Protective effect of α7 nicotinic receptor agonist AR-R17779 in TNBS-induced colitis

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BACKGROUND AND AIM

Inflammatory Bowel Diseases (IBDs) are characterized by chronic intestinal inflammation triggered by an aberrant immune response against luminal flora in genetically susceptible individuals (1). Evidence has been provided that IBD patients often show impaired efferent vagus nerve activity (2), and that activation of nicotinic acetylcholine receptors (*nAChRs*) expressed by immune cells could modulate intestinal inflammation (3).

Our aim was to pharmacologically investigate the role played by α7 and α4β2 *nAChRs* in the modulation of both local and systemic inflammatory responses in 2,4,6-Trinitrobenzene sulfonic acid (TNBS)-colitis in mice, one of the most widely used animal models for human IBDs.

METHODS

Colitis was induced in female Swiss mice, 7-11 weeks old, by enema administration (i.r.) of TNBS after skin sensitization. Mice were randomly assigned to:

SHAM: colitis was not induced (i.r. administration of 50 μL saline solution) + saline (0.9% NaCl, 10 mL/kg s.c.)
CTR: TNBS (5mg/mouse i.r. in 50% EtOH) + saline;
SULF: TNBS + sulfasalazine (50 mg/kg p.o.)
AR-R: TNBS + α7 agonist AR-R17779 (0.5; 1.5; 5 mg/kg s.c.)
MLA: TNBS + α7 antagonist methyllycaconitine (0.1; 0.5; 1 mg/kg s.c.)
TC: TNBS + α4β2 agonist TC 2403 (2; 5 mg/kg s.c.)
DBE: TNBS + α4β2 antagonist Dihydro-βerythroidine (0.5; 1.5 mg/kg s.c.)

Treatments started 8hrs after TNBS administration and were applied twice daily for 3 days. We determined clinical outcome as Disease Activity Index (DAI) (4), colonic local damage as Macroscopic Score (MS) (5), colonic thickness and edema, colonic and lung myeloperoxidase (MPO) activity. All animal experiments were performed according to the guidelines for the use and care of laboratory animals and were authorized by Ministero della Salute (DL 26/2014).

RESULTS AND CONCLUSIONS

Compared to **SHAM, CTR** mice showed markedly higher DAI ($p<0.001$), MS ($p<0.001$), colonic edema ($p<0.05$) and thickness ($p<0.01$), colonic and lung MPO ($p<0.01$).

Compared to **CTR**, **SULF** strongly decreased DAI ($p<0.01$), MS ($p<0.001$) and colonic MPO ($p<0.01$). **AR-R** showed a bell-shaped dose-response curve, significantly ameliorating local inflammatory parameters, like MS ($p<0.01$), colonic MPO and thickness ($p<0.05$) at 1.5 mg/kg, while the lowest and the highest doses tested were either ineffective or only weakly protective. On the contrary, body weight loss and stool consistency, expressed by DAI index, were improved at 0.5 mg/kg ($p<0.05$) and at 5 mg/kg. **MLA** dose-dependently augmented MS ($p<0.01$) and lung MPO ($p<0.05$) but did not significantly affect the other markers. **TC** 5 mg/kg significantly improved DAI ($p<0.01$) and colonic edema ($p<0.05$). **DBE** slightly worsened most of inflammation parameters but, at 1.5 mg/kg, significantly reduced colonic MPO and DAI ($p<0.05$).

Our data suggest that α7 *nAChRs*, tonically activated by an anti-inflammatory cholinergic reflex or exogenously stimulated, play a protective role against the TNBS-induced inflammatory response. Contradictory effects elicited by α4β2 agents seem to indicate a controversial role of this *nAChRs* subtype in this model of colitis thus requiring further investigations.

REFERENCES

1. Abraham C, Cho JH. N Engl J Med 2009; 361: 2066-78.
2. Pellissier S et al., Psychoneuroendocrinology 2010; 35: 653–62.
3. Van der Zanden EP et al., Neurogastroenterol. Motil. 2009; 21(1): 6-17
4. Qualls JE et al., J Leukoc Biol (2006) 80: 802-815.
5. Khan WI et al., Infect Immun (2002); 70: 5931-7