Hydrogen sulfide donors and their THERAPEUTIC potential as antipruritics and anti-inflAmmatory in MOUSE dorsal skin

Leandro Rodrigues[1], Eduardo Ekundi-Valentim[2], Juliana Florenzano[1], Simone Teixeira[1], Marcelo Muscará[1], Soraia Costa[1]

[1]Department of Pharmacology, Institute of Biomedical Sciences, University of São Paulo, São Paulo, 05.508-900, Brazil, [2]Institute of Health Sciences, University Agostinho Neto, Luanda, Angola, [3]Department of Pharmacology, Institute of Biomedical Sciences, University of São Paulo, São Paulo, 05.508-900, Brazil, [4]Department of Pharmacology, Institute of Biomedical Sciences, University of São Paulo, São Paulo, 05.508-900, Brazil, [5]Department of Pharmacology, Institute of Biomedical Sciences, University of São Paulo, São Paulo, 05.508-900, Brazil, [6]Department of Pharmacology, Institute of Biomedical Sciences, University of São Paulo, São Paulo, 05.508-900, Brazil

INTRODUCTION: Pruritus (itch) is a sensory modality that, similar to pain, acts as a protective mechanism for the organism. Despite advances in anti-pruritic therapy with histamine antagonists, these agents are ineffective in most patients. Recently, the gaseous mediator H_2S has been highlighted as an important modulator of inflammatory and nociceptive processing mechanisms, but its role in pruritus is unknown. This study was carried out to investigate the effects of H_2S donors Na_2S and Lawesson's Reagent (LR) in the pruritus and skin inflammation evoked by histamine and compound 48/80 (C48/80) intradermally (i.d.) injected in mouse dorsal skin. We have also evaluated the kinetics of H_2S generation in the naïve skin and its enzyme expression; cystathionine-□-synthase (CBS) and cystathionine-□-lyase (CSE).

METHODS: Under approval of the Institute of Biomedical Sciences Ethics Committee of University of Sao Paulo (no. 33, book 2/2010), male BALB/c mice (25-30 g) were anaesthetised with isoflurane, and either compound 48/80 (C48/80; 100 µg site^{-1}) or histamine (1µmol site^{-1}) alone or in addition to H_2S donors, Na_2S (1-100 nmol site^{-1}) and LR (3-300 nmol site^{-1}), was intradermally (i.d.) administered in a single (0.05 ml) injection to produce itching, that was measured as a bout of scratching during 30 min. Skin plasma protein extravasation and neutrophil accumulation was assessed in a separate set of animals by the extravascular accumulation of i.v. injected ^{125}I-albumin and increased myeloperoxidase (MPO) activity in the skin, respectively. Data are presented as mean ± SEM. Stats were performed by ANOVA followed by Bonferroni's test. $P<0.05$ was taken as significant.

RESULTS: Either C48/80 or histamine significantly increased itching frequency, and lead to a potent ($P<0.01$) plasma extravasation when compared to its vehicle Tyrode. The pruritus evoked by histamine, but not C48/80, was significantly inhibited by 67-77 % with the co-injection of Na_2S (1-3 nmol site^{-1}, respectively; n=5-9) and by 47-63 % following co-injection of LR (3-30 nmol site^{-1}, respectively; n=6). Either co-injection of Na_2S (10–100 nmol site^{-1}; n=5-6) or LR (10–300 nmol site^{-1}; n=5-8) significantly, but not dose-dependently, reduced plasma extravasation induced by C48/80 and histamine or increased MPO activity evoked by C48/80 in the mouse skin. Western blot analyzes revealed, for the first time, that CSE and CBS are constitutively expressed in murine naïve skin in parallel with basal production of H_2S.

DISCUSSION: We show for the first time that increasing bioavailability of H_2S in the murine skin, by local application of H_2S donors, exerts a protective role against histamine-induced pruritus and skin inflammation evoked by histamine and C48/80. We suggest that H_2S donors might represent a potential therapeutic class for treatment of pruritus associated with skin inflammation. We also show that there has been a regular production of H_2S in naïve skin along with constitutive expression of CSE and CBS.

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