NATIONAL CENTER FOR CASE STUDY TEACHING IN SCIENCE

What Happened to Beau?

How Amino Acids Affect Keratin Organization in Hair

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Handout 1 – Video, Quiz, and Questions

Step 1: Write down answers to these questions *before* watching the video:

Questions

1. What do you know already about proteins? How would you define a protein in your own words?
2. Where are proteins found in organisms? What structures in *your* body are made mainly of proteins?
3. What structures in organisms do *not* contain proteins?
4. From what material(s) are proteins made? Where does this material come from?

Step 2: Complete Video Quiz #1.

Step 3: Watch the video: *Protein Structure Part 1: Where Are Proteins? What Do They Do? <https://youtu.be/ooMG5Eqxe6k>*

Step 4: Complete Video Quiz #1 again.

Step 5: Write down answers to these questions *after* watching the video:

Questions

5. Go back to your pre-video responses.
   a. How would you change or expand your original answers to Questions 1–4 above?
   b. What *new* general principles or concepts did you learn from the video that you did not know already?
   c. What specific details about proteins did you learn that you did not know before?
6. What are the basic subunits from which cells make proteins?
   a. How are the basic subunits similar to each other?
   b. What makes each of them unique?
   c. Where do the basic subunits come from?
7. What is the basic shape of a protein molecule? What determines a protein's final 3-dimensional shape?

Step 6: Bring your answers to Questions 1–7 above with you to class.

Handout 2 – Case Scenario, Part I

A new puppy! Kayla was so excited she could hardly contain herself. Shortly after she left for college in August, her family's dog Cody had passed away in his sleep. Even though Cody had a long life and Kayla accepted he was gone, part of her dreaded being home for winter break without him. So she was delighted when her mother called to say they were bringing home a new dog.

When Kayla got home she hugged her mother and brother Karl, then immediately asked, "Where's our new puppy?" Her brother nodded towards the dog pillow next to the refrigerator.

"You are adorable! What's your name?"

Kayla's mother said, "We named him Beau."

Questions

1. Take a close look at Beau's coat. How would you describe it?
2. What is hair made up of? How much of it is protein, carbohydrate, nucleic acids, and lipids? (If you do not know, where could you find out?)

Figure 1. Puppy Beau.

Handout 3 – Case Scenario, Part II

Kayla spent nearly all of winter break playing with Beau. Just as he was starting his first growth spurt, it was time to go back to school.

After exams in May, Kayla drove home anxious to see how much Beau had grown. When she pulled in the drive, a dog came to greet her, but it definitely was not Beau. Worried something had happened to Beau, Kayla rushed inside and found her brother.

"Where's Beau?"

"Nice to see you too sis. He's outside. I heard him barking when you drove up."

"Karl, don't tease me. Where IS he? Did something happen?"

Kayla's brother didn't answer, but went to the door, whistled, then called "c'mon Beau." In bounded the same dog that had greeted Kayla outside.

Kayla was stunned. "What happened? He looks totally different!"

Questions

3. How has Beau's coat changed between puppyhood and adolescence?
4. What do you think is happening to make Beau's coat change? What are your initial hypotheses?

Figure 2. Beau at 7 months.

Handout 4 – Case Scenario, Part III

Figure 3. Beau's mother.

Figure 4. Beau's father.

After Kayla and Beau got reacquainted, Karl let Beau back outside. Karl grabbed his cell phone from the kitchen counter, then sat down with Kayla at the table.

"When we picked up Beau in November, he still had his puppy coat. The vet said all dogs have soft puppy coats at first. As they grow older, they shed their puppy fur, and it's replaced by their adult coat. The color and pattern can change a lot. Sometimes their coat changes to wiry hair, or switches from straight to curly, like Beau's. The vet said this is what he'll look like for the rest of his life."

"Well he's still cute. But why is his coat so different now?"

"I wondered that too, so I went back to the Tanners' farm where we got him. I took these photos of his parents, and a couple of his littermates that are still there. What do you see?"

"Well he must have inherited his curly coat from his dad. But his littermates still have straight fur. What's different about Beau's coat hair that makes it curly instead of straight?"

Karl replied, "I asked Mr. Hansen at school the same things. All he said was to think it through. He did give me a hint though. He said to think about what hair is, and how it forms, and what would have to change to make straight hair become curly."

Kayla rolled her eyes. "Yeah, I remember he always answered questions that way. So what do you think is going on?"

Figure 5. Two of Beau's littermates as puppies.

Questions

5. Based on what you know now about his parents and littermates, what do you think is happening to make Beau's hair curly? (Remember, your explanation has to allow for some of his littermates to be straight-haired.) Which of your earlier hypotheses are no longer likely to be true? Which ones can you keep as possibilities?
6. What additional information would you like to have to help you solve this puzzle? Where could you find that information?

