

Poor Devils: The Plight of the Tasmanian Devils

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Part I – Preliminary Research Jigsaw

This case is about an animal that is facing extinction. A previously unknown disease, which looks a lot like cancer, is decimating the population. This is alarming for many reasons, but understanding its cause also offers hope for conservation. Before you attempt this case study of the plight of an endangered species, you must do some research. You will work in teams of four. There are four research topics below. You should assign one topic to each member of your team. Your goal is to research your assigned topic and answer the questions. You might wish to use the suggested references to guide your research. Each student's research will be written up and shared among the other members of the team. You should therefore bring printed Research Reports for each member of your team next time you meet (next class).

You must submit the result of your research in the form of a Research Report. The format of a Research Report is as follows.

Name: Your complete name

Team: Names of all of the members of your team

Topic of Your Research: (e.g., Immune System, Cancer, Infectious Diseases, or Cytogenetics)

Question 1: Copy the First Question

Provide your answer. There are a few things you should keep in mind:

- Write in complete sentences.
- Write a complete answer to your question, including any information that is relevant. Be sure to define new or potentially difficult terms.
- You must write the information in your own words—not copy and paste from another source.
- You must judge the length of a response that is appropriate. Depending on the question, it may be a sentence, a paragraph, a page, or more.
- You can include a drawing, table, a diagram, or other figure, but it must be made by you, not copied from another source. Be sure to cite what other sources inspired your own creation, if appropriate.
- You must cite all your sources of information using APA source formatting (i.e., include in-text references and a proper References section at the end of your response). Each Question must have its own References section. You may *only* use trusted sources of information, i.e., textbooks, books of an academic nature, peer-reviewed journal articles. Use newspaper and magazine articles (e.g., *New York Times* or *Scientific American*) sparingly. For websites, you may use only two sources: government websites (e.g., the National Institutes of Health website) and websites created by academics (i.e., website sponsored by a university professor).

Repeat for each of the questions on your topic.

Example

Question 1: What are mitochondria?

Mitochondria are structures found within a cell that are bounded by a double lipid membrane (Freeman, 2005a). They have their own DNA, RNA, and protein, and were proposed by Lynn Margolis in the 1960s to be the descendants of bacteria that were engulfed or otherwise came to live in symbiotic relationship with the larger eukaryotic cell (this is called the Endosymbiotic Theory of mitochondrial evolution) (Margulis, 1981). They are the main site of cellular respiration, i.e., they are involved in the extraction of energy from sugars and in the production of ATP molecules (the energy currency of the cell) (Freeman, 2005b). Mitochondria are present in the cell but very few exist in the sperm. Consequently, humans inherit their mitochondria only from their mother. This can be used to trace back ancestry in the maternal line and has been used to identify mitochondrial Eve, the woman who is an ancestor shared by everyone alive today (Micklos, Freyer, & Crotty, 2003).

References

- Freeman, S. (2005a). Chapter 7: Inside the cell. In *Biological science* (2nd ed., pp. 128–158). Upper Saddle River, NJ: Pearson Prentice Hall.
- Freeman, S. (2005b). Chapter 9: Cellular respiration and fermentation. In *Biological science* (2nd ed., pp. 177–201). Upper Saddle River, NJ: Pearson Prentice Hall.
- Margulis, L. (1981). *Symbiosis in cell evolution*. New York, NY: WH Freeman.
- Micklos, D. A, Freyer, G. A, & Crotty, D.A. (2003). Chapter 8: Applying DNA science to human genetics and evolution. In *DNA science: A first course* (2nd edition, pp. 259–314). Cold Spring Harbor, NY: Cold Spring Harbor Laboratory Press.

Research Topics

Here are the four research topics and the questions associated with each.

Student 1: Infectious Diseases

- What are the causative agents of communicable diseases? (Identify at least four different types of infectious agents.)
- What are the characteristics of each infectious agent?
- How are infectious agents spread between organisms? Specifically, what are the differences between contact transmission, vehicle transmission, and vector transmission?
- What factors influence the introduction and transmission of communicable diseases (and how do they have an effect)?
- How do infectious agents replicate? What is their life cycle?
- How do we know that an organism is the infectious agent for a disease? What are Koch's postulates?
- How can infectious agents cause cancer? (Note: you may wish to consult your teammate working on cancer.)

Suggested References

Any chapter in a first year biology textbook on the topic of bacteria and viruses.

- Brock, T. D., Madigan, M.T., Martinko, J. M., & Parker, J. (1994). Chapter 6: Viruses. In *Biology of microorganisms* (7th ed., pp. 183–236). Englewood Cliffs, NJ: Prentice Hall.
- Phelan J. (2010). Chapter 13: Evolution and diversity among the microbes. In *What is life: A guide to biology* (pp. 482–513). New York, NY: WH Freeman.
- Alters, S., & Alters, B. (2006). Chapter 19: Viruses and bacteria. In *Biology: Understanding life* (pp. 307–323). Danvers, MA: John Wiley and Sons.
- Freeman, S. (2005). Chapter 34: Viruses. In *Biological science* (2nd ed., pp. 780–802). Upper Saddle River, NJ: Pearson Prentice Hall.

