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SHORT COMMUNICATION

Histamine Induced Decrease of Lecithin Levels in Broncho-Alveolar Lavage Fluid of Rats is Mediated by H_2 Receptor

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SUMMARY Lecithin, a major surface active substance of the surfactant system of the lung, was estimated in broncho-alveolar lavage (BAL) fluid in four groups of healthy adult male albino rats. Rats from group I were not administered any drug and acted as controls. Group II were administered histamine diphosphate. Group III were given H_1 blocker (pyrilamine maleate) followed by histamine diphosphate. Group IV received H_2 blocker (ranitidine hydrochloride) followed by histamine diphosphate. Lecithin content of BAL fluid in the control group was compared with that in the other three groups. A significant decrease in lecithin content was observed in the rats that received either histamine diphosphate or H_1 blocker followed by histamine diphosphate. However, compared to control rats no significant difference in lecithin content was seen in rats that received H_2 blocker followed by histamine diphosphate. The results clearly indicate that the decrease in surface active lecithin content in BAL fluid following administration of histamine diphosphate was unaffected by prior administration of H_1 blocker, but was blocked by prior administration of H_2 blocker. It was concluded that histamine induced decrease in lecithin content of BAL fluid is mediated through H_2 receptors. Since the predominant source of intra-alveolar lecithin are Type II cells of the alveolar epithelium, it is possible that Type II cells have H_2 receptors, stimulation of which resulted in decreased intra-alveolar lecithin.

The presence of surfactant in normal lungs to act against the surface tension forces was demonstrated by Pattle and Clements.[1,2] Pulmonary surfactant is essential in maintaining the patency of the alveoli.[3] It is also a factor acting against pulmonary edema.[4] There were many reports on regulation of pulmonary surfactant secretion by parasympathomimetics, sympathomimetics, anti-edema drugs, corticosteroids and other chemical mediators like substance P.[5-9] The effect of histamine on the secretion of lecithin from type II alveolar cell cultures of rat was studied.[10,11]

Broncho-alveolar lavage (BAL) is a standard procedure to assay various components of pulmonary surfactant system.[12] Lecithin is the major surface active phospholipid of the pulmonary surfactant system.[13] Thus, an assay of lecithin in BAL fluid was employed in the present study to learn about the *in vivo* effect of histamine on the lecithin content in broncho-alveolar lavage fluid of rats and to identify the type of histamine receptor responsible for the effect observed.

MATERIALS AND METHODS

Broncho-alveolar lavage:

Healthy adult male albino rats of Wistar strain weighing between 200-220 g were used. The animals were maintained in cages with free access to air, food and water. The rats were given pentobarbitone sodium intraperitoneally at a dose of 40 mg/kg. The anesthetized animals were incised from xiphisternum to chin. The thorax

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was opened and lungs along with trachea were isolated. The trachea was cannulated and alveoli were rinsed with normal saline via the airway. Ten milliliters of normal saline was introduced via the trachea and the fluid was retained in the lungs for one minute; then, it was rinsed back and forth and aspirated. The procedure was repeated till a volume of about 15 ml was extracted for each animal. Lungs which had abnormal appearance such as haemorrhagic spots or patches were not subjected to lavage. Samples from lungs which showed leakage of fluid during lavage, and lavage fluid which was contaminated with blood, or not clear, was not used for estimation of lecithin. Furthermore, samples collected with less than 70% of retrieval of instilled saline were also not used for determination of lecithin content.

Assay of lecithin

An assay of lecithin was performed by enzymatic method using chemicals and protocol supplied by Boehringer Mannheim Gmbh-Biochemica (Mannheim, Germany).

Experimental protocol

The animals were divided into 4 groups of 8 each. Rats from Group I were not administered any drug and lecithin content in BAL fluid in this group of animals acted as control. Individual rats from Group II were subcutaneously administered 0.06 mg of histamine diphosphate (Sigma Chemical Company, St. Louis, Mo.). This dose of histamine did not produce any untoward effects in pilot experiments. After 10 minutes of administration of histamine, BAL was performed and the lecithin content was estimated. Each rat in Group III was intraperitoneally given 0.25 mg pyrilamine maleate, a H_1 blocker, (Sigma Chemical Company) per animal. After 45 minutes, individual animals were administered 0.06 mg of histamine diphosphate subcutaneously. Following 10 minutes of administration of histamine, the BAL fluid was collected and the lecithin content was estimated. Each rat of Group IV was injected intraperitoneally 0.25 mg ranitidine hydrochloride, a H_2 blocker, (Sigma Chemical Company). Following 45 minutes of administration of ranitidine, each of the animals was given 0.06 mg of histamine diphosphate subcutaneously. After 10 minutes of injection of histamine, BAL was performed and the lecithin content was estimated. The doses of histamine blockers administered were equivalent to therapeutic doses employed in adult human beings.

