Anaerobic digestion of animal manure and crop residues may be employed to produce biogas as a climate-neutral source of energy and to recycle plant nutrients as fertilizers. However, especially organic farmers are concerned that fertilizing with the digestates may impact the soil microbiota and fertility because they contain more mineral nitrogen (N) and less organic carbon (C) than the non-digested input materials (e.g. raw animal slurry or fresh plant residues). Hence, an incubation study was performed where (1) water, (2) raw cattle slurry, (3) anaerobically digested cattle slurry/maize, (4) anaerobically digested cattle slurry/grass-clover, or (5) fresh grass-clover was applied to soil at arable realistic rates. Experimental units were sequentially sampled destructively after 1, 3 and 9 days of incubation and the soil assayed for content of mineral N, available organic C, emission of CO2 and N2O, microbial phospholipid fatty acids (biomass and community composition) and catabolic response profiling (functional diversity). Fertilizing with the anaerobically digested materials increased the soil concentration of NO3− ca. 30–40% compared to when raw cattle slurry was applied. Grass-clover contributed with four times more readily degradable organic C than the other materials, causing an increased microbial biomass which depleted the soil for mineral N and probably also O2. Consequently, grass-clover also caused a ~10 times increase in emissions of CO2 and N2O greenhouse gasses compared to any of the other treatments during the 9 days. Regarding microbial community composition, grass-clover induced the largest changes in microbial diversity measures compared to the controls, where raw cattle slurry and the two anaerobically digested materials (cattle slurry/maize, cattle slurry/grass-clover) only induced minor and transient changes.