Intro II: Obligatory tasks, two challenges and information on the group work and exam

Obligatory tasks:

Participants must deliver two texts prior to the course. Failure to do so will disqualify from participation. These texts are:

1) an **article** manuscript under preparation – including title, abstract, introduction and references, and 2) **a press release** that covers a part of the current research (see below for the format).

THE ARTICLE AND PRESS RELEASE MUST BE IN ENGLISH IF YOU ARE TAKING THE ENGLISH COURSE. At the Norwegian course participants may deliver a Norwegian article **if and only if** the article in your thesis will be in Norwegian.

There will also be a **home exam** consisting of **three parts** that should be submitted two weeks after the course.

You will find information about deadlines in the text below.

Challenges: Participants are invited to volunteer to give a 5 minute popularized talk about their research (5-6 people) and to bring a poster (12-15 people). The volunteers will receive constructive feedback on their talks/posters. Previous participants have found this opportunity to be very useful. There will also be a poster prize.

Day 1:

5-6 participants are given the opportunity to present **a 5 minute popularized talk about their research.** Define your target group «for interested 16 year olds» or similar.

Interested participants: send an e-mail to **adm-phd-emner@medisin.uio.no** . First come, first served.

Day 2:

Your own article manuscript under preparation – including Title, Abstract, Introduction and references. All participants must submit an article text for use in the group work

Deadline 23. October

For the group work on Tuesday - Participants must bring:

a. **4 printouts of** Title, Abstract, Introduction, References from your article text (ENGLISH!)

b. Computer or similar

Day 4:

Poster session

We need 12-15 posters for the poster session. Please send an email to adm-phd-emner@medisin.uio.no if you are interested.

Day 5:

Science and the Media. All participants must upload "a Press Release". Failure to do so will disqualify from taking the course. – Deadline 23. October

Write a PRESS RELEASE
Please adhere to the format, including the target audience and take home message. See box below, or the submission will be disqualified.

About the press release, format of this session on Friday:

This text will be used in the course to exemplify what a press desk is looking for in the multitude of incoming press releases. Selected examples from participants will be presented for the audience. In a plenary session, we will discuss to what extent these examples can be improved (what is fine, what can be improved, what may be lacking), specific suggestions will also be provided. It is suggested that participants use adequate time to provide the best possible "press release"-document from their own research. It will not be possible to discuss each and every submitted text in plenum, but participants may have the opportunity to discuss their submission in the breaks. For this purpose, participants are asked to bring a printout of their submitted text.

Your submission: Write a press release based on your research in the form of a **popular scientific text** of approximately half a page. (If you just copy paste an article abstract, this will disqualify.) More specifically, provide a presentation of your research for non-scientists in a form written for a newspaper or a magazine/periodical. The text should be structured with title, subtitles, a lead paragraph, and main text.

ALL PARTICIPANTS: Before you start writing, please also define the following. This information should be provided at the top of your page:
- Who is the main target group for your text? - Where would you like to see the text published? - Define ONE take-home message for the reader.

EXAM – deadline 17. November

The exam deadline will be Friday 2 weeks after the course.

You will submit:

Exam part 1: **Cover Sheet for own article text** and **Revised version** of own article with comments and track changes.

Exam Part 2: **Writing the medical article**. Based on unknown text (3 article choices) you will answer specific questions, write an abstract and propose a title. **You will deliver: Course Exam part II. Cover sheet and answer form**

Exam part 3: You will make a **Graphical abstract** in the correct format to illustrate one of the 6 paper options. You will deliver: Graphical abstract as a **Tif file** and the **Cover Sheet for the Graphical abstract**.

Exam deadline is Friday 2 weeks after course end.

EXAM Intro II. MF9030 and MF9030E

The exam deadline will be Friday 2 weeks after the course.

You will submit:

Exam part 1: **Cover Sheet for own article text** and **Revised version** of own article with comments and track changes.

Exam Part 2: **Writing the medical article**. Based on unknown text (3 article choices) you will answer specific questions, write an abstract and propose a title. **You will deliver: Course Exam part II. Cover sheet and answer form**

Exam part 3: You will make a **Graphical abstract** in the correct format to illustrate one of the 6 paper options. You will deliver: Graphical abstract as a **Tif file** and the **Cover Sheet for the Graphical abstract**.

Exam deadline 2 weeks after course end.

Course group work and Exam part 1 – Own paper

Participants work with their manuscript text, deliver a new edited version after completion of the week, to be uploaded within 2 weeks after the course.

Please name this file with your name "participantname.docx"

The new version must have the **Cover Sheet for own article text** that describes the revision process. The Word file changes must be trackable, i.e. "Track changes" (and/or yellowing/redding/comments in the text)

Note. YOU MUST INCLUDE THE COVER SHEET AND FILE WITH TRACK CHANGES/COMMENTS (and failure to do so will make you fail the exam).

Exam part 2 – Writing the Medical article (based on unknown text)

You can choose one of three options (article 1, 2 or 3) that you will work with. You will answer questions, write an abstract and propose a title. You will fill in **Course Exam part II. Cover sheet and answer form** where you indicate your name, your background, your choice of paper, your answers and your abstract title.

Note. YOU CAN NOT USE YOUR OWN ARTICLE TEXT (and if you do so you will fail the exam).

Exam part 3 – Graphical abstract

You can chose one of 6 options (article 1-6) that you work with. You will make a graphical abstract prepare the correct format and upload the tif file together with a **Cover Sheet for the Graphical abstract**.

Note. YOU CAN NOT USE YOUR OWN ARTICLE TEXT (and if you do so you will fail the exam).

Where to submit the texts: The texts and home exam should be submitted in Fronter.

Group Work in preparation for the revision and Exam parts 1, 2 and 3

The idea of the group work is to start a conversation between peers (most often outside your field) where you discuss your pre-submitted article and start an introspective revision process.

After working with fellow participants, you will continue with an experienced colleague – preferably your supervisor and revise your own article text.

The process that you develop, your discussions, experimentations and conclusions are to be presented as Exam part 1. You will upload a Cover sheet for the Exam part 1 where you take the sensor through your revision process.

Next you will be challenged with answering a set of key question that cover information presented in an article that you chose from a set of three papers, articles 1, 2 or 3. In this Exam part 2, you will also compose an abstract and propose a title to this article.

As an Exam part 3, you will deliver a graphical abstract in a specific format. Here, you will present a main finding from one of the articles 1, 2 or 3 (or if you like from 3 alternative articles that are provided). In addition to preparing this figure, you will submit a Cover sheet for the Graphical Abstract.

Group Work

Main point: have you managed to communicate efficiently with your reader? Is your language appropriate and do you meet the expectations of you reader (organization and content)? For example: Do you have the expected signals of content (see Figure below)? Is it absolutely clear to your reader what are the aims, the questions, the results/answers? Background-known-unknown (knowledge gap)- question/aim/hypothesis-message-results-conclusions-significance?

Look at your title

Is it descriptive, declarative (or interrogative)? Do you have a subject, action verb? Is it tagged with important key words? Conclusion? Can you convert it ->declarative, -> descriptive (or vice versa)? Is it catchy? Discuss with your group.

Look at your own abstract

Do you have research question(s)? Descriptive/hypothesis-testing? Do you have all the suggested elements, signals for your topics, see figure below? If you have a question/hypothesis: Is the question emphasized, made clear to the reader? If not – is it possible to make a question? Do you signal that you are presenting results? Do you present an answer to the question? Is it clear that this is the answer? Same words as in the question? Do you have a conclusion? Are broader implications included? Clear language and style? Compare/discuss with your group.

-> Signaling your topics

Signal the parts of an abstract both visually, by starting a new sentence, and verbally, by signaling the topic at the beginning of the sentence.

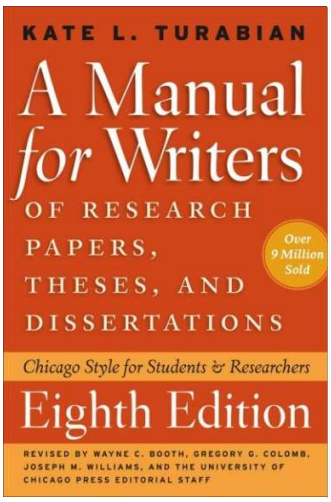
1. General information on the field	---	
2. Background information	The issue has yet to be resolved It has been hypothesized that	
A. Question or Hypothesis	We asked whether…	We hypothesized that …
B. Experiments that were done	To answer the question, we …	To test the hypothesis, we …
C. Answers to the question	We found, / Here we show	
3. General implications, importance for the field	These results suggest that	

Look at your own Introduction

Structure. Put labels on your paragraphs – how can each paragraph be described? Is the content of each of the same type?
How do you introduce the transitions (within paragraphs) and between paragraphs?
Known unknown, which comes first (see point 5 in the Figure below, familiar/unfamiliar)?
How do you describe the context, theme, several paragraphs?
Do you present a controversy? If not: can you do so? Have you answered the ”So what”-question, i.e. the significance, importance, consequences of the question and finding an answer?
Do you present a hypothesis (and if not - is this a descriptive paper)? Structured hypothesis PICO? Experimental approach?
Language and style. Sentence length, punctuations, tempo, parentheses? Unnecessary words? Verbs and their subjects? Voice active, passive – & why? Nominizations (verbal nouns): can these be substituted with verbs? Look at your subjects – do they all deserve to be subjects, are your subjects abstraction? Are they substituting for “We, us”? Are your subjects and verbs close together?

Language and style

Advice on what is important



Seven principles:

■ Avoid introducing more than a few sentences with long phrases and clauses; **get to the subject of your sentence quickly**.
■ Make **subjects short and concrete**, ideally **naming the character** that performs the action expressed by the verb that follows.
■ Avoid interrupting the **subject and verb** with more than a word or two.
■ Put key actions in **verbs**, not in nouns.
■ Put information **familiar** to readers at the beginning of a sentence, **new** information at the end.
■ Choose active or passive verbs to reflect the previous principles.
■ Use first-person pronouns appropriately.

Turabian, Kate L. (2013-04-09). A Manual for Writers of Research Papers, Theses, and Dissertations, Eighth Edition: Chicago Style for Students and Researchers (Chicago Guides to Writing, Editing, and) (Kindle Locations 3470-3480). University of Chicago Press. Kindle Edition.