Sadava, D., Hillis, D. M., Heller, H. C., and Berenbaum, M. R. (2011). Chapter 26: Bacteria and archaea: The prokaryotic domains. In *Life: The science of biology* (9th ed., pp. 536–559). Gordonville, VA: WH Freeman.

Bauman, R.W. (2011). Chapter 14: Infection, infectious diseases, and epidemiology. In *Microbiology with diseases by taxonomy* (3rd ed., pp. 401–434). Boston: Benjamin Cummings.

Student 2: Immune System

- How do organisms protect themselves against infectious agents? What are innate and adaptive immunity?
- What is a T lymphocyte?
- What different roles do B cells, T helper cells, and cytotoxic T cells play in immunity?
- What is the difference between humoral and cellular immunity?
- What is an MHC complex? What is its purpose?
- What is an HLA gene?
- How can organisms produce a defensive response to an almost infinite number of infectious agents?
- Why are organ grafts often rejected?
- How can the immune system help fight cancer? (Note: you may wish to consult your teammate working on cancer)

Suggested References

Any chapter in a first year biology textbook on the topic of the immune system.

Goldsby, R. A., Kindt, T. J., Osborne, B. A., & Kuby, J. (2003). Chapter 1: Overview of the immune system. In *Immunology* (5th ed., pp. 1–23). New York, NY: WH Freeman.

Campbell, N. A., & Reece, J. B. (2002). Chapter 43: The body's defenses. In *Biology* (6th ed., pp. 900–924). San Francisco: Benjamin Cummings.

Freeman, S. (2005). Chapter 49: The immune system in animals. In *Biological science* (2nd ed., pp. 1120–1142). Upper Saddle River, NJ: Pearson Prentice Hall.

Sadava, D., Hillis, D. M., Heller, H. C., & Berenbaum, M. R. (2011). Chapter 42: Immunology: Animal defense systems. In *Life: The science of biology* (9th ed., pp. 873–898). Gordonville, VA: WH Freeman.

Student 3: Cancer

- What is the difference between a tumor and cancer?
- What are the characteristics of cancerous cells? How are they different to normal cells?
- What are the factors that can cause cancer? By what mechanism do they cause cancer?
- How is cancer spread? Is it an infectious disease?
- What is different about the genes expressed in a cancer cell relative to a healthy cell?
- If you were presented with two cells, one which was cancerous and one which was not, how could you tell the two apart?
- Is cancer a homogeneous disease? In other words, if I take a breast cancer cell from one person and a breast cancer cell from another person, will the cancer cells be identical? What about different types of cancer (e.g., a lung cancer cell and a skin cancer cell)? What is similar and different about the cancer cells in each case?
- What is evolution by natural selection? What are the necessary steps required for this to occur?
- How is cancer an echo of evolution by natural selection?
- How can infectious agents cause cancer? (Note: you may wish to consult your teammate working on infectious agents)

Suggested References

Any chapter in a first year biology textbook on the topic of cell cycle, mitosis, or cancer.

Emory University (nd). *The Tumor Biology Series*. Retrieved on 10 January 2010 from <http://www.cancerquest.org/images/Documentary/English/DocInterfaceEng.swf>

Lane, N. (2005). Part 5: Murder or suicide: The troubled birth of the individual (Introduction and Chapter 11:

Conflict in the body). In *Power, sex, suicide: Mitochondria and the meaning of life* (pp. 189–214). Oxford: Oxford University Press.

Micklos, D. A., Freyer, G. A., & Crotty, D. A. (2003). Chapter 7: The DNA science of cancer. In *DNA science: A first course* (2nd ed., pp. 221–258). Cold Spring Harbor, NY: Cold Spring Harbor Laboratory Press.

Pierce, B. A. (2010). Chapter 15: Cancer genetics. In *Genetics essentials: Concepts and connections* (pp. 389–406). New York, NY: WH Freeman.

Griffiths, A. J. F., Wessler, S. R., Lewontin, R. C., Gelbart, W. M., Suzuki, D. T., & Miller, J. H. (2005). Chapter 17: Genetic regulation of cell number: Normal and cancer cells. In *Introduction to genetic analysis* (8th ed., pp. 545–574). New York, NY: WH Freeman.

Alters, S., & Alters, B. (2006). Chapter 12: Cell reproduction. In *Biology: Understanding life* (pp. 177–196). Danvers, MA: John Wiley and Sons.

Hanahan, D., & Weinberg, R. A. (2000). The hallmarks of cancer. *Cell*, 100, 57–70.

Student 4: Cytogenetics & Genetic Diversity

- What are the “anatomical parts” of a chromosome?
- How are whole chromosomes studied? What methods are available to study them?
- What can the study of chromosomes tell you about the organism you are studying?
- How are chromosomes prepared for examination?
- What is G banding? How is it performed, and what does it tell you?
- What sorts of mutations can occur that are visible at the chromosome level?
- What does the term “locus” (plural: “loci”) refer to in genetic parlance?
- What is genetic diversity in a population? How is it measured? Is it desirable? Why or why not?
- What is a microsatellite? How is it used in biotechnology?

Suggested References

Any chapter in a genetics textbook on the topic of chromosome variation.