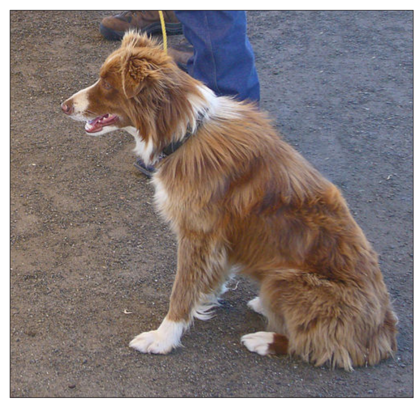


Figure 6 (left) and Figure 7 (right). Littermates at 7 months.

Now follow these steps:

Step 1: Complete Video Quiz #2.

Step 2: Watch the video, *Protein Structure Part 2: How and Why Do Proteins Fold? <https://youtu.be/vxlGEp-PCJQ>*

Step 3: Complete Video Quiz #2 again.

Step 4: Pick up Beau's story where you left off, and finish the remainder of this case.

Handout 5 – Case Scenario, Part IV

Kayla decided Beau's change of coat would be a great senior research project. Mr. Tanner let her collect hair from Beau's parents and two littermates, and Kayla pulled some samples from Beau's brush. She was all set to send the samples for analysis to Dr. Shugmann, a protein biochemist back at school. Then she realized she'd forgotten the most important sample of all! "Where will I get some of Beau's puppy coat?"

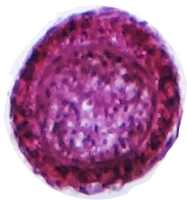
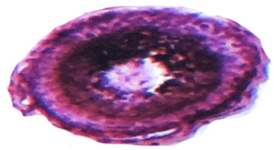
Kayla's mother came to her rescue. "Not a problem dear. Beau shed terribly as his new coat grew in. I've got his old dog bed downstairs, and I haven't washed it. You should have all the puppy hair you need!"

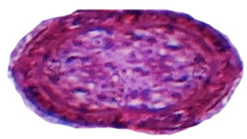
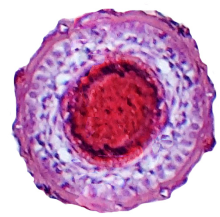
Kayla was elated. She collected the final sample of puppy hair, and then sent the samples to Dr. Shugmann, who promised to start protein analyses right away. "I'll leave the microscopy work for you to do when you get back to campus."

By November, Kayla and Dr. Shugmann had finished analyzing all of the hair samples. Their results are summarized below.

Chemical Analysis	*Source of Hair*					
	Beau		*Litter Mates*		*Beau's Parents*	
	Puppy	*7 Months*	*White*	*Brown*	*Father*	*Mother*
Total proteins (% dry wt.)	91.7%	96.3%	95.9%	97.4%	96.5%	96.1%
Keratins (% all proteins)						
Type I (acidic) keratins	42.4%	46.8%	43.1%	42.7%	45.6%	43.2%
Type II (basic) keratins	43.2%	40.1%	44.6%	43.1%	41.4%	44.0%
Total amino acid content	(%)	(%)	(%)	(%)	(%)	(%)
E (glutamic acid)	10.4	10.3	10.5	10.4	10.3	10.6
L (leucine)	10.1	10.2	9.9	10.0	10.1	10.1
S (serine)	9.7	9.6	9.5	9.5	9.7	9.6
A (alanine)	7.6	7.7	7.5	7.6	7.4	7.5
R (arginine)	7.4	6.6	7.4	7.3	6.5	7.5
V (valine)	6.2	6.1	6.3	6.2	6.2	6.4
G (glycine)	6.1	6.0	6.0	6.1	6.0	6.2
C (cysteine)	5.7	5.8	5.7	5.6	5.8	5.6
Q (glutamine)	5.1	5.0	5.0	5.2	5.1	5.0
N (asparagine)	4.7	4.6	4.8	4.7	4.8	4.7
T (threonine)	4.2	4.3	4.2	4.3	4.2	4.3
K (lysine)	4.1	4.2	4.1	4.1	4.3	4.1
I (isoleucine)	4.0	3.9	4.1	4.0	4.1	3.9
D (aspartic acid)	3.5	3.7	3.6	3.6	3.5	3.6
P (proline)	3.4	3.5	3.3	3.5	3.4	3.4
Y (tyrosine)	2.6	2.6	2.8	2.7	2.7	2.6
F (phenylalanine)	2.0	2.1	1.9	2.0	2.0	2.1
M (methionine)	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
H (histidine)	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
W (tryptophan)	n.d.	1.3	n.d.	n.d.	1.4	n.d.

