

1 - EVAN LAU, DANIEL LUKICH, and TUAN LE, Department of Natural Sciences and Mathematics, West Liberty University, West Liberty, WV, 26074. The evolution of the AMOA, PMOA and BMOA genes measured by k_a/k_s ratios.

The copper-containing membrane-bound monooxygenase enzyme family consists of enzymes facilitating the oxidation of methane (catalyzed by particulate methane

monooxygenase, pMMO) and ammonia (catalyzed by ammonia monooxygenase, AMO), in both aerobic and anaerobic methanotrophs and nitrifiers, respectively, and the oxidation of C2-10 alkanes (catalyzed by particulate butane monooxygenase, pBMO). Despite catalyzing different substrates, pMMO, AMO and pBMO form catabolic enzymes related structurally and evolutionarily to each other, and are encoded by the genes *pmoA*, *amoA* and *bmoA*, respectively. It has been suggested that oxygen-dependent methane and ammonia monooxygenases evolved from a substrate-promiscuous ancestor after horizontal transfer(s) into new hosts, which eventually became methanotrophs and nitrifiers. However, no extensive studies have been conducted on molecular adaptation of the beta-peptide structure of *pmoA*, *amoA* and *bmoA* gene products, which form transmembrane polypeptides, and on selective pressures acting on them. Over 80 near-complete *pmoA*, *amoA* and *bmoA* gene from database were downloaded and analyzed to assess whether the combined *amoA*, *pmoA* and *bmoA* gene tree is congruent to the 16S rRNA gene tree for these microbes. The gene was separated into 15 segments and pairwise K_a/K_s values were calculated to assess the evolutionary and selection pressure exerted on these taxa, based on the hypothesis that they evolved from a methanotrophic ancestor, as well as calculate K_a/K_s values between members of each family. Our analyses indicated a high level of negative (or purifying) selection between and amongst all taxa. K_a/K_s values were highest between methanotroph *pmoA* and the anammox, and that the *pmoA* and *amoA* genes are under purifying selection their clusters, and supports the hypothesis that one of these genes likely arose from the other via horizontal gene transfer.