Active or **passive voice** is often included in advice on writing (see also above). Passive voice may be useful when the object is more important than the subject – The figure below has red objects then verbs and unnecessary subjects hid away inside double parentheses, i.e. Object-verb-((subject)).

Merriam–Webster's Dictionary of English Usage (1994)

The store was robbed last night ((by a thief))

Kennedy was elected president ((by us))

Object	Subject
(receiver of the action)	(agent)

Recommends passive voice when identifying the object (receiver) of the action is more important than the subject (agent), and when the agent is unknown, unimportant, or not worth mentioning

Cancer killing cells were identified ((by us))

EMBO
reports

science & society

Inheritance of poor writing habits

To improve scientific writing we must break the chain of transmission of complex writing style from senior to junior scientists

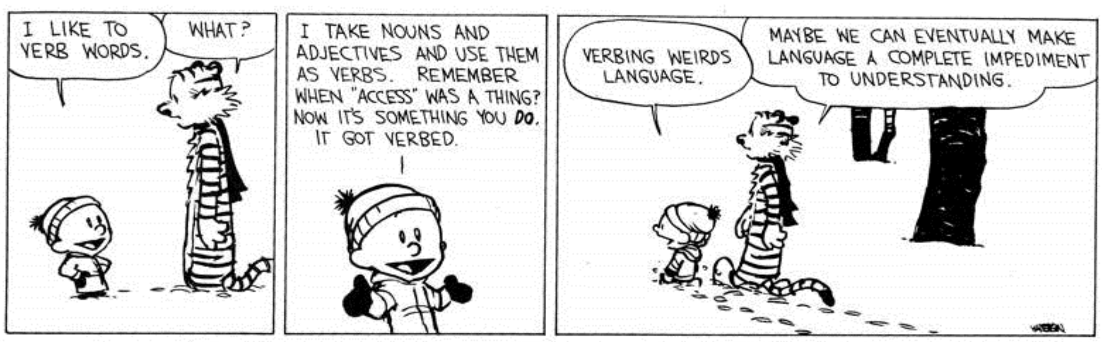
Amin Bredan

The volume of scientific literature is enormous, but it is largely inaccessible to non-expert readers, including scientists from other fields. This is not just because the content is highly specialized but also because scientific writing itself is far from simple and clear. Generations of editors, reviewers and readers have struggled to understand complex, exaggerated and often pompous prose that does little to enhance the reader's understanding but aims to demonstrate the scholarly prowess of the author. The causes go beyond an inadequate com- any writer may turn, if he will, into clear and accurate English". Over 120 years later, things have not improved; a recent editorial in *Nature Cell Biology* exhorts scientists to write their research clearly [2], whilst another in *Nature Structural & Molecular Biology* has the telling title 'Scientific writing 101' [3]. It is interesting to note that I have never come across an editorial, comment or article praising the quality of scientific writing. One senior editor of *Nature* bluntly stated "most papers are badly written" [4].

One of the best examples of awkward and convoluted writing that I have come across is the following sentence: "We adopt this broad-scale approach to determine that relationships occur both at the level of the population (and hence not confounded by [1] potential environmental variation and/or [2] statistical non-independence of individuals) and also across individuals (because [1] relatively recent colonization of the UK by rabbits […], and [2] previous work […] demonstrating extremely fine-scale genetic structuring in UK rabbits over

Take a look at the articles, advice and text uploaded into Fronter. Why should you care about writing? What is good language and style? Make sure that you don't inherit the abysmal habits of others… bust some myths and avoid

verbal zombies:

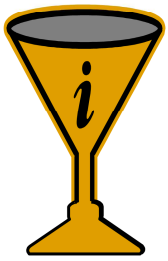


Nominalizations - Zombie nouns:

https://opinionator.blogs.nytimes.com/2012/07/23/zombie-nouns/

How to construct an abstract according to Mimi Zeiger

The Abstract (hypothesis testing)



1. General information on the field
2. Background information. Unresolved issue

A. Question or Hypothesis
B. Experiments that were done
C. Results. - Answers to the question/hypothesis

3. General implications, importance for the field

Verb tenses in the abstract should be the same as those in the paper:
Present (*presens*) tense for the question and the answer;
past (*preteritum*) tense for the experiment done and the results found.

Descriptive abstracts

Because there is no hypothesis in descriptive papers, the message is often stated at the beginning of the abstract.

1. General information on the field
2. Background information. **Unresolved issue**

A. Message. Experiments that were done
B. Results. Resolving the issue (?)

3. General implications, importance for the field

-> Signaling your topics

Signal the parts of an abstract both visually, by starting a new sentence, and verbally, by signaling the topic at the beginning of the sentence.

1. General information on the field	---	
2. Background information	The issue has yet to be resolved It has been hypothesized that	
A. Question or Hypothesis	We asked whether…	We hypothesized that …
B. Experiments that were done	To answer the question, we …	To test the hypothesis, we …
C. Answers to the question	We found, / Here we show	
3. General implications, importance for the field	These results suggest that	

How to construct an abstract according to Nature Publishing group

nature

How to construct a *Nature* summary paragraph

Annotated example taken from *Nature* **435**, 114-118 (5 May 2005).

*One or two sentences providing a **basic introduction** to the field, comprehensible to a scientist in any discipline.*

*Two to three sentences of **more detailed background**, comprehensible to scientists in related disciplines.*

*One sentence clearly stating the **general problem** being addressed by this particular study.*

*One sentence summarising the main result (with the words "**here we show**" or their equivalent).*

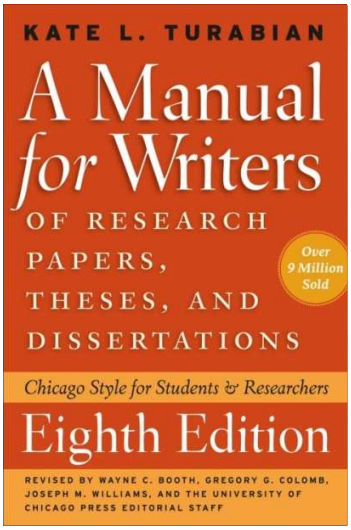
*Two or three sentences explaining what the **main result** reveals in direct comparison to what was thought to be the case previously, or how the main result adds to previous knowledge.*

*One or two sentences to put the results into a more **general context**.*

*Two or three sentences to provide a **broader perspective**, readily comprehensible to a scientist in any discipline, may be included in the first paragraph if the editor considers that the accessibility of the paper is significantly enhanced by their inclusion. Under these circumstances, the length of the paragraph can be up to 300 words. (The above example is 190 words without the final section, and 250 words with it).*

During cell division, mitotic spindles are assembled by microtubule-based motor proteins[1, 2]. The bipolar organization of spindles is essential for proper segregation of chromosomes, and requires plus-end-directed homotetrameric motor proteins of the widely conserved kinesin-5 (BimC) family[3]. Hypotheses for bipolar spindle formation include the 'push–pull mitotic muscle' model, in which kinesin-5 and opposing motor proteins act between overlapping microtubules[2, 4, 5]. However, the precise roles of kinesin-5 during this process are unknown. Here we show that the vertebrate kinesin-5 Eg5 drives the sliding of microtubules depending on their relative orientation. We found in controlled *in vitro* assays that Eg5 has the remarkable capability of simultaneously moving at ~ 20 nm s^{-1} towards the plus-ends of each of the two microtubules it crosslinks. For anti-parallel microtubules, this results in relative sliding at ~ 40 nm s^{-1}, comparable to spindle pole separation rates *in vivo*[6]. Furthermore, we found that Eg5 can tether microtubule plus-ends, suggesting an additional microtubule-binding mode for Eg5. Our results demonstrate how members of the kinesin-5 family are likely to function in mitosis, pushing apart interpolar microtubules as well as recruiting microtubules into bundles that are subsequently polarized by relative sliding. We anticipate our assay to be a starting point for more sophisticated *in vitro* models of mitotic spindles. For example, the individual and combined action of multiple mitotic motors could be tested, including minus-end-directed motors opposing Eg5 motility. Furthermore, Eg5 inhibition is a major target of anti-cancer drug development, and a well-defined and quantitative assay for motor function will be relevant for such developments.

What should you have in your Introduction?



The Introduction

1. **Opening context or background** - puts your project in the context of other research and sets up the next step.
2. **A statement of your research question**. This is typically a statement of **what isn't known** or understood or of what is flawed about the research you cited in step 1. It often begins with but, **however**, or another word signaling a qualification.
3. The **significance** of your question. **This answers So what?** It is key to motivating your readers.
4. **Your claim**. This answers your research question expressed in step 2.

(5. New broader significance/application/)

Turabian, Kate L. (2013-04-09). A Manual for Writers of Research Papers, Theses, and Dissertations, Eighth Edition: Chicago Style for Students and Researchers (Chicago Guides to Writing, Editing, and) (Kindle Locations 3274-3289). University of Chicago Press. Kindle Edition.

Cover sheet for Exam part 1 - revision of your own paper.

Please fill in this form and submit this as a cover sheet for your paper

Copy and paste this into your article file, as a cover sheet. Fill in the relevant info.

Participant name: _____

Department/institution: _____

You are required to have discussed this revision with an experienced colleague, preferably your supervisor. If this is not possible then indicate which other senior colleague you have discussed with.

I have discussed my revision process with an experienced senior colleague (at least senior postdoc-level) or my supervisor:

Name of experienced colleague/supervisor _____, and title:_____

or if more than 1, also with: _____

Describe your revision process

- Tell the evaluators about your manuscript, was it a first draft or was it submission ready.
- Inform the evaluators on the process in your group (the group-work tasks and the challenges), what feedback did you receive, how did you proceed, what was your process/focus. Did you work through all the group work challenges or did you proceed with these on your own with your supervisor/senior colleague?

Description of changes, short reasons

When you attempted the group work challenges, how did your revision evolve? E.g. did you change the format of the title? Did you change your writing style: unnecessary words, voice, sentence and paragraph structure, verbs, nominalizations, variation in sentence length? Did you have all the suggested signifiers and text types in your abstract? Was your aim or hypothesis clear? Did you manage to convey that your work matters (and have a "so what" sentence in the abstract and section I the Introduction?). Don't forget to explain why you ended up deciding on the changes.

