To determine the optimal dispersion energy that separated clay from quartz and silicate minerals we tested 300, 500, 900 and 1500 J ml^{-1}, and a control with deionised water and no ultrasonic dispersion. We would then select the dispersion energy that separated the minerals, whilst minimising and contamination of the MAOC fraction Dispersion energy applied to the sample was calculated according to equation 1 (Schmidt et al., 1999). Where

$$E = \frac{P\,t}{V},\tag{1}$$

where P is the power output in watts (W), t is the sonication time in seconds (s) and V is the volume of the suspension in millilitres (ml). The dispersed soil samples were run through the wet sieving procedure. Each dispersion treatment was replicated three times for both soil types. We then used MIR spectroscopy to semi-quantitatively asses the changes in composition of the soil fractions. We found that 500 J ml^{-1} was an optimal energy that balanced the separation of macroaggregates with minimal damage to mineral or microaggregate structures (See Supplementary Information). We used this dispersion energy for all fractionations reported in this study.

Assessment of dispersion with peak height

To test the effect of ultrasonic dispersion on the mineral components of the soil fractions we used the relative peak height from each spectra in regions assigned to key mineralogical absoprtions. We used the 913 cm^{-1} absorption for kaolinite, and 1158 cm^{-1} for quartz. Here we assume that the kaolinite minerals should be more prevalent in the finer fractions and quartz mineral more associated with the coarser fractions. Spectral replicates were averaged before analysis and the average spectra were not pre-processed. Once the region was selected the spectra were baselined to isolate the desired peaks and a relative peak height was calculated.

Selecting the optimal dispersion intensity

We found that the effects of ultrasonic dispersion were specific to soil type. Increasing dispersion energy did not alter the mean peak height of kaolinite mineral absorption (913cm^{-1}) in the POC$_{mac}$ fraction of the clay soil. Whereas in the sandy soils, this peak significantly decreased in the POC$_{mac}$ and increased in the MAOC as dispersion energy increased (Fig. S1). However, as dispersion energy increased over 500 J ml^{-1} peak height for kaolinite did not increase further in the MAOC fraction. Alternatively, the quartz peak (1158 cm^{-1}) increased in the MAOC fraction of the sandy soil at 900 J ml^{-1} (Fig S1). We therefore selected 500 J ml^{-1} as our dispersion energy for the subsequent analysis as it effectively breaks aggregates without further affecting the minerals within the soil.

A dispersion energy of 500 J ml^{-1} was selected as improved the break down of aggregates to increase the presence of kaolinite in the MAOC fraction, without breaking the mineral structures (Fig. S1). Applying excessive dispersion energy (>900 J ml^{-1}) to the sample increased the presence quartz minerals in the MAOC fraction (Fig S1).
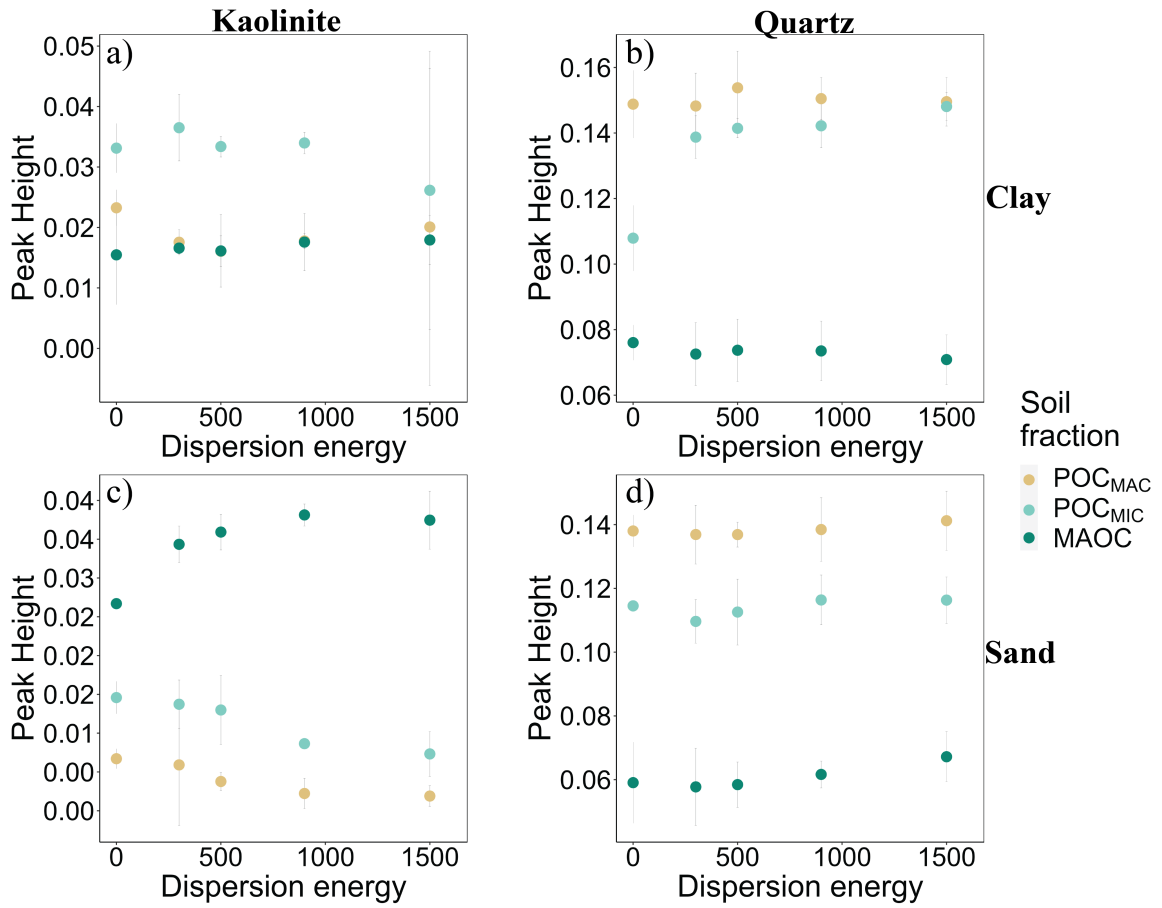
Figure S1: Mean peak heights of soil minerals kaolinite (913 cm^{-1}) and quartz (1158 cm^{-1}) used to determine the optimal dispersion energy for both SCL and S soil type. a) Mean peak height in clay soil at kaolinite absorption (913 cm^{-1}), b) mean peak height in clay soil at quartz absorption (1158 cm^{-1}), c) mean peak height in sand soil at kaolinite absorption, d) mean peak height in sand at quartz absorption. Bars represent the 95% confidence interval of the mean.