Statistical analysis

Student's unpaired *t*-test was used.

RESULTS

The means and standard deviations of lecthin contents in BAL fluids of all groups of rats are shown in Fig. 1. The contents were 0.133 ± 0.07 for Group I (control group), 0.036 ± 0.02 for Group II (received histamine diphosphate), 0.043 ± 0.02 for Group III (treated with H_1 blocker [pyrilamine maleate] followed by histamine diphosphate), and 0.118 ± 0.05 for

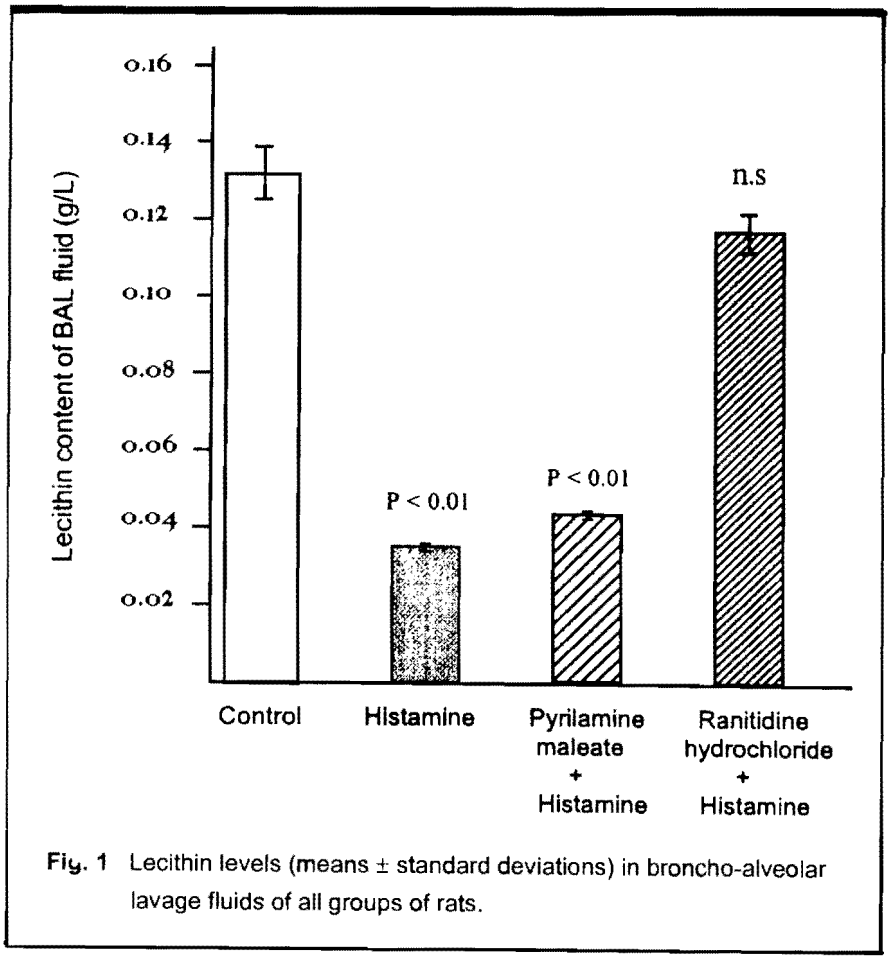

Fig. 1 Lecithin levels (means ± standard deviations) in broncho-alveolar lavage fluids of all groups of rats.

Group IV (received H_2 blocker [ranitidine hydrochloride] before administration of histamine diphosphate).

DISCUSSION

A significant reduction of the lecithin content in bronchoalveolar lavage (BAL) fluid was observed within 10 minutes of administration of histamine. Since lecithin is the predominant surface active substance in the pulmonary surfactant system, the finding indicates an acute decrease in pulmonary surfactant activity after administration of histamine.