Draft of your paper: Title, abstract, introduction, references, USE TRACK CHANGES and any necessary comments.

Course Exam part II. Cover sheet and answer form

Name:
Department:
Field of research:

My research is (tick those that apply):

clinical ☐ quantitive ☐ qualitative ☐ hypothesis driven ☐

basal science ☐ clinical intervention ☐ translational research ☐

Tick the appropriate option: I have answered

Article 1 ☐ **Article 2** ☐ **Article 3** ☐

1. Provide brief answers to each of these questions, a few lines:

What is the main subject or topic of this paper?

Which background information and context is provided for the reader?

What problem or problems does this article address?

What is the knowledge gap? *(Knowledge gap refers to the specific unknown)*

What were the aims? (or what were the hypotheses?)

What was/were the key finding(s)?

How were the results obtained? What methods were used?

Do the authors point out any "So whats", if so, which? *(This point is often raised by books on writing scientific articles. "So what"-sentences help the reader understand the implications of the questions that are asked – as in: why should anyone or someone or you care about this research question. Often used in the Introduction*).

What are the main conclusions or "take-home messages"?

What recommendations can be made based on the results? Broader implications or significance? *(This point may be similar to the so-what above, but in an abstract it is placed after having considered the results, the answer to the questions).*

2. Write <u>a <200 word</u> suggestion for an abstract (note the number of words in your contribution). As far as possible, conform with an accepted style, see first part of the exam booklet for examples (Mimi Zeiger's style, Nature publishing group abstract style. Please note which style you have used. Please also label elements with letters A, B, C, … and provide a key beneath the abstract e.g. A: general information, B … C …, etc.

Abstract:

Number of **words**:

Key for my labels in the abstract A: … B: …. Etc

I have used the following **style**:

If not the generic examples mentioned in this booklet, then which style guides have you used?

3.

A. Suggest a title.

B. Is your suggestion descriptive, declarative or interrogative?

Exam part 2 - Option. 1 Article 1 - Cystic Fibrosis

The efficiency of translation termination varies as a function of the sequence context surrounding stop codons in a variety of organisms. To determine whether context effects can cause the functional suppression of disease-causing premature stop mutations in human cells, we assayed for the production of full-length cystic fibrosis transmembrane conductance regulator (CFTR) from cDNAs containing two naturally occurring premature stop mutations that cause cystic fibrosis (CF). The mutations examined introduce an in-frame ochre (UGA) stop codon in place of glycine residue 542 (G542X) or arginine residue 553 (R553X) of CFTR. Each of these mutations occur near the end of the first nucleotide binding domain of CFTR'. HeLa cells infected with vaccinia-T7 were cotransfected with the plasmid vector pTM1 carrying the indicated CFTR allele under T7 promoter control'''. Following transfection of a wild-type CFTR cDNA into vaccinia-T7-infected HeLa cells, CFTR expression was readily observed by immunoprecipitation, and its function was detected with an anion permeability assay utilizing the halide-sensitive fluorophore, 6-methoxy-N-(3-sulfopropyl) quinolinium (SPQ)9. However, we were unable to detect either full-length CFTR or an increase in anion conductance from cells transfected with CFTR cDNAs containing either the G542X or the R553X mutations. This indicates that readthrough of these premature stop mutations (to the extent detectable by these assays) does not occur under normal conditions.

We next examined whether the suppression of these premature stop mutations could be induced by pharmacological treatment. Treatment with a low concentration of aminoglycosides can stimulate the suppression of stop codons in various organisms'''. To test initially whether aminoglycosides can stimulate the suppression of premature stop mutations within the CFTR messenger RNA, we incubated cells transfected with the CFTR R553X construct with different concentrations of the aminoglycoside G-418 for 8-12 hours. We observed a dose-dependent increase in the expression of full-length CFTR from the R553X mRNA as a function of G-418 concentration, indicating that G-418 stimulates readthrough of the R553X mutation (Fig. la). Quantification showed that the amount of full-length CFTR produced was as much as 25% of the level of protein expression obtained from the wild-type CFTR cDNA. Even more full-length CFTR (35% of wild type) was observed in cells transfected with the CFTR G542X cDNA (Fig. lb), indicating that G-418 also promotes readthrough of this second CFTR mutation. G542X is the most common premature stop mutation found in CF patients'''. In contrast, full-length CFTR was not detected in cells expressing a 5' portion of the CFTR R553X cDNA truncated after the codon for amino acid 699 of CFTR. This confirmed that the intact CFTR cDNA was required for expression of the translation product observed upon G-418 treatment.

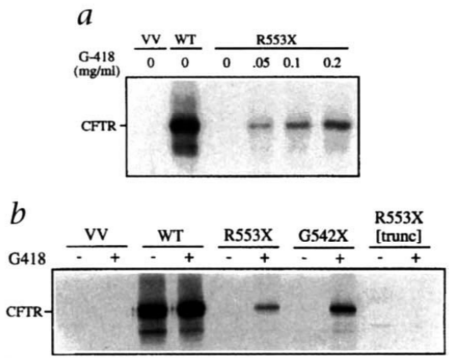


Fig. 1 Expression of full-length CFTR from *the CFTR* G542X and R553X cDNAs in the presence of G-418. a, Dose-dependent translational readthrough of the R553X mutation with increasing G-418 concentration. *b, Suppression of the* G542X and R553X mutations by 0.1 mg/m1 G-418.

Recent studies suggest that the suppression of premature stop mutations occurs through a mechanism of near-cognate mispairing of an aminoacyl-tRNA with the premature stop codon'. Because the amino acid inserted by this mechanism may differ from the amino acid encoded in the wild-type protein, we next used the SPQ assay to determine whether CFTR's function as a cAMP-activated chloride channel was also recovered upon G-418

treatment, cAMP treatment of cells transfected with the wild-type CFTR cDNA caused a rapid increase in SPQ fluorescence (Fig. 2), consistent with stimulation of CFTR-mediated halide efflux. This response required CFTR expression, as no increase in fluorescence was observed when cells infected with vaccinia-T7 alone were treated with cAMP. As discussed above, cells expressing either the G542X or R553X cDNAs in the absence of aminoglycosides showed no cAMP-dependent increase in anion permeability. However, after incubation with G-418, cAMP induced a significant anion efflux in cells transfected with either the G542X or R553X cDNA. This indicates that the full-length CFTR expressed from these mutant constructs following amino-glycoside treatment also functions as a cAMP-stimulated anion channel.

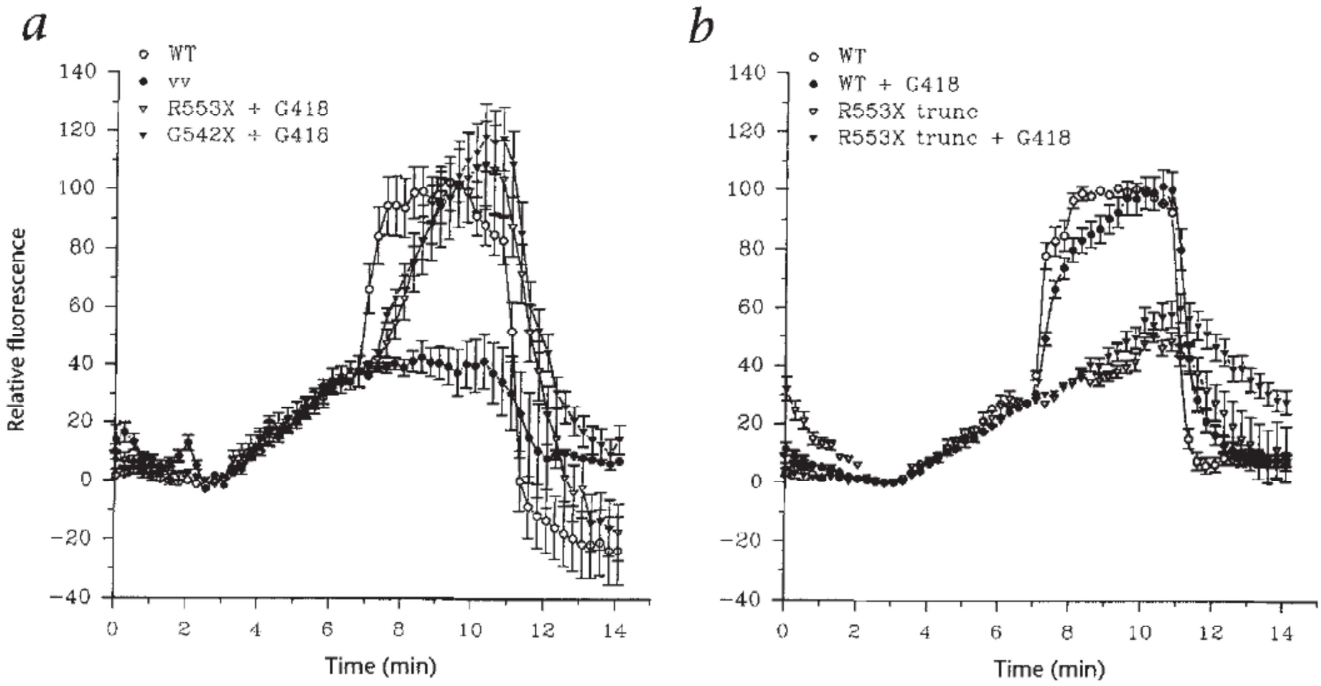


Fig. 2 Functional CFTR expression monitored as cAMP-induced anion efflux using the halide-sensitive fluorophore, SPQ. A cAMP stimulation cocktail was added to the bath at 6 min, and iodide was returned to the bath at 10 min (see Methods section for further details). a, G-418 increases cAMP-stimulated anion channel activity in cells expressing the G542X or R553X cDNAs. b, G418 stimulation of cAMP-stimulated anion channel activity requires an intact CFTR cDNA

To determine whether the truncated forms of CFTR produced by translation termination at either residue 542 or 553 might be activated to a functional state by aminoglycoside treatment, we next asked whether cAMP-activated anion efflux could be induced in cells transfected with the CFTR R553X cDNA truncated distal to the stop codon. We were unable to detect cAMP-dependent anion permeability in cells expressing this truncated cDNA in the presence of G-418 (Fig. 2b). Thus, the portion of the CFTR cDNA distal to the stop mutation is required for restoration of cAMP-activated chloride channel activity, indicating that this activity is attributable to the expression of full-length CFTR.

