Inflamm. res. 55, *Supplement 1* (2006) S15–S16
1023-3830/06/01S15-02
DOI 10.1007/s00011-005-0020-6

Inflammation Research

Effect of H_1-antihistamines on the oxidative burst of rat phagocytes

J. Kralova[1], M. Ciz[1], R. Nosal[2], K. Drabikova[2], A. Lojek[1]

[1] Institute of Biophysics AS CR, Brno, Czech Republic, Fax: ++420 541211293, e-mail: alojek@ibp.cz
[2] Institute of Experimental Pharmacology SAS, Bratislava, Slovak Republic

Published Online First 20 March 2006

Introduction

Phagocytosis is the essential defence against microbial pathogens. This function is performed by leukocytes, mainly neutrophils. As observed [1, 2], the histamine H_1-antagonist dithiaden inhibits production of reactive oxygen species (ROS) by phagocytes. Our aim was to compare the inhibitory effects of dithiaden with those of four second-generation H_1-antihistamines – loratadine, acrivastine, astemizole and ketotifen-fumarate.

Materials and methods

H_1-antihistamines were obtained from: dithiaden and loratadine (Zentiva, Czech Republic), acrivastine (Wellcome Foundation, UK), astemizole (Janssen, Belgium), ketotifen fumarate (VUFB, Czech Republic). Other chemicals were purchased from local distributors.

Experiments were performed in accordance with NIH guidelines for the care and use of laboratory animals. Leukocytes isolated from peripheral blood of Wistar rats were stimulated with opsonized zymosan (OZ) in the presence/absence of H_1-antihistamines (0.001–0.5 mmol/L). Kinetics of the production of ROS by leukocytes were analysed by luminol-enhanced chemiluminescence (CL) for 60 min [3]. Parameters recorded included the intensity of chemiluminescence expressed as relative light units (RLU) and the area under the obtained curve expressed as $RLU/60_{min}$.

Antioxidant activities of H_1-antihistamines against superoxide anion, hydroxyl radical and peroxyl radical were analysed as previously reported [4].

Data are expressed as means ± standard error of the mean of six experiments. Results were analysed by ANOVA followed by Newman-Keuls test.

Results and discussion

First-generation H_1-antagonists were effective antihistamines, but had side effects [5]. Second-generation antihistamines are characterized by reduced lipophilicity, large size, and extensive protein binding, and specificity for the H_1-receptor [6]. We compared effects of selected 2nd generation H_1-antihistamines on the respiratory burst of rat phagocytes with those of dithiaden, a first-generation H_1-antihistamine. Different effects were observed for individual antihistamines (Table 1). Ketotifen-fumarate had similar activity to dithiaden, both being effective at concentrations >0.01 mmol/L. This could be explained by the high affinity of ketotifen-fumarate for both H_1-receptors and other receptors [5] and because both drugs have similar chemical structures.

Astemizole was even more effective; only 0.001 mmol/L concentration did not affect the CL. In contrast, loratadine was a much less effective inhibitor of CL than dithiaden, being active only at 0.5 mmol/L (Fig. 1). As observed in human blood leukocytes [7], loratadine was also a less effective inhibitor of OZ-stimulated CL than dithiaden. This could be because of its higher selectivity but lower affinity for H_1-receptors [5]. Conversely, CL of human leukocytes was supressed significantly by loratadine at 0.02 mmol/L [7], a lower concentration than our study. This could be explained by an inverse ratio between neutrophils and lymphocytes in rat and human blood. Moreover, rat neutrophils contain less myeloperoxidase, responsible for generation of hypochlorous acid and related CL.

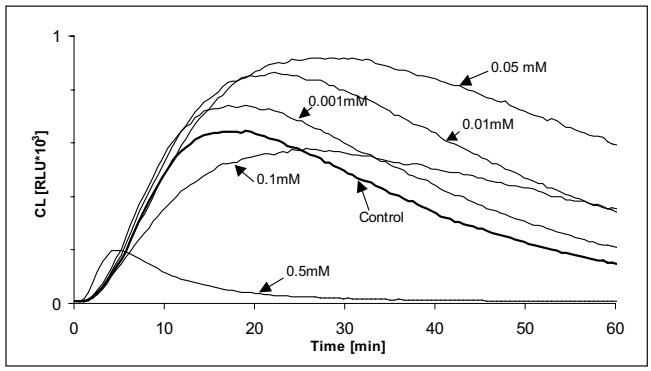

Fig. 1. Effects of loratadine on the kinetics of CL activity of rat leukocytes.