Microscopy Results

	Beau		*Littermates*	
	Puppy	7 months	White, 7 months	Brown, 7 months
*Hair in cross section**				

	Beau's Parents	
	Father	Mother
*Hair in cross section**		

** Photos represent the typical cross sections observed in at least five hairs per dog. All cross sections were taken from the middle 1/3 of the hair shaft. Only intact hairs were sampled; broken or obviously damaged hairs were not used.*

Questions

7. Look at the table describing the chemical composition of each hair sample. What patterns or changes stand out? Based on what you know about how proteins are synthesized and assemble together, what is probably happening at the level of the molecules in Beau's coat?
8. Look at the illustrations and photographs of the hair samples. What patterns or changes stand out? Is there any correlation between the microscopic evidence and the chemical analyses?
9. Based on what you know now, what do you think is happening to make Beau's hair curly?
10. You made several hypotheses earlier in the case about why Beau's hair became curly; did any of your earlier hypotheses match your current conclusions? If not, were there any important data, clues, or other information that you overlooked initially?
11. What additional information would you like to have now to help you solve this puzzle? Where could you find that information?
12. If you were Kayla, what would be your next experiment or question?

Handout 6 – Case Scenario, Part V (Optional)

Going Deeper

This introduction to keratins is adapted from: <http://www.ncbi.nlm.nih.gov/pmc/articles/PMC3258107/>.

Intermediate filament proteins are a family of 70+ proteins. The proteins in the family are fibrous proteins that organize into ~10 nm diameter intermediate filaments. Members in this family include desmin, vimentin, and GFAP (found in muscle, fibroblasts and endothelium, and astrocytes, respectively); neurofilament proteins in neurons; and nuclear lamin proteins, which form part of the nuclear envelope.

Keratins are the most common intermediate filament proteins. There are more than 57 unique keratin genes in both dogs and humans. They can be subdivided into two types:

- Type I, or acidic keratins. They are numbered K9–K20, K23–K28, and K31–K40.
- Type II or basic keratins. They are numbered K1–K8, K71–74, K81, and K85–K86.

There is a second way to categorize keratins: hard or soft. "Hard" keratins are resistant to both chemical and enzymatic breakdown. The most commonly occurring "hard" acidic keratins in hair are K31 and K35. The main "hard" basic keratins found in hair are K71–74, K81, K85– K86.

Individual hairs in humans or a dog's coat contain a mixture of 26+ different keratins. The specific type and ratio of hard and soft, or acidic vs. basic keratins, determines how pliable or stiff a particular hair is. Cells that form hairs can vary the ratio of keratins that are secreted depending on where they are located on the body, season, or stage of life. For example, human head hair tends to be much more flexible and softer than beard or moustache hair. Similarly, a dog's whiskers will be stiffer than hair in its coat. The amino acid sequence of the keratins also affects how the keratins pack together.

In 2009, Cadieu, et al. published a report explaining the genetics of long hair, wiry coats, and curly hair in dogs. Each of these three characteristics is controlled by a single gene. The R locus associated with curly coats contains the *KRT71* gene, which codes for keratin 71. Curly-coated dogs are homozygous (r/r) for a 1-nucleotide change from C to T in Exon 2 of K71. This single nucleotide polymorphism changes Arg-151 to Trp (R151W). Hair with Trp instead of Arg does not grow straight, but curls instead. Dogs that are heterozygous (R/r) deposit enough normal K71 to grow a coat with straight to slightly wavy hair.

Pure breed dogs that always have curly coats (poodles for example) are fixed for genotype r/r, while breeds that always have flat coats (like Labrador retrievers) are fixed for genotype R/R. Mixed breed dogs that are genotype R/r usually have straight coats, but if mated can produce both straight- and curly-coated offspring. Therefore Beau and his curly coated father both must be homozygous r/r. His straight-haired mother must be heterozygous R/r. If she was genotype R/R, she would not be able to produce curly-haired pups like Beau.

Question

13. How can a single amino acid change (R151W) in one protein produce such a dramatic change in phenotype?

Optional Questions

14. With your discussion group, design an experiment to test the explanation you just gave in Question 13. Be sure to include proper controls. What would be your variables? What observations would you make? How would you interpret the possible outcomes?
15. If you were to continue exploring this question, what is the next question you would ask? What additional information would you like to know to continue pursuing it?

Handout 7 – Post-Case Questions

Your instructor may ask you to complete these and turn them in as individual homework.

1. What is the general structure of an amino acid? As part of your answer, explain:
    a. Which parts of amino acids are different.
    b. The major types or categories of amino acids.
2. Where are proteins found in organisms? What structures in *your* body are made mainly of proteins?
3. What are the common shapes of protein molecules? What determines which of these shapes a protein will form?
4. What are the four main molecular interaction forces that determine protein shapes? Which forces are responsible for each of the following, and which *parts* of the amino acids experience these forces?
    a. Formation of alpha helices.
    b. The final 3-dimensional shape of a protein.
    c. How 2+ proteins come together in complex groups.
5. Based on the structure of proteins, how would you explain why Beau's coat hair changed from straight to curly? Be sure your explanation describes what is happening at the molecular level to proteins.

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