Pharmacologic analysis of LY188695 (KB-2413), 1-(2-ethoxyethyl)-2-(4-methyl-1-homopiperazinyl)-benzimidazole difumarate, a potent histamine$_1$ receptor antagonist$^1$


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Abstract

LY188695 was evaluated both in vitro and in vivo in the guinea pig to determine its pharmacologic profile. The compound antagonized histamine-induced contractions of ileum, aorta, and trachea with pK$_B$ values of 9.9, 9.9, and 9.2 respectively. In the lung parenchymal strip, LY188695 caused a rightward shift of the histamine concentration-response curve with a reduction in the maximal response at all antagonist concentrations tested. The reason for this effect is unknown, but it was not due to a nonspecific depressant action of the compound on the parenchyma. Selectivity was shown by its inactivity against leukotriene D$_4$, bradykinin, prostaglandin F$_{2a}$, acetylcholine, norepinephrine, and serotonin on various guinea pig and rat smooth muscles. Similarly, H$_2$ receptor-mediated relaxation of the rat uterus was unaltered by LY188695. Increases in total pulmonary impedance caused by i.v. histamine to anesthetized guinea pigs were reduced by as little as 3 µg/kg given orally 1 hour prior to histamine challenge. In this system, LY188695 was 15 times more potent than chlorpheniramine and 100 times more potent than terfenadine. Similar responses elicited by acetylcholine were not antagonized by LY188695. A duration of action greater than 4 hours was observed in this model. Ovalbumin given i.v. to sensitized guinea pigs increased total pulmonary impedance which was markedly decreased after oral administration of 30 or 100 µg/kg LY188695. These results indicate that LY188695 is a very potent antagonist of H$_1$-mediated responses and suggest that this agent might be useful in disease states characterized by an overproduction of histamine.

Histamine$_1$ (H$_1$) receptor antagonists (antihistamines) have been used for decades in the symptomatic treatment of allergic rhinitis, urticaria, conjunctivitis, and various other forms of atopy [1, 2]. Despite the ability of antigen to release histamine from sensitized human lung [3], the majority of antihistamines have shown a clear lack of effectiveness in ameliorating the symptoms of human allergic asthma [4]. The apparent inactivity of H$_1$ receptor antagonists in asthma suggests that either histamine plays a negligible role in asthma or that currently available antihistamines are not potent enough to interfere with the histamine-receptor interaction in the lung. This has set the stage for development of potent H$_1$ receptor antagonists. Fukuda et al. [5] have recently reported on the pharmacology of a new benzimidazole H$_1$ recep-
tor antagonist, KB-2413 (LY 188695, Figure 1). In their test systems, the compound appeared to be extremely potent and highly selective for the \( H_1 \) receptor. In the present communication, we describe our pharmacologic evaluation of LY 188695. The observations of Fukuda et al. [5] were confirmed and extended. Using a variety of smooth muscles from guinea pig and rat, we demonstrated that LY 188695 is an extremely potent \( H_1 \) receptor antagonist with a high degree of selectivity. In vivo experiments complemented the results obtained on isolated tissues and showed LY 188695 to be significantly more potent than some previously described antihistamines.

Methods and materials

Male Hartley guinea pigs (Murphy Breeding Laboratories, Plainfield, Indiana) weighing 200–400 grams and male or female Sprague Dawley rats weighing 200–400 grams (Harlan Industries, Cumberland, Indiana) were used in these studies.

LY 188695 was administered in vitro or in vivo as an aqueous solution. For the in vitro studies, the compound was initially dissolved in Krebs’ bicarbonate solution to a concentration of \( 10^{-3} \) M. Further dilutions were made in the same medium. A 1 mg/ml solution in 0.9% saline was made for the in vivo experiments. Subsequent dilutions were also prepared in saline.

In vitro experiments

Guinea pig ileum, trachea, parenchyma and aorta. Guinea pigs were killed by decapitation. A segment of terminal ileum was removed, the lumen cleaned, and the tissue cut into smaller segments of approximately 2 to 3 cm. Each segment was tied to the bottom of a tissue holder leaving the lumen open. The ilea were then transferred to tissue baths and attached to transducers by means of thread. Ilea were equilibrated for approximately 1 hr under a maintained resting tension of 0.5 g. Trachea and aortas were excised, cleaned, and cut into ring segments. The tissues were then placed on supports constructed from two 1-inch, 30-gauge disposable stainless-steel hypodermic needles [6] and transferred to organ baths. Approximately 1 hr was allowed for equilibration under a maintained resting tension of 2 g.

Strips of parenchyma were removed from the outer edge of the lung and the ends secured by cotton thread. The tissues were then placed in baths under a passive force of 0.5 g. The procedure essentially followed the descriptions by Lulich et al. [7] and Drazen and Schneider [8].

Rat stomach fundal strip. Rats were killed by decapitation. The stomachs were dissected out and placed in petri dishes containing Krebs’ bicarbonate solution. The fundi were separated from pylorus, opened into sheets, and cut into strips [9]. The strips were subdivided into segments which were suspended in isolated tissue baths by attaching, with thread, one end of the tissue to a stationary glass rod and the other end to a force-displacement transducer. A passive force of 4 grams was placed on each tissue and they were allowed to equilibrate for at least 1 hour before drugs were tested.

The tissues described above were suspended in 10 ml organ baths containing Krebs’ bicarbonate solution of the following composition in millimoles/liter: \( \text{KCl} \), 4.6; \( \text{KH}_2\text{PO}_4 \), 1.2; \( \text{MgSO}_4 \cdot 7 \text{H}_2\text{O} \), 1.2; \( \text{NaCl} \), 118.2; \( \text{NaHCO}_3 \), 24.8; and dextrose, 10.0. The concentration of \( \text{Ca}^{++} \), in the form of \( \text{CaCl}_2 \cdot \text{H}_2\text{O} \), varied with the tissue used: guinea pig ileum, 1.2 mM; guinea pig lung parenchyma, 1.8 mM; and guinea pig trachea, aorta, and rat stomach fundus, 2.5 mM. In those in vitro experiments using histamine or 2-(2-thiazolyl)-ethylamine (ThEA) as the agonists, indomethacin, a cyclooxygenase inhibitor, atropine, an anticholinergic, and cimetidine, an \( H_2 \) receptor antagonist, were incorporated into the Krebs’ solution at concentrations of \( 10^{-6} \) M.

Temperature of the isolated tissue baths was maintained at 37°C and the bathing solution aerated with 95% \( \text{O}_2 \) and 5% \( \text{CO}_2 \). Isometric measurements were made with Grass FT03C force-displacement transducers and recorded on a Grass polygraph as changes in grams of force.