Histamine release by vagal stimulation

P. Blandina, M. Barattini, R. Fantozzi, Emanuela Masini and P.F. Mannaioni
Department of Pharmacology and Toxicology, Florence University, School of Medicine, Viale G.B. Morgagni 65,
I-50134 Firenze, Italy

Abstract

The content of acetylcholine and histamine in the effluent of isolated, vagally innervated guinea-pig auricles was determined. Spontaneous or stimulation-induced overflow of acetylcholine was detected only in the presence of acetylcholinesterase inhibitors. The histamine overflow was measurable also in the absence of inhibition of cholinesterase, and neatly increased during vagal stimulation. The vagally evoked histamine overflow was blocked by atropine and potentiated by eserine. The stimulation-induced histamine overflow in the effluent is discussed, presumably assuming that acetylcholine may release histamine from cardiac histamine stores.

Introduction

The term 'cholinergic histamine release', firmly established to indicate clinical events such as 'cholinergic asthma' and 'cholinergic urticaria' is also entitled with experimental evidences [1]. In fact, vagal excitation leads to a depletion of gastric histamine stores [2-4] and stimulation of cholinergic nerves causes an histamine release from salivary glands [5-8]. Moreover, acetylcholine enhances plasma histamine levels in the dog [9] and the immunological histamine release from IgE sensitized tissues [10, 11]. Presumably mast cells are involved in this process, since stress degranulates gastric mast cells via a cholinergic pathway [12], carbamylcholine increases histamine release by 48/80 in isolated mast cells [13] and acetylcholine causes histamine secretion in isolated mast cells [14, 15].

To establish further a link between cholinergic nervous system and tissue histamine stores, in preparations physiologically more integrated than purified mast cells, we have measured the spontaneous overflow of endogenous acetylcholine and histamine and their overflows evoked by stimulation of the vagus nerve, in isolated, vagally innervated, guinea-pig auricles. We have chosen this preparation since it is known that guinea-pig atria contain histamine and mast cells, and they are provided with histamine H1- and H2-receptors and of specific enzymes necessary to maintain the biosynthetic pathway of histamine from synthesis, uptake, storage and metabolism [16, 17].

Materials and methods

Guinea-pig auricles with the right cervical vagus attached were prepared according to Dieterich et al. [18] and perfused at 37°C with oxygenated Tyrode solution at a flow rate of 10 ml per min. The nerve was stimulated (40 V, 1 msec) with platinum ring electrodes for 1 min at 20 Hz. Histamine and acetylcholine were measured in perfusates collected at 2-min intervals.

Histamine was measured fluorimetrically, using the method of Shore et al. [19], as modified by Kremzner and Wilson [20]. Authenticity of histamine was checked through the fluorescence spectra, thin-layer chromatography (propanol-0.2 N/ammonia (3:1) and iodine) and bioassay on guinea-pig ileum according to a 2 by 2 design.

Acetylcholine was measured biologically on the leech dorsal muscle.

The chemicals used for the fluorimetric assay were of Suprapur quality, E. Merck, AG. Acetylcholine chloride and O phthaldialdehyde were obtained by B.D.H. Chemicals Ltd. Other drugs used were: histamine dihydrochloride, Calbiochem.; atropine sulfate, Merck; and eserine sulfate, Sigma.

Results

The response of guinea-pig auricles to maximal vagal stimulation is reported in Fig. 1a, showing a typical experiment. Upon vagal stimulation there is a standstill of
both contraction and rate, although in some preparations vagal escapes could be observed within the stimulation period. Consistently, at the end of the stimulation a rebound phenomenon appears, characterized by a net increase in rate and amplitude of contraction. In the perfusates collected during the stimulation, acetylcholine overflow was detectable only in the presence of eserine, at an extent which amounts from 0.48 ng min⁻¹ mg⁻¹ wet weight (spontaneous overflow) to 0.67 ng min⁻¹ mg⁻¹ wet weight (stimulated overflow). Together with acetylcholine, histamine neatly appears in the perfusates collected during vagal stimulation with a time-course which is represented in Fig. 1b.

Treatment with atropine fully inhibits the dynamic response to vagal stimulation (Fig. 2a) and also decreases, in a dose-dependent fashion, the histamine overflow (Fig. 2b). Eserine prolongs the negative inotropic and chronotropic responses to vagal stimulation (Fig. 3a) and also extends in a parallel fashion, the time-course of histamine overflow (Fig. 3b).

Discussion

These results demonstrate that, during vagal stimulation of isolated guinea-pig auricles, acetylcholine and histamine appear in the perfusates, the ratio of their overflows roughly being one to one. It is worth mentioning that the measured amounts of acetylcholine and histamine represent only a fraction of their actual release, since acetylcholine is promptly inactivated by acetylcholin-