CO 2 -scrubbing, zero gases, Keeling plots, and a new mathematical approach to ameliorate the deleterious effects of ambient CO 2 during 13 C-breath testing in humans and animals

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Abstract

^{13}C-Breath testing is increasingly used in physiology and ecology research because of what it reveals about the different fuels that animals oxidize to meet their energetic demands. Here we review the practice of ^{13}C-breath testing in humans and other animals and describe the impact that contamination that ambient/background CO $_2$ in the air can have on the accuracy of ^{13}C-breath measurements. We briefly discuss physical methods to avoid sample contamination as well as the Keeling plot approach that researchers have been using for the past two decades to estimate δ ^{13}C from breath samples mixed with ambient CO $_2$. Unfortunately, Keeling plots are not suited for ^{13}C breath testing in common situations where 1) a subject's VCO $_2$ is dynamic 2) ambient [CO $_2$] may change, 3) a subject is sensitive to hypercapnia, or 4) in any flow-through indirect calorimetry system. As such, we present a mathematical solution that addresses these issues by using information about the instantaneous [CO $_2$] and the δ ^{13}CO $_2$ of ambient air as well as the diluted breath sample to back-calculate the δ ^{13}CO $_2$ in the CO $_2$ exhaled by the animal. We validate this approach by titrating a sample of ^{13}C-enriched gas into an air stream and demonstrate its ability to provide accurate values across a wide range of breath and air mixtures. Researchers can now instantaneously calculate the δ ^{13}C of alveolar gas of humans or animals in real time without having to scrub ambient CO $_2$ or rely on estimated values.

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