Currently, some aminoglycosides are aerosolized into the lungs of CF patients to treat bacterial infections. To determine whether these clinical aminoglycosides are also capable of inducing readthrough in human cells, we next used our vaccinia-based readthrough assay system to ask whether two commonly used compounds, tobramicin and gentamicin, could stimulate readthrough of the G542X or R553X mutations in HeLa cells. We were unable to detect full-length CFTR following treatment with tobramicin, but a small amount of full-length CFTR was observed by immunoprecipitation following treatment with gentamicin (Fig. 3). However, we were

unable to reproducibly detect an increase in the cAMP-stimulated anion permeability by SPQ fluorescence, possibly because this assay is not sufficiently sensitive to quantify the small amount of CFTR produced.

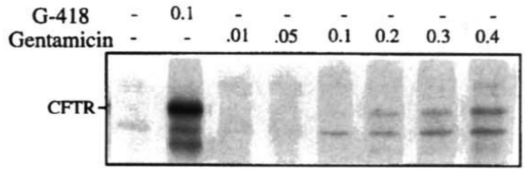


Fig. 3 Dose-dependent stimulation of full-length CFTR synthesis from ***the CFTR*** R553X cDNA with gentamicin. Gentamicin concentrations used are indicated in milligrams per milliliter.

It is possible that a general increase in the suppression of stop codons might lead to the accumulation of toxic, nonfunctional readthrough products. However, in these studies we did not find evidence that low level G-418 treatment significantly impaired normal cellular functions. Exposure of HeLa cells expressing wild-type CFTR to G-418 did not affect the total amount of CFTR synthesized (Fig. lb), their functional response to cAMP stimulation (Fig. 2b), or total protein synthesis rates (data not shown).

Furthermore, it is well documented that relatively efficient suppressor tRNAs that promote readthrough of stop mutations can be maintained in several organisms (including human cell lines) without adverse affects'.

Premature stop mutations account for approximately 5% of the total mutant alleles in CF patients'''. However, in certain subpopulations the incidence of this class of mutation is much higher. For example, the W1282X mutation is the most common CF-causing mutation in the Ashkenazi Jewish population, where it is present on 60% of all CF chromosomes". Our findings raise the possibility that the aerosolized delivery of aminoglycosides to the airway may promote the production of full-length CFTR through the suppression of premature stop mutations in lung epithelia. If successful, this approach would represent the first clinical treatment capable of correcting CF by restoring the expression of functional, endogenous CFTR in a specific genotypic subgroup of CF patients.

Methods

Infections and transfections. HeLa cells were infected with vaccinia-T7 (cTF7-3) at a multiplicity of infection (M01) of 10. Transfection of vaccinia-T7 infected cells with pTM1 carrying the indicated CFTR allele were carried out using lipofectin (BRL) as described'''.

Immunoprecipitation of CFTR. Cells were treated with aminoglycosides for 8 h at the concentrations indicated, then labeled with ["S]methionine for 1 h before cell lysis. CFTR was immunoprecipitated using a monoclonal antibody to the extreme carboxy terminus (Genzyme). The proteins were then resolved by SDS-PAGE and visualized by fluorography.

Halide efflux assays. HeLa cells grown on glass coverslips were loaded for 10 min in a hypotonic iodide buffer containing 10 mM SPQ; iodide quenches SPQ fluorescence'. This hypotonic buffer was made by diluting an isotonic iodide buffer (130 mM Nal, 4 mM KNO„ 1 mM Ca(NO3)2, 1 mM Mg(NO3)2, 1 mM Na2HPO4, 10 mM glucose, 20 mM Hepes, pH 7.4) 1:1 with water. Cells were then returned to the isotonic iodide buffer to recover for 5 min. The experiment measuring SPQ fluorescence was initiated in this same buffer. Nal in the bath was replaced by NaNO, at 2 min; because nitrate does not interact with SPQ fluorescence increases as cell iodide is lost to the bath'. A cAMP stimulation cocktail (10 sM forskolin, 100 uM cpt-cAMP and 100 uM IBMX) was added at 6 min. Fluorescence was then quenched again by returning Nal to the bath at 10 min. Functional CFTR expression was monitored as the dequenching of SPQ fluorescence caused by cAMPinduced iodide efflux.

Truncation of the CFTR cDNA. The plasmid carrying the CFTR R553X cDNA was truncated distal to the premature stop mutation by digestion with EcoRI and Sad. The cohesive ends were removed by treatment with the Klenow fragment of DNA polymerase I and the plasmid was then religated. This treatment removed the CFTR structural gene from the EcoRI site at position 2230 through the Sacl Site at position 4651. This resulted in the loss of the distal 2346 nucleotides of the coding sequence in the CFTR cDNA.

Exam part 2 - Option 2, i.e. article 2. Hypothermia, surgery and transfusion

Introduction

Mild perioperative hypothermia (core temperature 34-36°C) results from intraoperative heat loss and anaesthetic-induced inhibition of normal thermoregulatory control.' Postoperative restoration of a normal core temperature typically requires several hours 2 increasing the duration of hypothermia well beyond the time in surgery. Although intraoperative hypothermia can easily be prevented,3 it remains common;4 no prospective randomised study has shown adverse outcomes as a result of mild hypothermia. In-vitro studies suggest that perioperative hypothermia may aggravate surgical bleeding by impairing platelet function and directly reducing clotting factor enzyme function.5,6 Hypothermia increases the bleeding time, an inhibition apparently related to defective thromboxane Az release, upregulation of platelet surface protein GMP- 140, and downregulation of platelet glycoprotein Ib-IX complex .5 Furthermore, hypothermia prolongs both the prothrombin (PT) and partial thromboplastin (PTT) times-most likely via direct inhibition of clotting factor enzyme function.6 Despite in-vitro evidence that hypothermia impairs coagulation, the extent to which mild perioperative hypothermia increases bleeding during surgery remains unknown. Accordingly we tested the hypothesis that a policy of maintaining normothermia reduces blood loss and allogeneic transfusion requirements during hip arthroplasty. This is a relatively standardised operation associated with considerable microvascular blood loss.

Methods

We evaluated blood loss and transfusion requirements in patients undergoing initial, unilateral total hip arthroplasties at the Hospital of Amstetten, Austria. The study was approved by review boards at the Hospital of Amstetten, the University of Vienna, and the University of California at San Francisco; written informed consent was obtained from participating patients. We studied 60 patients because a preliminary study indicated that this number would provide about an 80% chance of identifying a significant hypothermia-induced increase in blood loss (two-tailed (xO-05). Under the pilot study, 28 patients at the University of Vienna had been randomly assigned to normothermia (Teare 36-6 [0'4]OC, n=13) or hypothermia (Teare 34-9 [0'8]OC, n=15). Blood loss was 2-3 [0'8] and 1-8 [0-61] L in the respective groups (p=007). In this study, patients were aged 40 to 80 yr, had American Society of Anesthesiologists physical status 1-3, and weighed 50-100 kg. None of the arthroplasties was for treatment of tumour. Patients having a history of excessive bleeding or bruising were excluded as were those having PTT of more than 35 s, PT less than 70% clot formation, fibrinogen less than 200 mg/dL, a platelet count less than 100 000/,L, or any contraindication to red-cell scavenging. Patients reporting ingestion of aspirin or non-steroidal anti-inflammatory drugs within 2 weeks of surgery also were excluded. Per surgical protocol, all patients were given low-molecular weight heparin (5000 IU every 8 h) starting 2 h before surgery (Depot Heparin Immuno, Immuno Inc, Vienna, Austria).

Protocol

Ambient temperature was maintained near 21°C. The patients were premedicated with 10 mg oral diazepam 1-2 h before surgery. General anaesthesia was induced by administration of thiopental sodium 3-5 mg/kg, fentanyl 250 lig, and vecuronium 0-1-0-15 mg/kg. The trachea was intubated, and the lungs ventilated

mechanically. Anaesthesia was subsequently maintained with nitrous oxide 60%, isoflurane 0-4-0-8% endtidal concentration, and fentanyl: these drugs were administered in doses sufficient to keep arterial blood pressure within 20% of pre-induction values. The patients were assigned to normothermia (core temperature maintained near 36-5°C) or mild hypothermia (core temperature allowed to decrease to about 35°C). Randomisation was based on computer-generated codes sealed in sequentially numbered, opaque envelopes. Patients assigned to normothermia were actively warmed with an upper-body forced-air cover and a warmer set to "high" (Bair-Hugger, Augustine Medical, Eden Prairie, MN, USA). Additionally, intravenous fluids in these patients were warmed to 37°C. In contrast, active skin and fluid warming was avoided in patients assigned to hypothermia. Target minimum haematocrits were prospectively determined based on ages and cardiovascular status. The target haematocrit was 26% in patients aged less than 65 yr having no significant cardiovascular disease. The haematocrit was allowed to decrease to 28% in patients aged 65 yr or more or having cardiovascular disease. Significant cardiovascular disease was defined as previous myocardial infarction, angina, congestive heart failure, cardiomyopathy, hypertension (a diastolic blood pressure exceeding 90 mm Hg or requiring chronic drug treatment), or peripheral vascular disease. Haematocrit was maintained at 30% or more in patients having both cardiovascular disease and an age 65 yr or more. Preoperative acute normovolemic haemodilution, to a haematocrit of about 30%, was used in all patients. Removed blood was immediately replaced with an equal volume of colloid plasma expander (Haemaccell, Behring Werke AG, Marburg, Germany). Nearly all intraoperative blood loss was scavenged using a Shiley Stat autotransfusion system (Dideco, Mirandola, Italy). The system was primed with 40 000 IU heparin (Immuno Inc) in 1000 mL solution, of which patients received 300 mL. Crystalloid was infused throughout surgery at a rate of 10 mL/kg/h. The first 500 mL estimated blood loss were replaced with additional crystalloid at a ratio of 3 mL/mL blood loss. Additional blood loss was replaced with colloid, haemodilution blood, scavenged red cells, and allogeneic transfusions, if necessary, to maintain target haematocrit. All blood initially removed from the patients during haemodilution and all unused scavenged blood was re-infused by the end of recovery. Allogeneic packed red-blood cells were administered postoperatively as necessary to maintain the target haematocrit.

