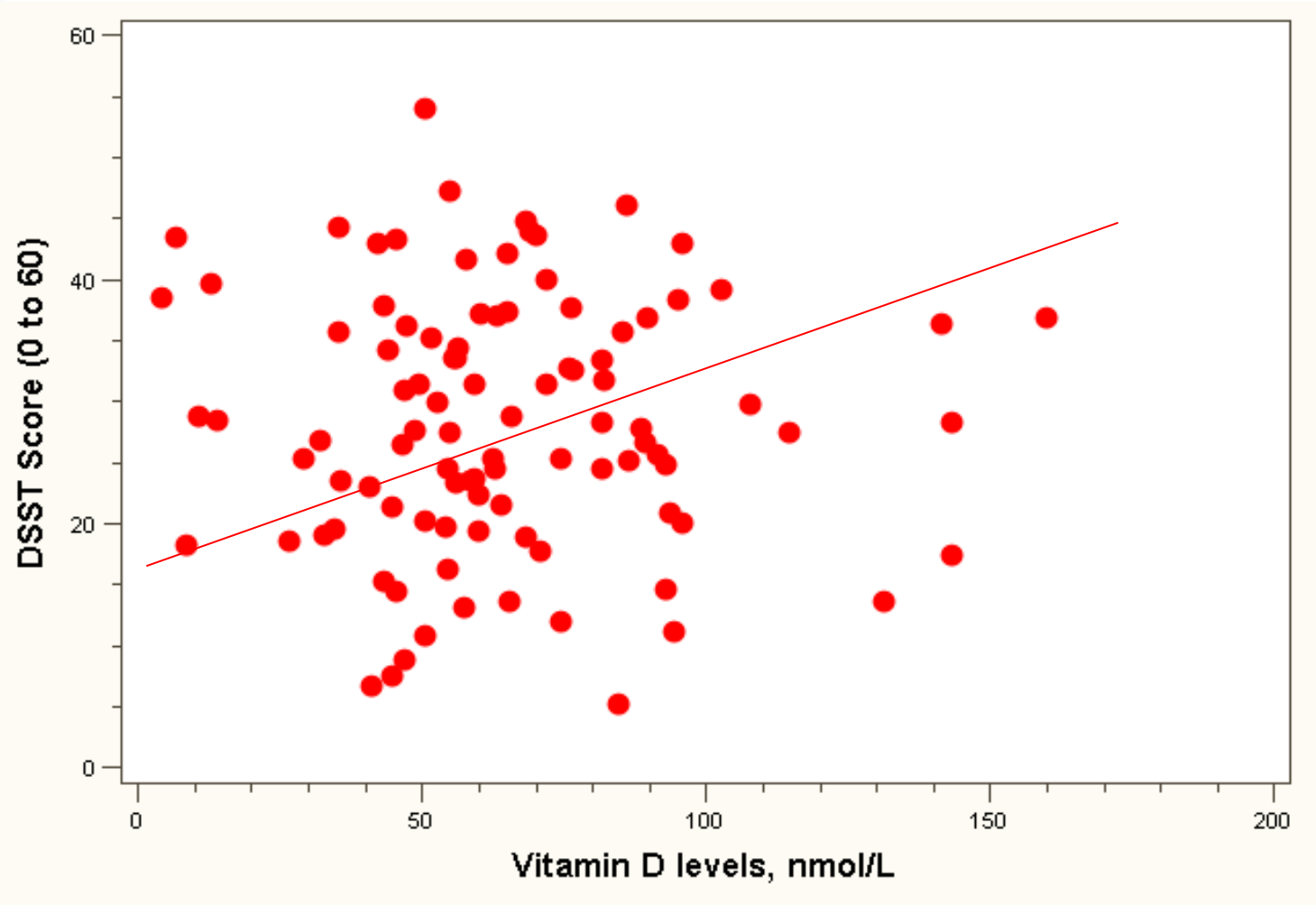
Cheng W-Y, Kandel JJ, Yamashiro DJ, Canoll P, et al. (2012) A Multi-Cancer Mesenchymal Transition Gene Expression Signature Is Associated with Prolonged Time to Recurrence in Glioblastoma. PLoS ONE 7(4): e34705. doi:10.1371/journal.pone.0034705
http://www.plosone.org/article/info:doi/10.1371/journal.pone.0034705

Figure 4. Theoretical fitting of E. coli metabolite concentrations by chemical properties.

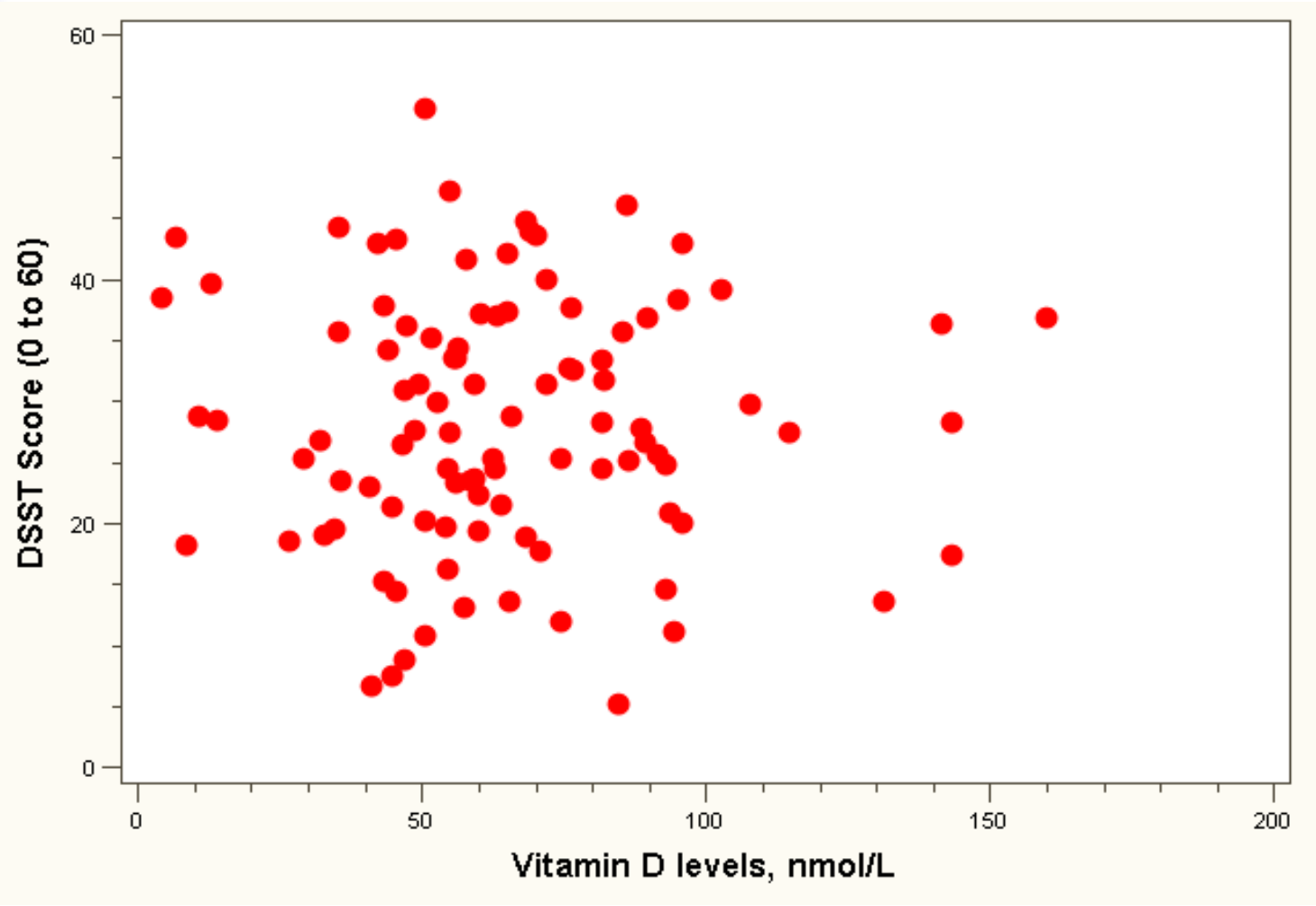


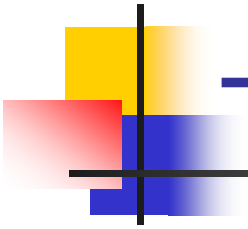
Zhu Q, Qin T, Jiang Y-Y, Ji C, et al. (2011) Chemical Basis of Metabolic Network Organization. PLoS Comput Biol 7(10): e1002214. doi:10.1371/journal.pcbi.1002214
http://www.ploscompbiol.org/article/info:doi/10.1371/journal.pcbi.1002214

Lines can draw your eye!



Lines can draw your eye!

