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Localization of muscarinic acetylcholine receptor subtype 2 (M2) in immune tissues of Balb/C mice and Wistar rats – An immunohistochemical study

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Cholinergic input of different immune tissues through nicotinic and muscarinic acetylcholine receptors has been documented. In most instances the expression of muscarinic acetylcholine receptors (MAChRs) has been from isolated lymphocytes of blood. The presence of the exact type of receptor in different immune tissues is yet to be identified. The aim of this study was to localize the expression and compare the distribution of M2AChRs of different immune tissues of Balb/C mice and Wistar rats using immunohistochemistry. Thymus, spleen, lymph nodes, Peyer's patches of terminal ileum and liver tissues were processed for Hematoxylin and Eosin staining and were labeled by polyclonal anti-M2AChR and biotinylated anti-rat IgG. The labeled StreptAvidin Biotin technique was used, with diaminobenzedene to detect the antigens of receptors. Cardiac muscle was processed as the positive control and skeletal muscle tissue was used as the negative control. Slides were observed and digitalized using advanced Olympus FSX100 and the staining intensity was evaluated within a score of 0 – 3+. The intensity of immunoreactivity (IR) in distinct compartments of immune tissue has been analyzed by one way ANOVA using SAS software. The splenic capsule and red pulp expressed a significantly higher mean value of M2AChR immunoreactivity, while in lymph nodes the M2AChR distribution was observed in the capsule and subcasular sinus. The thymic capsule and septa expressed significantly ($P \leq 0.05$) high values. The rest of the tissues showed a low or absence of M2AChR IR. The above regions with high IR predominantly have T cells. Similar sites localized with α1 & α7 nicotinic acetylcholine receptors and their cholinergic neural control which has been shown in our previous studies, expressed M2AChRs. The mean values of M2 subtype IR was significantly higher in the capsules of spleen and thymus of mice tissues than that of Wistar rats, while both animals expressed relatively similar distribution of M2AChRs in immune tissues. The thymus, spleen and lymph nodes showed significant cholinergic innervation through M2 subtype of AChRs in their T cell and macrophage abundant compartments and a low or absence of innervation in the liver and Peyer's patches.

Keywords: M2 subtype of muscarinic acetylcholine receptors, immunoreactivity, innervation

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