Microbial protein production from biogas: developing methanotrophic enrichments with potential for aquafeed

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Methanotrophic bacteria (MOB) are great allies in the fight against climate change and crucial in the carbon cycle by reducing methane (CH_4) emissions. Moreover, they are valuable for sustainable biotechnology, such as producing protein-rich biomass for animal feed. Current processes use natural gas, however, biogas from anaerobic digestion of organic wastes is a renewable, low-cost alternative. Biogas-derived MOB production represents an alternative biogas valorisation route with circular economy potential, promoting waste treatment and generating products valuable for feed. Developing microbial cultures able to use raw biogas and optimizing mass transfer rate are the key steps toward viability.

Aiming high biogas affinity, four MOB enrichments were developed at 30°C using DNMS medium and a headspace mixture of biogas and air (50/50%) at moderate pressure (200kPa) for better mass transfer between phases. Four inocula were used: anaerobic suspended sludge (SS), and sediment samples of São Miguel Island lagoons (Azores, PT): Seven Cities (7C), Fire (FO) and Furnas (FU). Enrichments were characterized in terms of performance, protein content, and microbial communities' composition (16S rRNA sequencing).

All enrichments have similar protein content (~50%, g protein g-1 dry weight) but differentiated microbial communities. SS is 97.7% enriched in Type I MOB from the genus *Methylococcus* spp. being taxonomically close to strain *Methylococcus geothermalis* IM1. 7C, FO, and FU are respectively 98.7%, 41.1% and 46.5% enriched in Type II MOB, but only FO was taxonomically identified (*Methylosinus sporium*).

Four promising MOB enrichments were developed for microbial protein production intended for aquafeeds, showing strong adaptation to biogas as carbon and energy source. Their distinct microbial compositions highlight the potential to magnify underutilized microorganisms for inclusion in aquafeeds and enhance process productivity.