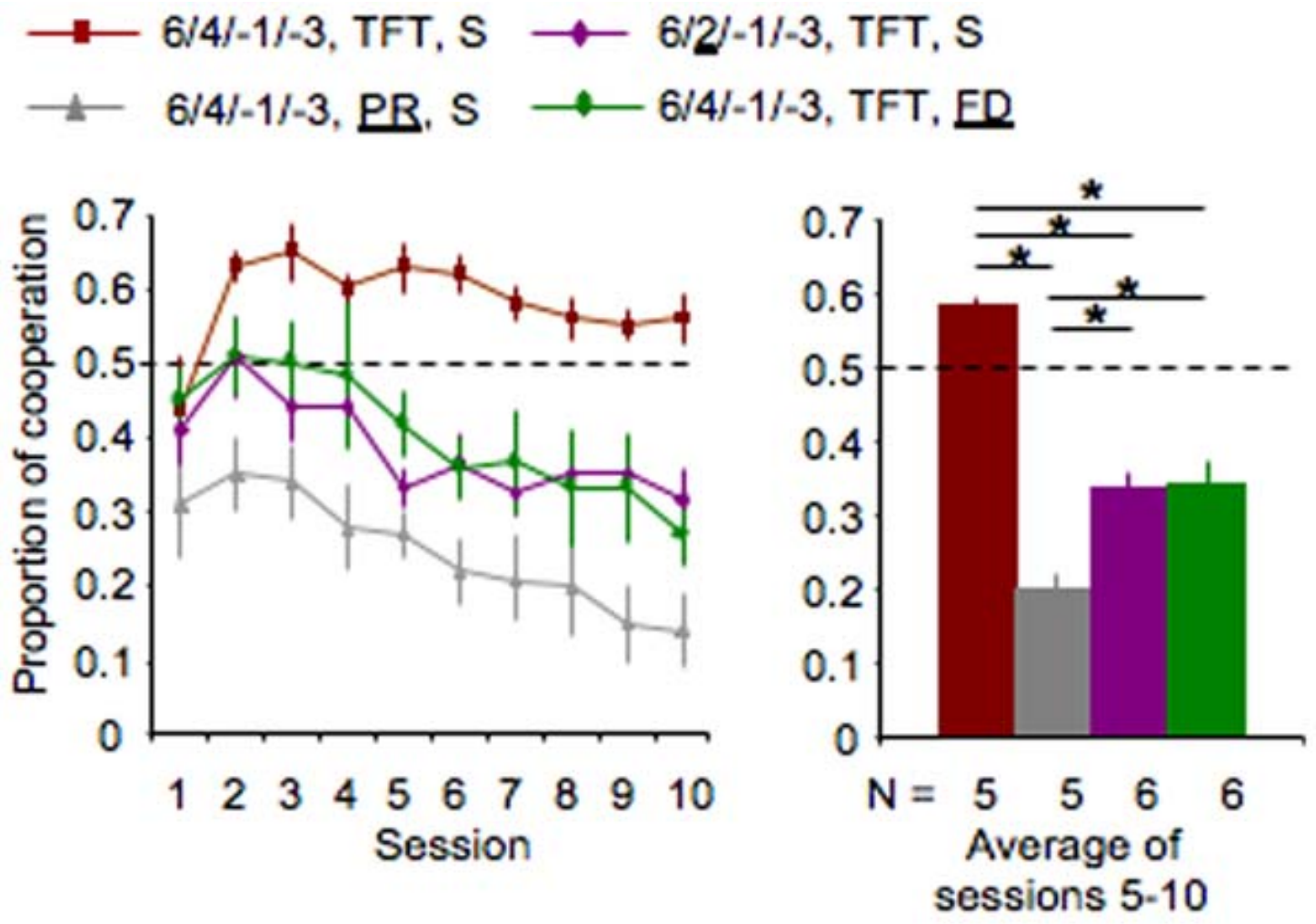




Tips for Graphs

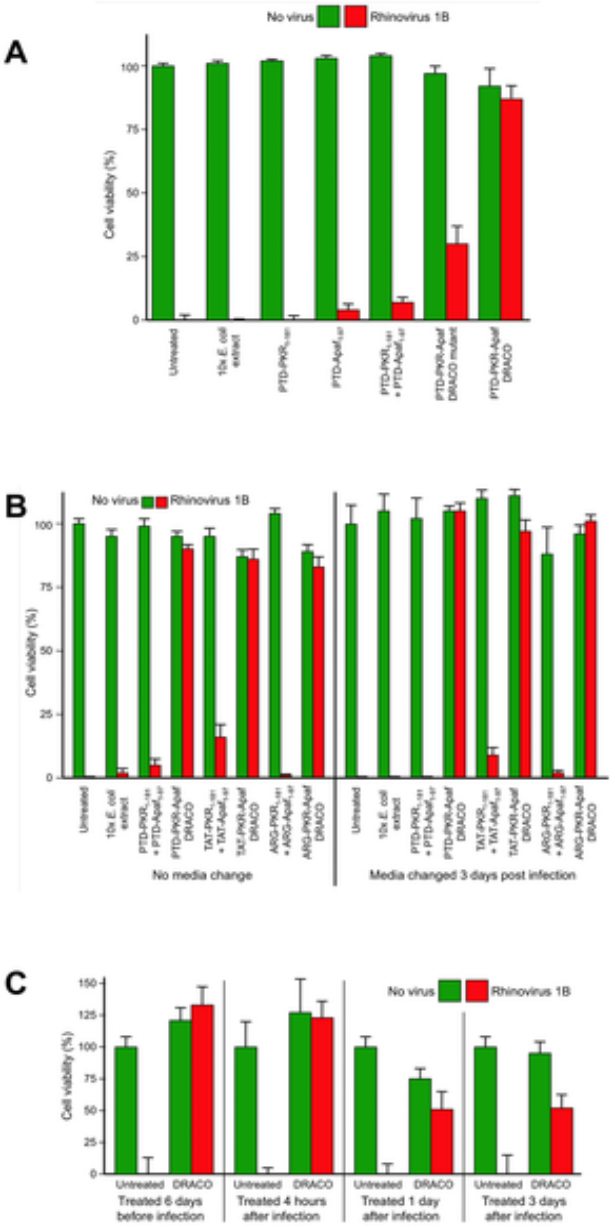
- Tell a quick visual story
- Keep it simple!
- Make it easy to distinguish groups (e.g., triangles vs. circles vs. squares is not easy!)
- If it’s too complex, maybe it belongs in a table

Figure 5. Cooperation levels vary with the different iPD games.



Viana DS, Gordo I, Sucena É, Moita MAP (2010) Cognitive and Motivational Requirements for the Emergence of Cooperation in a Rat Social Game. PLoS ONE 5(1): e8483. doi:10.1371/journal.pone.0008483
http://www.plosone.org/article/info:doi/10.1371/journal.pone.0008483

Figure 4. DRACOs were effective against rhinovirus 1B in NHLF cells.

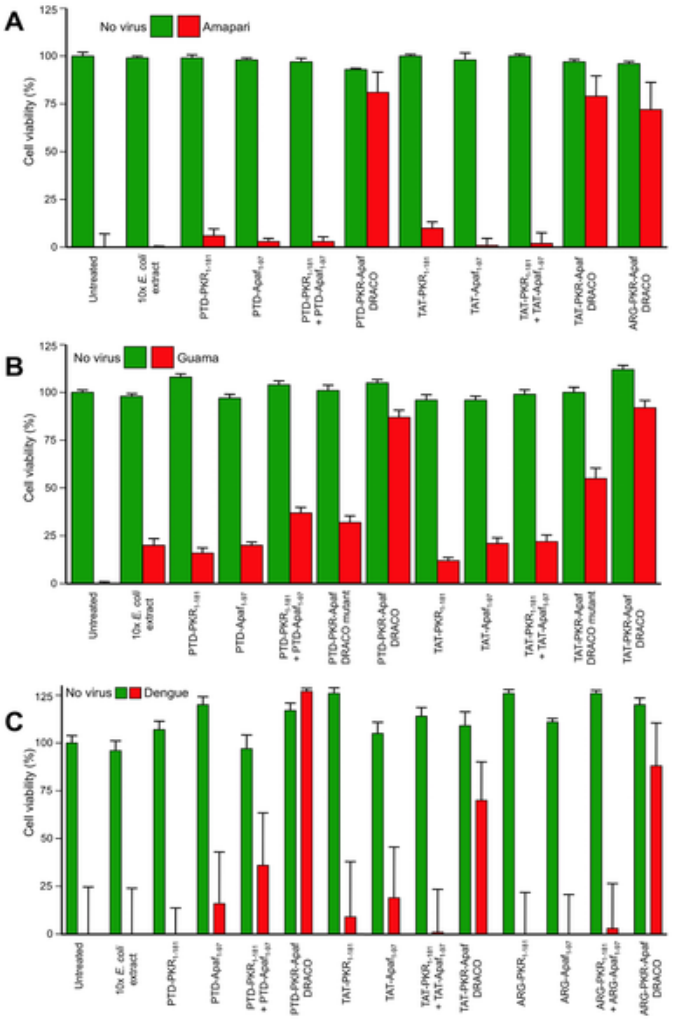


Rider TH, Zook CE, Boettcher TL, Wick ST, et al. (2011) Broad-Spectrum Antiviral Therapeutics. PLoS ONE 6(7): e22572. doi:10.1371/journal.pone.0022572

http://www.plosone.org/article/info:doi/10.1371/journal.pone.0022572

PLoS one

Figure 8. DRACOs were effective against arenaviruses, bunyaviruses, and flaviviruses.

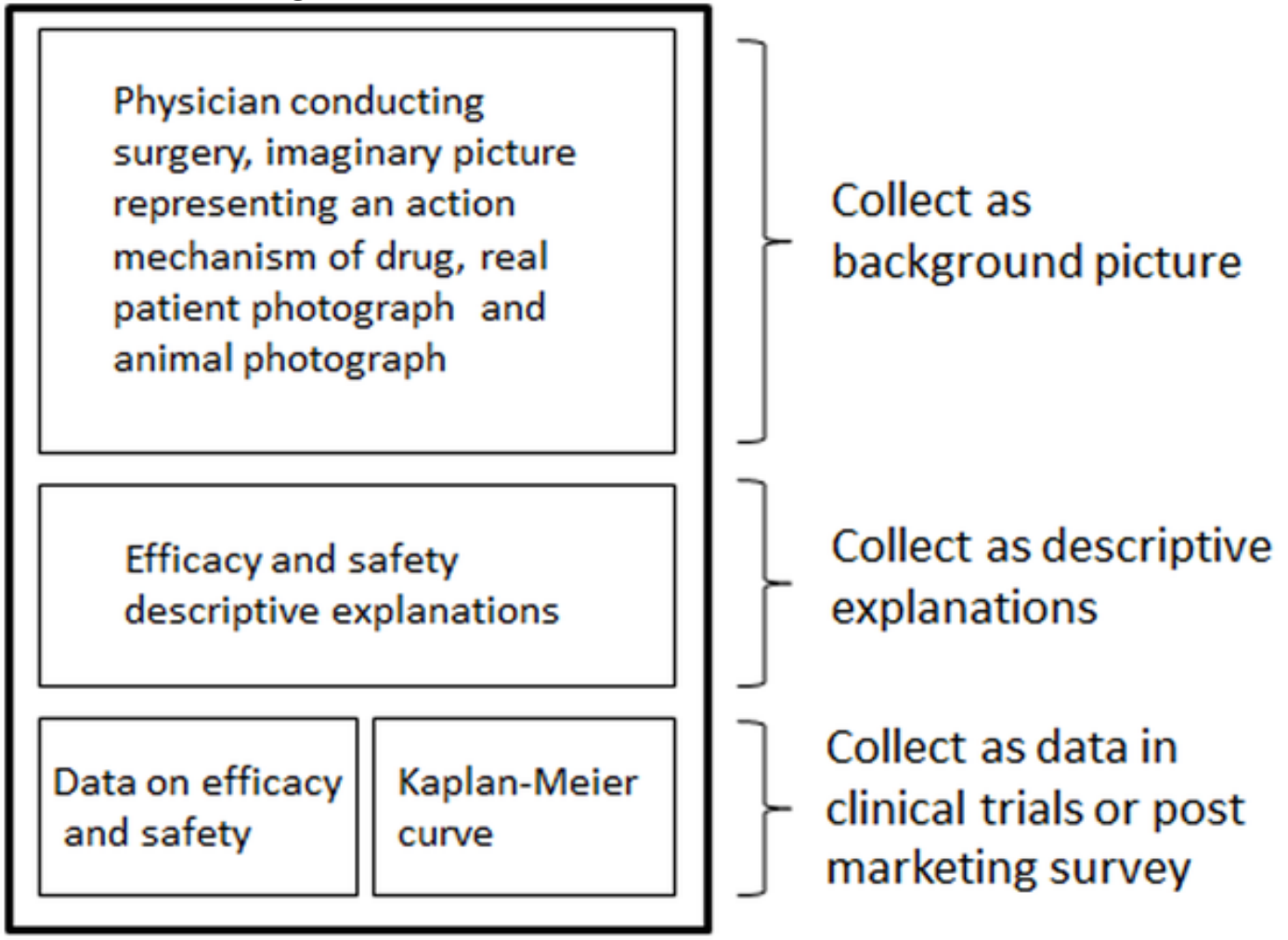


Rider TH, Zook CE, Boettcher TL, Wick ST, et al. (2011) Broad-Spectrum Antiviral Therapeutics. PLoS ONE 6(7): e22572. doi:10.1371/journal.pone.0022572
http://www.plosone.org/article/info:doi/10.1371/journal.pone.0022572

Diagrams and Drawings

- illustrate an experimental set-up or work-flow
- indicate flow of participants
- illustrate cause and effect relationships or cycles
- give a hypothetical model
- represent microscopic particles or microorganisms as cartoons

- **Figure 1. Example of the content of a typical print advertisement for a pharmaceutical drug and the data collection method.**



Yonemori K, Hirakawa A, Ando M, Hirata T, et al. (2012) Content Analysis of Oncology-Related Pharmaceutical Advertising in a Peer-Reviewed Medical Journal. PLoS ONE 7(8): e44393. doi:10.1371/journal.pone.0044393
http://www.plosone.org/article/info:doi/10.1371/journal.pone.0044393