Measurements

Core temperatures (T core) were recorded from the tympanic membrane (Mallinckrodt Anesthesia Products Inc, St Louis, MO, USA). Intraoperative temperatures, end-tidal PCOz and isoflurane concentrations, heart rates, and oscillometric blood pressures were recorded at 20-min intervals. Postoperatively, temperatures were recorded at 30-min intervals for 2 h. Intraoperative fluid balance was tabulated at 20-min intervals, using aspirated suction volume, return to the cell scavenger, irrigation volume, and blood returned to the patients from the cell scavenger. Surgeons at the Hospital of Amstetten do not routinely use sponges; thus, most shed blood volume could be accurately-and objectively-recorded from aspirated volume. Similar methods have been used in previous studies.' Blood loss from the wound drains was subsequently recorded 3 and 12 h postoperatively, and the following morning. Spun haematocrits were determined at 30-min intervals throughout surgery and used to guide intraoperative fluid and blood administration, as above. Blood haemoglobin concentrations were determined preoperatively, after haemodilution, at the end of surgery, and the next morning. Prothrombin and plasma thrombin times and blood fibrinogen, anti-thrombin 3, platelet, and haemoglobin concentrations were also determined by the clinical laboratory preoperatively, immediately after surgery, and on the first postoperative day. The PT and PTT were determined at 37°C, the PTT is reported in seconds (normal 27-35) and the PT is reported as percent clot formation (normal 70-140). Bleeding times were not measured because the correlation between bleeding time and blood loss in individuals is poor.

Data analysis

Results were analysed after completion of data collection and an audit confirming integrity of the randomisation process. An intention-to-treat analysis was used, ie, patients were considered to be in the temperature group to which they were assigned (even when target temperatures were not reached). 9 Time-dependent results were evaluated using one-way ANOVA with Dunnett's test for comparison to preoperative

values. Results in the two treatment groups were compared using unpaired, two-tailed t tests. The number of patients in each group requiring allogeneic blood transfusion were compared using a Fisher exact test. Data are presented as means (SD); $p<0.005$ identified statistically significant differences.

Results

The morphometric characteristics, duration of surgery, anaesthetic management, and haemodynamic responses were comparable in the two groups. By design, final intraoperative core temperature was approximately 1 ’5°C warmer in the patients assigned to extra warming. Among those assigned to extra warming, final intraoperative core temperature exceeded 36°C in all but one; among the unwarmed patients, all but two had final core temperature less than 36°C. 2 h postoperatively, Tcore remained significantly cooler in the unwarmed patients (table 1).

	Normothermic	Hypothermic
Gender (M/F)	11/19	12/18
Age (yr)	63 (10)	63 (10)
Weight (kg)	79 (13)	75 (13)
Height (cm)	170 (11)	168 (9)
Duration of surgery (min)	85 (31)	87 (24)
Mean arterial blood pressure (mm Hg)	97 (13)	99 (17)
Heart rate (bpm)	65 (12)	64 (10)
End-Tidal PCO_2 (mm Hg)	38 (4)	36 (4)
Isoflurane (%)	0·5 (0·2)	0·5 (0·1)
T_{core} (°C)		
Pre-operative	36·8 (0·3)	36 9 (0·7)
Final interoperative	36 6 (0·4)	35·0 (0·5)*
2 h after surgery	36·9 (0·3)	35·9 (0·6)*

Only final intraoperative and 2-h postoperative core temperatures differed significantly in the two groups (two-tailed, unpaired *t*-tests; *indicates $p<0·05$). Results are presented as mean (SD).

	Normothermic	Hypothermic	p
Haemoglobin (mg/dL)			
Pre-operative	12·7 (1·3)	12·2 (1·2)	NS
After haemodilution	10·9 (1·1)	10·8 (1·4)	NS
End of surgery	10·6 (1·3)	10·4 (1·2)	NS
Next morning	10 8 (1·5)	10·2 (1·3)	NS
Cumulative blood loss (mL)			
End of surgery	690 (230)	920 (400)	0·008
3 h postoperatively	1310 (330)	1700 (510)	<0·001
12 h postoperatively	1500 (310)	1970 (560)	<0·001
Next morning	1670 (320)	2150 (550)	<0·001
Intraoperative fluid administration (mL)			
Colloid (haemodilution)	880 (260)	870 (290)	NS
Crystalloid	2500 (500)	2900 (600)	0·007
Colloid (additional)	80 (173)	217 (303)	0·036
Blood (haemodilution)	450 (320)	470 (340)	NS
Postoperative fluid administration (mL)			
Crystalloid	960 (390)	1150 (290)	0·04
Colloid	12 (19)	40 (12)	<0·001
Blood (haemodilution)	320 (330)	320 (340)	NS
Cell scavenger	400 (240)	520 (260)	0·07
Allogeneic blood			
Patients (no/total)	1/30	7/30	0·06
3 h postoperatively (mL/patient)	10 (55)	60 (120)	0·04
Within 24 h (mL/patient)	10 (55)	80 (154)	0·02

Total blood loss was significantly greater in the hypothermic patients at each measured time (two-tailed, unpaired *t*-tests). The hypothermic patients required more intraoperative crystalloid and heta-starch. Seven of the 30 hypothermic patients required transfusion of 8 units of allogeneic blood within 24 h of surgery, whereas only 1 unit of allogeneic blood was required in 1 of the 30 normothermic patients. The section labelled "postoperative fluid administration" refers to the first 3 h after surgery. Results are presented as means (SD). NS indicates $p \geq 0{\cdot}05$. *P* values for the volume of intraoperative colloid administration and allogeneic blood requirement were virtually identical when calculated using a non-parametric (Wilcoxon) statistic.

Table 2: **Haemoglobin, blood loss, administered fluid, and allogeneic transfusion requirements**

Partial thromboplastin times and blood fibrinogen and anti-thrombin 3 concentrations were normal and similar preoperatively in both groups, and did not change significantly during the study. The prothrombin time decreased significantly from 94 (17)% clot formation preoperatively, to 65 (10)% immediately after surgery, and remained 67 (9)% the following morning. Blood platelet number decreased significantly from 244 000 (59 000)/RL preoperatively to 195 000 (44 000)/IiL immediately after surgery and then to 162 000 (48 000)/ ILL the following morning. However, values in the two groups never differed significantly. Blood loss was significantly greater in the hypothermic patients at the end of surgery, and 3, 12, and 24 h after surgery. Eight units of allogeneic packed red cells were required in seven of the 30 hypothermic patients, whereas only a single normothermic patient required a unit of allogeneic blood ($p < 0.05$ for administered volume). Most blood loss occurred after surgery, and all allogeneic blood was given postoperatively. Initial haemoglobin concentrations were comparable in the two groups, but were somewhat (although not significantly) less in the hypothermic patients on the first postoperative day (table 2).

Discussion

Consistent with in-vitro data,5,6 our results indicate that mild hypothermia significantly increased bleeding. Less than 2°C perioperative core hypothermia increased blood loss about 500 mL (1 unit). Because we used haemodilution and returned scavenged red cells to our patients, relatively few required allogeneic

transfusions. Nonetheless, many more hypothermic patients required allogeneic transfusions and the overall volume transfused was significantly greater in the unwarmed patients.

In addition to its direct cost (excess cost of blood in our hypothermic patients was about$US1050), the administration of allogeneic blood carries risks of infection, transfusion reaction, immune suppression, and may violate religious dictates of some patients. 10

There are three major pathways by which hypothermia might augment surgical blood loss: impaired platelet function, reduced intrinsic and extrinsic clotting, and increased fibrinolysis. Hypothermia significantly impairs platelet function, an inhibition apparently related to defective thromboxane Az release, upregulation of platelet surface protein GMP-140, and downregulation of platelet glycoprotein Ib-IX complex.' The platelet function defect (as assessed by bleeding time) is related to local temperature, rather than core temperature." Wound temperature, however, is largely determined by core temperature and will be distinctly higher in normothermic patients.

The prothrombin and partial thromboplastin times remain nearly normal in hypothermic patients when tested in the usual manner.'2'3 The difficulty with these studies, however, is that the tests were performed at 37°C. There is considerable evidence that both the prothrombin and partial thromboplastin times are highly sensitive to the temperature at which the tests are performed.6,14These tests may, therefore, fail to accurately assess individual clotting potential unless conducted at the patient's temperature. Most likely, both intrinsic and extrinsic clotting is substantially impaired by hypothermia in vivo.

The fibrinolytic system normally regulates the balance between formation of haemostatic plugs and restoration of blood flow after clot formation. The conversion of plasminogen to plasmin is the core of this mechanism, and is largely enhanced by tissue-type plasminogen activator. In contrast to platelet function and the coagulation cascade, fibrinolysis remains normal during mild hypothermia;15 these data suggest that hypothermiainduced coagulopathy does not result from excessive clot lysis.

We used two methods to reduce blood loss in our patients: haemodilution and red-cell scavenging. Surgical bleeding and allogeneic transfusion requirements might have been further reduced had our management included other methods such as regional anaesthesia, deliberate hypotension, or use of autologous blood donated before surgery.

Excessive bleeding is only one reason to maintain intraoperative normothermia. It is well established that mild hypothermia causes postoperative shivering and decreases comfort. Recent evidence suggests that mild intraoperative hypothermia may also predispose toward myocardial ischaemia,16 aggravate surgical wound infections,17,18 and prolong drug action.19,zo Taken together, these factors indicate that surgical patients should be kept normothermic (core temperature =36°C) unless hypothermia is specifically indicated. We suspect that a policy of maintaining normothermia will also decrease blood loss during other surgical procedures.

