



Calibration of microBIOMETER® to units of μg microbial carbon / gram soil

The gold standard of laboratory soil microbial biomass testing is Chloroform Fumigation and Extraction (CFE). The multiple steps, time, and labor involved with CFE require pricing at up to \$500 per sample. CFE works by comparing the difference of chemically extractable carbon between two portions of a soil sample: One that has been treated to break open microbial cell membranes and expose the carbon-containing biological molecules to extraction, and one that has not. The difference in carbon for the two portions is reported as microbial biomass carbon (MBC), in units of $\mu\text{g C} / \text{g}$ soil.

microBIOMETER® is calibrated to the same units by a different method. Estimates of bacterial dry mass converge at around one trillionth (1×10^{-12}) of a gram (1 pg) for a 1 μm bacterium. We measured the area of microbes in known volumes of microBIOMETER® extract (both by manual counting on a hemocytometer and by digital analysis of micrographs) and calculated total microbial mass, which was then converted to $\mu\text{g} / \text{g}$ for the whole 0.5 ml sample of soil in the extract. We found that on average, 0.5 ml of soil weighs 0.6 g when fully dried, independent of starting moisture content. The 1 pg dry mass per bacterium is 50% carbon, so we also had to account for that in our calibration.

Here's an example of the conversion.

Let's say that in 1×10^{-8} liter (10 nl) of microBIOMETER® extract we measured $240 \mu\text{m}^2$ of microbes. $240 \mu\text{m}^2 = 240$ bacteria equivalents (BE). $240 \text{ BE} \times 1 \times 10^{-12} \text{ g per BE} = 240 \times 10^{-12} \text{ g}$ of dry microbes. The volume of original extract is 10 ml (1×10^{-2} liter), and 10 nl of microscopically examined extract represents $1 \times 10^{-8} / 1 \times 10^{-2} = 1 \times 10^{-6}$ of the total mass of the microbes in the extract. So $240 \times 10^{-12} \text{ g microbes} / 1 \times 10^{-6} = 240 \times 10^{-6} \text{ g microbes}$ in the whole extract. 50% of the $240 \times 10^{-6} \text{ g}$ of microbes is carbon, so we have $120 \times 10^{-6} \text{ g microbial carbon}$. We started with 0.5 ml = 0.6 grams of dried soil in the extraction process, therefore $120 \times 10^{-6} \text{ g microbial carbon} / 0.6 \text{ g soil} = 200 \times 10^{-6} \text{ g microbial carbon} / \text{gram soil}$, or $200 \mu\text{g microbial carbon} / \text{gram soil}$.

While we arrived at μg microbial carbon / gram soil through a different method than CFE, it turns out our methods are on par with the ~\$500 CFE test. We compared measurements of μg carbon / gram soil via CFE and microBIOMETER® from 28 soils from across the U.S.

The slope of ~ 1 of the regression line indicates our units are on par with CFE, and the 95% correlation indicates that users can be confident that the \$10 or less microBIOMETER® test gives results as accurate and informative as one priced \$500.