Snustad, D. P., & Simmons, M.J. (2009). Chapter 6: Variation in chromosome number and structure. In *Principles of genetics* (5th ed., pp. 115–139). Danvers, MA: John Wiley and Sons.

Pierce, B.A. (2010). Chapter 7: Chromosome variation. In *Genetics essentials: Concepts and connections* (pp. 167–192). New York: WH Freeman.

Mertens, T.L., & Hammersmith, R.L. (2007). Chapter 10: Human chromosomes. In *Genetics laboratory investigations* (13th ed., pp. 90–122). Boston: Benjamin Cummings.



Part II – Disseminating the Information

Come to class with a hard copy of your Research Report for every member of your team. In this section of the case, you will share the results of your research. The instructor will provide instructions.

Part III – Tasmanian Devil Facial Tumor Disease

You may recall the cartoon character the Tasmanian Devil of *Looney Tunes* fame from your childhood memories. It may surprise you to learn that this character is based on a real animal that lives in Tasmania, a large island-state off the southeast coast of Australia. The devils used to be found in Australia, but they were extirpated some 3,000 years ago (Brown, 2006). They now only exist in Tasmania.

The Tasmanian devil (*Sarcophilus harrisii*) is the world's largest carnivorous marsupial. It feeds on small prey, such as birds, snakes, and even insects. They are also scavengers, feasting on carrion. As marsupials, the young develop in a protective pouch like the kangaroo (Guiler, 1970). The devil looks like a cross between a rat and a dog, and it weighs between 10–20 pounds. The animal gets its name for the ferocious cries and growls it emits and the threat displays it produces when faced with a predator, fighting for mate, or defending a meal (Pemberton & Renouf, 1993). Tasmanian devils often bite one another while fighting, particularly on the face and neck.

In 1996, conservation authorities in Tasmania noticed that some animals had peculiar ulcers on the face. Further investigation determined that the ulcers were cancerous in nature. The tumors grew relatively rapidly, became friable, and eventually prevented the animal from eating, causing death. This disease was called Tasmanian Devil Facial Tumor Disease (DFTD). Alarmingly, many animals were affected; it is estimated that the population has been halved as animals die from this disease and that the species could go extinct within the next 25–30 years (Quammen, 2008).

This latter fact should trouble you for many reasons. First, from a conservation point of view, the disappearance of the largest carnivorous marsupial represents a loss of the biodiversity for the planet. When animals go extinct, they don't come back. As molecular biologists, this case concerns us for an entirely different reason. An animal population seems to be decimated by cancer. And not just any form of cancer, but the same type: facial cancer. But wait. How could this be? Cancer isn't infectious, is it? How is it then that many animals are dying of facial cancer?

You will now watch an introductory documentary from *National Geographic* on this disease.

<http://news.nationalgeographic.com/news/2007/06/070608-devils.html>

(Note: Windows Media Player is required to view this clip.)

Questions

1. Hypothesize what could be causing many animals in a population to be affected by a similar type of cancer. Describe the causes and how it is spread. Try to come up with as many hypotheses as you can.
2. How would you test one of your hypotheses? Design an experiment to determine whether your hypothesis is correct.



Photo of Tasmanian devil with Devil Facial Tumour Disease (DFTD).

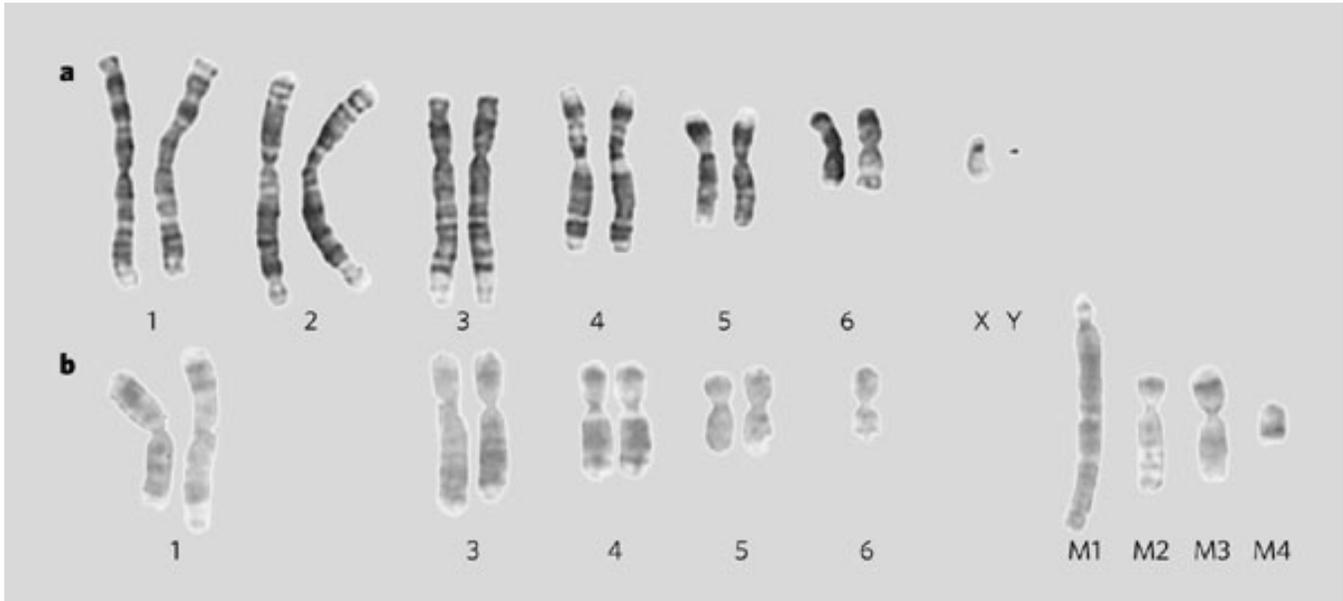
Source: "To Lose Both Would Look Like Carelessness: Tasmanian Devil Facial Tumour Disease," H. McCallum and M. Jones, *PLoS Biology* Vol. 4/10/2006, e342. <http://dx.doi.org/10.1371/journal.pbio.0040342>. Photo by Menna Jones. Image licensed under the Creative Commons Attribution 2.5 Generic license.

