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MODULATION OF ConA-INDUCED INFLAMMATORY ASCITES BY HISTAMINE – SHORT COMMUNICATION

KÁROLY BAINTNER*

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The early phase of the ConA-induced inflammatory ascites was studied, with special reference to histamine. Concanavalin A (ConA), a cell-surface binding lectin was injected i.p. (25 mg/kg bw) to mice. After 1 h the animals were killed, the ascitic fluid collected and measured. Other agents were injected s.c., 10 min before the ConA-challenge. Exogenous histamine markedly inhibited the ConA-induced ascites. Release of endogenous vasoactive agents from the mast cells by Compound 48/80 had a similar, but slight effect. Cromolyn, a mast cell stabilizing agent, and chloropyramine, a histamine H1 receptor antagonist was ineffective. Although histamine increases endothelial permeability, it did not enhance the formation of ascitic fluid, on the contrary, it inhibited the ConA-induced ascites, presumably due to its known hypotonic effect. It is concluded that ConA-induced ascites is not mediated by mast cell histamine.

Keywords: histamine, ConA, ascites, mast cell, cromolyn, chloropyramine

Introduction

Earlier experiments [1] showed that non-covalent cross-linking of cell-surface glycoproteins by plant lectins or polykationic macromolecules induces accumulation of inflammatory (protein-rich) ascitic fluid in the peritoneal cavity still before the major influx of leukocytes [2]. However, the cellular mediation of ascites formation has remained unexplored.

Concanavalin A (ConA) is a plant lectin which recognizes α-mannosyl residues in complex glycosyl side chains of cell surface glycoproteins. If injected into the abdominal cavity, ConA binds to the mesothelial lining [3], mast cells [4–6] and leukocytes [7].

* E-mail: rakiab22@gmail.com

In the present experiments ConA was used for the induction of ascites, and the modulation by exogenous or mast cell histamine was investigated. The role of macrophages and the mesothelial lining in the mediation of ascites will be dealt with in another paper.

Materials and Methods

Chemicals

ConA was Sigma L7647, type VI. Chloropyramine Cl (Suprastin inj.™) was a commercial product and the other chemicals were bought from Sigma.

Animals and protocol

Female NMRI mice (24 to 25 g, Charles River) were used in groups of 6. The experiments complied with the Hungarian Animal Welfare Act XVIII/1998 and Edict 243/1998 and were approved by the local ethical committee and the Veterinary Office of Somogy County.

ConA (25 mg/kg bw) was dissolved in physiological saline and injected intraperitoneally to mice in 0.1 ml volume. After 1 h the animals were decapitated under ether anesthesia, the abdomen was opened and the ascitic fluid carefully collected. The volume was measured by pipetting the fluid into another tube and was expressed as % of body weight.

Histamine, Compound 48/80 and sodium cromoglycate (cromolyn) was dissolved in physiological saline and injected s.c. to mice in 0.1 ml volume, 10 minutes before the ConA challenge. Histamine was neutralized with HCl before use and the site of s.c. injection was treated locally with lidocaine.

Negative control groups were treated with i.p. Compound 48/80 or physiological saline (0.1 ml) for 1 h.

Statistics

Significance of differences was calculated with one-way ANOVA using the SPSS program.