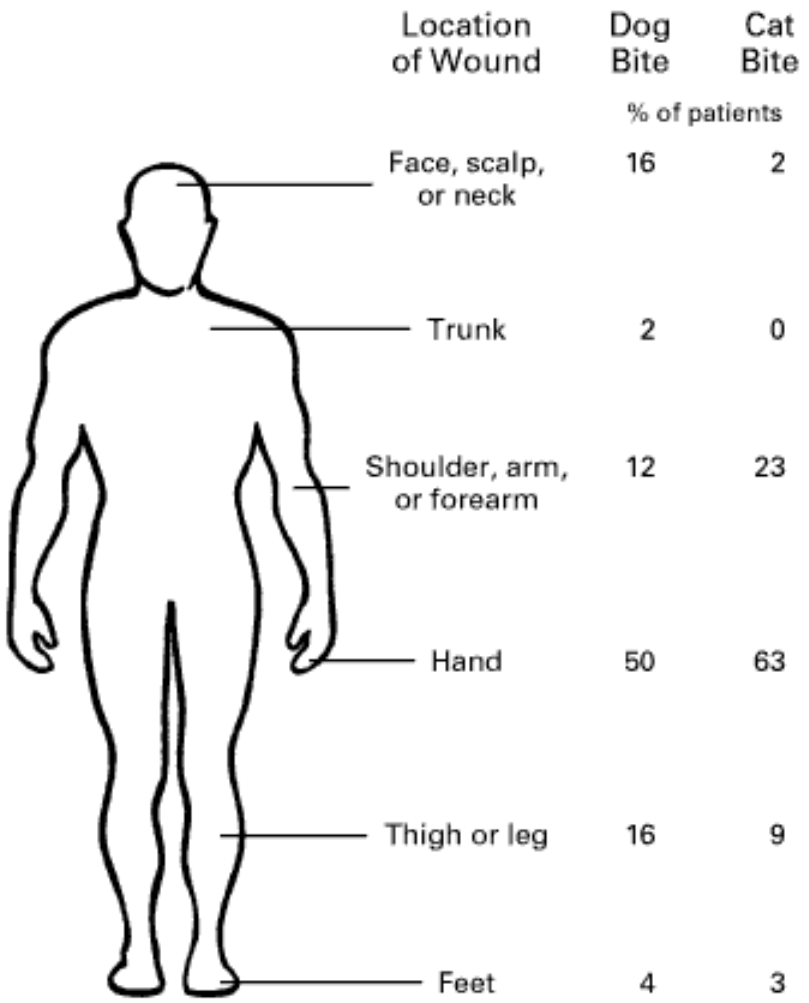
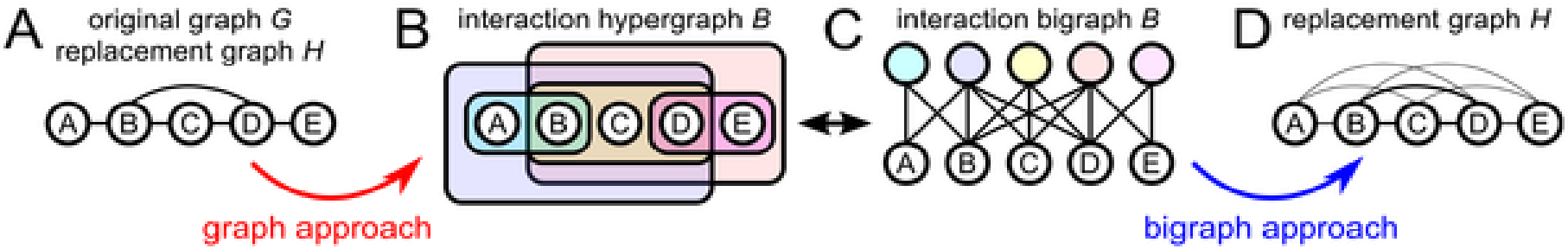


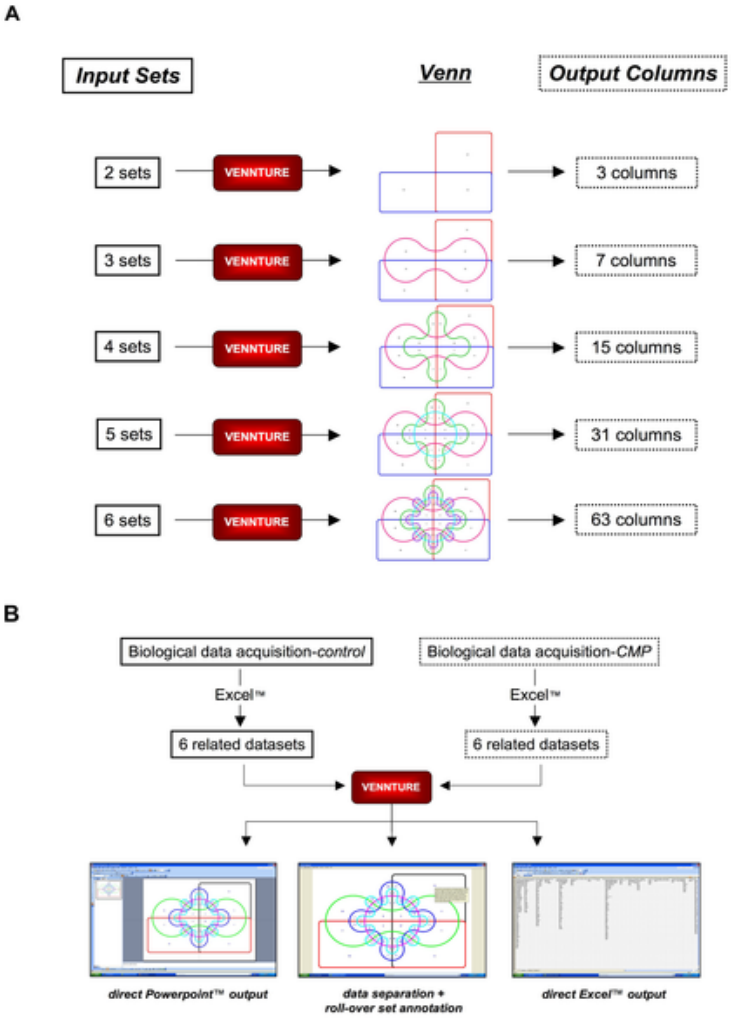
Figure 1. Location of Wound Infections in 50 Patients Bitten by Dogs and 57 Patients Bitten by Cats.

Figure 1. Modeling population structures in evolutionary multiplayer games.



Peña J, Rochat Y (2012) Bipartite Graphs as Models of Population Structures in Evolutionary Multiplayer Games. PLoS ONE 7(9): e44514. doi:10.1371/journal.pone.0044514
http://www.plosone.org/article/info:doi/10.1371/journal.pone.0044514

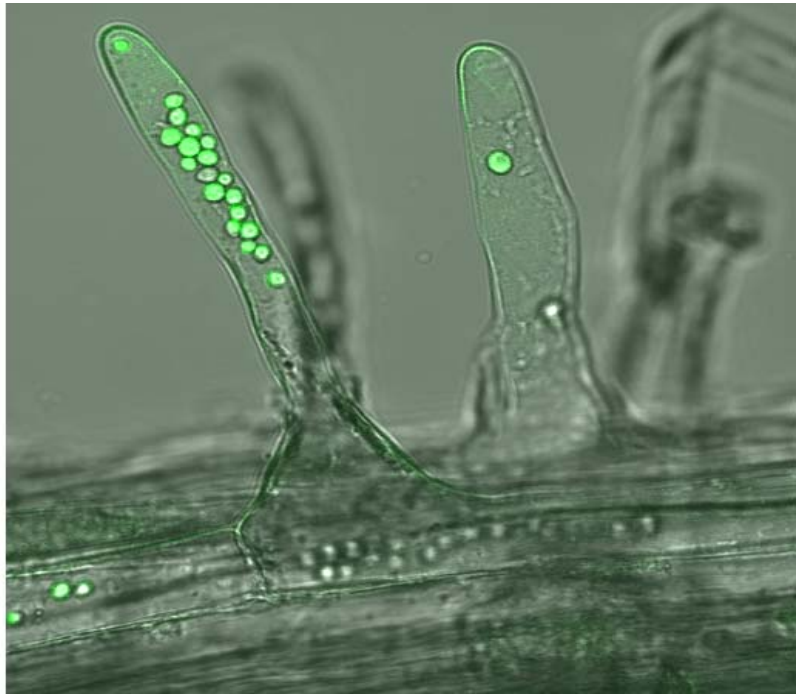
Figure 4. Multiple set VENNTURE data input.



Martin B, Chadwick W, Yi T, Park S-S, et al. (2012) VENNTURE–A Novel Venn Diagram Investigational Tool for Multiple Pharmacological Dataset Analysis. PLoS ONE 7(5): e36911. doi:10.1371/journal.pone.0036911
http://www.plosone.org/article/info:doi/10.1371/journal.pone.0036911

Besides tables and figures... Movies!

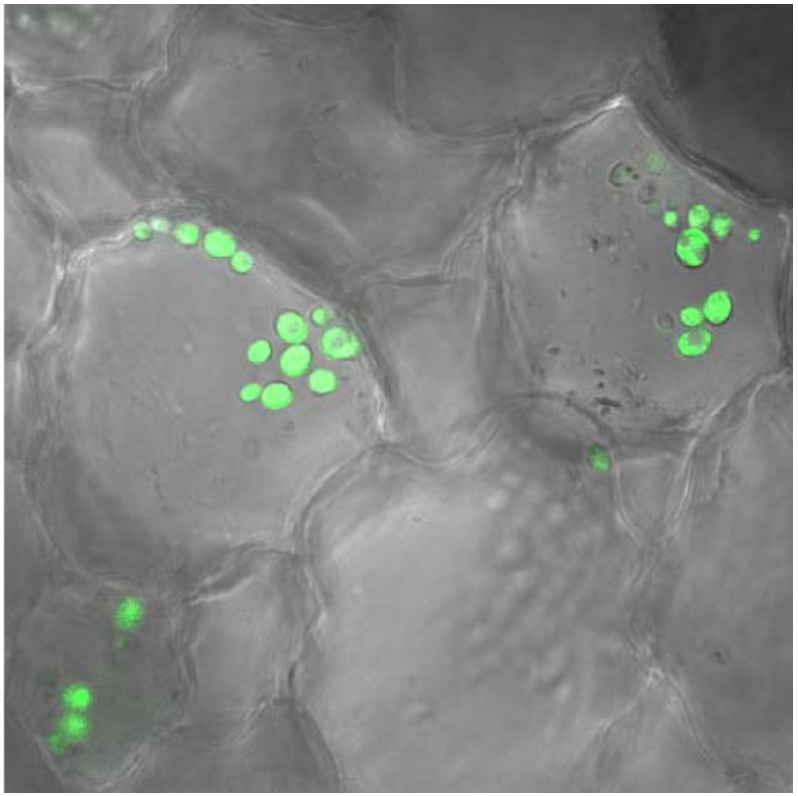
- Allowed as supplemental material.



Paungfoo-Lonhienne C, Rentsch D, Robatzek S, Webb RI, et al. (2010) Turning the Table: Plants Consume Microbes as a Source of Nutrients. PLoS ONE 5(7): e11915. doi:10.1371/journal.pone.0011915
http://www.plosone.org/article/info:doi/10.1371/journal.pone.0011915

PLoS one

Movies!



Paungfoo-Lonhienne C, Rentsch D, Robatzek S, Webb RI, et al. (2010) Turning the Table: Plants Consume Microbes as a Source of Nutrients. PLoS ONE 5(7): e11915. doi:10.1371/journal.pone.0011915
http://www.plosone.org/article/info:doi/10.1371/journal.pone.0011915

PLoS one

Writing in the Sciences

Unit 5.2: Results

Results ≠ Raw Data

- The results section should:
  - *Summarize* what the data show
    - Point out simple relationships
    - Describe big-picture trends
    - Cite figures or tables that present supporting data
  - Avoid simply repeating the numbers that are already available in tables and figures.

Examples

“Over the course of treatment, topiramate was significantly more effective than placebo at improving drinking outcomes on drinks per day, drinks per drinking day, percentage of heavy drinking days, percentage of days abstinent, and log plasma -glutamyl transferase ratio (Table 3).”

“The total suicide rate for Australian men and women did not change between 1991 and 2000 because marked decreases in older men and women (Table 1) were offset by increases in younger adults, especially younger men.[7]”

Arch Intern Med. 2008;168(11):1188-1199;
BMJ. 2003 May 10; 326(7397): 1008.

Hypothetical Example

Table 1. Descriptive characteristics of the study groups, means ± SD or N (%).

Characteristic	Bad Witches	Good Witches
N	13	12
Age (yrs)	45 ± 5	36 ± 6*
Female	11 (85%)	10 (83%)
BMI (kg/m²)	21 ± 6	23 ± 3
Systolic BP (mmHg)	140 ± 10	120 ± 9*
Exercise (min/day)	30 ± 20	60 ± 30*
Employment status		
Unemployed	4 (31%)	0 (0%)
Part time	3 (23%)	4 (33%)
Full time	6 (46%)	8 (66%)
Smoker (yes/no)	6 (50%)	0 (0%)*

*p<.05, ttest or Fisher's exact test, as appropriate.