Exam part 2 - Option 3, i.e. article 3 Multiple sclerosis and perceived disease state

Multiple Sclerosis (MS) is a neurological disease which is associated with a varied symptomatology and uncertain course. Symptoms include numbness, loss of sensation, fatigability, visual impairment, lack of co-ordination, spasticity, bladder and bowel disturbances, impediments to speech, physical imbalance, heat

sensitivity, intellectual disturbances, physical weakness and sexual impairment. However, despite the obvious impact of MS upon the day-to-day lives of persons suffering this disease, insufficient research has been completed with respect to the impact of symptoms upon psycho-social well-being. Instead, most research has focused on the neuropsychology of MS [4, 5]. Research in other areas of psychology related to MS is limited. Some have examined the organic basis for depression and mood disturbance among MS patients [6, 7]. Models of depression, especially the helplessness [8,9] and hopelessness [10] models, suggest that the psycho-social outcomes of MS may interact with depression. Others have focused on the impact of psycho-social issues and "adaptive demands of the disease" [11]. With respect to the latter group, increased disease activity was found to be related to increased emotional disturbance [11, 12]. Matson and Brooks [13] went one step further and proposed a model for adjustment to chronic illness based upon a series of interviews with patients with MS. This model, based on the Kubler-Ross [14] stage model of death and dying, incorporated four stages: denial, resistance, affirmation, and integration. I:ach successive stage was viewed as psychologically healthier than the previous stage. Within this model there is the inherent acceptance of MS as a disease dependent upon the length of time one has been subjected to the disease. More recently, Walsh and Walsh [15] provided support to the validity of this model by reporting that self-esteem in persons with MS varied according to the stage of adaptation.

However, not only is there limited research investigating the psycho-social effects of MS, such research is further restricted by a number of methodological shortcomings. Most important of these is the inclination to treat persons with MS as an homogenous group. This is evidenced by a brief look at the literature. For example, while Wassem [16] reported that severity of MS significantly influenced adjustment, the sample was regarded as homogenous according to rating of disability. Similarly, Walsh and Walsh [17] did not distinguish between the differential characteristics of their sample in their investigation of the relationship of love to self-esteem in persons with MS. Instead, they assumed that the response rate to their mail-out of questionnaires was high enough to avoid significant biases. Finally, Zeldow and Pavlou [18] gave token appreciation to the notion that differential disease state impacts upon psycho-social well-being but did not specifically attend to the problem.

The population of persons with MS is composed of several distinct sub-groups according to differentiated disease state. Yet studies not accounting for such differences often generalise results to the population of persons with MS. Specifically, this study attends to the heterogeneity of symptoms associated with MS and examines subjects in separate groups according to disease state. This supports the notion of Rudick et al. [19] that diseases are characterised by different profiles, and goes further to acknowledge that each state of MS may be differentially characterised by unique psycho-social profiles. Along with the problems associated with the treatment of persons with MS as an homogenous group, most studies have also been restricted to relatively small numbers of subjects. For example, Beulow [20] used only 20 hospitalised MS patients as subjects while Walsh and Walsh (I 989), in one of the more expansive studies, accessed 135 MS patients as subjects. Beulow acknowledged difficulties associated with small sample sizes and suggested that "a larger sample size with greater variability of disability might more accurately describe the stress of disabilities" [20] (p. 251). Small sample sizes, particularly with clinical groups, tend not to be as representative of the larger population and therefore generalisation of results to the larger group is limited. The present study examines the psycho-social ramifications of MS using a relatively large sample of persons, wherein "psycho-social" refers to the social and emotional aspects of everyday life, in particular, how individuals interpret their well-being with respect to everyday life in regard to their ability to handle social and emotional life on a daily basis. Specifically, the study attends to the heterogeneity of symptoms associated with MS and examines subjects in separate groups according to disease state. The two disease states examined include persons in the chronic progressive state of the disease and those experiencing remission*. The progressive form of MS is characterised by steadily worsening symptoms and is more common later in the disease course. However, in rare cases, MS can present as a severely progressive form from the outset. The remitting/relapsing form of the disease is characterised by erratic, periodic onset or worsening of symptoms. Remission is the cessation of these erratic exacerbations. There are distinct disease states in MS and these states may be differentially

associated with different symptoms, experiences, and psycho-social ramifications. Therefore, it is appropriate that one should consider disease state as an important predictor of psycho-social well-being. The present study addresses these issues and enhances the work of previous researchers. It is hypothesised that attitude to MS is indicative of adaptation and, therefore, varies according to disease state. Specifically, this study attempts to clarify the profile of persons with MS by examining adaptation to disease according to perceived disease state.

METHOD

Subjects Subjects (N = 243) were obtained through the Multiple Sclerosis Society of Victoria (MSSV), Melbourne, Australia, data base and were formally diagnosed as having MS. The sample consisted of 180 females (74.1%) and 63 males (25.9%) and ranged in age from 11 to 83 years (M = 48.09, SD = 13.39). Most lived at home with others (N = 196, 80.7%) and were taking medication (N = 138, 56.8%). With an overall population of persons with MS (registered with the MSSV) of 1590, the sample size achieved is consistent with the needed sample size at ct = 0.05 [21]t. Subjects were divided into two diagnostic groups consisting of those experiencing either (a) remission (N = 108) or the (b) chronic progressive form of the disease (N = 135). Patients' subjective experience of symptoms was used as the basis for inclusion into one or the other of the diagnostic categories$. This was completed by subjects answering the following question:

My MS is
chronic progressive []
In remission []
Experiencing relapse []
Benign []

Apparatus and procedure Data was obtained via a questionnaire (Appendix) constructed according to procedures determined by Fishbein and Azjen [! 7] and based upon their Theory of Reasoned Action. The Fishbein and Azjen model incorporates two components§, attitude and subjective norm, as the basis for predicting the intention to perform a specific behaviour. While the questionnaire developed for the current study (Appendix) elicited data on both components of this model, the current study is primarily concerned with the attitudinal

component. In this instance "a person's attitude towards a specific behaviour is proposed to be a function of the perceived consequences of performing that behaviour and of the person's evaluation of those consequences" [18]. Therefore,

$$A_B = \Sigma b_i e_i$$

where b is the belief that performing B* leads to the consequence or outcome i and e is the person's evaluation of outcome i. During the first stage of the questionnaire development a pilot group of participants was asked to list the advantages and disadvantages, and general ideas they held in relation to their MS (salient beliefs). To identify salient beliefs a content analysis was performed. Selecting the beliefs totalling 75% of all beliefs recorded provided 11 for inclusion in 1:he questionnaire. The selected salient beliefs were:

1. I am frustrated because I can't do what I used to do or want to do.
2. I will have to cope with symptoms.
3. I won't be able to work.
4. I would like more information and better progress in research.
5. I won't be able to get around.
6. I will be isolated and lonely.
7. I will be bored because I won't have anything

to do.
8. I will make new friends.
9. I will feel depressed.
10. I will be dependent on others.
I I. I found God.

Thus the inclusion of particular attitudinal beliefs is grounded in the data resulting from the elicitation process. It can be seen that there is unlimited potential in the process for participants to include any issues or attitudes and that these may include attitudes manifested in several ways (e.g. humour, spirituality). However, the process is such that the resulting questionnaire can only include those dimensions of attitudes which arise from the elicitation data. Participants have the opportunity to express either positive or negative attitudes to each of the salient beliefs. These salient beliefs formed the basis for items pertaining to attitude on the questionnaire. This attitudinal component was measured by seven-point bipolar scale closed-format questionnaire items and subjects were asked to respond toward beliefs associated with MS. For example, the relevance cf a specific belief with respect to a consequence or outcome was measured according to the subject's response to the following question:

I will be depressed
likely____:____:____:____:____:____:____unlikely
extremely quite slightly neither slightly quite extremely

The subject's evaluation of this belief was measured according to the response to the following question:

Being depressed is
good ____ : ____ : ____ : ____ : ____ : ____ : ____ bad
extremely quite slightly neither slightly quite extremely

A mail-out of questionnaires was completed in four waves during the months of July, August and September, YEAR with a useable questionnaire return rate of 59%. 31 questionnaires were completed as structured interviews due to the physical incapacities of some subjects and the current reduced staffing levels in some facilities.

Data analysis

The dependent variables, attitudinal beliefs associated with MS, were measured by summating the products of the strength of a behavioural belief multiplied by the evaluation of the outcome of this behavioural belief. The overall measure of attitude was completed by summating each of the measures of attitudinal belief. The independent variable, disease state, consisted of two levels: remission and chronic progressive. Multivariate analysis of variance (MANOVA) and multiple discriminant analysis (MDA) were the principal inferential techniques. MANOVA was used to indicate overall group differences between the predictor groups. MDA was used in conjunction with MANOVA to determine the direction and intensity of each criterion variable's impact on overall group differences. In addition, an independent measures t-test was used to test for differences between levels of the independent variable in relation to the overall measure of attitude.

