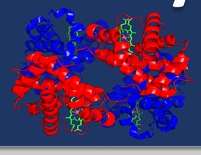
Meta-analysis of residue substitutions adapted in endothermic hemoglobin



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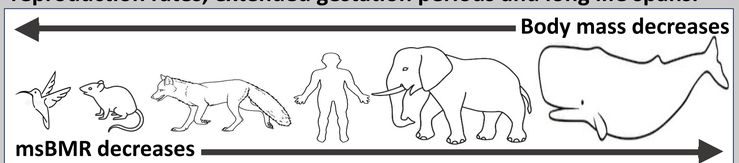
Background

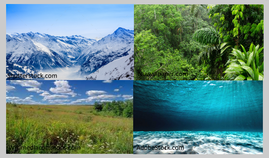
Allometry influences all aspects of life[1]

Figure 1. Mammals that display distinct characteristics such as quick reproduction rates, extended gestation periods and long life spans.

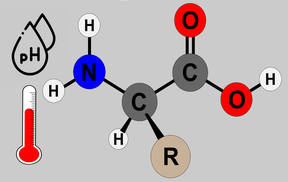


Mass-specific O_2 requirements for greater energy expenditure in small-bodied endotherms

Hemoglobin (Hb) is the species-specific O_2-carrying pigment[2]



External



Internal

These factors work together to maintain Hb function and stability

Hb isoelectric point (pI): pH value at which the protein molecule exerts a neutral net charge[3]

- Positive and negative charges are equal
- Altered by charged amino acid residue interactions

Hb structure

Methods

Body masses and pI values obtained from literature sources

Globin sequences downloaded from Genbank

Objectives & Hypothesis

To compare the amino acid sequences of endotherms with different body sizes and determine if the positive charges differ between small and large endotherms

Hypothesis: positively charged residues are favoured in the α- and β-chains of small endotherms to keep an overall positive net surface charge of Hb

Predictions: positive amino acids will be dominant in the globin chains of smaller species

Preliminary Results

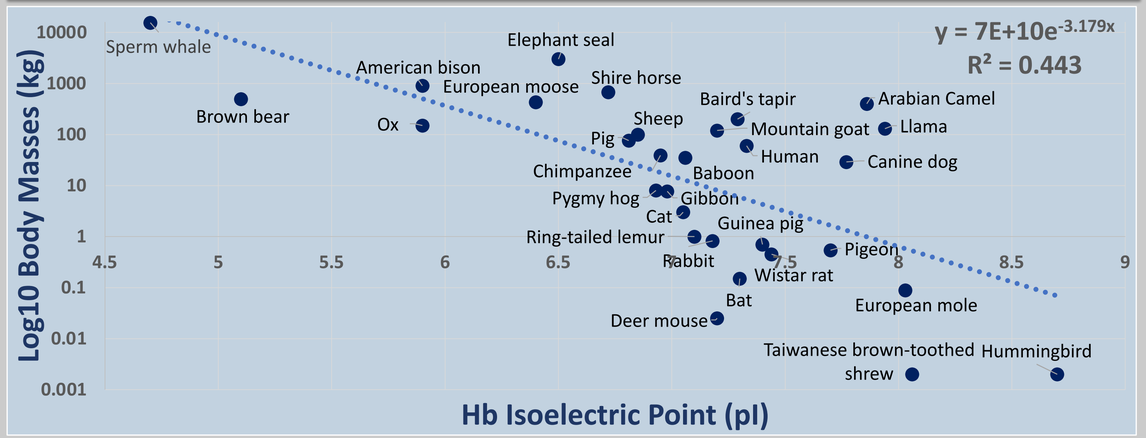


Figure 2. The high pI suggests small endotherms favour positive charges as a potential mechanism to aid in increased O_2 unloading within the tissues.