The characteristics of the bad witches and the good witches are shown in Table 1. There was a significant difference in age between the groups. The mean age of the bad witches was 45 ± 5; and the mean age of the good witches was 36 ± 6. There was no significant difference in gender between the groups, with the bad witches having 85% females and the good witches having 83% females. BMI was not significantly different between the groups, which both had normal BMIs. Systolic blood pressure and exercise were significantly different. The bad witches had a mean blood pressure of 140 ± 10, whereas the good witches had a mean blood pressure of 120 ± 9. Estimated daily exercise was higher in the good witches (60 ± 30) than the bad witches (30 ± 20). Employment was not significantly different between the two groups…

Edited version…

Original:

The characteristics of the bad witches and the good witches are shown in Table 1. There was a significant difference in age between the groups. The mean age of the bad witches was 45 ± 5; and the mean age of the good witches was 36 ± 6. There was no significant difference in gender between the groups, with the bad witches having 85% females and the good witches having 83% females. BMI was not significantly different between the groups, which both had normal BMIs. Systolic blood pressure and exercise were significantly different. The bad witches had a mean blood pressure of 140 ± 10, whereas the good witches had a mean blood pressure of 120 ± 9. Estimated daily exercise was higher in the good witches (60 ± 30) than the bad witches (30 ± 20). Employment was not significantly different between the two groups…

Revised:

The witches were, on average, lean and predominantly female (Table 1). Bad witches were significantly older, had higher blood pressures, exercised less, and were more likely to smoke than good witches. More bad witches were unemployed, but this difference did not reach statistical significance.

Tips for writing Results

- **Break into subsections, with headings (if needed)**
- **Complement the information that is already in tables and figures**
  - **Give precise values that are not available in the figure**
  - **Report the percent change or percent difference if absolute values are given in the table**
- **Repeat / highlight only the most important numbers**

Tips for writing Results

- **Don’t forget to talk about negative and control results**
- **Reserve the term “significant” for statistically significant**
- **Reserve information about what you did for the methods section**
  - **In particular, do not discuss the rationale for statistical analyses within the Results section.**
- **Reserve comments on the meaning of your results for the discussion section**

Example...

TABLE 2. Summary of running during pregnancy and breastfeeding.

Running during pregnancy and breastfeeding	Mean± SD or Percent (n)
Ran ever during pregnancy	77 (70.0%)
Ran during the first trimester	69 (62.7%)
Ran during the second trimester	57 (51.8%)
Ran during the third trimester	34 (30.9%)
Running during pregnancy (n=77):	
Average weekly mileage	20.3 ± 9.3
Average running intensity (percent of normal)	47.9% ± 21.0%
Sustained a running injury	3 (3.9%)

The majority of runners ran during pregnancy (70.0%, 77/110), with 62.7% running during the first trimester, 51.8% during the second trimester, and fewer than one third (30.9%) during the third trimester (Table 2). From the 77 women who ran during pregnancy, we observed the average weekly mileage during pregnancy for those who ran to be 20.3 ± 9.3 miles. Average running intensity was reported to be 47.9% ± 21.0% as a percent of non-pregnant running effort. A small number (3.9%, 3/77) reported sustaining a running injury while pregnant.

Edited version...

Original:

The majority of runners ran during pregnancy (70.0%, 77/110), with 62.7% running during the first trimester, 51.8% during the second trimester, and fewer than one third (30.9%) during the third trimester (Table 2). From the 77 women who ran during pregnancy, we observed the average weekly mileage during pregnancy for those who ran to be 20.3 ± 9.3 miles. Average running intensity was reported to be 47.9% ± 21.0% as a percent of non-pregnant running effort. A small number (3.9%, 3/77) reported sustaining a running injury while pregnant.

Edited:

Seventy percent of runners (n=77) ran sometime during pregnancy, and almost a third ran through the third trimester (Table 2). On average, women who ran during pregnancy greatly curtailed their training—cutting their weekly mileage and running intensity to about half of pre-pregnancy efforts. Only 3 (3.9%) reported sustaining a running injury while pregnant.

What verb tense do I use?

***Use past tense for completed actions:**

We found that…
Women were more likely to…
Men smoked more cigarettes than…
The average reaction time was…

***Use the present tense for assertions that continue to be true, such as what the tables show, what you believe, and what the data suggest:**

Figure 1 shows…
The findings confirm…
The data suggest…
We believe that this shows…

Example: verb tense

Example:

Information was available for 7766 current cigarette smokers. Of these, 1216 (16%) were classified as hardcore smokers. Table 1 gives characteristics of all the smokers. The most striking difference was that hardcore smokers were about 10 years older on average and tended to be more dependent on tobacco. Significantly more hardcore smokers had manual occupations, lived in rented accommodation, and had completed their full time education by the age of 16 years. There was no difference by sex.

Jarvis et al. Prevalence of hardcore smoking in England, and associated attitudes and beliefs: cross sectional study *BMJ* 2003;326:1061 (17 May)

Use the active voice!

- **More lively!**
- **Since you can talk about the subjects of your experiments, “we” can be used sparingly while maintaining the active voice!**

Use the active voice!

Comparison with Californian estimates

Using the same definition of hardcore smoking as adopted in the Californian study, we found a prevalence of 17% across all age groups and 19% among smokers aged 26 compared with a figure of 5% for this group in the US study. When we added the Californian requirement of 15 cigarettes a day to our criteria we found a prevalence of 10% among smokers aged 26, still twice the prevalence in California

Jarvis et al. Prevalence of hardcore smoking in England, and associated attitudes and beliefs: cross sectional study *BMJ* 2003;326:1061 (17 May)

Example Continued…

Differences in attitudes and beliefs by level of dependence

To test whether it was appropriate to exclude a measure of cigarette dependence from our criteria for defining hardcore smoking, we compared attitudes and beliefs by dependence in hardcore and other smokers (table 4). For most items, beliefs were similar in low and high dependence hardcore smokers but strikingly different from those of other smokers. For example, almost 60% of both low and high dependency non-hardcore smokers agreed that improved health would be a major benefit from quitting whereas among hardcore smokers only 27% of low dependency and 32% of high dependency smokers agreed. Similar differentiation in beliefs by hardcore smoking status, but not dependence level, emerged for other items, especially those related to health

Writing in the Sciences

Unit 5.3: Methods

Methods and Materials

- Give a clear overview of what was done
- Give enough information to replicate the study (like a recipe!)
- Be complete, but make life easy for your reader!

1. Break into smaller sections with subheadings
2. Cite a reference for commonly used methods
3. Display in a flow diagram or table where possible

- You *may* use jargon and the passive voice more liberally in the methods section

Who, what, when, where, how, and why...

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?

Reprinted, with permission, from: Annesley TM. Who, what, when, where, how, and why: The ingredients in the recipe for a successful methods section. *Clinical Chemistry*. June 2010 vol. 56 no. 6, 897-901.

Materials and Methods

- **Materials**
  - **Drugs, buffers, chemicals, gases, reagents, cell lines, etc.**
- **Participants/subjects**
  - **Animals (state that the research was approved by the appropriate committee at your institution)**
  - **Humans (state that the research was approved by the appropriate committee at your institution)**
- **Experimental protocol/study design**
- **Measurements**
  - **How were the dependent and independent variables measured**
    - **Instruments (telescope, microscope, weighing scale, questionnaire, etc.)**
- **Analyses**

Make life easy for your reader!

1. Break into sub-sections with informative subheadings

Example subheadings

METHODS

- **General Approach**
- **Biosafety**
- **Isolation of Virus**
- **Serologic Analysis**
- **Pathological and Immunohistochemical Studies**
- **Molecular Analyses**

e.g., Ksiazek et al. A Novel Coronavirus Associated with Severe Acute Respiratory Syndrome *NEJM* 348:1953-1966, May 15, 2003

Example subheadings

METHODS

- **Subjects and experimental protocols**
- **Hardware**
- **GPS data processing**
- **Wind**

Sachs G, Traugott J, Nesterova AP, Dell'Omo G, et al. (2012) Flying at No Mechanical Energy Cost: Disclosing the Secret of Wandering Albatrosses. PLoS ONE 7(9): e41449. doi:10.1371/journal.pone.0041449
http://www.plosone.org/article/info:doi/10.1371/journal.pone.0041449

Example subheadings

MATERIALS AND METHODS

- **Cell culture and transfections**
- **Antibodies**
- **Plasmids**
- **Recombinant virus production and infection**
- **Metabolic labeling and immunoprecipitation**
- **Immunoblotting**
- **Subcellular fractionation**
- **Electron microscopy**

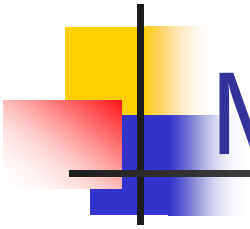
The Journal of Cell Biology, Volume 161, Number 3, 521-533
Proprotein convertase cleavage liberates a fibrillogenic fragment of a resident glycoprotein to initiate melanosome biogenesis
Joanne F. Berson1, Alexander C. Theos1, Dawn C. Harper1, Danielle Tenza2, Graça Raposo2 and Michael S. Marks1

Example subheadings

MATERIALS AND METHODS

- **Materials**
- **Antibodies**
- **Plasmids**
- **Recombinant virus production and infection**
- **Metabolic labeling and immunoprecipitation**
- **Immunoblotting**
- **Subcellular fractionation**
- **Electron microscopy**

The Journal of Cell Biology, Volume 161, Number 3, 521-533
Proprotein convertase cleavage liberates a fibrillogenic fragment of a resident glycoprotein to initiate melanosome biogenesis
Joanne F. Berson1, Alexander C. Theos1, Dawn C. Harper1, Danielle Tenza2, Graça Raposo2 and Michael S. Marks1



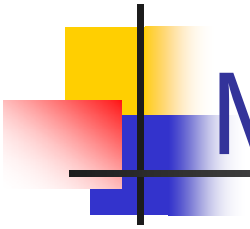
Make life easy for your reader!

2. Cite a reference for commonly used methods or previously used methods rather than explaining all the details...

Cite commonly/previously used methods

Each peptide was covalently coupled to agarose (AminoLink Kit, Pierce Chemical), and 30-to-200-ml quantities of each crude polyclonal antiserum were affinity-purified with the use of the appropriate immobilized peptide, ***as previously described.***[13]

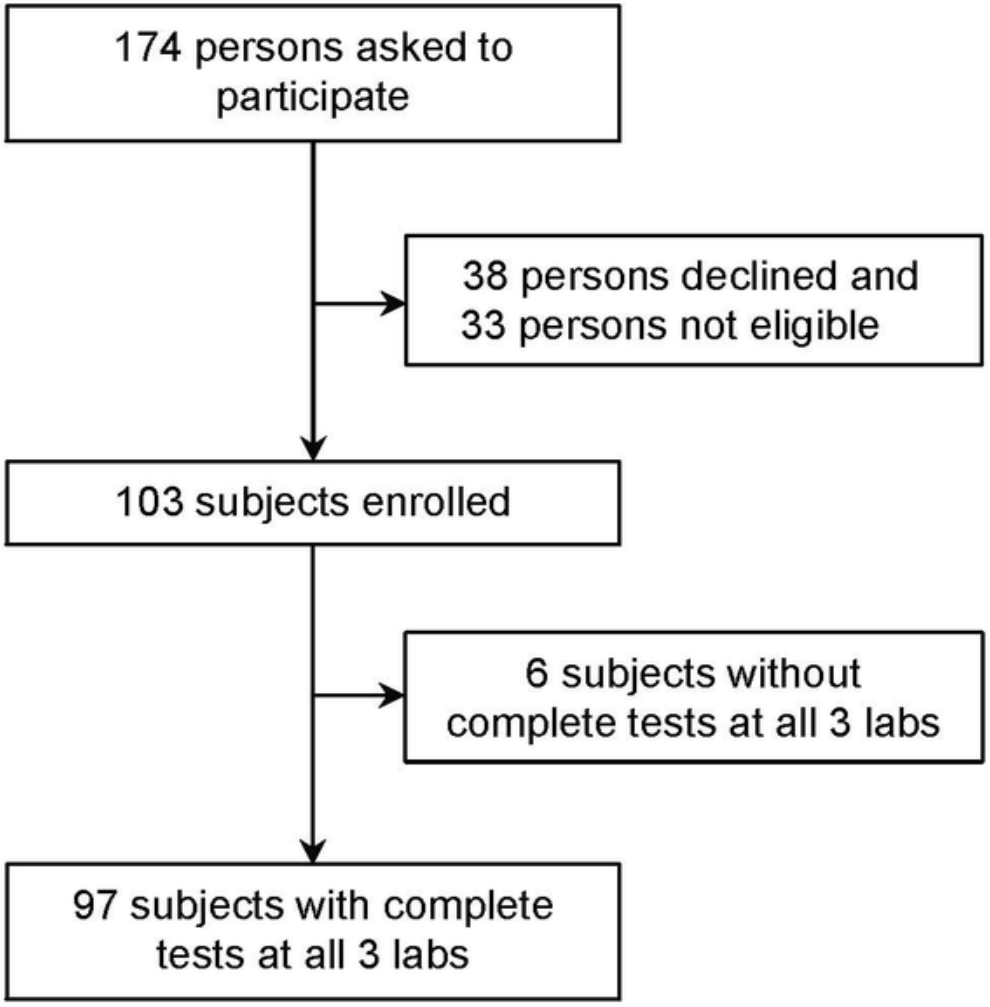
Immunoprecipitations, SDS-PAGE on 10% polyacrylamide gels, and phosphorimaging analysis were performed **as described previously** (Berson et al., 2000).



