首届致远学术节
学生科研成果展示

The role of ASIC3 ion channel in pathogenesis of psoriasis

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RESEARCH BACKGROUND & PURPOSE

Inflammation is a common clinical pathologic process characterized by activation of vessel response. Chronic inflammation is prone to result in inflammatory diseases, which do harm to human body, affect peoples' quality of life and become major threats of human health as various inflammatory diseases prove to be associated to carcinogenesis.

Traditionally, people focused on immune system while studying inflammation and immunology. More and more recent studies reveal that interaction between nervous system and immune system also plays significant role. Publications in Prof. Xu Lab before also indicate that certain ion channels (acid-sensing ion channel 3, ASIC3) in nervous system are involved in process of inflammation (1, 2). Psoriasis is a typical skin inflammatory disease with excess keratinocyte proliferation, inflammatory cytokine release and inflammatory cell infiltration. However, whether and how nervous system participates in psoriasis development is not yet fully understood. ASIC3 ion channel is mainly distributed in peripheral nervous system and serve for sensory transduction (pain, mechanical sense, etc.), yet it is not reported whether ASIC3 is related to peripheral inflammation.

Here we report that ASIC3 channel may play an important role in pathogenesis of psoriasis, as phenotype in ASIC3 KO and WT animals are different. We hope this research could help us understand the interaction of nervous and immune system better and point out potential target for treating psoriasis.

METHODS

Psoriasis modeling in animal: we do this by focal application of imiquimod（IMQ）cream on shaved back of C57/B6 female mice (7~10W) for 7 consecutive days (62.5 mg/d). Vasoline is used for control.

Pathological analysis: we do this by collecting back skin tissue of model animals and conduct H&E and immunohistochemistry staining to verify and evaluate the psoriasis phenotype.

Biochemical analysis: we do this by collecting back skin tissue of model animals and conduct enzyme-linked immunosorbent assay (ELISA) to detect the secretion level of certain cytokine (IL-17, IL-22, IL-23, etc.).

ASIC3 knock down: we do this by i.p. injecting AAV-GFP-ASIC3 shRNA or NC-control virus in P3 C57/B6 mice and allow them 6 weeks for virus expression.

Devervation experiment: see the reference.

Other methods are mentioned in **FUTURE PLAN** part.

RESULTS

In the study we induced psoriasis-like skin inflammation in model animals and investigated whether the phenotypes are different with or without ASIC3. After IMQ application for 7 days (Figure 1.A, B), we observed splenauxe in model animals, which can be the result of inflammation. Also we found that splenauxe in ASIC3 KO mice is less significant than that in WT mice(Figure 1.C, D). In a reference it is reported that nociceptive sensory neurons drive IL-23 mediated psoriasis-form skin inflammation (3). Based on another reference (4), we used different methods to achieve peripheral denervation and compare spleen weight in these groups(Figure 1.E, F). Our results show that ASIC3 in TRPV1$^+$ neurons may be a key molecular in driving inflammation.

At morphological level we found that ASIC3 knock out or knock down results in thinner psoriasis skin and less ki-67 staining intensity, which represent the keratinocyte proliferation in skin tissue. Also ASIC3 KO can reverse the "anti-psoriasis" effect of peripheral denervation (Figure 2).

IL-23, IL-17, and IL-22 are key cytokine in the process of psoriasis. IL-23 can be released by dendritic cells in dermal tissue and activates γδ-T17 lymphocytes to release IL-17 and IL-22. IL-17 recruits neutrophils and IL-22 induces keratinocyte proliferation. Consistently, by ELISA we found that the level of these cytokines are lower in ASIC3 KO model animals (Figure 3), which further support our hypothesis that ASIC3 is involved in the process of psoriasis.

CONCLUSIONS

Our experiment results provide basic evidence that ASIC3 in peripheral sensory neurons contributes to the process of psoriasis. Although the underlying mechanism is still not delineated, it is reasonable to suppose that ASIC3 channel can sense the change in affected tissue (as its major function) and interact with immune cells, thus influence the level of certain cytokines.

FUTURE PLAN

To further investigate the mechanism of how ASIC3 channel contribute to process of psoriasis, we plan to do transcriptome analysis of dorsal root ganglion (DRG) of model animals to find out the changes of mRNA before and after psoriasis modeling, and in WT and ASIC3 KO model animals, to screen out potential molecular intermediate (neurotransmitter, etc.) between nervous and immune system. Electrophysiological analysis of DRG can help study whether DRG response to signaling molecular from immune cells. Flow-cytometrty is also necessary for understanding which subset of immune cells are regulated by ASIC3 channel. These data will help fulfill the working model of our hypothesis.