RESULTS

Means and standard deviations for modal salient behavioural beliefs (Table 1) indicate differential strengths associated with the various beliefs. The central hypothesis contends that attitude to MS varies according to disease state. Specifically, persons in the chronic progressive state of MS are expected to differ in their attitudes in comparison to persons experiencing remission. As reported in Table 2, the multivariate F-ratio of MANOVA is significant at the 0.000 level, indicating overall differences between the 2 groups. The subsequent univariate ANOVA procedures and MDA reveal that at the 0.05 level, the groups are significantly different on 6 of the !1 behavioural beliefs. The group means and the discriminant loadings for these 6 beliefs are supportive of the hypothesis. The differences in means are quite large and the effect sizes are between 2% and 7%. In addition, the predictive validity of the discriminant function was assessed by comparing the hit ratio (70.04%)

with the proportional chance criterion (50.56%). To have confidence in the predictive validity of the MDA function, the classification accuracy reflected in the hit ratio is

Table 1. Means and standard deviations of modal salient beliefs

	Outcome evaluation		Behavioural belief		Product†	
Behavioural belief	M	SD	M	SD	M	SD
Frustration with daily activities	2.1	1.1**	− 1.3	2.0**	− 3.3	5.1**
Coping with symptoms	1.7	1.8**	− 0.7	2.3**	− 2.0	5.8**
Ability to work	1.8	1.5**	− 0.3	2.6	− 0.6	6.4
Research into MS	0.3	2.0*	− 0.3	2.1*	2.6	4.0**
Information on MS	1.4	1.3**	− 0.1	2.0	− 0.5	4.4
Personal mobility	0.8	2.3**	− 0.8	2.4**	0.3	6.4
Isolation and loneliness	1.5	1.5**	0.9	2.0**	− 0.7	4.9
Activity and non-boredom	1.8	1.7**	0.3	2.3*	1.2	5.9**
Developing friendships	1.3	1.5**	0.0	2.1	− 0.0	4.3
Feeling depressed	2.0	1.3**	0.1	2.1	1.0	5.1**
Comfort of religious beliefs	1.2	1.4**	− 0.6	2.0**	− 2.7	3.9**

†Refers to the product of the multiplication of the outcome evaluation by the strength of the behavioural belief over all subjects.
**Significantly different from zero ($P \leqslant = 0.0001$).
*Significantly different from zero ($P \leqslant = 0.05$).

more precise if it is at least 25% higher than the proportional chance criterion (i.e. 63.20% or higher). Because this criterion is met, the research hypothesis is supported. Persons in the chronic progressive state of MS, in comparison to persons experiencing remission, were significantly more negative in their frustration with completing daily activities, coping with the symptoms of MS (e.g. memory loss and fatigue), belief with respect to their ability to work, personal mobility, isolation and loneliness, and being active and not bored. Finally, an assessment of differences between groups associated with the overall estimate of attitude was completed. Means and standard deviations for the estimates of overall attitude in each disease state (Table 3) indicate both positive and negative scores of attitude in each disease state. The most positive scores were associated with remission and the least positive scores were associated with those experiencing the chronic progressive form of the disease. An independent measures t-test for attitude by disease state indicated that attitude differed significantly according to disease state [t(225)= -6.91, P< 0.000]. The test indicated that subjects in remission had significantly more positive attitudes toward their disease state than subjects in the chronic progressive state of the disease.

Table 2. MANOVA and MDA results for differences in attitudinal beliefs across patients with MS experiencing remission vs patients with MS defined as chronic progressive

Attitudinal beliefs	*F*-Ratio	*P*-value	η^2	Discriminant* loadings	Group means: Experiencing remission	Group means: Chronic progress
Frustration with daily activities	16.0	0.000	0.07	0.517	−1.84	−4.47†
Coping with symptoms	14.0	0.000	0.06	0.484	−0.46	−3.31†
Ability to work	4.3	0.039	0.02	0.268	0.44	−1.34†
Research into MS	0.1	0.724	0.00	−0.045	2.47	2.66
Information on MS	3.0	0.085	0.01	0.224	0.04	−0.97
Personal mobility	13.0	0.000	0.06	0.480	1.97	−1.16†
Isolation and loneliness	4.4	0.036	0.02	0.272	1.52	0.16†
Activity and non-boredom	14.9	0.000	0.06	0.498	3.09	0.14†
Developing friendships	0.0	0.926	0.00	−0.012	−0.17	−0.11
Feeling depressed	0.1	0.767	0.00	0.038	1.17	0.96
Comfort of religions beliefs	1.5	0.213	0.00	0.161	−2.27	−2.90
Multivariate significance level		0.000				
% *correctly classified* (*hit ratio*)						
Analysis group				70.04		
Maximum chance criterion				55.94		
Proportional chance criterion				50.56		
Press's Q				21.58		

*Pooled within-groups correlations between discriminating variables and canonical discriminant functions.
†Significant difference in attitudinal belief between patients with MS experiencing remission and patients within the chronic progressive state. A negative value indicates a negative assessment of the attitudinal belief while a positive value indicates a positive assessment of the attitudinal belief.

Table 3. Means and standard deviations for overall estimates of attitude for each disease state

Disease state	M	SD	N
Chronic progressive	−10.3	19.76	127
Remission	6.0	15.73	100

DISCUSSION

Changes in attitude in persons with MS were associated with different disease states. Differences within factors for each disease state indicated varying characteristics between each disease state. Those experiencing the chronic progressive state are likely to have had the disease for the longest time. This suggests that those in the chronic progressive group should be better adapted and therefore report the most positive overall attitude*. This was not the case in the present study. Thus, the current findings suggest that factors other than duration since onset are important in the psycho-social profile of persons with MS.

The psycho-social profile of persons with MS presented in the literature and available to professionals and patients tends to be inordinately skewed and presents an unduly negative picture for the newly diagnosed. However, the results indicated that persons with MS were not homogenous in terms of their psycho-social characteristics. This disease consists of separate disease states recognised by differing psycho-social profiles. Acknowledging the varying characteristics of each disease state provides a more accurate picture of the psycho-social symptomatology of this disease for the benefit of both doctors and patients.

Importantly, this picture of the nature of the disease is not the negative one produced when MS patients are treated and reported as a homogenous group. The identification and recognition of different psycho-social profiles for each disease state provides optimism in those patients who are newly diagnosed and for doctors faced with informing patients of the diagnosis of MS. Furthermore, an examination of the perceptions of

patients is a fertile ground for understanding the impacts of chronic disease and for enabling patients to gain insights and develop strategies which will enhance day-to-day emotional and social well-being.

Finally, this study provides a useful springboard for future study. It is limited to the extent that it examines the perceptions of patients toward their disease state within a confined period of time. Future studies would be more informative if they were longitudinal in nature, for example, a study of patients and their attitude as they experience the various disease states and stages over time.

Exam PART 3 Graphical Abstract

A **Graphical Abstract** is a single, concise, pictorial and visual summary of the main findings of an article or a poster.

This could either be the concluding figure from the article/poster or a figure that is specially designed for the purpose, which captures the content of the article/poster for readers at a single glance.

A Graphical Abstract should allow readers to quickly gain an understanding of the **main take-home message** of the paper and is intended to encourage browsing, promote interdisciplinary scholarship, and help readers identify more quickly which papers are most relevant to their research interests.

Cell Press and Elsevier journals are examples of publishing houses that use this format in published papers.

A graphical abstract can be very useful communications tool on **Posters** and be a useful **eye catcher**.

https://www.elsevier.com/authors/journal-authors/graphical-abstract

Make a graphical abstract for one of the exam texts that you have used (paper 1, 2, 3- Not your own paper! or Option 4, 5 and 6 below).
Upload a Tif picture file in the resolution of **3000 x 3000 pixels.** This should have the format "MyNameFirst-Surname.tif"

Format: Upload the illustration as a Tif file with the following resolution: 3000 pixels x 3000 pixels (square), so that the figure can be shown at 600DPI in a 5 inch + 5 inch format.

Also upload the cover sheet in your worldfile (see next page):

About my graphical abstract "MyNameFirst-Surname.tif"

I chose paper 1/2/3/option 4/5.
I have decided to convey this take home message: _____ I therefore have illustrated ____ I use these devices to convey my message _____ (drawing, color, arrows, pictures, font serif/sans serif-typography)
Other comments, difficulties/dilemmas/choices? _____

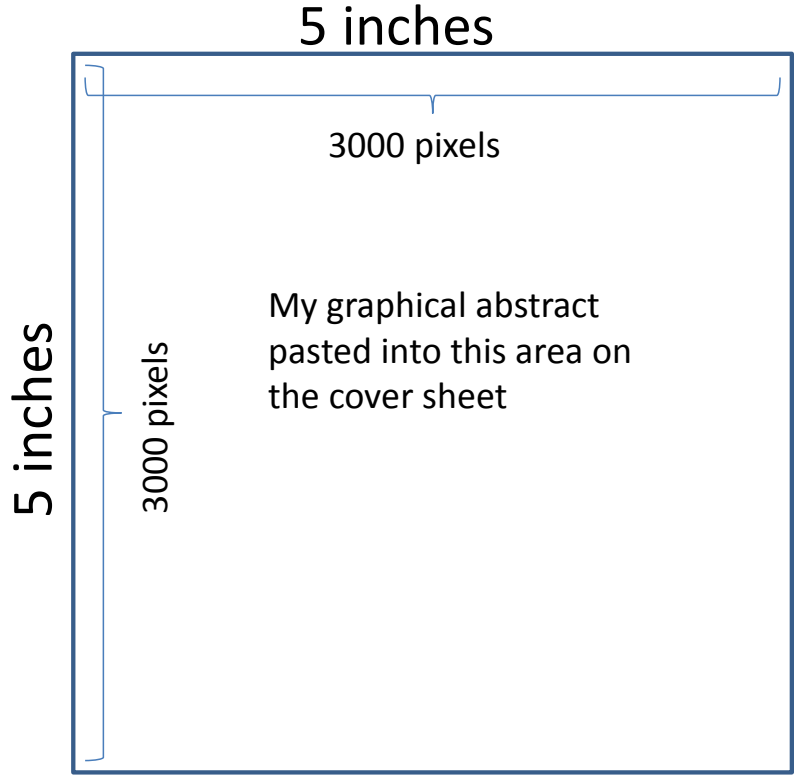
Tools: You can chose to use Servier Medical art powerpoint picture (see Fronter) – where you can pick and modify elements by ungrouping illustrations.

Copy Right: As you are not publishing this work – just using it as teaching tools, you can use material from the internet as long as you include the source

COVER SHEET FOR THE GRAPHICAL ABSTRACT- EXAM PART 3

MY NAME
FILENAME: *.tif**

Insert from file (or paste) your Tif file here, i.e. Replace the figure below with your own graphical abstract (Make sure that you also upload your Tif file to Fronter. If you paste the image (and not insert from file) then make sure that the file is represented properly with an appropriate dpi resolution):



About my graphical abstract

I chose paper 1/2/3/ option 4/5/6.

I have decided to convey this take home message: _____ I therefore have illustrated ____ I use these devices to convey my message _____ (drawing, color, arrows, pictures, font serif/sans serif- typography)

Other comments, difficulties/dilemmas/choices? _____

Advice from Cell press - The graphical abstract should:

Have a clear start and end, "reading" from top-to-bottom or left-to-right

Provide a visual indication of the context of the results depicted (for example tissue, subcellular location, tissue or cell type, species, etc.)