Make life easy for your reader!

3. Use flow diagrams or tables to help simplify explanations of methods!

Figure 1. Study participation diagram.



Whitworth WC, Hamilton LR, Goodwin DJ, Barrera C, et al. (2012) Within-Subject Interlaboratory Variability of QuantiFERON-TB Gold In-Tube Tests. PLoS ONE 7(9): e43790. doi:10.1371/journal.pone.0043790
http://www.plosone.org/article/info:doi/10.1371/journal.pone.0043790

Verb tense

Report methods in past tense (“we measured”),

But use present tense to describe how data are presented in the paper (“data are summarized as means ± SD”)

It’s OK to use passive voice (or even to use a combination)!

Passive:

E.g., Oral temperatures were measured.

Emphasizes the method or variable.

Active:

E.g., We measured oral temperatures

More lively, but sacrifices having the material/method/variable as the subject of the sentence

Requires creativity to avoid starting every sentence with We!

Passive voice and jargon are OK!

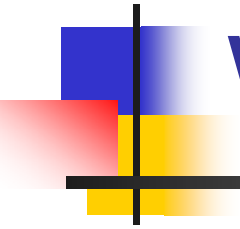
For sequencing, *amplicons* were purified with *ExoSAP-Codes*. The partial nucleotide sequences of the polymerase gene were aligned with published coronavirus sequences, using *CLUSTAL W for Unix (version 1.7)*.

From: Ksiazek et al. A Novel Coronavirus Associated with Severe Acute Respiratory Syndrome *NEJM* 348:1953-1966, May 15, 2003

Passive voice and jargon are OK!

Peptides were synthesized by the Biopolymer Core Facility, Massachusetts General Hospital, Boston. Peptides representing portions of the FGF-23 precursor — [Cys70]FGF-23(51–69)amide, [Tyr185] FGF-23(186–206)amide, [Tyr223]FGF-23(206–222)amide, and [Tyr224]FGF-23(225–244)amide — were coupled to keyhole limpet hemocyanin, emulsified with complete Freund's adjuvant, and used for subcutaneous immunization of eight goats (with approximately 100 µg per animal); each…

From Jonsson et al. Fibroblast Growth Factor 23 in Oncogenic Osteomalacia and X-Linked Hypophosphatemia NEJM 348:1656-1663; April 24, 2003.



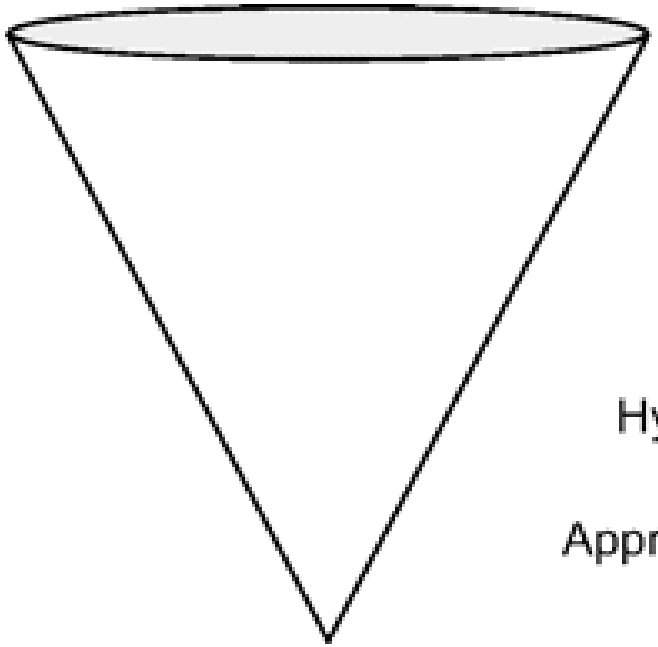
Writing in the Sciences

Unit 5.4: Introduction

Introduction

- Good News: The introduction is easier to write than you may realize!
- Follows a fairly standard format
- Typically 3 paragraphs long
  - Recommended range: 2 to 5
- It is **not** an exhaustive review of your general topic
  - should focus on the specific hypothesis/aim of your study

Introduction



Background, known information

Knowledge gap, unknown information

Hypothesis, question, purpose statement

Approach, plan of attack, proposed solution

Reproduced with permission from: Annesley TM. "It was a cold and rainy night." Set the scene with a good introduction. *Clinical Chemistry*. May 2010 56: 708-713. (Figure 1)

Introduction

1. What's known
2. What's unknown
   - limitations and gaps in previous studies
3. Your burning question/hypothesis/aim
4. Your experimental approach
5. Why your experimental approach is new and different and important (fills in the gaps)

Mimi Zeiger. *Essentials of Writing Biomedical Research Papers*, McGraw Hill Professional, 1999

Corresponds to roughly 3 paragraphs...

1. What's known

≈ Paragraph 1

2. What's unknown
   - limitations and gaps in previous studies

≈ Paragraph 2

3. Your burning question
4. Your experimental approach
5. Why your experimental approach is new and different and important (fills in the gaps)

≈ Paragraph 3

Tips for writing an Introduction

- Keep paragraphs short
- Write for a general audience
  - clear, concise, non-technical
- Take the reader step by step from what is known to what is unknown. End with your specific question.
  - Known→Unknown→Question/hypothesis
- Emphasize how your study fills in the gaps (the unknown)
- Explicitly state your research question/aim/hypothesis:
  - "We asked whether"; "Our hypothesis was"; "We tested the hypothesis that"; "Our aim/s were"
- Do not answer the research question (no results or implications).
- Summarize at a high level! Leave detailed descriptions, speculations, and criticisms of particular studies for the discussion.

Introduction, Example

Exposures to secondhand tobacco smoke, road vehicle traffic, and diet are some of the most prevalent modifiable risk factors for asthma in children. The effect of parental smoking on wheezing illness and diagnosed asthma in children is well established (1, 2), but evidence that these outcomes are more common in children living close to a main road (3–5)has not been confirmed in all studies (6, 7). Several dietary factors have been linked to asthma (8), and one of the most consistent observations is of an inverse association with fruit intake (9–13).

The National Schools Fruit Scheme is a government initiative that aims to provide each child aged 4–6 years with free fruit in school every day by winter 2004. As part of an evaluation of the health benefits of this scheme, we have taken the opportunity to investigate the relative importance of fruit intake, exposure to secondhand smoke, and road vehicle traffic in determining the prevalence of asthma in over 11,000 children.

Lewis SA, Antoniak M, Venn AJ, Davies L,Goodwin A, Salfield N, Britton J, Fogarty AW. Secondhand Lewis SA et al. Smoke, Dietary Fruit Intake, Road Traffic Exposures, and the Prevalence of Asthma: A Cross-Sectional Study in Young Children. *Am. J. Epidemiol* 2005; 161: 406-411.

Introduction, Example

What's known

Exposures to secondhand tobacco smoke, road vehicle traffic, and diet are some of the most prevalent modifiable risk factors for asthma in children. The effect of parental smoking on wheezing illness and diagnosed asthma in children is well established (1, 2), but evidence that these outcomes are more common in children living close to a main road (3–5)has not been confirmed in all studies (6, 7). Several dietary factors have been linked to asthma (8), and one of the most consistent observations is of an inverse association with fruit intake (9–13).

The National Schools Fruit Scheme is a government initiative that aims to provide each child aged 4–6 years with free fruit in school every day by winter 2004. As part of an evaluation of the health benefits of this scheme, we have taken the opportunity to investigate the relative importance of fruit intake, exposure to secondhand smoke, and road vehicle traffic in determining the prevalence of asthma in over 11,000 children.

Lewis SA, Antoniak M, Venn AJ, Davies L,Goodwin A, Salfield N, Britton J, Fogarty AW. Secondhand Lewis SA et al. Smoke, Dietary Fruit Intake, Road Traffic Exposures, and the Prevalence of Asthma: A Cross-Sectional Study in Young Children. *Am. J. Epidemiol* 2005; 161: 406-411.

Introduction, Example

What's unknown

Exposures to secondhand tobacco smoke, road vehicle traffic, and diet are some of the most prevalent modifiable risk factors for asthma in children. The effect of parental smoking on wheezing illness and diagnosed asthma in children is well established (1, 2), but **evidence that these outcomes are more common in children living close to a main road (3–5)has not been confirmed in all studies (6, 7). Several dietary factors have been linked to asthma** (8), and one of the most consistent observations is of an inverse association with fruit intake **(9–13).**

The National Schools Fruit Scheme is a government initiative that aims to provide each child aged 4–6 years with free fruit in school every day by winter 2004. As part of an evaluation of the health benefits of this scheme, we have taken the opportunity to investigate the relative importance of fruit intake, exposure to secondhand smoke, and road vehicle traffic in determining the prevalence of asthma in over 11,000 children.

Lewis SA, Antoniak M, Venn AJ, Davies L,Goodwin A, Salfield N, Britton J, Fogarty AW. Secondhand Lewis SA et al. Smoke, Dietary Fruit Intake, Road Traffic Exposures, and the Prevalence of Asthma: A Cross-Sectional Study in Young Children. *Am. J. Epidemiol* 2005; 161: 406-411.

Introduction, Example

What's known

Exposures to secondhand tobacco smoke, road vehicle traffic, and diet are some of the most prevalent modifiable risk factors for asthma in children. The effect of parental smoking on wheezing illness and diagnosed asthma in children is well established (1, 2), but evidence that these outcomes are more common in children living close to a main road (3–5)has not been confirmed in all studies (6, 7). Several dietary factors have been linked to asthma (**8**), and **one of the most consistent observations is of an inverse association with fruit intake (9–13).**

The National Schools Fruit Scheme is a government initiative that aims to provide each child aged 4–6 years with free fruit in school every day by winter 2004. As part of an evaluation of the health benefits of this scheme, we have taken the opportunity to investigate the relative importance of fruit intake, exposure to secondhand smoke, and road vehicle traffic in determining the prevalence of asthma in over 11,000 children.

Lewis SA, Antoniak M, Venn AJ, Davies L,Goodwin A, Salfield N, Britton J, Fogarty AW. Secondhand Lewis SA et al. Smoke, Dietary Fruit Intake, Road Traffic Exposures, and the Prevalence of Asthma: A Cross-Sectional Study in Young Children. *Am. J. Epidemiol* 2005; 161: 406-411.

Introduction, Example

Our question/aim

Exposures to secondhand tobacco smoke, road vehicle traffic, and diet are some of the most prevalent modifiable risk factors for asthma in children. The effect of parental smoking on wheezing illness and diagnosed asthma in children is well established (1, 2), but evidence that these outcomes are more common in children living close to a main road (3–5)has not been confirmed in all studies (6, 7). Several dietary factors have been linked to asthma (8), and one of the most consistent observations is of an inverse association with fruit intake (9–13).

The National Schools Fruit Scheme is a government initiative that aims to provide each child aged 4–6 years with free fruit in school every day by winter 2004. **As part of an evaluation of the health benefits of this scheme, we have taken the opportunity to investigate the relative importance of fruit intake, exposure to secondhand smoke, and road vehicle traffic in determining the prevalence of asthma in over 11,000 children.**

Lewis SA, Antoniak M, Venn AJ, Davies L,Goodwin A, Salfield N, Britton J, Fogarty AW. Secondhand Lewis SA et al. Smoke, Dietary Fruit Intake, Road Traffic Exposures, and the Prevalence of Asthma: A Cross-Sectional Study in Young Children. *Am. J. Epidemiol* 2005; 161: 406-411.

