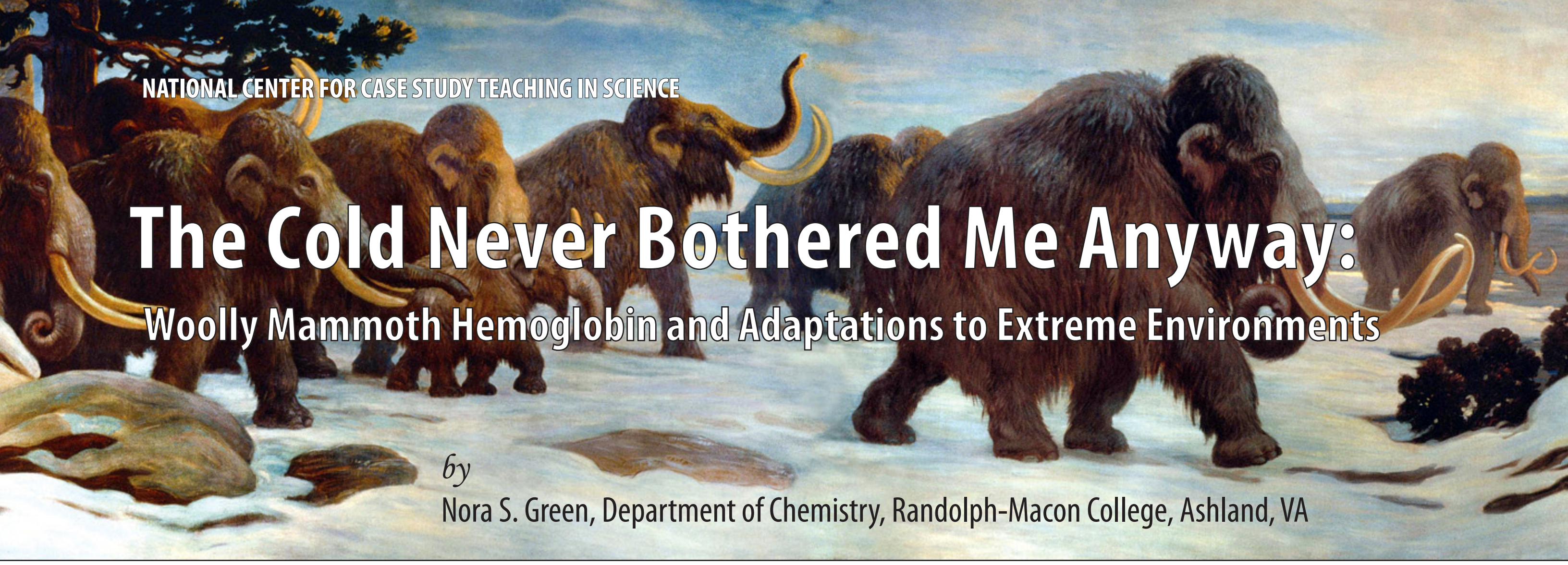

NATIONAL CENTER FOR CASE STUDY TEACHING IN SCIENCE

The Cold Never Bothered Me Anyway:

Woolly Mammoth Hemoglobin and Adaptations to Extreme Environments

by

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Daniel shivered as he ran across the snowy lawn in front of the Natural History Museum. Even with his new winter boots, his feet were almost numb from the subfreezing temperatures and his lungs ached from the cold air. He opened the door to the museum and let the warm air wash over him. He stomped the snow from his boots and stepped into the large domed entrance hall and into the shadow of the giant stuffed woolly mammoth. This mammoth was one of the most popular exhibits in the museum and one of Daniel's favorites.

"On a day like today," Daniel thought, "I sure am glad I'm not a woolly mammoth."

Glancing around, Daniel spotted the familiar information panel detailing several adaptations that allowed the great beast to survive in extreme cold weather, including small ears, long hair, and long tusks. In biochemistry class, Daniel's teacher had recently mentioned that scientists had used DNA from a preserved mammoth bone to produce and study the woolly mammoth's hemoglobin. Daniel wondered if this form of hemoglobin was another adaptation that helped the animal survive in the brutal cold. He wanted to know more.