Lecithin constitutes 60-70% of the surface active materials found in the pulmonary surfactant system. These surface active materials reduce surface tension forces of alveolar lining and prevent collapsing of the alveoli. Reduction in pulmonary surfactant activity leads to increased work of breathing and respiratory distress. A significant reduction in lecithin level was observed in the present study even at a dose that did not produce any untoward effects in the animals. Histamine is a common mediator in many allergic disorders. It is possible that reduction in lecithin levels may be one of the factors responsible for the respiratory distress in allergic diseases like bronchial asthma and pulmonary aspergillosis.. An assay of lecithin of the BAL fluid in the above patients may elucidate the effect of histamine on surfactant system of the lungs in adult human beings.

Decrease in lecithin content in BAL fluid following administration of histamine was unaffected by prior administration of H_1 blocker (pyrilamine maleate), but was blocked by prior administration of H_2 blocker (ranitidine hydrochloride). This indicates that the histamine induced decrease in lecithin content of BAL fluid observed in the present study was mediated through H_2 receptors. Since the predominant source of intra-alveolar lecithin is type II cells of alveolar epithelium, it is possible that type II cells have H_2 receptors, stimulation of which resulted in decreased intra-alveolar lecithin. Cheng and Brown[11] have reported stimulation of phosphatidyl choline secretion by histamine in alveolar type II cell cultures of adult rats which could be blocked by both H_1 antagonist (pyrilamine) and H_2 antagonist (cimetidine). Gilfillan *et al.*[10] observed stimulation of phosphatidyl choline secretion in type II cell cultures of adult rats by thiazinamium chloride which has antihistaminic properties. They have also observed stimulation of phosphatidyl choline secretion by H_1 antagonists (promethazine and pyrilamine) but not by H_2 antagonist (cimetidine). However, they were unable to demonstrate any inhibitory effect of histamine on *in vitro* cultures of type II pneumocytes. In our study which adult rats were used, a significant reduction in intra-alveolar lecithin levels was observed which could be blocked by H_2 antagonist (ranitidine) but not by H_1 antagonist (pyrilamine). The findings in the present study emphasize the fact that *in-vitro* and *in-vivo* effects may be different in view of the complexity of responses in the whole animal. Further, it appears that histamine has a role in the regulation of secretion of pulmonary surfactant.

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REFERENCES

1. Pattle RE. Properties, function and origin of the alveolar lining layer. Nature 1955; 175: 1125-6.
2. Clements JA. Surface tension of lung extracts. Proc soc Exp Biol Med 1957; 95: 170-2.
3. Avery ME, Mead J. Surface properties in relation to atelectasis and hyaline membrane disease. AMA J Dis Child 1959; 97: 517-23.
4. Rao GJ, Narayan KR, Rao AK, Nalini K. Experimental production of high surface tension pulmonary edema. Indian J Pathol Microbiol 1988; 31: 1-7.
5. Massaro D, Clerck L, Massaro GD. Surfactant secretion: Evidence that cholinergic stimulation of secretion is indirect. Am J Physiol 1982; 243: C39-45.
6. Rao GJ, Aroor AR, Rao AK, Sreedhar S. Effect of bronchodilators on surfactant system of lung. Indian J Med Res 1989; 90: 266-9.
7. Rao GJ, Aroor AR, Nalini K, Rao AK. Effect of drugs used in pulmonary edema on surfactant system of lung. Indian J Chest Dis Allied Sci 1991; 33: 59-62.
8. Rao GJ. Decreased lecithin in lung lavage fluid following intravenous hydrocortisone. Asia Pac J Pharmacol 1996; 11: 37-9.
9. Rice WR, Singleton FM. Regulation of surfactant secretion from isolated type II pneumocytes by substance P. Biochem Biophys Acta 1987; 889: 123-7.
10. Gilfillan AM, Lewis AJ, Rooney SA. Effect of thiazinamium chloride and other antihistamines on phosphatidylcholine secretion in rat type II pneumocyte cultures. Biochem Pharmacol 1987; 36: 277-81.
11. Chen M, Brown LA. Histamine stimulation of surfactant secretion by rat type II pneumocytes. Am J Physiol 1990; 258: L195-200.
12. Pattle RE. Surface lining of lung alveoli. Physiol Rev 1965; 45: 48-79.
13. King RJ, Clements JA. Lipid synthesis and surfactant turnover in the lungs. In: Fishman AP, Fisher AB eds. Handbook of Physiology: American Physiological Society, 1985; p. 309-36.