Introduction. Example

Would also like to know: how is this study going to do better than previous studies?

Exposure [illegible] some of the most prevalent modifiable risk factors for asthma in children. The effect of parental smoking on wheezing illness and diagnosed asthma in children is well established (1, 2), but evidence that these outcomes are more common in children living close to a main road (3–5)has not been confirmed in all studies (6, 7). Several dietary factors have been linked to asthma (8), and one of the most consistent observations is of an inverse association with fruit intake (9–13).

The National Schools Fruit Scheme is a government initiative that aims to provide each child aged 4–6 years with free fruit in school every day by winter 2004. As part of an evaluation of the health benefits of this scheme, we have taken the opportunity to investigate the relative importance of fruit intake, exposure to secondhand smoke, and road vehicle traffic in determining the prevalence of asthma in over 11,000 children.

Lewis SA, Antoniak M, Venn AJ, Davies L,Goodwin A, Salfield N, Britton J, Fogarty AW. Secondhand Lewis SA et al. Smoke, Dietary Fruit Intake, Road Traffic Exposures, and the Prevalence of Asthma: A Cross-Sectional Study in Young Children. *Am. J. Epidemiol* 2005; 161: 406-411.

Introduction, Example

The relations between excess body weight and mortality, not only from all causes but also from cardiovascular disease, are well established.[1,2,3,4,5,6] Although we have known for some time that excess weight is also an important factor in death from cancer,[7] our knowledge of the magnitude of the relation, both for all cancers and for cancers at individual sites, and the public health effect of excess weight in terms of total mortality from cancer is limited. Previous studies have consistently shown associations between adiposity and increased risk of cancers of the endometrium, kidney, gallbladder (in women), breast (in postmenopausal women), and colon (particularly in men).[8,9,10,11,12] Adenocarcinoma of the esophagus has been linked to obesity.[11,13,14] Data on cancers of the pancreas, prostate, liver, cervix, and ovary and on hematopoietic cancers are scarce or inconsistent.[7,8,9,10,11,15,16,17] The lack of consistency may be attributable to the limited number of studies (especially those with prospective cohorts), the limited range and variable categorization of overweight and obesity among studies, bias introduced by reverse causality with respect to smoking-related cancers, and possibly real differences between the effects of overweight and obesity on the incidence of cancer and on the rates of death from some cancers.[18,19]

We conducted a prospective investigation in a large cohort of U.S. men and women to determine the relations between body-mass index (the weight in kilograms divided by the square of the height in meters) and the risk of death from cancer at specific sites. This cohort has been used previously to examine the association of body-mass index and death from any cause.[5]

Eugenia E. Calle, Ph.D., Carmen Rodriguez, M.D., M.P.H., Kimberly Walker-Thurmond, B.A., and Michael J. Thun, M.D. Overweight, Obesity, and Mortality from Cancer in a Prospectively Studied Cohort of U.S. Adults. N Engl J Med 2003; 348:1625-1638.

What's known

What's unknown

The relations between excess body weight and mortality, not only from all causes but also from cardiovascular disease, are well established.[1,2,3,4,5,6] Although we have known for some time that excess weight is also an important factor in death from cancer,[7] our knowledge of the magnitude of the relation, both for all cancers and for cancers at individual sites, and the public health effect of excess weight in terms of total mortality from cancer is limited. Previous studies have consistently shown associations between adiposity and increased [illegible]e endometrium, kidney, gallbladder (in women), breast (in [illegible]omen), and colon (particularly in men).[8,9,10,11,12] [illegible] the esophagus has been linked to obesity.[11,13,14] Data on [illegible]reas, prostate, liver, cervix, and ovary and on hematopoietic [illegible]or inconsistent.[7,8,9,10,11,15,16,17] The lack of consistency may be [illegible]imited number of studies (especially those with prospective [illegible]l range and variable categorization of overweight and obesity [illegible]s introduced by reverse causality with respect to smoking-related cancers, and possibly real differences between the effects of overweight and obesity on the incidence of cancer and on the rates of death from some cancers.[18,19]

"This study will answer the question with better methods."

We conducted a prospective investigation in a large cohort of U.S. men and women to determine the relations between body-mass index (the weight in kilograms divided by the square of the height in meters) and the risk of death from cancer at specific sites. This cohort has been used previously to examine the association of body-mass index and death fro[illegible]

Gaps/limitations of previous studies

Introduction, Example

One of the most intriguing findings in systems biology is that despite the varied constituents and metabolic pathways of three domains of life, their metabolic networks exhibit the same scale-free organization. That is, a small part of metabolites participate in a large number of reactions (which are also termed hubs), while others are involved in a few reactions [1]. As the scale-free architectures are robust and error-tolerant, this finding provides meaningful insights into the design principle of metabolic networks.

The scale-free organization of metabolic networks has been hypothetically explained in terms of evolution that the new-recruited metabolite members attach preferentially to those that are already well connected (rich get richer, also known as preferential attachment principle) [2]–[4]. This implies that the metabolic network hubs originated relatively earlier than others in evolutionary history [5]. However, several issues about this evolutionary explanation remain elusive. First, the molecular basis of preferential attachment principle has not been fully elucidated, as it is inexplicable how the new metabolites “know” which metabolites are well connected. Second, the evolutionary explanation to the metabolic network organization has little implications for network design, because we do not know how to choose metabolites as hubs to construct a new metabolic network. Since most metabolites are small molecules and metabolic processes are basically chemical reactions, we speculate that the metabolic network organization may have a chemical basis, which stimulated our interest to address these issues by combining bioinformatics and chemoinformatics. The latter is a discipline devoted to encoding, storing, managing, searching and analyzing all kinds of chemical data by information technology [6], [7].

Zhu Q, Qin T, Jiang Y-Y, Ji C, et al. (2011) Chemical Basis of Metabolic Network Organization. PLoS Comput Biol 7(10): e1002214.

Background/What's known

One of the most intriguing findings in systems biology is that despite the varied constituents and metabolic pathways of three domains of life, their metabolic networks exhibit the same scale-free organization. That is, a small part of metabolites participate in a large number of reactions (which are also termed hubs), while others are involved in a few reactions [1]. As the scale-free architectures are robust and error-tolerant, this finding provides meaningful insights into the design principle of metabolic networks.

The scale-free organization of metabolic networks has been hypothetically explained in terms of evolution that the new-recruited metabolite members attach preferentially to those that are already well connected (rich get richer, also known as preferential attachment principle) [2]–[4]. This implies that the metabolic network hubs originated relatively earlier than others in evolutionary history [5]. However, several issues about this evolutionary explanation remain elusive. First, the molecular basis of preferential attachment principle has not been fully elucidated, as it is inexplicable how the new metabolites "know" which metabolites are well connected. Second, the evolutionary explanation to the metabolic network organization has little implications for network design, because we do not know how to choose metabolites as hubs to construct a new metabolic network. Since most metabolites are small molecules and metabolic processes are basically chemical reactions, we speculate that the metabolic network organization may have a chemical basis, which stimulated our interest to address these issues by combining bioinformatics and chemoinformatics. The latter is a discipline devoted to encoding, storing, managing, searching and analyzing all kinds of chemical data by information technology [6], [7].

What's unknown

One of the most intriguing findings in systems biology is that despite the varied constituents and metabolic pathways of three domains of life, their metabolic networks exhibit the same scale-free organization. That is, a small part of metabolites participate in a large number of reactions (which are also termed hubs), while others are involved in a few reactions [1]. As the scale-free architectures are robust and error-tolerant, this finding provides meaningful insights into the design principle of metabolic networks.

The scale-free organization of metabolic networks has been hypothetically explained in terms of evolution that the new-recruited metabolite members attach preferentially to those that are already well connected (rich get richer, also known as preferential attachment principle) [2]–[4]. This implies that the metabolic network hubs originated relatively earlier than others in evolutionary history [5]. However, **several issues about this evolutionary explanation remain elusive. First, the molecular basis of preferential attachment principle has not been fully elucidated, as it is inexplicable how the new metabolites "know" which metabolites are well connected. Second, the evolutionary explanation to the metabolic network organization has little implications for network design, because we do not know how to choose metabolites as hubs to construct a new metabolic network.** Since most metabolites are small molecules and metabolic processes are basically chemical reactions, we speculate that the metabolic network organization may have a chemical basis, which stimulated our interest to address these issues by combining bioinformatics and chemoinformatics. The latter is a discipline devoted to encoding, storing, managing, searching and analyzing all kinds of chemical data by information technology [6], [7].

Our study

One of the most intriguing findings in systems biology is that despite the varied constituents and metabolic pathways of three domains of life, their metabolic networks exhibit the same scale-free organization. That is, a small part of metabolites participate in a large number of reactions (which are also termed hubs), while others are involved in a few reactions [1]. As the scale-free architectures are robust and error-tolerant, this finding provides meaningful insights into the design principle of metabolic networks.

The scale-free organization of metabolic networks has been hypothetically explained in terms of evolution that the new-recruited metabolite members attach preferentially to those that are already well connected (rich get richer, also known as preferential attachment principle) [2]–[4]. This implies that the metabolic network hubs originated relatively earlier than others in evolutionary history [5]. However, several issues about this evolutionary explanation remain elusive. First, the molecular basis of preferential attachment principle has not been fully elucidated, as it is inexplicable how the new metabolites “know” which metabolites are well connected. Second, the evolutionary explanation to the metabolic network organization has little implications for network design, because we do not know how to choose metabolites as hubs to construct a new metabolic network**.** Since most metabolites are small molecules and metabolic processes are basically chemical reactions, **we speculate that the metabolic network organization may have a chemical basis, which stimulated our interest to address these issues by combining bioinformatics and chemoinformatics.** The latter is a discipline devoted to encoding, storing, managing, searching and analyzing all kinds of chemical data by information technology [6], [7].

Introduction Example

Exogenous estrogens prevent or substantially retard the decrease in bone mineral density (BMD) that accompanies menopause [1]. However, it is unclear whether exogenous estrogens, administered as oral contraceptives (OCs), can modify premenopausal BMD. Several studies suggest that exposure to OCs during the premenopausal years has a favorable effect on BMD [2-10], whereas other studies show no effect [11-18].

Past studies of the relationship between OC use and BMD have several limitations. Studies have focused primarily on crude measures of OC use, such as current, past and never. These categories combine diverse types of OC use and may reduce the power to detect an effect. Many studies also failed to take into account lifestyle characteristics of study participants. Finally, few studies have considered an effect of OCs on BMD in women of races other than white.

The aim of this study was to evaluate the associations of OCs with spine, hip and whole body BMD in black and white premenopausal women. Our primary hypothesis was that there would be an association between cumulative exposure to estrogen from OCs and BMD.

Osteoporos Int. **2002 Nov;13(11):893-900.**

Gaps in previous research

The lit. review

What's unknown/the research question

What's known

Exogenous estrogens prevent or substa[illegible]crease in bone mineral density (BMD) that accompanies menopause [1]. However, it is unclear whether exogenous estrogens, administered as oral contraceptives (OCs), can modify premenopausal BMD. Several studies suggest that exposure to OCs during the premenopausal years has a favorable effect on BMD [2-10], whereas other studies show no effect [11-18].

Past studies of the relationship between OC use and BMD have several limitations. S[illegible] primarily on crude measures of OC use, such as current, pa[illegible] categories combine diverse types of OC use and may reduce the power to detect an effect. Many studies also failed to take into account lifestyle characteristics of study participants. Finally, few studies have considered an effect of OCs on BMD in women of races other than white.

This study

The aim of this study was to evaluate the associations of OCs with spine, hip and whole body BMD in black and white premenopausal women. Our primary hypothesis was that there would be an association between cumulative exposure to estrogen from OCs and BMD.