Emphasize the new findings from the current paper without including excess details from previous literature

Avoid the inclusion of features that are more speculative (unless the speculative nature can be made apparent visually)

Not include data items of any type; all the content should be in a graphical form

KEEP IT SIMPLE

The graphical abstract should:

Use simple labels

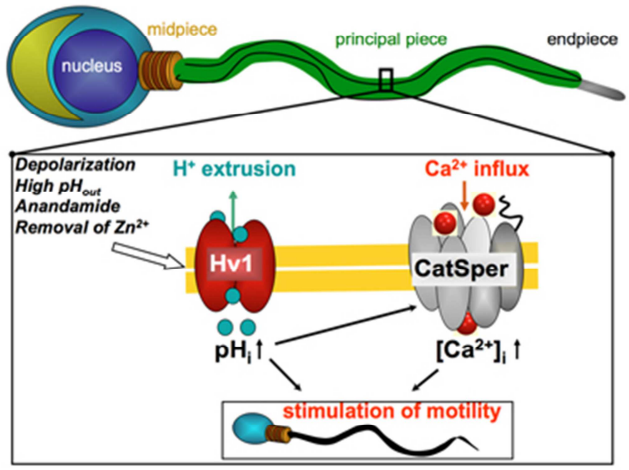
Use text sparingly

Highlight one process or make one point clear

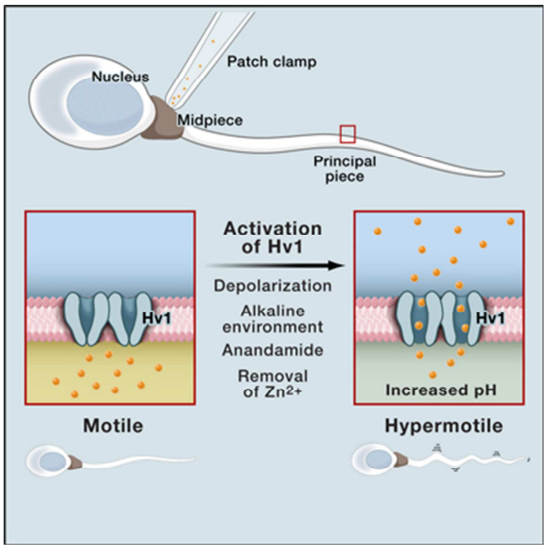
Be free of distracting and cluttering elements

Examples of graphical abstracts that have been modified by professional illustrators (Cell press)

Before



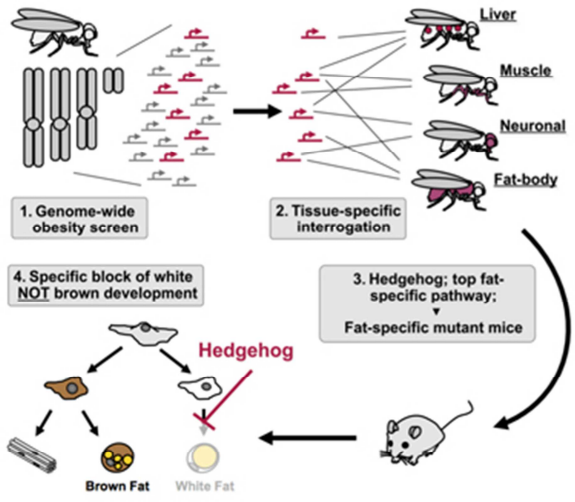
After



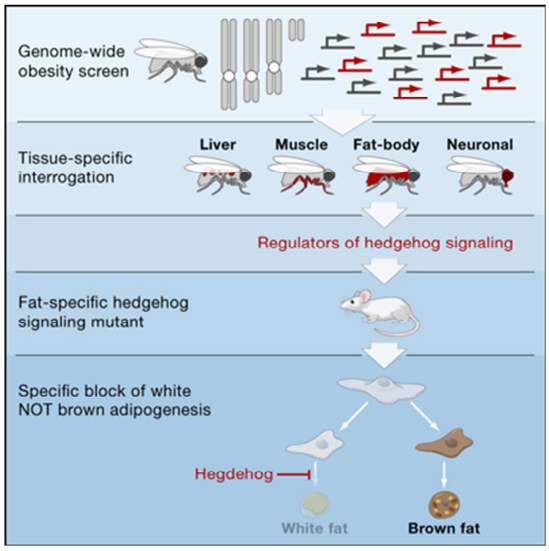
EXAMPLE 1

The image's components have been reoriented to tell the story from left to right. Some arrows and text were removed for simplicity. The color palate was softened. The paper's take-away message and new findings ("Activation of Hv1") were set as the focal point of the abstract.

Before



After



EXAMPLE 2

The image's components have been reoriented and condensed to tell the story from top to bottom. The colors have been adjusted to highlight and direct focus toward the most relevant information. See also pinterest (“graphical abstract”), Cell press or Elsevier.

In addition to article 1-3, you can choose from these three options om
Option 4:
https://www.nature.com/articles/ncomms6621

The antimicrobial peptide LL37 is a T-cell autoantigen in psoriasis

Psoriasis is a common T-cell-mediated skin disease with 2–3% prevalence worldwide. Psoriasis is considered to be an autoimmune disease, but the precise nature of the autoantigens triggering T-cell activation remains poorly understood. Here we find that two-thirds of patients with moderate-to-severe plaque psoriasis harbour $CD4^+$and/or $CD8^+$ T cells specific for LL37, an antimicrobial peptide (AMP) overexpressed in psoriatic skin and reported to trigger activation of innate immune cells. LL37-specific T cells produce IFN-γ, and $CD4^+$ T cells also produce Th17 cytokines. LL37-specific T cells can infiltrate lesional skin and may be tracked in patients blood by tetramers staining. Presence of circulating LL37-specific T cells correlates significantly with disease activity, suggesting a contribution to disease pathogenesis. Thus, we uncover a role of LL37 as a T-cell autoantigen in psoriasis and provide evidence for a role of AMPs in both innate and adaptive immune cell activation.

Option 5:
http://www.nejm.org/doi/full/10.1056/NEJMc1703047

Reversal of Autoimmune Toxicity and Loss of Tumor Response by Interleukin-17 Blockade

N Engl J Med 2017; 376:1989-1991May 18, 2017DOI: 10.1056/NEJMc1703047

Patients with autoimmune diseases have typically been excluded from immunotherapy clinical trials, although recent evidence suggests that the benefits of checkpoint therapy may outweigh the risks of autoimmune flare-ups in carefully selected patients who have a high probability of response.1

We report the case of a 50-year-old man who had metastatic colon cancer with a mismatch-repair deficiency and who had been treated with third-line pembrolizumab after disease progression while he was receiving FOLFOX (folinic acid, fluorouracil, and oxaliplatin) and FOLFIRI (folinic acid, fluorouracil, and irinotecan). The patient had mild psoriasis, for which he received topical treatment, and Crohn's disease, which had been treated with small-bowel resection and ileostomy in 2012. He was not receiving glucocorticoids or other immunomodulating agents at the start of his treatment with pembrolizumab. Written informed consent was obtained for the use of a checkpoint inhibitor in a patient with autoimmune disease.

The patient had a severe adverse reaction after the third cycle of pembrolizumab, with a psoriatic rash involving more than 75% of the body-surface area, including on the hands and legs (Figure 1A and 1B Reversal of Autoimmune Toxicity and Tumor Response)

The rash was associated with intense arthralgia and pruritus. He also had increased abdominal pain, which was associated with up to six stools per day and occasional bloody output in his ileostomy bag. Given the predominance of his skin symptoms, in consultation with dermatologic specialists, the patient received once-weekly subcutaneous administration of 150 mg of secukinumab, a human monoclonal antibody that selectively binds to circulating interleukin-17A cytokine and inhibits its interaction with the interleukin-17 receptor. After four doses, there was complete resolution of the skin psoriasis (Figure 1C and 1D) and the gastrointestinal symptoms. Pembrolizumab was then continued

for another two cycles, in conjunction with the continuation of the same once-monthly dose of secukinumab. The patient had initially had a biochemical response of 50% in the level of carcinoembryonic antigen, which was lost after the introduction of secukinumab (Figure 1E); progression was confirmed by means of two serial computed tomographic scans.

Although there is some evidence from murine models that proinflammatory cytokines that are secreted by helper T-cell type 17 (Th17) cells can promote tumor growth and metastasis,2 other studies in mice and humans have suggested that Th17 cells and interleukin-17 enhance tumor surveillance and immunity.3-5 In our patient, the inhibition of the interleukin-17 pathway was associated with tumor growth and escape from immune attack.

In conclusion, interleukin-17 blockade provided dramatic relief of immune-mediated skin and gastrointestinal toxic effects. The subsequent loss of antitumor efficacy suggests that interleukin-17 may play a role in the antitumor effects of checkpoint inhibitors such as pembrolizumab.

Option 6

http://www.sciencedirect.com/science/article/pii/S027795361730031X?via%3Dihub

Hour-glass ceilings: Work-hour thresholds, gendered health inequities

https://doi.org/10.1016/j.socscimed.2017.01.024

Abstract

Long workhours erode health, which the setting of maximum weekly hours aims to avert. This 48-h limit, and the evidence base to support it, has evolved from a workforce that was largely male, whose time in the labour force was enabled by women's domestic work and care giving. The gender composition of the workforce has now changed, and many women (as well as some men) combine care-giving with paid work, a change viewed as fundamental for gender equality. However, it raises questions on the suitability of the work time limit and the extent it is protective of health. We estimate workhour–mental health thresholds, testing if they vary for men and women due to gendered workloads and constraints on and off the job. Using six waves of data from a nationally representative sample of Australian adults (24–65 years), surveyed in the Household Income Labour Dynamics of Australia Survey (N = 3828 men; 4062 women), our study uses a longitudinal, simultaneous equation approach to address endogeneity. Averaging over the sample, we find an overall threshold of 39 h per week beyond which mental health declines. Separate curves then estimate thresholds for men and women, by high or low care and domestic time constraints, using stratified and pooled samples. We find gendered workhour-health limits (43.5 for men, 38 for women) which widen further once differences in resources on and off the job are considered. Only when time is ‘unencumbered’ and similar time constraints and contexts are assumed, do gender gaps narrow and thresholds approximate the 48-h limit. Our study reveals limits to contemporary workhour regulation which may be systematically disadvantaging women's health.