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A paracrine mechanism promotes kidney stone formation in a simulated metabolic syndrome environment using in vitro and in vivo studies

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Purpose: This study aimed to investigate the molecular communication mechanism in kidney stone formation under metabolic syndrome (MetS) condition using *in vitro* co-culture system and *in vivo* obesity stone model.
Materials and Methods: 1) *in vitro* study; Mouse renal tubular cells (M-1) were cocultured with adipocytes (3T3-L1) and/or macrophages (RAW264.7), and they were divided into four groups; (1) control; M-1s, (2) M-T; co-culture of M-1s and 3T3-L1s, (3) M-R; co-culture of M-1s and RAW264.7s, and (4) M-T-R; co-culture of whole type of the cells. Calcium oxalate monohydrate (COM) crystals were exposed to M-1s after 48h of co-culture and the COM crystals attachment to the M-1s' surface were quantified. 2) *in vivo* study; wild-type (+/+) and ob/ob mice (having a disorder to produce leptin) were administered daily doses of 50 mg/kg glyoxylate (GOx) for 6 days.
Results: 1) *in vitro* study; The amount of COM crystals attached to M-1s showed significantly higher value with M-T, M-R and M-T-R than control. qPCR for M-1s demonstrated significantly increased expression of *Ccl2* and *Tnf* in M-T, *Spp1* in M-R, and *Spp1*, *Ccl2* and *Tnf* in M-T-R compared to control. ELISA for the co-cultured medium showed significant increase of MCP-1 and TNF-α in M-T, OPN in M-R, and OPN, MCP-1, TNF-α and IL-6 in M-T-R compared to control. 2) *in vivo* study; the stone formation was determined only in ob/ob on the sixth administration day, and the expression of 4 genes and the macrophage migration indicated significantly higher value than +/+.
Conclusions: This study indicated the possibilities that tubular cells under MetS environment caused inflammation by the coexistence with the adipocytes and increased OPN expression by the coexistence with the macrophages. Furthermore, the coexistence of adipocytes and macrophages could promotes these changes which lead to stone formation by paracrine via IL-6 expression.