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Changes of Histamine Contents and Mast Cell Population in the Gastric Mucosa of Carbon Tetrachloride-Induced Cirrhotic Rats

Rokuroh SEIKOH, Osamu KODAMA, Yoshiteru OGAWA*, Nobuaki ITOH, Hitoshi TAKEUCHI, Tsuneo TANAKA, Yasuyuki ICHIBA and Kiyohiko DOHI

The 2nd Department of Surgery, Hiroshima University School of Medicine, 1-2-3, Kasumi, Minami-ku, Hiroshima 734, Japan
** Hiroshima Hospital of JNR, 3-1-36, Futabanosato, Higashi-ku, Hiroshima 730, Japan*
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ABSTRACT

For the purpose of clarifying the mechanism of gastric mucosal lesion complicated with liver cirrhosis, the authers examined the changes of histamine contents and mast cell population in the gastric mucosa of carbon tetrachloride-induced cirrhotic rats.

The histamine contents in the gastric mucosa and the mast cell count in the superficial area of the mucosa were significantly increased spontaneously and reduced with cold restraint stress in cirrhotic rats. However, no reductions in the both factors were observed in normal rats.

The incedence of gastric mucosal lesions in cirrhotic rats was 38%, while it was 0% in normal rats, and rose to 100% in the former against 28% in the latter with stress.

The hexosamine contents in the gastric mucosa were significantly lower in cirrhotic rats than in normal rats.

It is considered that in cirrhotic rats easy release of gastric mucosal histamine with stress, which is associated with degranulation of mast cells in superficial layer, results in development of gastric mucosal lesions, and that the reduction in defensive capacity may be one of the causes for the increase in mast cells.

Our examination of gastric mucosal lesions in 74 patients who underwent non-shunting operation for esophagial varices revealed that the incidence of gastric mucosal lesions was high and inclined to increase with operative treatment[3] and postoperative bleeding from gastric mucosal lesions constituted one of the causes of operative death in cases of liver cirrhosis.

In this study, carbon tetrachloride-induced cirrhotic rats were prepared and examined the changes in histamine contents and mast cell population in the gastric mucosa, with a view to experimental clarification of the pathogenesis of gastric mucosal lesions complicated with liver cirrhosis.

MATERIALS AND METHODS

1. Administration of carbon tetrachloride

Male Wistar rats weighing about 200g were used. Rats were administered 50% solution of carbon tetrachloride in olive oil at a dose of 0.3ml per 100g body weight intramuscularly twice a weak for four months.

2. Determination of histamine contents

After the fast for 24hr, rats were subjected to the cold restraint (4~6°C) in 30min or not, and then sacrificed by decapitation. The stomach was removed at once and opened along the greater curvature. Evidence of gastric mucosal lesions was examined grossly, then the fundic mucosa was removed by scratching.

Histamine was extracted from the mucosal tis-

sue by the method of Wada et al[8] and determined by the fluorometric method of Shore et al[6].

3. Mast cell counting

The stomach was pinned to a board and fixed for 24hr in a solution containing 0.6% formaldehyde and 0.5% acetic acid which was discribed by Enerback[1]. Then the tissue was dehydrated, embedded in paraffin, and sectioned at 7 microns. After staining with toluidine blue in a 0.05% aqueous solution at pH 4.1, mast cells were counted under oil immersion (x1000 magnification) in 40 fields from the superficial area immediately below the epithelium and the submucosal area.

4. Determination of hexosamine contents

Hexosamine contents in the fundic mucosal tissue were measured with a technique described by Robert[4].

RESULTS

1. Pathological findings of the liver

Large or small sized nodules were observed grossly on the surface of the liver, and the histological picture showed pseudolobule formation and fatty degeneration in the rats administered carbon tetrachloride (cirrhotic group).

2. Histamine contents in the gastric mucosa

As showed in Table 1, the histamine contents in the gastric mucosa were significantly higher in the cirrhotic group than in the normal control group. During cold restraint stress, the level of histamine contents in the cirrhotic group was significantly reduced, whereas no significant reduction was noted in the control group.

3. Mast cell population in the gastric mucosa

Table 2 shows the mast cell population in the gastric mucosa. In the superficial area of the mucosa, the number of mast cells in the cirrhotic group increases significantly as compared with in the normal control group. During the stress, the number of mast cells was significantly reduced in the cirrhotic group, while there was no significant change in the control group.

On the other hand, in the submucosa, no significant difference of mast cell counts was found between the cirrhotic group and the control group, and there were no significant changes of mast cell counts during the stress in the both groups.

4. Incidence of gastric mucosal lesions

As shown in Table 3, the incidence of gastric mucosal lesions was higher in the cirrhotic group than in the control group. During the stress, the incidence increased remarkably in the cirrhotic group, but the slight increase was noted in the control group.