Part IV – DFTD Paper 1

Researchers became interested in this mysterious cancer. Read the following passage from a landmark paper on DFTD (Pearse & Swift, 2006). It should help you elucidate what's happening to the devils.

“We studied tumours that included early neoplasms, huge primary cancers and secondary cancers. The cancers were sampled from animals throughout eastern Tasmanian, Australia, over a 12-month period.

The normal number of chromosomes in the devil is 14, including the XX or XY sex chromosomes (Fig 1a).



*Figure 1. Chromosomes of facial tumors from Tasmanian devils. A, Normal karyotype for a male Tasmanian devil (14 chromosomes, including XY). B, Karyotype of a cancer cell found in each of the facial tumors of all 11 animals studied. Source: Pearse, A-M, & Swift, K. (2006). Allograft theory: Transmission of devil facial-tumour disease. *Nature*, 439, 549. Figure 1. Reprinted by permission from Macmillan Publishers Ltd: *Nature*, copyright 2006.*

[...] We made the fortuitous observation of a pericentric inversion of chromosome 5 in the constitutional karyotype of one animal. This constitutional anomaly was found in all cultures of that devil's normal tissues, but was not present in either of the chromosomes 5 of his facial-tumour cells.”

Questions

- Hypothesis:** What do you suppose were the aims of the investigators? What were they trying to assess with this research? What is their hypothesis?
(*Hint:* Look at the figure. What are the researchers comparing? Why might they be doing this? What are they trying to learn from this line of investigation?)
- Materials & Methods:** The Materials and Methods section is omitted from this paper. Instead, they appear in the “Supplementary Information” document available on the journal’s website. Given the figure and the information provided in the text, retrace their experimental steps. What did they do in this experiment? Be as specific as you can.
- Results:** Given your hypothesis and the experimental method, what results would you expect if your hypothesis is right? What if your hypothesis is incorrect?
 - What do the images in the top part of Figure 1 represent?
 - What do the images in the bottom part of Figure 1 represent?
 - Complete the following template form with appropriate terms. Duplicate this form as many times as you need.
If we compare _____ and _____, we learn that _____.

- d. What is unusual about the images in the bottom part of Figure 1? What is unexpected? If you were to repeat this experiment with tumors taken from different humans, would you expect similar findings?
 - e. Overall, what we learn from Figure 1 is _____.
 - f. What do the results reported in the bottom paragraph (“We made the fortuitous...”) strongly suggest? Why is this fortuitous? What’s unusual about it? What does it tell you about the mode of infection?
4. *Discussion:*
- a. Summarize the results of this paper using one sentence to describe the conclusions and one sentence to describe the evidence used to support this conclusion.
 - b. Provide one criticism of the methods used. What is one area of concern? What could you do better?
 - c. Is there anything you didn’t understand or are still unclear about?
 - d. What are some outstanding issues? What sort of questions do these results raise? What would you still want to know about DFTD that this study doesn’t answer?

Part V – What Comes Next?

Experiments are seldom done in isolation. Research is a continuous process. If you were the researchers that published the paper you just analyzed, what questions would you still have about DFTD? In this section, you will work with your team to propose the next experiment that the researchers should do. Start with a question you still have about the research, propose a hypothesis, and then an experiment. You must complete the following Research Proposal form and bring it to class the next time we meet. Your team will present your proposal to the rest of the class. The other students may question your proposal, so be ready to defend your plan. You do not need to know the names of specific molecular biology techniques (although you should feel free to research them if you want), but you should describe what you hope an experiment would do. After the class presentations, the class will vote on the best proposal, simulating a research grant panel that allocates funds for research.

Research Proposal (should be about 1–2 pages in length)

Names: Name of each group member

Title: Title of your research proposal. Pick a title that summarizes the research you are proposing in one sentence or less

Abstract: Summary of your research proposal. Essentially, summarize each of the sections below into one sentence each and compose a paragraph from this information.

Background: What information should a person evaluating your proposal know in order to be able to evaluate the merit of this work? What topic are you proposing to study? Why is this important or interesting? What previous research has been done on this topic? What remains to be investigated?