Daniel had learned that the oxygenation of normal adult hemoglobin in humans is an exothermic process, which has been considered adaptive for humans as well as many other animal species.

Questions

1. Oxygen binding to hemoglobin in humans is an exothermic process. Write the equilibrium expression for this binding and explain what happens to oxygen affinity in both warm and cold temperatures.
2. Why is this useful for humans?
3. Why might this be disruptive to cold-adapted mammals (e.g., reindeer, musk ox, woolly mammoths)?
4. Would you expect the binding of oxygen to woolly mammoth hemoglobin to be more or less exothermic than human hemoglobin?

Of course, humans are very different from woolly mammoths. Daniel knew from the information panel that the closest living relative of the woolly mammoth was the Asian elephant.

5. Would you expect the binding of oxygen to woolly mammoth hemoglobin to be more or less exothermic than Asian elephant hemoglobin?

Exploring the differences between the hemoglobin in Asian elephants (AE) and woolly mammoths (WM) might shed further light on this important adaptation.

6. The primary sequence of Hb WM differs from Hb AE in only four positions: K5N (α chain), T12A, A86S, and E101Q (β-like chain). What kind of mutation occurs at each of these positions (i.e., charged to uncharged, polar to nonpolar, etc.)?

7. The T12A mutation occurs near the 2,3-bisphosphoglycerate (2,3-BPG) binding site. What is the role of 2,3-BPG binding on the function of Hb?
8. Examine the oxygen equilibrium curves below (37° C and pH 7.0). The solid lines represent the Hb protein in the absence of any allosteric effectors. Which animal has a higher oxygen affinity? Explain.

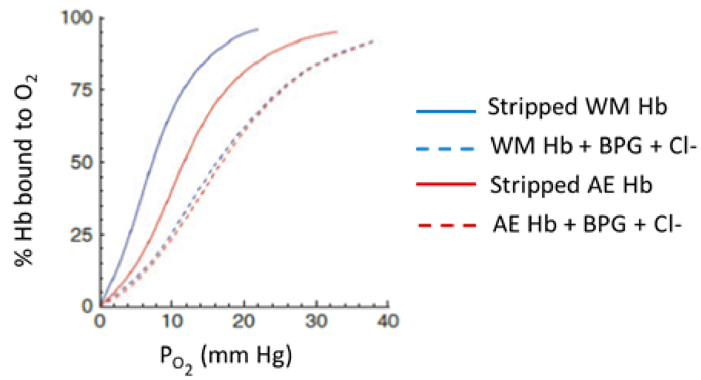

Figure 1. Adapted from: *Nature Genetics* 2010, 42, 536–40.

9. The dotted lines represent the proteins in the presence of Cl^- and 2,3-BPG. What can you conclude about the affinities of Asian elephant and woolly mammoth Hb with respect to Cl^- and 2,3-BPG?
10. In human and Asian elephant Hb, the threonine in position 12 forms a hydrogen bond with the carbonyl of a Lys residue (position 8), leaving an Asp side chain to project into the 2,3-BPG pocket. What is the effect of this on binding of 2,3-BPG?
11. In the woolly mammoth, position 12 is an Ala residue instead of threonine. What is the effect of this mutation on 2,3-BPG binding? Explain your reasoning.
12. What is the Bohr effect? Structurally, what leads to this effect?
13. The hemoglobins from humans and most mammals exhibit a stronger Bohr effect at lower temperatures. Why might this be beneficial for humans?
14. In contrast, the Bohr effect in woolly mammoths occurs at a higher pH_{max} (the pH at which the maximum protons are released) and is less dependent on temperature. How is this adaptive for the woolly mammoth?
15. Given the information so far, what evidence supports that hemoglobin may have been important in the cold tolerance of the woolly mammoth? What counter-arguments could be made?

References

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