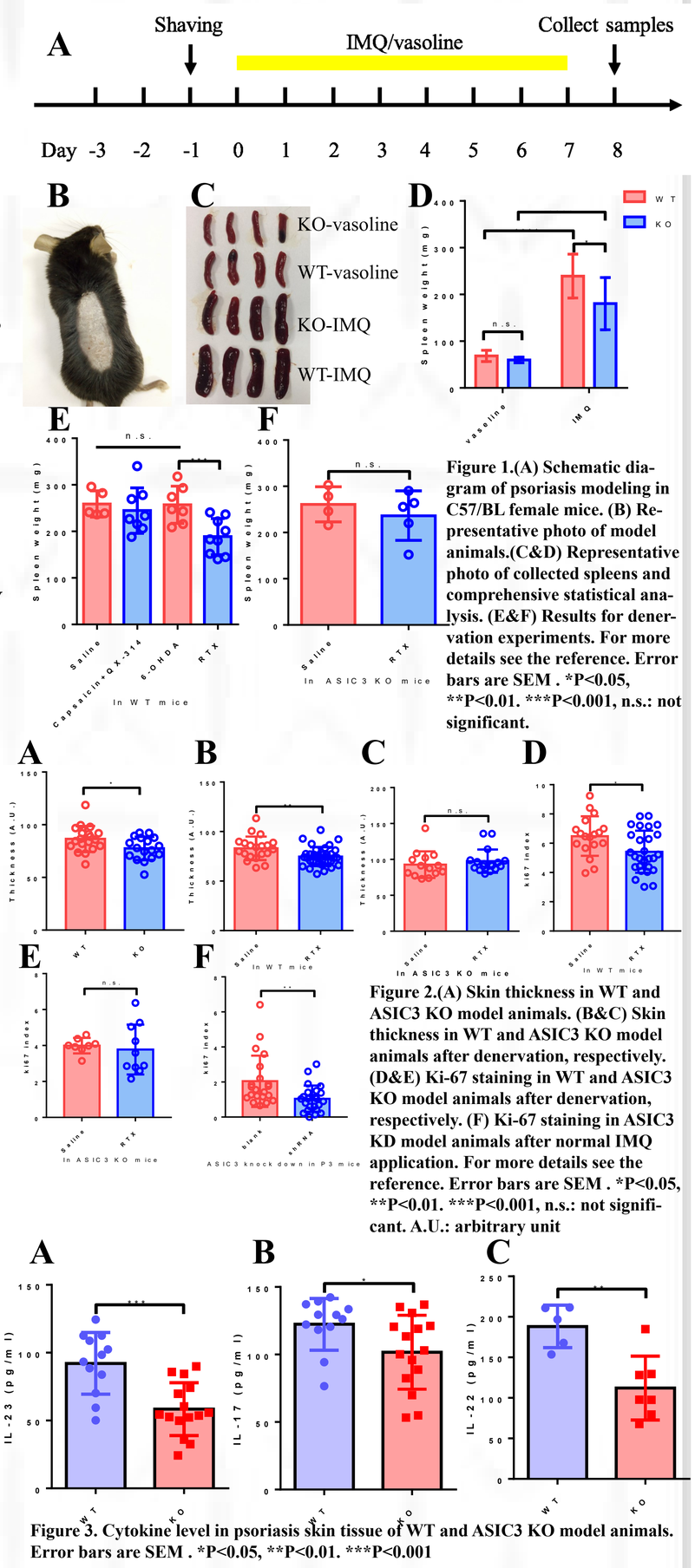

Figure 1.(A) Schematic diagram of psoriasis modeling in C57/BL female mice. (B) Representative photo of model animals.(C&D) Representative photo of collected spleens and comprehensive statistical analysis. (E&F) Results for denervation experiments. For more details see the reference. Error bars are SEM . *P<0.05, **P<0.01. ***P<0.001, n.s.: not significant.

Figure 2.(A) Skin thickness in WT and ASIC3 KO model animals. (B&C) Skin thickness in WT and ASIC3 KO model animals after denervation, respectively. (D&E) Ki-67 staining in WT and ASIC3 KO model animals after denervation, respectively. (F) Ki-67 staining in ASIC3 KD model animals after normal IMQ application. For more details see the reference. Error bars are SEM . *P<0.05, **P<0.01. ***P<0.001, n.s.: not significant. A.U.: arbitrary unit

Figure 3. Cytokine level in psoriasis skin tissue of WT and ASIC3 KO model animals. Error bars are SEM . *P<0.05, **P<0.01. ***P<0.001

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