Hypothesis: What is the question you wish to investigate in this proposal and what is your hypothesis?

Materials and Methods: How might you go about investigating your question or hypothesis? What would constitute a good experiment? What would be a good control? What conditions would you impose on an experimental group to assess your hypothesis? What are possible conflicting variables and how do you plan to control for them? What will you do to ensure reproducibility? You do not need to detail the specific procedures and methods, but rather describe the general experimental approach you would take to evaluate an answer to your question.

Results: What results would you expect if your hypothesis is correct? What if your hypothesis is incorrect?

Significance of the Work: How will performing this experiment increase our knowledge of the world or help to find a cure or address a problem in the world?

Limits of this Experimental Design: What are some of the challenges or pitfalls of this experimental design that you foresee? Are there some variables you will not be able to control for? Are there only a limited number of subjects you will be able to test? Are there ethical limitations?

Part VI – DFTD Paper 2, Part I

A second paper on DFTD was published shortly after the one you just analyzed (Siddle et al., 2007). In this paper, the researchers first confirmed, at the molecular level, the results of the first paper. In this experiment, the researchers examined the DNA of cancerous and healthy devil cells. They discovered that the genes of cancerous cells from any animal were much more similar to one another than to the genes of healthy cells from the animals from which the cancerous cells were isolated. This confirmed that the cancer originated with a single cell and is transmissible between animals. The researchers hypothesized that the behavior of devils, notably the biting on the face, served as a means for the cancerous cells to spread between animals (remember the observation that the tumors are friable).

That should strike you as odd. Cancer is not usually a transmissible disease. Humans don't fear being exposed to people who have cancer.

Question

1. Propose at least two hypotheses that could account for the transmission of cancer between animals of the same species. For each proposed mechanism, how can this occur (think about the molecular mechanism that could explain this—at the genetic or immunological level)? What usually prevents this from happening, and why might devils be susceptible to the transmission?

Part VII – DFTD Paper 2, Part II

The authors state that the immune system of the devils is functional (Siddle et al., 2007). The authors propose two hypotheses to explain the transmission of cancerous cells:

- The cancerous cells down-regulate their own expression of MHC genes during tumor growth to escape notice by the immune system;
- The devils are so genetically similar to one another that there is no diversity among their MHC genes. Consequently, the cells of another devil are not recognized as foreign by the immune system.

Transmissible cancers have been reported in other species. The canine transmissible venereal tumor is a type of cancer passed between dogs during coitus. Research has shown that it evolved approximately 200 years ago from a single cell. Such tumors escape the immune system by down-regulating the MHC genes in tumor cells (Murgia et al., 2006). In Syrian hamsters, a contagious cancer has also been observed. In this case, the transmission is thought to be due to the fact that these animals are extremely inbred and lack MHC diversity (Streilen & Duncan, 1983).

The researchers set up a series of experiments to investigate which mechanism might be at play in devils.

In the first experiment, the researchers took the following types of cells from two affected devils:

- Tumor biopsy (T)
- Healthy liver (L)
- Healthy kidney (K)
- Healthy spleen (S)

The researchers extracted all the RNA from these cells, and used RT-PCR to make a lot of DNA copies of specific RNA sequences. In particular, the procedure amplified the RNA encoding MHC Class I (panel A) and MHC Class II (panel B) RNA. The amplified DNA was separated on a gel and stained as shown in Figure 2. Lane M represents a lane with molecular weight markers (DNA of known sizes), and lane N is a negative control where no RNA was placed in the sample.

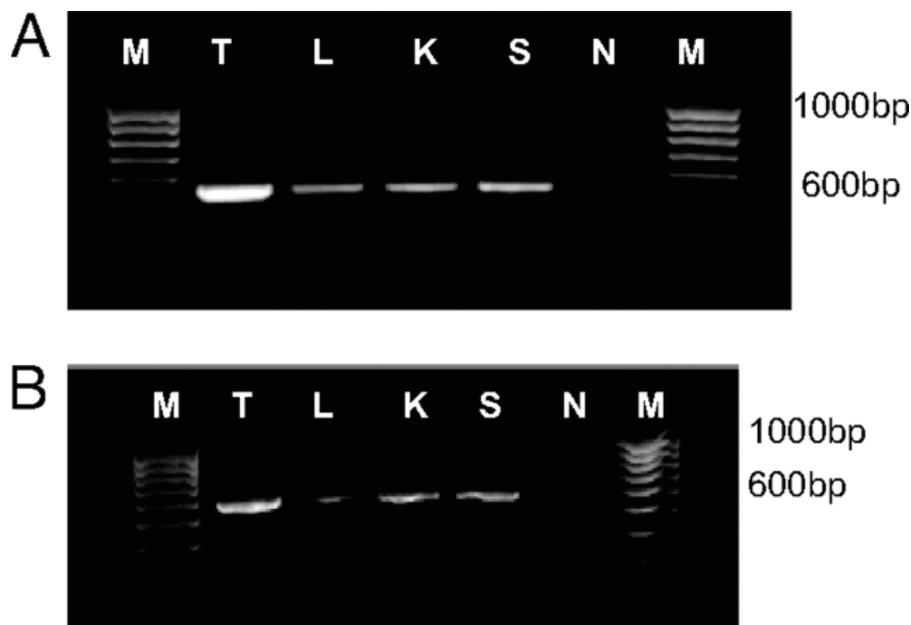


Figure 2. Source: Siddle, H. V., et al. (2007), *PNAS*, 104(41), 16221–26, Figure 1. Copyright © by the National Academy of Sciences. Used in accordance with educational, non-commercial terms of use as defined at <http://www.pnas.org/site/misc/rightperm.shtml>.