5. Hexosamine contents in the gastric mucosa

The hexosamine contents were significantly lower in the cirrhotic group (20.8 ± 2.0 μg/mg, n=7) than in the control group (29.1 ± 2.1 μg/mg, n=6). ($p<0.01$)

Table 1. Histamine contents in the gastric mucosa in carbon tetrachrolide-induced cirrhotic rats and in control rats

Group	Histamine, μg/g (mean ± SD)	
	Non-stressed	Stressed
Cirrhosis	25.6 ± 3.0*,** (13)	17.5 ± 5.0* (9)
Control	16.0 ± 3.4** (15)	15.4 ± 4.0 (12)

Numbers in parenthesis indicates the number of animals. Statistical significance of differences was ditermined by t test.

* $p<0.01$: significance of difference between the non-stressed rats and stressed rats

** $p<0.01$: significance of difference between the cirrhotic rats and control rats

Table 2. Mast cell populations in the gastric mucosa in cirrhotic rats and in control rats

Group	Superficial area		Submucosa	
	Non-stressed	Stressed	Non-stressed	Stressed
Cirrhosis	390 ± 159*,** (7)	234 ± 74* (7)	33 ± 8 (7)	31 ± 6 (7)
Control	135 ± 40** (7)	162 ± 79 (7)	31 ± 7 (7)	32 ± 9 (7)

*$p<0.05$: significance of difference between the non-stressed rats and stressed rats (t test)

**$p<0.01$: significance of difference between the cirrhotic rats and control rats (t test)

Table 3. Incidences of gastric mucosal lesions in cirrhotic rats and control rats

Group	Non-stressed	Stressed
Cirrhosis	6/16 (37.6%)	15/15 (100%)
Control	0/26 (0%)	5/18 (27.8%)

DISCUSSION

Histamine is an ulcerogenic agent, and the liver is an important organ for histamine metabolism. In liver cirrhosis, liver function disorder and portosystemic collateral circulation may result in histamine inactivation. Shastin[5] reported that in patients with liver cirrhosis the blood histamine levels were elevated. But the relation of gastric mucosal histamine to gastric mucosal lesions complicated with liver cirrhosis has not clarified yet.

It was found in the present study that the histamine contents in the gastric mucosa of cirrhotic rats were increased, and that mucosal histamine was released easily with stress, presumably causing the onset and the aggravation of gastric mucosal lesions.

Histamine in the gastric mucosa of rats is considered to exist in mast cells and enterochromaffin-like cells. Guth et al[2] reported that the degranulation of the mast cells in the gastric mucosa occured with stress and the released vasoactive amines might induce stress ulcers. Therefore the investigation of mucosal mast cells is important to the research of the alteration of histamine contents. The mast cells in the gastric mucosa of rats are numarous in the superficial area and the submucosa. Examination of mast cells in these two areas revealed that in cirrhotic rats the mast cells in the superficial area were increased spontaneously, and decreased easily with stress.

It was considered from these findings that in cirrhotic rats the increase in the number of mast cells in the superficial area of the mucosa was one of the causes for the increase in the histamine contents rats, and that the degranulation of the mast cells in the superficial area with stress played an important role for the release of histamine.

Mast cells have been thought to be associated with the repairing process of tissue, and an increase in mast cell counts in the gastric wall was reported with inflammatory changes including gastritis and ulcer[7]. It is speculated that the reduction in hexosamine contents in the gastric mucosa in cirrhotic rats may result in damage of the epithelium, and induce the increase in the number of mast cells in the superficial area of the mucosa.

REFERENCES

1. **Enerback, L.** 1966. Mast cells in gastrointestinal mucosa. 1. Effect of fixation. Acta. Path. et Microbiol. Scandinav. **66**: 289-302.
2. **Guth, P. H. and Hall, P.** 1966. Microcirculatory and mast cell changes in restraint-induced gastric ulcer. Gastroenterol. **50**: 562-570.
3. **Kodama, O., Seikoh, R., Tanaka, T., Matsuyama, T., Nishiki, M., Dohi, K. and Ezaki, H.** 1985. Gastric mucosal lesions in cases of nonshunting procedures for esophageal varices. Hiroshima J. Med. Sci. **34**: 151-154.
4. **Robert, A., Bayer, R.B. and Nezamis, J.E.** 1963. Gastric mucus content during development of ulcers in fasting rats. Gastroenterol. **45**: 740-751.
5. **Shastin, N.N.** 1972. Peculiarities of histamine metabolism in chronic hepatitis and liver cirrhosis. Klin. Med. (Mosk.) **50**: 39-42.
6. **Shore, P.A., Burkhalter, A. and Cohn, V.H.** 1959. A method for the fluorometric assay of histamine in tissue. J. Pharm. Exp. Ther. **127**: 182-186.
7. **Siurala, M. and Sundberg, M.** 1958. Occurence of mast cells in the gastric mucosa under normal and pathological conditions. Ann. Med. Exp. et Biol. Fenn. **36**: 271-277.
8. **Wada, H., Yamatodani, A. and Ogasawara, S.** 1977. Systematic determination of biogenic amines. Seitai no kagaku. **28**: 215-222.