Response to “Comments on ‘Evaluation of the Microwave Irradiation Method for Measuring Soil Microbial Biomass’”

We welcome the response from Dr. Weil and Dr. Islam regarding the microwave (MW) irradiation method for measuring soil microbial biomass (MBC).

Islam and Weil (1998) initially determined the optimum level of MW irradiation with 100 g soil packed in a 50-mL (original number) beaker, but recommended using twelve 10-g sample tubes per batch for routine operation, without description on how the tubes were arranged in the rack. We found that MW delivery to 24 tubes arranged in a square rack had significant corner and edge heating effect and the sample temperatures after irradiation varied from 60 to 88°C (Wang et al., 2001). Contrary to the critiques by Weil and Islam (2003), we cannot see any inappropriateness in the irradiation time for this test (e.g., no boiling), and the spatial variation is irrelevant to MW oven calibration. Islam and Weil (1998) calibrated a manufacturer-rated 650-W MW oven with a procedure that differed from the International Standard, IEC 705, and obtained an output of 640 W (J s⁻¹). But they irradiated 100 g soil for 60 s to achieve 400 J g⁻¹ soil MW application, which could only have been achieved using a 670-W MW oven. In fact, we calibrated our MW ovens using the method of IEC 705 as well as that described by Islam and Weil (1998) and found that the latter gave lower wattage values than the former.

Islam and Weil (1998) found that sample temperatures after each MW treatment at 400 J g⁻¹ soil were ≈82°C, regardless of soil water content that varied from 15 to 32% for three different soils and from 80% water-filled porosity (WFP) to ~11% and obtained resultant temperatures of 96 to 97°C at 400 J g⁻¹ (for 45 s) in a 100-mL beaker covered with a Petri dish and obtained resultant temperatures of 96 to 97°C as measured with an inserted mercury thermometer (MT). Abrupt steam emission from the beaker was sometimes observed. The lower temperature measured by Islam and Weil (1998) might have resulted from the use of a thermistor (see below).

Islam and Weil (1998) operationally used 80% WFP as the standard moisture for MW irradiation of all soils, but did not give any rationalization. The absolute amount of water at the same percentage WFP or water-holding capacity could differ considerably for different soils. Because the amount of water in soil significantly affects the resultant temperature when the samples were exposed to the same MW dosage (J g⁻¹ soil), we operationally moistened soils with same amount of water (50%) on the basis of mass instead of percentage WFP. Weil and Islam (2003) were concerned by this modification because many of the samples were irradiated at >100% WFP. However, there seems no reason to keep soil moisture at <100% measured with a thermistor and an IRT by Weil and Islam (2003) might have resulted from a few factors: (i) the temperature of 10-g sample could decrease markedly because a reacting thermistor probe gave a stable reading; (ii) the 10-g sample might be insufficient for the probe to give a correct reading; (iii) the metal probe could conduct heat from the surrounding soil and thus underestimate temperature; (iv) the operator failed to keep a reasonably close distance between the sample and the IRT sensor for MW readings at different spots of an irradiated MW tube could vary a few degrees, due to uneven MW delivery to the sample rather than the problem of the IRT temperature should be determined by twisting the tube quickly.

Weil and Islam (2003) reprocessed our data on the 21 of 30 soils with clay content <50% and pH <7.5. Analyses with the 21 soils showed: rCFE vs. MWE was 0.985 (P<0.01) > rMWE vs. CFI = 0.31, and rCFE vs. SIR (0.01) > rMWE vs. SIR = 0.67 (P<0.01) (SIR = substrate-induced respiration). Contrary to their claim, CFI was superior to MWE in terms of their correlations with MBC. However, the practice of excluding the nine or 10 soils by the standard CFI (CFE) would be an approximate for predicting MBC. Weil and Islam (2003) criticized these soils as being “highly unique” and it is very important to test the MW method using soils having a wide range of properties.

Weil and Islam (2003) assessed our MBC estimates of the percentage of total organic C (TOC) as MBC they did not exclude the soils that had >250 mg kg⁻¹ as estimated with CFI. Averaged across all soils, conventional methods, CFE, CFI, and SIR, consistently give values lower than the MWE-MBC. Because the soil was air-dried, stored, and remoistened, it was normal for some MBC > 250 mg kg⁻¹ as estimated with CFI. Weil and Islam (2003) criticized these soils as being “highly unique” and it is very important to test the MW method using soils having a wide range of properties.

The optical densities at 410 nm of the 0.5 M KOH following MW irradiation were significantly higher in counterparts following chloroform fumigation (CF) number of soils (Wang et al., 2001). Weil and Islam (2003) that the difference in optical density between the two (0.072) and CF treatments (0.052 ± 0.067) was averaged across all soils. However, the numbers indicate variations among soils. Assuming that soil microbial constituents MW and CF were equally extractable in 0.5 M KOH, the difference in the amounts of organic C extracted by MW and after CF (C_MWE - C_CFE) would be an indication of the extra amount of nonbiomass C present in MW. Figure 1 shows that C_MWE was larger than that.