Questions

8. What specific question or hypothesis is the experiment depicted in the above figure attempting to answer?
9. Fill in the following template to analyze the figure. Repeat as often as you feel is insightful.
If we compare lane _____ and lane _____, we learn that _____.
10. What are the main trends in the data? Are the trends in both MHC classes consistent?
11. My simplified (decoded, in regular language) title for this figure is: _____.
12. Overall, what we learn from this figure is: _____.
13. Does this finding support previous work or does it contradict it?
14. The following issues are of concern to me (these can be things you do not understand, or criticisms of the method, questions for the authors, or anything else that comes to mind).
15. Given the stated hypotheses of the authors at the beginning of the paper, which have now been addressed and which remain to be studied?

Part VIII – DFTD Paper 2, Part III

In the second experiment, a mixed lymphocyte reaction (MLR) was performed. This test is performed to assess the reactivity of T-cells. In this assay, lymphocytes are mixed with a substance or a cell type that may or may not stimulate them. When a lymphocyte recognizes an antigen as foreign, it replicates. Cell replication can be monitored by including radioactive nutrients in the medium surrounding the cells. When the cells replicate, they take up the radioactivity. At the end of the assay, the cells are washed of the surrounding nutrients and the radioactivity inside the cells counted. The more radioactivity is measured, the more cells there are, and therefore the more reactive lymphocytes were to foreign antigens.

Three mixed lymphocyte reactions were performed (Figure 3):

- Devil lymphocytes were mixed with Con A
- Devil lymphocytes from different devils were mixed together
- Eastern quoll lymphocytes from 2 animals were mixed together (only data point shown is at 96 hrs)

Notes:

- Concanavalin A (Con A) is a protein that causes lymphocytes to proliferate.
- Like devils, eastern quolls are dasyurid marsupials, and they share a similar habitat to devils. However, they are not endangered.
- Marsupials are known to have a low immune reaction.

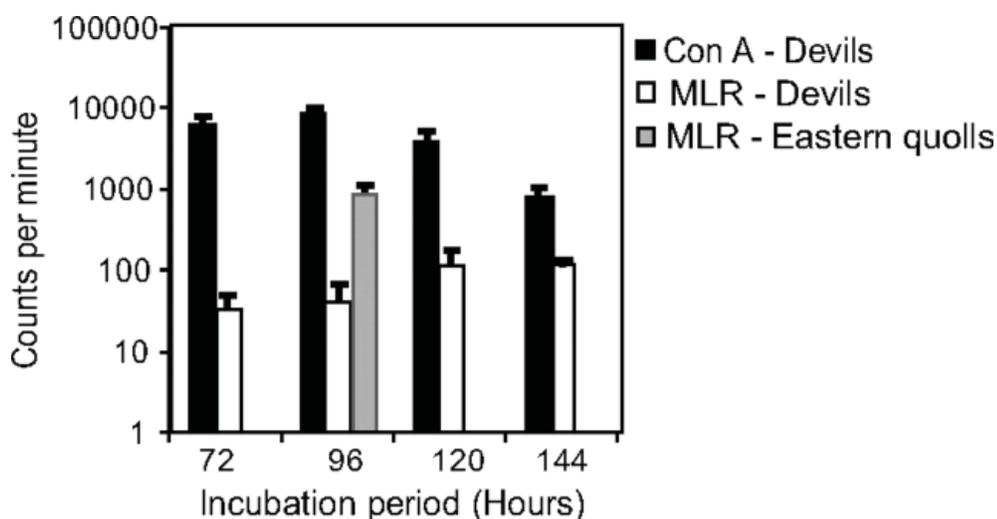


Figure 3. Mixed lymphocyte reaction and Con A stimulation of lymphocytes from 30 devils and 2 eastern quolls obtained from the same region of Tasmania. Source: Siddle, H. V., et al. (2007). Transmission of a fatal clonal tumor by biting occurs due to depleted MHC diversity in a threatened carnivorous marsupial. *PNAS*, 104(41), 16221–26, Figure 2. Copyright © by the National Academy of Sciences. Used in accordance with educational, non-commercial terms of use as defined at <http://www.pnas.org/site/misc/rightperm.shtml>.

In the third experiment, a region of MHC Class I gene known to be highly polymorphic between individuals of a species was sequenced in 25 devils, gir lions (a wild population that has undergone severe population reduction), and humans. The researchers were interested in determining how many different variants of the MHC class I gene existed in the population. They converted the gene sequence into an amino acid sequence, and focused their attention to 11 amino acid sites in the protein. They looked at the number of variations in amino acids that were found at each of these 11 sites in the three species. The results are shown in Figure 4.