Comparing the amino acid sequences of 2 small and 2 large species

1. *Suncus murinus* (House shrew)

α: VLSANDKANVKAAWDKVGGQAANYGAEALERTFASFPTTKTYFPHYDLSPGSAQVKAHGKKVADALTKAVHSMDDLPGALSALSDLHAHKLRVDPVNFKLLS HCLLVTLAAHHPADFTPAVHASLDKFLASVSTVLTSKYR

β: VHLSGEEKACVTGLWGKVNEDEVGAEALGRLLVVYPWTQRFFDSFGDLSSASAVMGNPKVKAHGKKVLHSLGEGVANLDNLKGTFAKLSELHCDKLHVDPEN FRLLGNVLVVVLASKFGKEFTPPVQAAFQKVVAGVANALAHKYH

2. *Talpa europaea* (European mole)

α: VLSGTDKSNIKAAWDKVGAHAGEYGAEALERTFTSFPTTKTYFPHFDLSHGSAQVKAHGKKVADALTNAVGHLDDLPGAMSALSDLHAHKLRVDPVNFKLLS HCLLVTLACHHPNDFTPAVHASLDKFLATVSTVLTSKYR

β: VHLSGEEKGLVTGMWGKVNVDEVGGEALGRLLVVYPWTQRFFDSFGDLSSASAIMGNAKVKAHGKKVANSITDGVKNLDNLKGTYAKLSELHCDKLHVDPEN FRLLGNVLVCVLARNLGKEFTPQAQAAFQKVVLGVATALAHKYH

3. *Elephas maximus* (Indian elephant)

α: VLSDKDKTNVKATWSKVGDHASDYVAEALERMFFSFPTTKTYFPHFDLSHGSGQVKGHGKKVGEALTQAVGHLDDLPSALSALSDLHAHKLRVDPVNFKLLS HCLLVTLSSHQPTEFTPEVHASLDKFLSNVSTVLTSKYR

β: VNLTAAEKTQVTNLWGKVNVKELGGEALSRLLVVYPWTRRFFEHFGDLSTADAVLHNAKVLAHGEKVLTSFGEGLKHLDNLKGTFADLSELHCDKLHVDPEN FRLLGNVLVIVLARHFGKEFTPDVQAAYEKVVAGVANALAHKYH

4. *Phyester catodon* (Sperm whale)

α: VLSPADKTNVKAAWAKVGNHAADFGAEALERMFMSFPSTKTYFSHFDLGHNSTQVKGHGKKVADALTKAVGHLDTLPDALSDLSDLHAHKLRVDPVNFKLLS HCLLVTLAAHLPGDFTPSVHASLDKFLASVSTVLTSKYR

β: VHLTGEEKSGLTALWAKVNVEEIGGEALGRLLVVYPWTQRFFEHFGDLSTADAVMKNPKVKKHGQKVLASFGEGLKHLDNLKGTFATLSELHCDKLHVDPEN FRLLGNVLVVVLARHFGKEFTPELQTAYQKVVAGVANALAHKYH

Indicates positive charges with the following labelled: R = Arginine (Arg); H = Histidine (His); K = Lysine (Lys)

Preliminary Results Cont.

Based on residue substitutions, we find:

1. Conservative substitutions in similar body masses (overall minor effect on Hb structure)
2. Positive charges had little variation across globin chains
3. Small endotherms had a pattern of picking up positive charges (Table 1)

Compared species	β-chain position
Sperm whale vs House shrew	21Asp → 21Arg 43Asp → 43His
Indian elephant vs European mole	65Glu → 65Lys

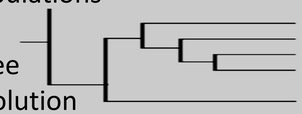
Table 1. Substitutions made on beta chain positions. Aspartic (Asp) and glutamic (Glu) acid are negatively charged residues.

Conclusion

Hb may have convergently evolved in small endotherms for greater O_2 affinity
- Positive charges may indicate more Hb concentration in the red blood cells for increased Hb-O_2 binding

The next step in examining residue substitutions:

1. Take large sampling populations
2. Compare substitutions
3. Plot on phylogenetic tree
4. Look for convergent evolution



References

[1]Reiss, Michael J. 1991. The Allometry of Growth and Reproduction. Cambridge University.

[2]Prisco, Di G., Giardina, B., & Weber, R. E. 2000. Hemoglobin Function in Vertebrates: Molecular Adaptation in Extreme and Temperate Environments, Springer.

[3]Briehl, Robin W. 1970. Relations between aggregation of subunits and the oxygen equilibrium of human hemoglobin. *Journal of Biological Chemistry* 245.3:538-543.

Acknowledgements

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