Identifying Abnormal Hemoglobin in Human Blood

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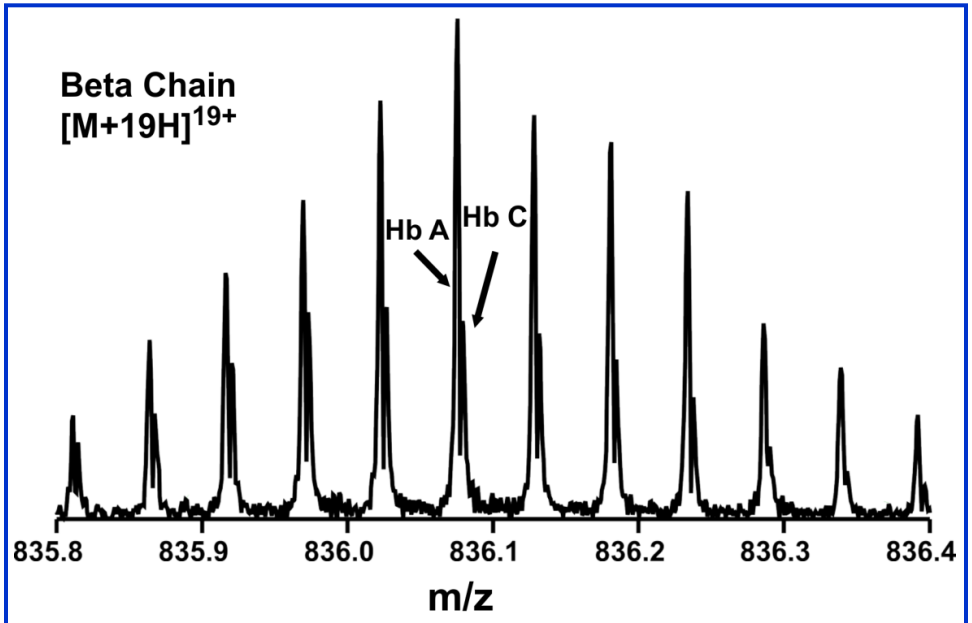
Human hemoglobin (Hb) is a protein responsible for oxygen transport in human blook. Hb consists of two α and two β chains. More than 1,500 structurally abnormal Hbs have been identified, some of which cause debilitating diseases.

"Homozygous" people (e.g., 3.2 million for sickle cell) have mutations on both chains, and "heterozygous" (46 million for sickle cell) have one normal and one abnormal chain. *There is currently no rapid way to confidently identify unknown Hb variants. Here, we report comprehensive characterization of Hb in human blood by 21 T FT-ICR MS/MS for fast and accurate clinical diagnosis.*

Aqueous buffer is added to red blood cells to break the cells and release the Hb. After centrifugation to remove cell debris, the solution is electrosprayed to electrically charge the Hb molecules. The resulting Hb ions are directly infused into MagLab's custom-built 21T Fourier Transform - Ion Cyclotron Resonance (FT - ICR) mass spectrometer and broken into small fragments. The masses of the fragments reveal the identity and location of the mutated amino acid(s) within the sequence of amino acids that form the Hb subunit chains.

The method has successfully identified both heterozygous and homozygous Hb variants in a few minutes, including a newly discovered variant. The method promises to provide clinicians with a valuable new tool for personalized medicine.



21T FT - ICR mass spectrum for the human "AC Hb" β chain 19+ charge state (i.e., neutral β-chain plus 19 additional protons). "Hb AC" denotes heterozygous β-chains: one normal, and one with a single mutation of glutamic acid to lysine at amino acid sequence position 6. Resolution of the overlapping isotopic distributions (representing various numbers of heavy isotopes in the β-chain) is possible only with ultrahigh-resolution FT - ICR Mass Spectrometry. This resolution identifies Hb that features the sickle cell variant.

Facilities and instrumentation used: Ion Cyclotron Resonance (21 T FT-ICR Mass Spectrometer)

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