Introduction, Example

- Defatted flours, a by-product of the oil industry, constitute an important source of proteins. In general, the industrial process of oil extraction leads to denaturation and diminished solubility of proteins [*1*, *2*], thus affecting the yield of protein extraction and its economical benefit.
- In 2009, Argentina was the second largest sunflower oil producer in the world and the largest exporter of sunflower refined oil and oil cake [*3*]. Among sunflower oil manufacturing by-products, the sunflower oil cake is underused, being almost exclusively employed for animal feeding in spite of its high content of highly digestible proteins with an important content of essential amino acids (except for lysine and sulfur amino acids) [*2*]. The high concentration of phenolic compounds, of which the majority is chlorogenic acid with small amounts of caffeic acid [*2*, *4*, *5*], is the main reason for the underutilization of sunflower oil cake. In addition, these compounds reduce protein solubility and cause unwanted organoleptic characteristics [*2*, *4*, *5*]. Thus, different methods to remove phenolic compounds have been proposed, among them extraction with aqueous alcoholic solutions which has been shown to be very effective [*4*–*6*]. In parallel, however, in the last few years there has been increasing interest in keeping these phenolic compounds, and even in adding them to the formulations, due to their antioxidant activity [*7*] and their benefits for preventing diseases and delaying aging [*8*]. In this context, it is unclear whether these compounds should be removed or not when protein concentrates and isolates are prepared from the sunflower oil cake.
- The aims of this study were: (1) to develop methods for obtaining, from the residual pellet of the local oil industry, sunflower protein concentrates and isolates with different contents of phenolic compounds, and (2) to assess the effect of such compounds on the structural and physicochemical properties (particularly water solubility of the proteins and surface hydrophobicity) and the antioxidant capacity of the products.

A.-D. M. Sørensen, N. S. Nielsen, E. A. Decker, M. B. Let and X. Xu, *et al.* The Efficacy of Compounds with Different Polarities as Antioxidants in Emulsions with Omega-3 Lipids *Journal of the American Oil Chemists' Society*, 2011, 88: 489-502.

Background/what's known

- **Defatted flours, a by-product of the oil industry, constitute an important source of proteins. In general, the industrial process of oil extraction leads to denaturation and diminished solubility of proteins [1, 2], thus affecting the yield of protein extraction and its economical benefit.**
- **In 2009, Argentina was the second largest sunflower oil producer in the world and the largest exporter of sunflower refined oil and oil cake [3]. Among sunflower oil manufacturing by-products, the sunflower oil cake is underused, being almost exclusively employed for animal feeding in spite of its high content of highly digestible proteins with an important content of essential amino acids (except for lysine and sulfur amino acids) [2]. The high concentration of phenolic compounds, of which the majority is chlorogenic acid with small amounts of caffeic acid [2, 4, 5], is the main reason for the underutilization of sunflower oil cake. In addition, these compounds reduce protein solubility and cause unwanted organoleptic characteristics [2, 4, 5]. Thus, different methods to remove phenolic compounds have been proposed, among them extraction with aqueous alcoholic solutions which has been shown to be very effective [4–6].** In parallel, however, in the last few years there has been increasing interest in keeping these phenolic compounds, and even in adding them to the formulations, due to their antioxidant activity [7] and their benefits for preventing diseases and delaying aging [8]. In this context, it is unclear whether these compounds should be removed or not when protein concentrates and isolates are prepared from the sunflower oil cake.
- The aims of this study were: (1) to develop methods for obtaining, from the residual pellet of the local oil industry, sunflower protein concentrates and isolates with different contents of phenolic compounds, and (2) to assess the effect of such compounds on the structural and physicochemical properties (particularly water solubility of the proteins and surface

What's unknown/controversy

- Defatted flours, a by-product of the oil industry, constitute an important source of proteins. In general, the industrial process of oil extraction leads to denaturation and diminished solubility of proteins [1, 2], thus affecting the yield of protein extraction and its economical benefit.
- In 2009, Argentina was the second largest sunflower oil producer in the world and the largest exporter of sunflower refined oil and oil cake [3]. Among sunflower oil manufacturing by-products, the sunflower oil cake is underused, being almost exclusively employed for animal feeding in spite of its high content of highly digestible proteins with an important content of essential amino acids (except for lysine and sulfur amino acids) [2]. The high concentration of phenolic compounds, of which the majority is chlorogenic acid with small amounts of caffeic acid [2, 4, 5], is the main reason for the underutilization of sunflower oil cake. In addition, these compounds reduce protein solubility and cause unwanted organoleptic characteristics [2, 4, 5]. Thus, different methods to remove phenolic compounds have been proposed, among them extraction with aqueous alcoholic solutions which has been shown to be very effective [4–6]. **In parallel, however, in the last few years there has been increasing interest in keeping these phenolic compounds, and even in adding them to the formulations, due to their antioxidant activity [7] and their benefits for preventing diseases and delaying aging [8]. In this context, it is unclear whether these compounds should be removed or not when protein concentrates and isolates are prepared from the sunflower oil cake.**
- The aims of this study were: (1) to develop methods for obtaining, from the residual pellet of the local oil industry, sunflower protein concentrates and isolates with different contents of phenolic compounds, and (2) to assess the effect of such compounds on the structural and physicochemical properties (particularly water solubility of the proteins and surface hydrophobicity) and the antioxidant capacity of the products.

This study aims to help answer the controversy…

- Defatted flours, a by-product of the oil industry, constitute an important source of proteins. In general, the industrial process of oil extraction leads to denaturation and diminished solubility of proteins [1, 2], thus affecting the yield of protein extraction and its economical benefit.
- In 2009, Argentina was the second largest sunflower oil producer in the world and the largest exporter of sunflower refined oil and oil cake [3]. Among sunflower oil manufacturing by-products, the sunflower oil cake is underused, being almost exclusively employed for animal feeding in spite of its high content of highly digestible proteins with an important content of essential amino acids (except for lysine and sulfur amino acids) [2]. The high concentration of phenolic compounds, of which the majority is chlorogenic acid with small amounts of caffeic acid [2, 4, 5], is the main reason for the underutilization of sunflower oil cake. In addition, these compounds reduce protein solubility and cause unwanted organoleptic characteristics [2, 4, 5]. Thus, different methods to remove phenolic compounds have been proposed, among them extraction with aqueous alcoholic solutions which has been shown to be very effective [4–6]. In parallel, however, in the last few years there has been increasing interest in keeping these phenolic compounds, and even in adding them to the formulations, due to their antioxidant activity [7] and their benefits for preventing diseases and delaying aging [8]. In this context, it is unclear whether these compounds should be removed or not when protein concentrates and isolates are prepared from the sunflower oil cake.
- **The aims of this study were: (1) to develop methods for obtaining, from the residual pellet of the local oil industry, sunflower protein concentrates and isolates with different contents of phenolic compounds, and (2) to assess the effect of such compounds on the structural and physicochemical properties (particularly water solubility of the proteins and surface hydrophobicity) and the antioxidant capacity of the products.**

Introduction, Example

Road traffic collisions are an important cause of death and disability worldwide. Every year around the world 1.2 million people are killed and up to 50 million are injured or disabled as a result of road traffic collisions.[1] Morbidity from road traffic collisions is expected to increase in future years, and it is estimated that road traffic collisions will move from ninth to third place in the global burden of disease ranking, as measured in disability adjusted life years.[2 3]

Measures to reduce traffic speed are considered essential to reducing casualties on the road.[1 4 5] Speed cameras are increasingly used to help to reduce traffic speeds in the belief that this will reduce road traffic collisions and casualties, and an expansion in the use of speed cameras is under way in many countries, most notably the United Kingdom.[6] The use of speed cameras is controversial, however. Vociferous opponents, including some motoring associated organisations, oppose their use, and cameras are often criticised in the media.[7-9] The lack of readily available evidence of the effectiveness of cameras has made it difficult for road safety and health professionals to engage in an informed debate about the effectiveness of speed cameras.

A previous small non-systematic review of six studies found a 17% reduction in collisions after introduction of speed cameras.[10] Non-systematic reviews can, however, be limited by bias. We aimed, therefore, to systematically assess the evidence for the effectiveness of speed cameras in reducing road traffic collisions and related casualties.

Pilkington P. Effectiveness of speed cameras in preventing road traffic collisions and related casualties: systematic review. *BMJ* 2005;330:331.

Statement of problem. What's known.

Road traffic collisions are an important cause of death and disability worldwide. Every year around the world 1.2 million people are killed and up to 50 million are injured or disabled as a result of road traffic collisions.[1] Morbidity from road traffic collisions is expected to increase in future years, and it is estimated that road traffic collisions will move from ninth to third place in the global burden of disease ranking, as measured in disability adjusted life years.[2 3]

Measures to reduce traffic speed are considered essential to reducing casualties on the[4 5] help to reduce traffic speeds in the and casualties, and an expansion in the use of speed cameras is under way in many countries, most notably the United Kingdom.[6] The use of speed cameras is controversial, however. Vociferous opponents, including some motoring associated organisations, oppose their use, and cameras are often criticised in the media.[7-9] The lack of readily available evidence of the effectiveness of cameras has made it difficult for road safety d debate about the effectiveness of speed cameras.

What's unknown/controversial.

Limitations of previous research.

What we did to answer this question better.

A pr on in collisions after introduction of speed cameras.[10] Non-systematic reviews can, however, be limited by bias. We aimed, therefore, to systematically assess the evidence for the effectiveness of speed cameras in reducing road traffic collisions and related casualties.

Writing in the Sciences

Unit 5.5: Discussion

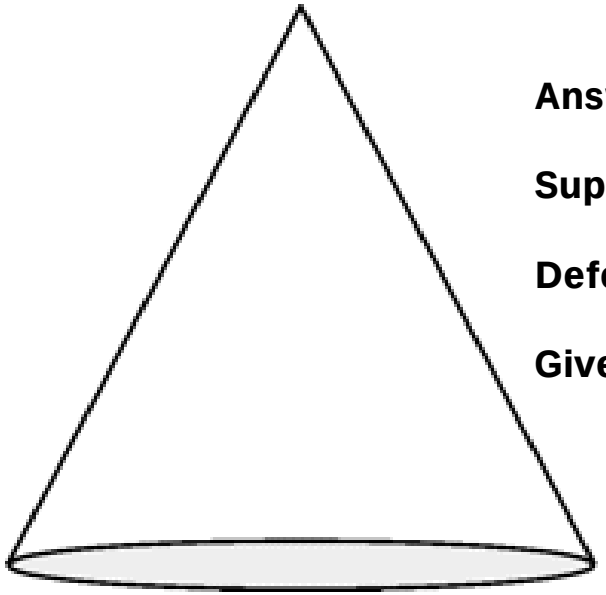
The Discussion section...

- Gives you the most freedom
- Gives you the most chance to put good writing on display
- Is the most challenging to write

The Discussion

Follow your rules for good writing!

Invert the cone!



Answer the question asked.

Support your conclusion (your data, others' data)

Defend your conclusion (anticipate criticisms)

Give the "big-picture" take-home message

I.e., what do my results mean and why should anyone care?

Key finding (answer to the question(s) asked in Intro.)	• Start with: "WE FOUND THAT…" (or something similar) • Explain what the data mean (big-picture!) • State if the findings are novel
Key secondary findings	
Context	• Give possible mechanisms or pathways • Compare your results with other people's results • Discuss how your findings support or challenge the paradigm
Strengths and limitations	• Anticipate readers' questions/criticisms • Explain why your results are robust
What's next	• Recommended confirmatory studies ("needs to be confirmed") • Point out unanswered questions and future directions
The "so what?": implicate, speculate, recommend	• Give the big-picture (human) implications of basic science findings • Tell readers why they should care
Strong conclusion	• Restate your main finding. • Give a final take-home message.

