Analysis of bacterial communities and bacterial pathogens in a biogas plant by the combination of ethidium monoazide, PCR and Ion Torrent sequencing

The present study investigated the changes of bacterial community composition including bacterial pathogens along a biogas plant, i.e. from the influent, to the biogas reactor and to the post-digester. The effects of post-digestion temperature and time on the changes of bacterial community composition and bacterial pathogens were also studied. Microbial analysis was made by Ion Torrent sequencing of the PCR amplicons from ethidium monoazide treated samples, and ethidium monoazide was used to cleave DNA from dead cells and exclude it from PCR amplification. Both similarity and taxonomic analysis showed that the bacterial community composition in the influent was changed after anaerobic digestion. Firmicutes were dominant in all the samples, while Proteobacteria decreased in the biogas reactor compared with the influent. Variations of bacterial community composition in the biogas reactor with time were also observed. This could be attributed to varying composition of the influent. Batch experiments showed that the methane recovery from the digested residues (obtained from biogas reactor) was mainly related with post-digestion temperature. However, post-digestion time rather than temperature had a significant effect on the changes of bacterial community composition. The changes of bacterial community composition were also reflected in the changes of relative abundance of bacterial pathogens. The richness and relative abundance of bacterial pathogens were reduced after anaerobic digestion in the biogas reactor. It was found in batch experiments that bacterial pathogens showed the highest relative abundance and richness after 30days' post-digestion. Streptococcus bovis was found in all the samples. Our results showed that special attention should be paid to the post-digestion since the increase in relative abundance of bacterial pathogens after post-digestion might reflect regrowth of bacterial pathogens and limit biosolids disposal vectors. © 2014 Elsevier Ltd.