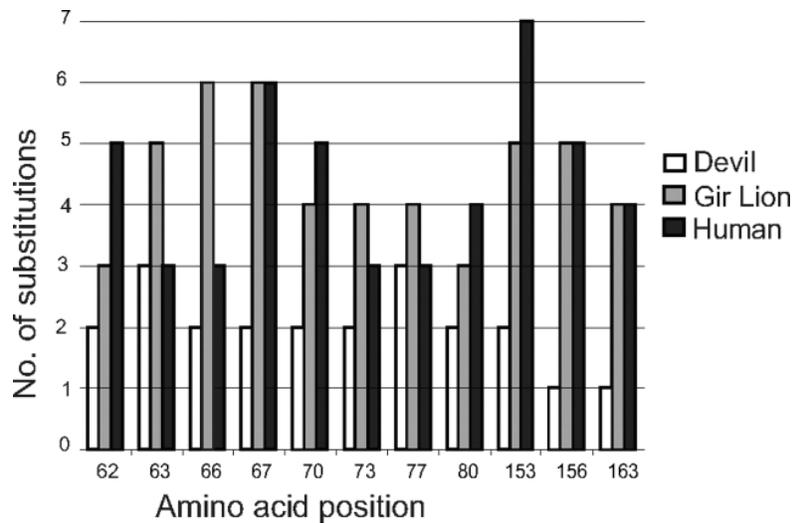


Figure 4. Graphical representation of polymorphism in the Peptide-Binding Region of class I sequences from humans (HLA-A, HLA-B, and HLA-C) (Bjorkman PJ, Parham P (1990) *Annu Rev Biochem* 59:253–288.), wild Gir lions (multiple loci) (n = 25), and Tasmanian devils (multiple loci) (n = 25). Source: Siddle, H. V., et al. (2007). Transmission of a fatal clonal tumor by biting occurs due to depleted MHC diversity in a threatened carnivorous marsupial. *PNAS*, 104(41), 16221–26, Figure 4. Copyright © by the National Academy of Sciences. Used in accordance with educational, non-commercial terms of use as defined at <http://www.pnas.org/site/misc/rightperm.shtml>.

Questions

16. What specific question or hypothesis are the experiments depicted in these figures attempting to answer?
17. For the experiment performed to obtain Figure 3:
 - a. Describe the data that would support the author’s hypothesis. Describe the results that would contradict the author’s hypothesis.
 - b. What is the purpose of adding Con A to the reaction tube for one experimental condition?
 - c. Why were cells taken from eastern quoll used in this experiment?
 - d. Fill in the following template to analyze the figure. Repeat as often as you feel is insightful.
If we compare _____ and _____, we learn that _____.
 - e. What are the main trends in the data? Are all the data consistent with this trend?
 - f. My simplified (decoded, in regular language) descriptive title for this table is: _____.
 - g. Overall, what we learn from this figure is: _____.
18. For the experiment performed to obtain Figure 4:
 - a. Describe the data that would support the author’s hypothesis. Describe the results that would contradict the author’s hypothesis.
 - b. The amino acid sequence of a specific protein is being analyzed. Which one? Why was this protein selected for analysis?
 - c. Why are only certain amino acid positions shown?
 - d. Why were protein sequences analyzed from humans, gir lions, and devils?
 - e. Fill in the following template to analyze the figure. Repeat as often as you feel is insightful.
If we compare _____ and _____, we learn that _____.
 - f. What are the main trends in the data? Are all the data consistent with this trend?
 - g. My simplified (decoded, in regular language) descriptive title for this figure is: _____.
 - h. Overall, what we learn from this figure is: _____.

19. Do the two experiments support or contradict one another?
20. The following issues are of concern to me (these can be things you do not understand, criticisms of the method, questions for the authors, or anything else that comes to mind).
21. Given the stated hypotheses of the authors at the beginning of the paper, which have now been answered and which remain to be studied?
22. Write three bullet points of the conclusions you have reached as a result of analyzing the figures from this paper.
23. How convincing was the evidence that support these conclusions?
24. Write a one-paragraph synopsis (an abstract) of this paper. Be sure to include the question addressed in this paper, the methodology, results, and conclusions.
25. What would you like to investigate next? If you were these researchers, what would you test next? What questions does this research open up? What questions remain unanswered about devil tumor facial disease?

Assignments

Tomorrow, we will hold a discussion on transmissible cancer. To prepare, please do the following two assignments.

1. Read the following three review articles on DFTD:
 - Jones, M.E., & McCallum, H. (2011). The devil's cancer. *Scientific American* (June 2011), 72–77.
 - Quammen, D. (2008). Contagious cancer: The evolution of a killer. *Harper* (April 2008), 33–43.
 - Dingli, D., & Nowak, M.A. (2006). Infectious tumour cells. *Nature*, 443, 26.
2. Make a concept map of all that has been learned. Include at least 25 concepts in your map; be sure to include descriptions on the links between concepts. Try to incorporate the figures analyzed in this case at appropriate places in the concept map.