Discussion section, tips

- **Showcase good writing!**
  - **Use the active voice**
  - **Tell it like a story**
- **Start and end with the main finding**
  - **“We found that…”**
- **Don’t travel too far from your data**
  - **Focus on what your data do prove, not what you had hoped your data would prove**
- **Focus on the limitations that matter, not generic limitations**
- **Make sure your take-home message is clear and consistent**

Discussion, example

LAST PARAGRAPH OF INTRODUCTION:

The differences in health benefits between a carbohydrate-restricted diet and a calorie- and fat-restricted diet are of considerable public interest. However, there is concern that a carbohydrate-restricted diet will adversely affect serum lipid concentrations.[1] Previous studies demonstrating that healthy volunteers following a low-carbohydrate diet can lose weight have involved few subjects, and few used a comparison group that followed consensus guidelines for weight loss.[2,3] The reported effects of a carbohydrate-restricted diet on risk factors for atherosclerosis have varied.[2,3,4] **We performed a study designed to test the hypothesis that severely obese subjects with a high prevalence of diabetes or the metabolic syndrome [a] would have a greater weight loss, [b] without detrimental effects on risk factors for atherosclerosis, while on a carbohydrate-restricted (low-carbohydrate) diet than on a calorie- and fat-restricted (low-fat) diet.**

Samaha FF, Iqbal N, Seshadri P, et al. A low-carbohydrate as compared with a low-fat diet in severe obesity. *N Engl J Med* 2003;348:2074-2081

The Discussion

1. <u>We found that</u> severely obese subjects with a high prevalence of diabetes and the metabolic syndrome lost more weight in a six-month period on a carbohydrate-restricted diet than on a fat- and calorie-restricted diet. [answer to a] The greater weight loss in the low-carbohydrate group suggests a greater reduction in overall caloric intake, rather than a direct effect of macronutrient composition. [mechanisms] However, the explanation for this difference is not clear. Subjects in this group may have experienced greater satiety on a diet with liberal proportions of protein and fat. However, other potential explanations include the simplicity of the diet and improved compliance related to the novelty of the diet. [possible mechanisms/unanswered questions]

2. Subjects in the low-carbohydrate group had greater decreases in triglyceride levels than did subjects in the low-fat group; nondiabetic subjects on the low-carbohydrate diet had greater increases in insulin sensitivity, and subjects with diabetes on this diet had a greater improvement in glycemic control. No adverse effects on other serum lipid levels were observed.

[answer to b] Most studies suggest that lowering triglyceride levels has an overall cardiovascular benefit.[14,15,16] Insulin resistance promotes such atherosclerotic processes as inflammation,[17] decreased size of low-density lipoprotein particles,[18] and endothelial dysfunction.[19] Impaired glycemic control in subjects with other features of the metabolic syndrome markedly increases the risk of coronary artery disease.[20] As expected, we found that the amount of weight lost had a significant effect on the degree of improvement in these metabolic factors. [comparison to previous studies and paradigms] However, even after adjustment for the differences in weight loss between the groups, assignment to the low-carbohydrate diet predicted greater improvements in triglyceride levels and insulin sensitivity.

[unexpected] Subjects who lost more than 5 percent of their base-line weight on a carbohydrate-restricted diet had greater decreases in triglyceride levels than those who lost a similar amount of weight while following a calorie- and fat-restricted diet. [supporting details]

3. There was a consistent trend across weight-loss strata toward a greater increase in insulin sensitivity in the low-carbohydrate group, although these changes were small and were not significant within each stratum. **[supporting details: dose/response]** Although greater weight loss could not entirely account for the greater decrease in triglyceride levels and increase in insulin sensitivity in the low-carbohydrate group, we cannot definitively conclude that carbohydrate restriction alone accounted for this independent effect. **[mechanisms]** Other uncontrolled variables, such as the types of carbohydrates selected (e.g., the proportion of complex carbohydrates or the ratio of carbohydrate to fiber), or other unknown variables may have contributed to this effect. In addition, more precise measurements of insulin sensitivity than we used would be needed to confirm this effect of a carbohydrate-restricted diet. **[limitations/future studies]**

4. Many of our subjects were taking lipid-lowering medications and hypoglycemic agents. Although enrolling these subjects introduced confounding variables, it allowed the inclusion of subjects with the obesity-related medical disorders typically encountered in clinical practice. Analyses from which these subjects were excluded still revealed greater improvements in insulin sensitivity and triglyceride levels on a carbohydrate-restricted diet than on a fat- and calorie-restricted diet. [limitations and how they were addressed]

5. Our study included a high proportion of black subjects a group previously underrepresented in lifestyle-modification studies. [strength] As compared with the white subjects, the black subjects had a smaller overall weight loss. Future studies should explore whether greater weight loss in this population can be achieved by more effective incorporation of culturally sensitive dietary counseling. [future directions]
6. The high dropout rate in our study occurred very early and affected our findings. The very early dropout of these subjects may indicate that attrition most closely reflected base-line motivation to lose weight, rather than a response to the dietary intervention itself. [limitation]

7. Taken together, our findings demonstrate that severely obese subjects with a high prevalence of diabetes and the metabolic syndrome lost more weight during six months on a carbohydrate-restricted diet than on a calorie- and fat-restricted diet. The carbohydrate-restricted diet led to greater improvements in insulin sensitivity that were independent of weight loss and a greater reduction in triglyceride levels in subjects who lost more than 5 percent of their base-line weight. **[conclusion; restate answers to a and b]** These findings must be interpreted with caution, however, since the magnitude of the overall weight loss relative to our subjects' severe obesity was small, and it is unclear whether these benefits of a carbohydrate-restricted diet extend beyond six months. Furthermore, the high dropout rate and the small overall weight loss demonstrate that dietary adherence was relatively low in both diet groups. **[big picture] This study proves a principle and does not provide clinical guidance; given the known benefits of fat restriction, future studies evaluating long-term cardiovascular outcomes are needed before a carbohydrate-restricted diet can be endorsed. [take-home message]**

Example, discussion

- 1. In the present work, we give closed formulae for the generating function of the cumulants of the current in the open TASEP. [Answers the research question/aim] These results hold for all values of the boundary parameters α and β and all values of the system size L. We emphasize the fact that our expressions are exact and not only asymptotic: they are of combinatorial nature. [What's novel/important] They allow us to describe the ASEP at all points of its phase diagram, including the phase-transition lines. The cumulant generating function is given in the form of a parametric representation—equations (15)–(19). A similar mathematical structure can be found in other works [15, 18, 34] and it can be related physically to the additivity principle [3, 10] [Context] .

- 2. The statistics of the current in the open TASEP had remained a challenging open problem for many years and no exact solution for the full distribution of the current was known for finite-size systems [Context] . The BetheAnsatz equations for the open ASEP were studied in [7] but they are valid only on some surfaces in the parameter space: this restriction seemed to be a major obstruction to the computation of the large-deviation function. Only recently, a very subtle analysis of the Bethe equations, valid in the $L \to \infty$limit, together with some conjectures on the asymptotic locations of the Bethe roots, was carried out in [8], leading to an expression for the cumulant generating function. However, the result in [8] can only be established deep inside the low- and the high-density phases and it is an asymptotic expression, valid only when $L \to \infty$] [Context] . In our work, we have followed a different path and used the matrix product representation [1, 16] to calculate the cumulants of the current order by order [What's novel] . We have performed some explicit calculations and uncovered the general structure of the solution. From the mathematical point of view, our formulae are only a conjecture but we have verified it in dozens of cases and derived from it all the previously known results [Limitations/defending the conclusion]. We have absolutely no doubt that our expressions are true. In particular, the main expression obtained in [8] can be derived as a limiting case of our results [Defending the conclusion].

3. We have decided to present the final formulae (15) and (16) before completing the proof, because we find the results elegant and sufficient by themselves. Furthermore, they allow us to draw some interesting physical consequences and to open new problems [Implications/future directions]. We emphasize that although equations (15) and (16) (and also equations (8) and (9)) are conjectures, all the results that we have drawn from them are established rigorously [Limitations/defending the conclusion]. We think that the proof is a question of carrying out a very long computation rather than having some deep mathematical insight. We are presently working on this aspect [Future studies]. We hope to have given enough details to the reader to clarify what kind of assumptions were made, which allowed us to jump to the final result and to guess the full structure of the solution. Another possibility, now that the final formula is known, is to search for a direct method to check it: after all, the cumulant generating function is nothing but the largest eigenvalue of a known operator [Future studies].

4. Besides completing the derivation, we also intend to extract from equations (15) to (19) the scaling limit of the large deviation function in the $L \to \infty$ limit. It would be interesting to compare that scaling form with recent numerical results obtained by Monte Carlo and DMRG methods [25, 31, 33] and also to investigate the crossover with theoretical results derived on the infinite lattice [21, 39] [What's next/future studies].

- 5. Finally, we have considered here only the TASEP. But the matrix method can also be applied to the partially asymmetric case (PASEP), and we have checked that the tensor products of the PASEP quadratic algebra that appeared in multispecies PASEP models [35] allow us to solve the hierarchy of equations (30) for the cumulants [wider implications]. We believe that the parametric representation still holds in the PASEP case and that combinatorial tree structures akin to those found for the PASEP on a periodic ring in [34] will probably play an important role [wider implications]. Besides, complex integral representations for matrix elements analogous to those we have used here also appear in the PASEP [1, 2, 37, 38]. There must exist a general and hopefully elegant structure that encompasses all the cases, though we are well aware that such a structure may be difficult to discover [take-home message/big picture].

What NOT to do...

Don’t start your discussion like this!

Discussion

- This meta-analysis is subject to a number of limitations. The estimates of risk for melanoma subsequent to using sunlamps/sunbeds are based on published data in a series of 10 articles over a period of 20 years. A pooled analysis of original observations taken in the 10 studies would have provided a more powerful approach ...

The Discussion: verb tense

Past, when referring to study details, results, analyses, and background research:

- We found that
- Subjects may have experienced
- Miller et al. found

Present, when talking about what the data suggest:

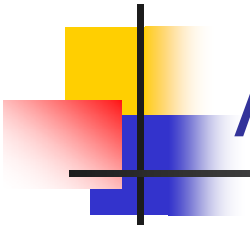
The greater weight loss suggests

The explanation for this difference is not clear.

Potential explanations include

Writing in the Sciences

Unit 5.6: Abstract



Abstract

Abstracts (*ab*=out, *trahere*=pull; “to pull out”)

- Overview of the main story
- Gives highlights from each section of the paper
- Limited length (100-300 words, typically)

- Stands on its own
- Used, with title, for electronic search engines
- Most often, the only part people read

Abstract

1. Background
2. Question/aim/hypothesis
   - "We asked whether," "We hypothesized that,"...etc.
3. Experiment(s)
   - Quick summary of key materials and methods
4. Results
   - Key results found
   - Minimal raw data (prefer summaries)
5. Conclusion: The answer to the question asked/take-home message
6. Implication, speculation, or recommendation

Mimi Zeiger. *Essentials of Writing Biomedical Research Papers*, McGraw Hill Professional, 1999

The Abstract

Abstracts may be structured (with subheadings) or free-form.

Abstract, Example

Background

Question asked

INTRODUCTION: Avian H5N1 influenza viruses currently circulating in southeast Asia could potentially cause the next pandemic. However, currently licensed human vaccines are subtype-specific and do not protect against these H5N1 viruses. We aimed to develop an influenza vaccine and assessed its immunogenicity and efficacy to confer protection in BALB/c mice.

METHODS: We developed an egg-independent strategy to combat the avian influenza virus, because the virus is highly lethal to chickens and the maintenance of a constant supply of embryonated eggs would be difficult in a pandemic. We used a replication-incompetent, human adenoviral-vector-based, haemagglutinin subtype 5 influenza vaccine (HAd-H5HA), which induces both humoral and cell-mediated immune responses against avian H5N1 influenza viruses isolated from people.

Experiments done

The Lancet, 2006 Feb 11;367(9509):475-81.

Abstract

Results found

FINDINGS: Immunisation of mice with HAd-H5HA provided effective protection from H5N1 disease, death, and primary viral replication ($p<0.0001$) against antigenically distinct strains of H5N1 influenza viruses. Unlike the recombinant H5HA vaccine, which is based on a traditional subunit vaccine approach, HAd-H5HA vaccine induced a three-fold to eight-fold increase in HA-518-epitope-specific interferon-gamma-secreting CD8 T cells ($p=0.01$)

Answer to the question asked

INTERPRETATION: Our findings highlight the potential of an Ad-vector-based delivery system, which is both egg-independent and adjuvant-independe for the development of a pandemic

Wider implication

Abstract: unstructured example

Empirical research with nonhuman primates appears to support the view that causal reasoning is a key cognitive faculty that divides humans from animals. The claim is that animals approximate causal learning using associative processes. The present results cast doubt on that conclusion. Rats made causal inferences in a basic task that taps into core features of causal reasoning without requiring complex physical knowledge. They derived predictions of the outcomes of interventions after passive observational learning of different kinds of causal models. These competencies cannot be explained by current associative theories but are consistent with causal Bayes net theories.

Science. 2006 Feb 17;311(5763):1020-2. Causal reasoning in rats. Blaisdell AP, Sawa K, Leising KJ, Waldmann MR.