Proatherosclerotic effect of trimethylamine-N-Oxyde: in vitro preliminary evidences

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Aim: following the ingestion of red meat, chicken, eggs and dairy products, L-carnitine and phosphatidylcholine undergo microbic and hepatic metabolism, leading to the formation of trimethylamine-N-oxide (TMAO). Recent metabolomic studies positively associated plasma levels of TMAO with incident and prevalent cardiovascular disease, but the mechanisms accounting for this relation are still lacking. The aim of this work is to evaluate TMAO capacity to impair lipid metabolism in cultured macrophages.

Methods: cultured peritoneal macrophages from C57BL/6 mice (MPM) were exposed to TMAO 10-100µM for 2-24h in presence or absence of acetylated LDL 25µg/ml. Cell cholesterol content was evaluated by a fluorimetric technique. Protein expression of the scavenger receptor CD36 was assesed by western blot. Cholesterol efflux was quantified through a radioisotopic method upon cell exposure to apoA-I 10µg/ml, HDL 12.5µg/ml or murine plasma 0.5% as lipid acceptors.

Results: TMAO 100µM, but not 10 µM, incubated for 24h prior to AcLDL significantly increased cell cholesterol content compared to control MPM: 30.85±6.77 vs 16.20±0.86 (µg cholesterol/mg protein, mean ± standard deviation; $p<0.05$) and induced a significant raise of CD36 expression. Shorter times of pre-incubation did not produce any effect. Similarly, when simultaneous incubation of TMAO 100µM and AcLDL was carried out, no changes in cholesterol content were observed. TMAO did not affect cholesterol efflux from MPM in any tested conditions.

Conclusions: TMAO exerts a proatherosclerotic activity in vitro by increasing CD36-mediated uptake of cholesterol. Further studies are necessary to establish the molecular mechanisms accounting for this effect.