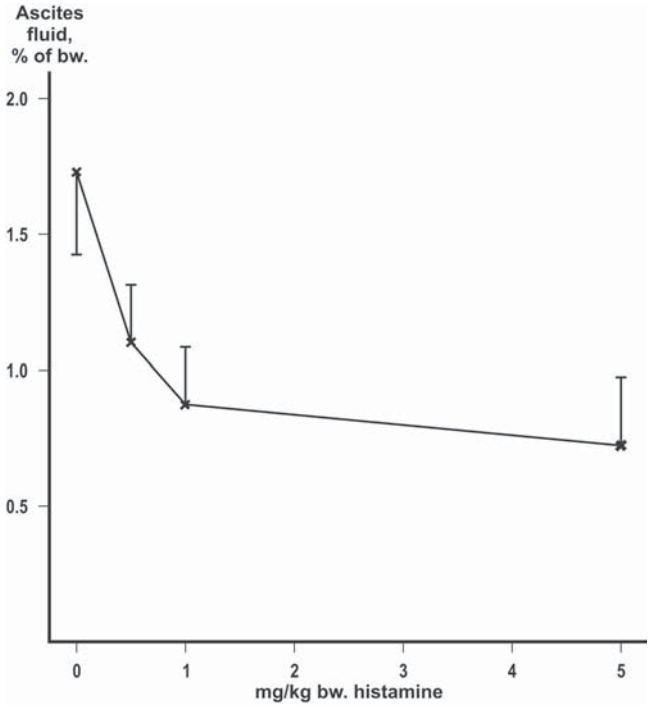

Figure 1. Effect of pre-treatment with different doses of histamine (s.c.) on ConA-induced ascites (i.p., 25 mg/kg bw, 1 h). Mean ± SD, n = 6

Results

The effect of exogenous histamine on the ConA-induced ascites

Exogenous histamine significantly ($p < 0.01$) suppressed the ConA-induced ascites (Fig. 1) as compared to the positive control (ConA alone). The effect was dose-dependent, but complete inhibition could not be achieved.

ConA-induced ascites as affected by the functional state of mast cells

S.c. administration of Compound 48/80 (1.5 mg/kg bw) significantly ($p < 0.05$) inhibited the ascites formation as compared to the positive control group (i.p. ConA alone), whereas the groups treated with Na cromoglycate (cromolyn, 60 mg/kg bw) or chloropyramine chloride (4 mg/kg bw) did not differ significantly (Fig. 2).

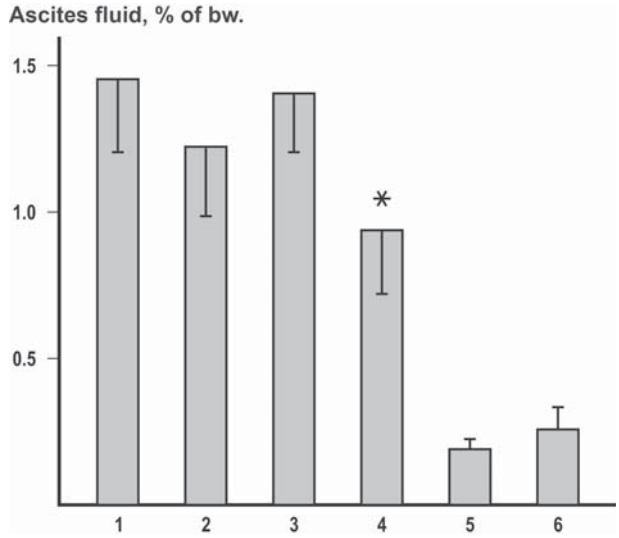

Figure 2. ConA-induced ascites as affected by the functional state of mast cells

Mean±SD, n = 6. The asterisk indicates significant ($p < 0.05$) difference
1: I.p. ConA, positive control
2: S.c. chloropyramine and i.p. ConA
3: S.c. cromoglycate and i.p. ConA
4: S.c. Compound 48/80 and i.p. ConA
5: I.p. Compound 48/80 alone
6: Physiological saline, negative control

When Compound 48/80 was injected i.p. as a control (1.5 mg/kg bw, without ConA-challenge), the volume of the peritoneal fluid did not differ significantly from that of the saline-treated animals (Fig. 2).

Discussion

Mast cell histamine, being a vasoactive agent, was considered a possible mediator of ConA-induced ascites. However, ascites could not be enhanced by the s.c. administration of histamine, on the contrary, it markedly inhibited it (Fig. 1), presumably due to its known hypotonic effect.

In a second experimental series the release of endogenous histamine by treatment with Compound 48/80 slightly, but significantly ($p < 0.05$) suppressed the volume of ConA-induced ascites (Fig. 2). The mast cell stabilizer cromolyn [8] and the H1 receptor antagonist chloropyramine were ineffective in our experimental arrangement (Fig. 2).

During peritoneal dialysis, permeability appears to be regulated by the vascular endothelium and not by the layer of the mesothelial cells [9–10]. It is also known that histamine increases the permeability of the venous endothelium [11–12]. Mast cells produce several agents together with histamine. However, both exogenous histamine and mast cell degranulation counteracted the ConA-induced ascites.

Acknowledgement

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