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REGULATION OF TLR9/MYD88/NF-KAPPA B SIGNALING PATHWAY ON REGULATORY B CELLS (BREG) IN PATIENTS WITH THYMOMA AND MYASTHENIA GRAVIS

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Objective: To investigate the regulatory mechanism of TLR9/MyD88/NF-kappa B signaling pathway on regulatory B cells (Breg) in thymoma patients with myasthenia gravis.

Methods: 60 clinical specimens cases were collected and grouped according to the normal control group (control group), simple thymoma group (Tm group), thymoma combined with myasthenia gravis group (MG group), the percentage of Breg in the three groups was detected by flow cytometry, and the percentage of CD19+IL10+B cells in thymoma tissues was detected by immunohistochemical double staining. Utilize Real-Time PCR and Western blot to detect the gene and protein expression levels of TLR-9, MyD88, NF- kappa B; peripheral blood B cells were cultured by immunomagnetic beads sorting technique (MACS), co-cultured *in vitro* and contacted with Thy0517 thymoma cells, and TLR9 signaling pathway stimulating agent CpG oligodeoxynucleotides (CpG ODN) was added simultaneously, then, divided into separate B cell group (group A), B cell + thy0517 co-culture group (group B), B+ cell stimulating agent group (group C), and B cells co-cultured with + thy0517 + stimulation agent group (D group). At last, to detect the expression of Breg in different groups by flow cytometry is used. After CpG-ODN stimulated, the expression levels of TLR-9, MyD88 and NF- kappa B were detected by Real-Time PCR and Western-Blot.

Results: Breg was the most expressed in MG patients, and the level of peripheral flow cytometry was consistent with that of immunohistochemical double staining. The expression of TLR9, MyD88, NF- kappa B gene and protein level in thymoma tissues increased gradually in group control, group Tm and group MG; and the co-culture of B cells *in vitro* and thymoma cells Thy0517 indicated that the expression of Breg gradually increased in accordance with the order of group A, B, C to group D, and the gene and protein expression levels of TLR-9, MyD88, NF- kappa B gradually increased.

Conclusion: Thymoma may stimulate the development of Breg cells by up regulating the TLR9/MyD88/NF- kappa B signaling pathway; and Breg cells may be involved in the regulation of the pathogenesis of thymoma with myasthenia gravis.

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