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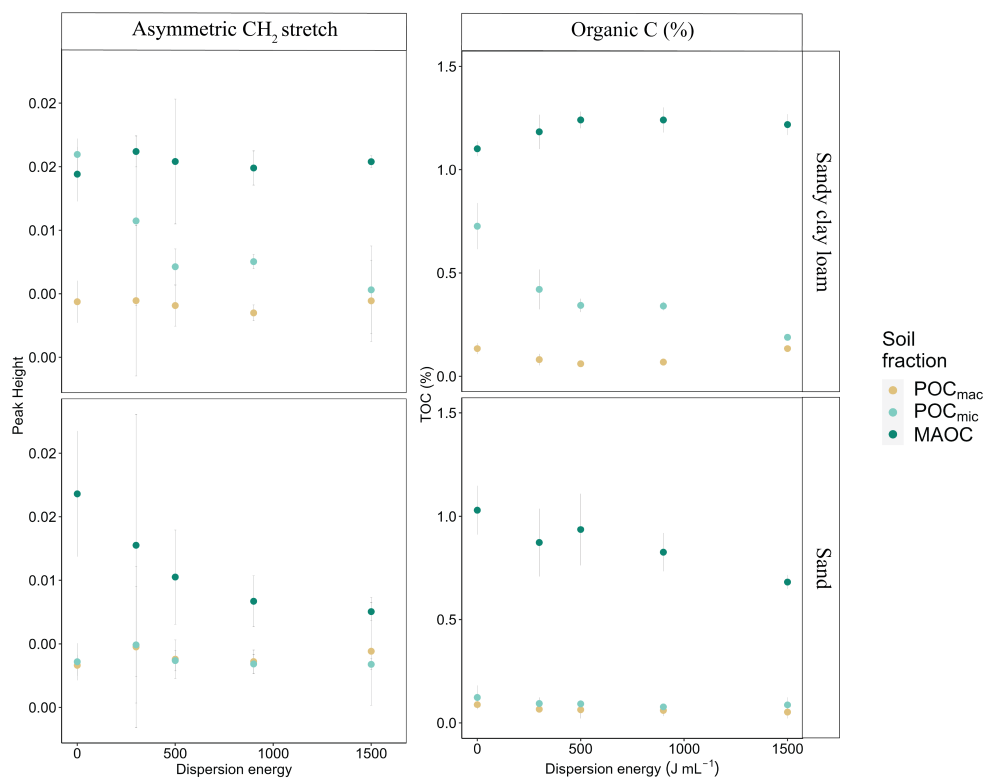
Figure S2: Mean peak heights of soil minerals asymmetrich CH2 stretch (2980 cm^{-1}) and total organic C (%) used to determine the optimal dispersion energy for both SCL and S soil type. Bars represent the 95% confidence interval of the mean.

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References

Schmidt, M. W., Rumpel, C., and Kögel-Knabner, I.: Evaluation of an ultrasonic dispersion procedure to isolate primary organomineral complexes from soils, European Journal of Soil Science, 50, 87–94, https://doi.org/10.1046/J.1365-2389.1999.00211.X, 1999.