Thimerosal Induces Skin Pseudo-allergic Reaction via Mas-related G-protein-coupled Receptor B2

Bin Peng[1], Delu Che[2], Yong Hao[1], Yi Zheng[1], Ye Qian[3], Langchong He[2], Songmei Geng[1]

1. Department of Dermatology, Northwest Hospital, The Second Hospital Affiliated to Xi'an Jiaotong University, Xi'an, Shaanxi, China
2. School of Pharmacy, Xi'an Jiaotong University, Xi'an, Shaanxi, China
3. Department of Dermatology, University of North Carolina at Chapel Hill, Chapel Hill, NC, USA

Correspondence: Songmei Geng, MD, PhD, gsm312@yahoo.com

Introduction

Thimerosal has been used as a preservative in many products which may cause contact dermatitis. Thimerosal-induced contact dermatitis is generally considered to be a delayed-type hypersensitivity reaction, but it is difficult to explain the fact that most patients develop an allergic reaction upon first encounter with thimerosal. Recent studies have demonstrated the association between Mas-related G-protein-coupled receptor X2 (MRGPRX2) and pseudo-allergic reactions which occur at the first contact with stimulation. This suggests the possibility that thimerosal may cause contact dermatitis via MRGPRX2 mediated mechanism.

Objectives

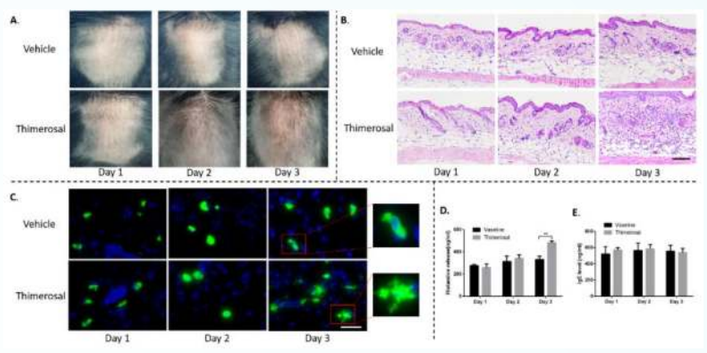
To investigate the role of Mas-related G-protein-coupled receptors (MrgprB2 and MRGPRX2 in mouse and human, respectively) in contact dermatitis induced by thimerosal.

Methodology

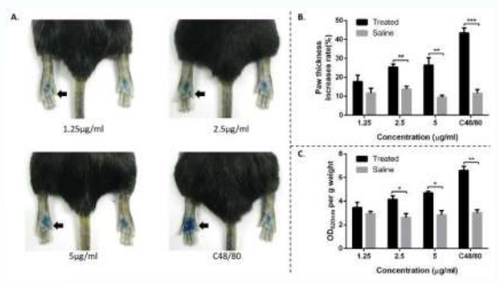
Thimerosal induced pseudo-allergic reactions via MrgprB2/ MRGPRX2 were investigated using a novel skin pseudo-allergic reaction mouse model, footpad swelling and extravasation assays *in vivo* and mast cell degranulation assay *in vitro*.

Results

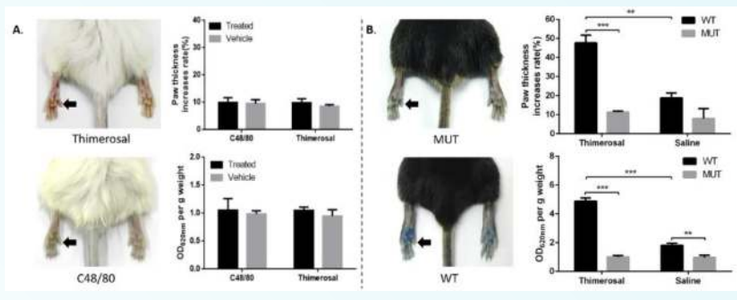
1. Thimerosal induces skin pseudo-allergic reaction in mice which is IgE-independent: Thimerosal application to WT mice induced redness and swelling of dorsal skin (Fig 1A). Profound inflammatory cell infiltration and vasodilation were observed when compared to vehicle group(Fig 1B), and the degranulation of mast cells increased dramatically in thimerosal treated skin (Fig 1C). While serum histamine increased following thimerosal application in a dose-dependent manner, there was no significant change in IgE levels (Fig 1D, E).



