Agents and Actions, vol. 20, 3/4 (1987)

0065-4299/87/040172-02 $ 1.50+0.20/0

Study of the inhibition of histamine release by inosine pranobex

R. Ziesche, D. Eichelberg and W. Schmutzler

Abteilung Pharmakologie, Medizinische Fakultät, Rheinisch-Westfälische Technische Hochschule Aachen, D-5100 Aachen, FRG

Abstract

The effects of Inosine pranobex and its parent compounds inosine and dimethylamino-2-propanol-p-acetamidobenzoic acid (DipPacBa) on the Concanavalin A (ConA)-induced histamine release from human adenoidal mast cells were investigated.

Inosine pranobex inhibited the ConA-induced histamine release at concentrations $> 10^{-3}$ *M*. Inosine itself (10^{-5}–10^{-2} *M*) enhanced the ConA-induced histamine release whereas DipPacBa inhibited the release significantly at concentrations between 10^{-6} and 10^{-2} *M*.

These results are consistent with the assumption that the effect of Inosine pranobex is due to the DipPacBa moiety.

Introduction

Inosine pranobex is a combination compound consisting of 1 part inosine and 3 parts dimethylamino-2-propanol-p-acetamidobenzoic acid (DipPacBa). It is reported to influence the immune system, particularly T cell dependent functions [4–6]. Nothing is known about any effects of Inosine pranobex on allergic states. We [7] observed a marked inhibition by Inosine pranobex (10^{-3} *M*) of the Concanavalin A (Con A)-induced histamine release from human adenoidal mast cells (HAMC). Since in some other models [3] the DipPacBa moiety appeared to be the carrier of the observed effects, we compared Inosine pranobex with both its parent compounds, inosine and DipPacBa.

Methods

The human adenoidal mast cells were isolated according to [7].

Only preparations of at least 1.8 g (wet weight) were used in order to obtain about 300000 to 400000 mast cells per ml. After mechanical dissection using a McIlwain and Buddle chopper the cells were washed and filtered twice, stained with alcian blue (pH 2.7) and counted in a Neubauer chamber. After distribution of the cells, each sample contained 30000 to 40000 mast cells in a final volume of 500 µl.

The cells were equilibrated with the test substances for 10 minutes at 37 °C. After the addition of Con A (50 µg/ml) the cells were incubated for a further 5 minutes. The reaction was stopped by placing the samples in an ice bath and centrifugation.

The histamine contents were determined separately in supernatants and pellets by the double isotope assay [1].

Inosine pranobex and DipPacBa were gifts from Newport Chemicals, USA. Inosine was purchased from Serva, Heidelberg.

Table 1

Controls, % histamine release, uncorrected		Effect of Inosine pranobex, Inosine or DipPacBa on the Con A-induced histamine release from human adenoidal mast cells in vitro (% change of uninhibited release)						
Spont.	Con A (50 µg/ml)	Test substance (M) + Con A (50 µg/ml)						
		10^{-8}	10^{-7}	10^{-6}	10^{-5}	10^{-4}	10^{-3}	10^{-2}
		Inosine pranobex						
9.3±1.2 (13)	22.2±2.9 (13)	–	–	–	− 6.7±18.3 (9) n.s.	+29.9±48.9 (8) n.s.	− 16.9±34.7 (9) $p<0.05$	− 90.3±20.0 (8) $p<0.01$
		Inosine						
8.2±1.2 (11)	23.9±1.8 (11)	–	–	–	+52.1±29.7 (5) n.s.	+17.8±26.5 (7) n.s.	+51.9±22.6 (8) $p<0.025$	− 23.3±21.3 (7) n.s.
		DipPacBa						
7.5±1.2 (17)	26.6±2.4 (17)	+7.9±3.0 (5) n.s.	+12.2±16.6 (5) n.s.	−21.1±0.8 (5) $p<0.05$	−29.2± 0.9 (7) $p<0.05$	−30.1±12.5 (8) $p<0.025$	−43.0± 0.7 (10) $p<0.01$	−26.0± 0.7 (10) $p<0.01$

Results

Inosine pranobex caused a marked decrease of the Con A-induced histamine release at concentrations $> 10^{-3}$ *M*. Lower concentrations had no significant effect (Table).

Inosine itself enhanced the histamine release in the concentration range 10^{-5} to 10^{-3} *M*. Only a small and not significant inhibition was observed at 10^{-2} *M*.

DipPacBa, on the other hand, inhibited the Con A-induced histamine release significantly at concentrations $> 10^{-6}$ *M*. The maximum inhibition was observed at 10^{-3} *M*.

Discussion

The results show that Inosine pranobex inhibits the histamine release from human adenoidal mast cells at concentrations which can be easily reached under therapeutic conditions.

The inosine moiety obviously increases the histamine release in a similar manner to adenosine [2]. Therefore, the inosine moiety may partially mask the marked inhibitory effect of the DipPacBa moiety which is probably responsible for the inhibition by the combination compound Inosine pranobex (molar ratio of DipPacBa and inosine 3 : 1). It is noteworthy that DipPacBa was found to be similarly effective to other degranulation inhibitors, i.e. disodium cromoglycate (DSCG) which causes under comparable conditions a 30–50% inhibition of release at concentrations $> 10^{-5}$ *M* [8].

References

[1] M. A. Beaven, S. T. Jacobsen and Z. Horakova, *Modification of the enzymic isotopic assay of histamine and its application to measurement of histamine in tissues, serum and urine.* Clin Chim. Acta *37,* 91–103 (1972).

[2] K. Delmich, D. Eichelberg and W. Schmutzler, *The effects of adenosine and some adenosine analogues on the concanavalin A- or acetylcholine-induced histamine release from human adenoidal mast cells.* Agents and Actions *16,* 141–143 (1985).

[3] A. M. Dräger, K. Ziehe, J. Malms and W. Schmutzler, *Effect of in vivo administration of Levamisole and Inosine pranobex on murine T cell functions.* Naunyn Schmiedeberg's Arch. Pharmacol. *330* (suppl.), R53 (1985).

[4] T. Ginsberg and A. J. Glasky, *Inosiplex: an immunomodulation model for the treatment of viral disease.* Ann. N.Y. Acad. Sci. *284,* 128–138 (1977).

[5] J. W. Hadden, E. M. Hadden and R. G. Coffey, *Isoprinosine-augmentation of phytohemagglutinin-induced lymphocyte proliferation.* Infect. Immun. *13,* 382–387 (1976).

[6] G. Renoux, M. Renoux, J. M. Guillaumin and C. Gouzien, *Isoprinosine as immunopotentiator.* Int. J. Immunopharmac. *1,* 337–356 (1979).

[7] W. Schmutzler, K. Delmich, D. Eichelberg, S. Glück, T. Greven, H. Jürgensen, K. P. Riesener, G. Risse and P. Pult, *The human adenoidal mast cell. Susceptibility to different secretagogues and secretion inhibitors.* Int. Arch. Allergy appl. Immunol. *77,* 177–178 (1985).

[8] W. Schmutzler, A. Kortemeier and K. P. Riesener, *Pharmakologische Grundlagen der Therapie obstruktiver Atemwegserkrankungen unter besonderer Berücksichtigung der β_2-Adrenergika und der DNCG.* Pharmakotherapie *7,* 1–8 (1984).