INTESTINAL INFLAMMATION INDUCED CHANGES IN THE EXPRESSION AND DISTRIBUTION OF MAS-RELATED GENE RECEPTORS IN THE MURINE ILEUM

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INTRODUCTION/OBJECTIVES:
Mas-related gene receptors (Mrgs) constitute a complex family of orphan GPCRs, some of which are predominantly expressed in spinal sensory neurons. Some Mrgs are suggested to participate in the regulation of inflammatory responses to IgE-independent mast cell activation, as well as in the neuroimmune communication.

AIMS & Methods:
The lack of data concerning Mrgs in the gastrointestinal tract, in both healthy and inflamed conditions, led us to investigate the expression and putative function of Mrgs in the non-inflamed and *Schistosoma mansoni*-infected murine ileum. Immunohistochemical analyses were performed on cryosections and whole-mounts of non-inflamed and inflamed ileum using custom-made polyclonal antisera against MrgA4, MrgB2, MrgB8 and MrgB10, as well as a commercially available antiserum against MrgD. Real-Time PCR was performed on non-inflamed and inflamed ileum to detect and quantify the expression levels of MrgA4, MrgB2, MrgB8, MrgB10 and MrgD mRNAs.

RESULTS:
In the non-inflamed ileum, immunohistochemistry revealed no MrgB10 or MrgD immunoreactivity (IR), whereas moderate MrgA4, MrgB2 and MrgB8 IR was detected in some neuronal somata in the enteric plexuses. In the inflamed ileum, MrgA4, MrgB2, MrgB8, MrgB10 and MrgD IR was observed in a significantly increased number of neuronal somata in both enteric plexuses (Table 1), as well as in nerve fibres in the tunica muscularis and the lamina propria.

Table 1. Proportional expression of Mrgs in enteric neurons in the non-inflamed and inflamed ileum. *= *P-value*<0.05

	Non-inflamed ileum	Inflamed ileum
MrgA4	0.5%	7.0%*
MrgB2	0.4%	4.0%*
MrgB8	1.5%	12.0%*
MrgB10	0.0%	6.0%*
MrgD	0.0%	5.0%*

Colocalisation studies using antibodies against neurochemical markers demonstrated that MrgA4-, MrgB2-, MrgB8-, MrgB10- and MrgD-expressing neurons were predominantly intrinsic sensory neurons. In the inflamed ileum, MrgB10 and MrgD were also detected in mucosal mast cells (MMCs). These results were corroborated by Real-Time PCR which demonstrated the upregulation of MrgA4, MrgB2, MrgB8, MrgB10 and MrgD mRNAs in the inflamed ileum.

CONCLUSION:
The increased expression of MrgA4, MrgB2, MrgB8, MrgB10 and MrgD in neurons and nerve fibres, as well as the presence of MrgB10 and MrgD in MMCs during inflammation indicate the involvement

of Mrgs in neuronal and mast cell responses during intestinal inflammation. Future work should aim at elucidating the (patho)physiological role of these Mrgs.

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