Part I – Genetics

It was a sunny afternoon in early September and the fall semester was getting underway at the university. Casey was a sophomore who had just declared her biology major and was excited to be enrolled in a genetics class. Even though the weather outside was beautiful, Casey found herself in her dorm immersed in the subject. She was fascinated by the fact that common phenotypic traits could be inherited yet variably expressed through generations and was eager to learn how certain medical conditions were acquired genetically. A little bit of background reading eased the nerves that Casey and many students experience in the first week of class.

The next day, Casey’s genetics professor got right to work talking about some prevalent genetically induced disorders. The first condition the class reviewed was Duchenne muscular dystrophy (DMD) and Casey was intrigued. She had heard of sex-linked disorders in high school, but was struggling to remember how a person could be at a higher risk for a disease simply because of their biological sex: didn’t both parents typically contribute an equal number of chromosomes? Her interest grew as her professor explained the symptoms of the disease. DMD directly affects skeletal muscles starting in the early stages of life, often disabling children to the point that they are no longer able to walk by the age of 10. As the disease progresses, other types of muscles are affected, resulting in the need for breathing machines and other treatments. Unfortunately heart problems often lead to death by the age of 25. It sounded like a horrible condition, and Casey wondered what the chances were that one day she might have children with the disease.

Having learned that Duchenne muscular dystrophy is a genetically inherited condition resulting in a lack of the dystrophin protein, Casey lay in bed that night wondering how various types of gene variations could lead to DMD.

Questions

1. Given that Duchenne muscular dystrophy is a sex-linked recessive disorder, explain why biological males are more likely than females to be affected by this condition.

2. Identify some types of mutations you’re familiar with. Knowing that the DMD gene produces dystrophin, and cells of DMD patients often lack adequate amounts of dystrophin, what type(s) of mutation(s) is most likely to be responsible for the DMD phenotype? Why?

3. Scientific journals are an excellent resource for learning about previous research on a wide variety of topics. Refer to the abstract of the article below and identify the type of gene mutation that is most closely associated with DMD.

Part II – Cell Biology

Following a successful fall semester excelling in genetics, Casey was reinvigorated after winter break and was ready to begin the next chapter in her biology undergraduate education. The new semester provided the challenge of a cell biology class. In addition to her recently acquired foundation in genetics, Casey had some introductory knowledge of cells from her high school curriculum. She knew the basic structure of cells and that there are countless types of cells in the body responsible for specific functions. Cells of similar types make up tissues, which can be bounded by a membrane and connected to other cells by the extracellular matrix. Over break, Casey familiarized herself with muscle cells as part of looking into Duchenne muscular dystrophy. She saw that while muscles can vary in size, they share a common structural organization.

Duchenne muscular dystrophy targets muscles, but Casey wanted to take her research a step farther and see how it specifically impacts the muscle cells. Casey knew that DMD is an inherited disorder characterized by progressive muscular degeneration and weakness, though she was not fully content with her level of understanding and decided to do more research. She remembered having previously using the Online Mendelian Inheritance in Man (OMIM) database to find preliminary information on the impacts of DMD on the cellular level and went back to the site to refresh her memory. Knowing that the main protein associated with DMD is dystrophin, she looked it up and found that it has a major structural role in muscle, as it links the internal cytoskeleton to the extracellular matrix (Nowak & Davies, 2004). From there, Casey realized that the absence of this protein in DMD patients must therefore impact cytoskeleton organization and disrupt the integrity of muscle membranes. She knew this was problematic but wanted more specific information on the connection between the dystrophin protein and these issues. She emailed her professor, who graciously answered her questions and provided some articles for Casey to read during her free time, including the following:


Casey’s professor told her to focus on page 2801, including the full section under the subheading “Dystrophin” and the schematic depicting the dystrophin-associated protein complex. This would help her better understand the role of dystrophin in maintaining muscles and the problems that result when it is absent in DMD patients.

Questions

1. In order to understand how muscle weakness manifests in DMD patients, we must first explore the structural organization of muscles. Using textbook or appropriate internet resources, identify and discuss the role of the following components: sarcolemma, sarcomere, cell cytoskeleton (with attention to actin filaments), and extracellular matrix.

2. Use the Ehmsen *et al.* (2002) article to answer the following:
   a. Describe the role of the dystrophin-associated protein complex (DAPC) in maintaining muscle integrity. How does it stabilize the sarcolemma of muscles?
   b. The DAPC is comprised of various proteins that are vital for maintaining muscle integrity. Here we will focus on the protein dystrophin, which is lost in most DMD patients. How does the absence of dystrophin result in the manifestation of the DMD phenotype and muscle degeneration in these patients? What is its specific role within the DAPC? *(Hint: refer to schematic.*)
Part III – Biochemistry

Like many of her peers, Casey couldn’t believe how quickly her sophomore year had flown by. She had successfully completed her cell biology course and her confidence in her developing skills in the sciences continued to grow. She could feel herself becoming a well-rounded student and was starting to identify connections between other subjects under the science umbrella. Casey knew she was interested in Duchenne muscular dystrophy from her previous studies and was intrigued to investigate other aspects of the condition. She decided to take biochemistry in the fall of her junior year. She knew the course examined biology with a chemistry perspective but was unsure how the two subjects intertwined enough to have a course dedicated to this overlap.

A few weeks into the semester, the class had covered the concept of biochemical pathways and the impact of intermediate steps on the way to a desired product in cells. As an example, the professor of the class asked the students to look up the following article and review Figure 3A in particular:


As Casey examined the sphingolipid synthesis pathway in the figure, she also noted that the suppression of the \textit{wunen} gene contributed to the reduction of DMD phenotypes observed in \textit{Drosophila} (Pantoja et al., 2013). This instantly reminded her of the work of a research student on campus who had annotated the \textit{wunen} gene earlier during the semester and then learned about its ortholog in humans, the \textit{PLPP3} gene. The research student found that \textit{PLPP3} is responsible for the conversion of sphingosine-1-phosphate (S1P) to sphingosine, which is identical to the role of \textit{wunen} in \textit{Drosophila}. Casey also remembered the research student mentioning that the suppression of \textit{PLPP3} can have the potential to reduce DMD symptom severity in humans, similar to the impact of \textit{wunen} suppression on DMD phenotypes in \textit{Drosophila}. These connections helped Casey better understand the concept of orthologous genes and realize the importance of biochemical pathways in the manifestation of DMD.

Questions

1. Refer to the following article and navigate to the “Ortholog Identification” section on the first page.

   Define the term \textit{orthologous genes} and identify how they are related in terms of biological function. Is this in agreement with why \textit{wunen} and \textit{PLPP3} are considered orthologs?

2. Read the abstract of the Pantoja et al. (2013) article and examine the first schematic on top of page 141, Figure 3A. How does the suppression of \textit{wunen} lead to suppressed DMD phenotypes? Consider the role of sphingosine-1-phosphate (S1P) in reducing the symptoms of DMD in your answer as well.

3. Based on the gene ortholog and biochemical information you have collected thus far, suggest a therapeutic approach that could have the potential to reduce DMD phenotypes in humans. Provide the rationale behind your suggestion.