Correspondence to: A. Lojek

Concentration [mmol/L]	Integral of CL [RLU/60_{min} * 10^5]				
	Dithiaden	Acrivastine	Astemizole	Loratadine	Ketotifen-f.
0 (control)	20.3 ± 2.3	13.5 ± 1.8	24.9 ± 9.5	13.5 ± 1.8	14.8 ± 0.7
0.001	23.5 ± 3.1	14.5 ± 1.7	39.5 ± 10.3	16.5 ± 1.9	13.4 ± 1.8
0.01	19.8 ± 2.4	18.9 ± 0.9	0.4 ± 0.09*	20.8 ± 2.6	13.0 ± 1.8
0.05	1.5 ± 0.3*	16.5 ± 1.6	0.2 ± 0.01*	24.8 ± 2.4*	2.9 ± 0.6*
0.1	0.5 ± 0.2*	17.6 ± 1.8	0.2 ± 0.01*	15.4 ± 3.1	0.3 ± 0.02*
0.5	0.3 ± 0.01*	26.3 ± 0.6*	0.2 ± 0.01*	1.2 ± 0.2*	0.3 ± 0.01*

Table 1. Effects of H_1-antihistamines on CL activity of rat leukocytes. Values are means ±sem, n = 6. Asterisks show significant differences from control ($p < 0.05$, ANOVA and Newman-Keuls test).

Acrivastine did not inhibit CL, despite high selectivity for H_1-receptors [5]. Possibly the inhibition of leukocyte-derived CL by some antihistamines is caused partly via non H_1-receptor pathway.

None of these antihistamines showed direct scavenging properties against superoxide anion, hydroxyl radical and peroxyl radical (data not shown), thus this could not contribute to the inhibition of CL. Other possible mechanisms for this inhibitory effect of antihistamines could be via interference with calcium ion movement, enzymatic pathways or second messengers [6].

It can be concluded that the inhibition of oxidative burst of leukocytes observed in tested drugs was mediated neither via H_1-receptor pathway nor by direct antioxidative properties. Based on our results, antihistamines which do not interfere with microbicidal mechanisms of leukocytes could be used preferentially in infections. Conversely, antihistamines inhibiting CL activity of leukocytes should be used preferentially under pathological conditions accompanied by the overproduction of ROS.

Acknowledgement. Work was supported by grants GA CR 305/04/0896 and VEGA 2/4003/04.

References

[1] Nosál R, Drábiková K, Číž M, Lojek A, Danihelová E. Effect of H_1-antagonist. Dithiaden on Human PMN-leukocyte aggregation and chemiluminiscence is stimulus-dependent. Inflamm Res 2002; 51: 557–62.

[2] Drábiková K, Nosál R, Jančinová V, Číž M, Lojek A. Reactive Oxygen Metabolite Production is Inhibited by Histamine and H_1-antagonist Dithiaden in Human PMN Leukocytes. Free Radic Res 2002; 36: 975–80.

[3] Pavelková M, Kubala L. Luminol-, isoluminol- and lucigenin-enhanced chemiluinscence of rat blood phagocytes with different activators. Luminescence 2004; 19: 37–42.

[4] Nosál R, Jančinová V, Číž M, Drábiková K, Lojek A, Fabryová V. Inhibition of chemiluminescence by carvedilol in the cell-free system, whole human blood and blood cells. Scand J Clin Lab Invest 2005; 65: 55–64.

[5] Kiss R, Kovari Z, Keseru GM. Homology modelling and binding site mapping of the human histamine H1 receptor. Eur J Med Chem 2004; 39: 959–67.

[6] Lieberman P. Preclinical evidence of azelastine hydrochloride activity. Curr Ther Res Clin Exp. 2002; 63: 556–71.

[7] Nosál R, Drábiková K, Jančinová V, Petríková M, Fábryová V. Antiplatelet and antiphagocyte activity of H_1-antihistamines. Inflamm Res 2005; 54: S19–S20.