The green ciliate Paramecium bursaria contains several hundred symbiotic Chlorella species. We previously reported that symbiotic algal carbon fixation is enhanced by P. bursaria extracts and that the enhancing factor is a heat-stable, low-molecular-weight, water-soluble compound. To identify the factor, further experiments were carried out. The enhancing activity remained even when organic compounds in the extract were completely combusted at 700 °C, suggesting that the factor is an inorganic substance. Measurement of the major cations, K\(^+\), Ca\(^{2+}\), and Mg\(^{2+}\), by an electrode and titration of the extract resulted in concentrations of 0.90 mM, 0.55 mM, and 0.21 mM, respectively. To evaluate the effect of these cations, a mixture of the cations at the measured concentrations was prepared, and symbiotic algal carbon fixation was measured in the solution. The results demonstrated that the fixation was enhanced to the same extent as with the P. bursaria extract, and thus this mixture of K\(^+\), Ca\(^{2+}\), and Mg\(^{2+}\) was concluded to be the carbon fixation-enhancing factor. There was no effect of the cation mixture on free-living C. vulgaris. Comparison of the cation concentrations of nonsymbiotic and symbiotic Paramecium extracts revealed that the concentrations of K\(^+\) and Mg\(^{2+}\) in nonsymbiotic Paramecium extracts were too low to enhance symbiotic algal carbon fixation, suggesting that symbiotic P. bursaria provide suitable cation conditions for photosynthesis to its symbiotic Chlorella.