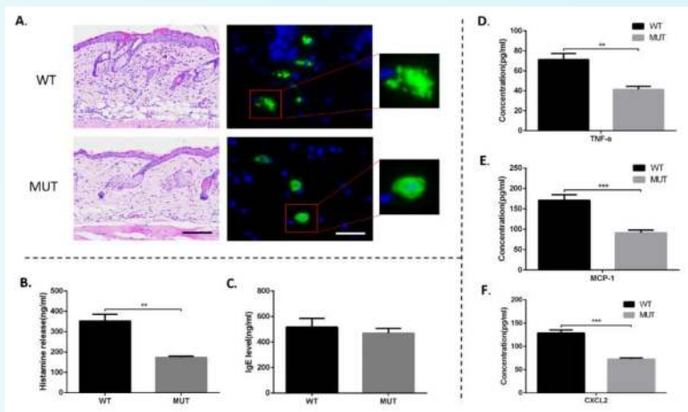
2. Pseudo-allergic reaction induced by thimerosal in mice is dose-dependent: The footpad swelling was observed after thimerosal injection, which occurred in a dose-dependent manner (Fig 2A, B). The Evans blue dye exudation also indicated that the vasodilation reaction is in a dose-dependent manner (Fig 2C).



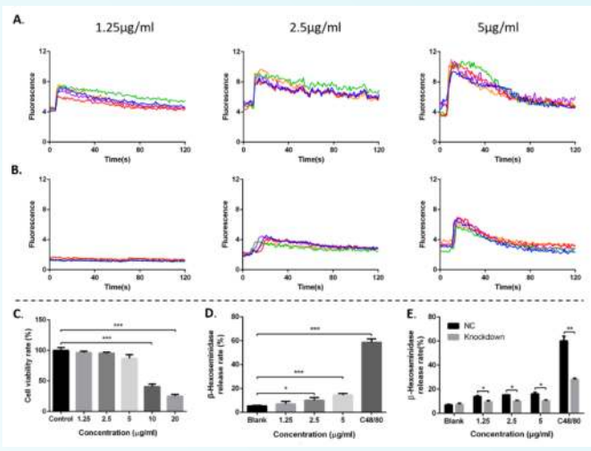
3. MrgprB2-mediated mast cells activation contributes to pseudo-allergic reaction induced by thimerosal: The footpad swelling and EBD exudation of the $Kit^{W\text{-}sh/W\text{-}sh}$ mice induced by thimerosal were similar to those observed in the negtive control (Fig 3A), which indicated that mast cells play a major role in thimerosal-induced pseudo-allergic reaction. The footpads of MrgprB2-knockout mice injected with thimerosal did not swell and showed no obvious Evans blue dye exudation (Fig 3B).



4. Skin pseudo-allergic reaction induced by thimerosal was reduced in MrgprB2 knockout mice: Infiltrated inflammatory cells were much lower in the skin of MrgprB2-knockout mice, degranulation of mast cells reduced as well (Fig 4A). The levels of inflammation related cytokines, including TNF-α, MCP-1, and CXCL2, in serum of MrgprB2-knockout mice were much lower than those of WT mice (Fig 4D, E, F)



5. Thimerosal induced human mast cell degranulation via MRGPRX2 in a dose-dependent manner: Thimerosal increased the intracellular calcium mobilization in MrgprB2/MRGPRX2-HEK293 cells(Fig 5A, B). Thimerosal induced LAD2 cells degranulation in dose-dependent manner(Fig 5D). The degranulation was much lower in MRGPRX2-knockdown LAD2 cells(Fig 5E).



Conclusions

1. MrgprB2 mediates thimerosal-induced mast cell degranulation and pseudo-allergic reaction in mice.
2. MRGPRX2 may be a key contributor to human contact dermatitis.

Acknowledgements

This work was supported by the National Natural Science Foundation of China [grant number 81171490]; the Fundamental Research Funds for the Central Universities [grant number PY3A0241001016].