Part IX – Discussion on Transmissible Cancers

In this section, there will be a whole-class discussion on transmissible cancer, taking all that you have learned about devils to assess threats to human life, and to consider what can be done to save the devils from extinction. Your instructor will provide more instructions.



Part X – Summary Essay

To wrap up this extensive case study, you must write a paper on the topic of your choice (select one topic from the three proposed below). To write this paper, you will need to synthesize all that you have learned thus far about DFTD. You may need to research some additional areas in the scientific literature, reference it appropriately, and you will need to think independently and critically. This is an individual assignment. Essays should be 2 pages, single-spaced.

Choice of Essay Topic (choose one of the following):

Genetic Diversity and Conservation Strategy

Why do the devils have a low genetic diversity? How has low genetic variability impacted the susceptibility of Tasmanian devils to facial tumor disease? How is this low diversity affecting the chances that devils will adapt to this pathogen? How is the low diversity likely to impact the chances of recovery of Tasmanian devils to the emergence of a new disease? How should this knowledge influence management practices in dealing with the disease and preserving the species? What strategy would you suggest for dealing with this situation and ensuring the long-term survival and success of this species?

Suggested article to consult: McCallum & Jones (2006).

Cancer and Evolution

How can the mechanism of evolution by natural selection be applied to the study and understanding of the emergence, progression and spread of cancer? In the case of DFTD, how could you support the claim that the disease represents a step forward in the evolution of cancer? Can we expect other forms of cancer (in devils and in other organisms such as humans) to evolve along these lines? Why or why not? How can thinking of DFTD as an evolutionary disease help to treat it or find ways of curing it? Are there lessons to be learned from this paradigm for dealing with human cancers?

Suggested articles to consult: Merlo et al. (2006); Goymer (2008).

Comparison of Transmissible Cancers

Devil facial tumor disease is not the only form of transmissible cancer: a similar form of disease occurs in dogs, called the canine transmissible venereal tumor (CTVT). There are many differences between the two transmissible cancers, notably how incapacitating they are (how aggressive), how cancer cells evade the immune system, how they are transmitted, and how long ago the disease first emerged. Research the differences and commonalities between the two cancers and draw conclusions about what is necessary for a cancer to become transmissible, how each can be treated, and how DFTD is likely to evolve in the future.

Suggested article to consult: Das & Das (2002).

References

- Bjorkman, P.J., & Parham, P. (1990). Structure, function, and diversity of class I major histocompatibility complex molecules. *Annu Rev Biochem*, 59, 253–88.
- Brown O. (2006). Tasmanian devil (*Sarcophilus harrisii*) extinction on the Australian mainland in the mid-Holocene:

- multicausality and ENSO intensification. *Alcheringa: An Australasian Journal of Paleontology*, 31, 49–57
- Das, U., & Das, A. K. (2002). Review of canine transmissible venereal sarcoma. *Vet Res Commun.*, 24, 545–56.
- Dingli, D., & Nowak, M. A. (2006). Infectious tumour cells. *Nature*, 443, 26.
- Goymer, P. (2008). Natural selection: The evolution of cancer. *Nature*, 454(7208), 1046–48.
- Guiler, E. R. (1970). Observations on the Tasmanian devil, *Sarcophilus harrisii* II. Reproduction, breeding and growth of pouch young. *Australian Journal of Zoology*, 18, 63–70.
- Jones, M.E., & McCallum, H. (2011). The devil's cancer. *Scientific American* (June 2011), 72–77.
- McCallum, H., & Jones, M. (2006). To lose both would look like carelessness: Tasmanian devil tumour disease. *PLOS Biology* 4(10), e342.
- Merlo, L. M., Pepper, J. W., Reid, B. J., & Maley, C.C. (2006). Cancer as an evolutionary and ecological process. *Nat Rev Cancer*, 6(12), 934–35.
- Murgia, C., Pritchard, J. K., Kim, S. Y., Fassati, A., & Weiss, R. A. (2006). Clonal origin and evolution of a transmissible cancer. *Cell*, 126(3), 477–87.
- National Geographic. (2007). (Online video clip.) Tasmanian devils battle gruesome cancer. *Wild Chronicles*, PBS. Retrieved 27 July 2010 from <http://news.nationalgeographic.com/news/2007/06/070608-devils.html>
- Pearse, A-M, & Swift, K. (2006). Transmission of devil facial-tumour disease, *Nature*, 439, 549.
- Pemberton, D., & Renouf, D. (1993). A field-study of communication and social behaviour of Tasmanian Devils at feeding sites. *Australian Journal of Zoology*, 41, 507–526.
- Quammen, D. (2008). Contagious cancer: The evolution of a killer. *Harper* (April 2008), 33–43.
- Siddle, H. V., Kreiss, A., Eldridge, M. D. B., Nooman, E., Clarke, C. J., Pyecroft, S., Woods, G. M., & Belov, K. (2007). Transmission of a fatal clonal tumor by biting occurs due to depleted MHC diversity in a threatened carnivorous marsupial. *PNAS*, 104(41), 16221–26.
- Streilen, J., & Duncan, W. (1983). On the anomalous nature of the major histocompatibility complex in Syrian hamsters, Hm-1. *Transplant Proc*, 15(2), 1540–45.



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