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Glicentin-containing cells in intestinal metaplasia, adenoma and carcinoma of the stomach

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Summary. Glicentin-containing cells (Glic. cells) in intestinal metaplasia, adenoma and carcinoma of the stomach were examined using immunohistochemical techniques. Glic. cells first occurred in the gastric mucosa of the transitional area between metaplastic and intact gastric glands. They frequently showed hyperplasia or micronoduli in the budding area of the deeper metaplastic glands, but in completely intestinalized mucosa these endocrine cells decreased remarkably. Gastric adenomas with mild dysplasia had a good number of glicentin-immunoreactive cells which were located in the deeper adenoma glands. Gastrin- and somatostatin-positive cells were also detected in the adenomas. The incidence of glicentin-positive tumor cells was significantly higher in well differentiated adenocarcinoma than in poorly differentiated adenocarcinoma. Among the seven cases of scirrhous argyrophil cell carcinoma, three showed glicentin- and glucagon-immunoreactivity in the same area of the tumor. These findings suggest that the selective increase of Glic. cells in intestinal metaplasia may be closely related to the development of gastric adenoma. Glicentin positive tumor cells in gastric carcinomas can be regarded to be an expression of intestinal or fetal markers.

Key words: Endocrine cells-Glicentin-Intestinal metaplasia-Adenoma-Argyrophil cell carcinoma

Glicentin or enteroglucagon, which is also called glucagon-like immunoreactivity (GLI), was first extracted by Sundby in 1976 from the porcine lower intestinal tract. The structure of glicentin is nearly analogous to proglucagon and consists of 69 amino acid residues (Thim and Moody 1981). Moreover, glicentin has been shown to have a trophic action on the mucosal growth of the intestine (Bloom and Polak 1981; Besterman 1982).

Glicentin containing cells (Glic. cells), which are normally distributed

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in the ileum, have also been detected in the intestinal metaplasia of the stomach (Bordi 1979; Nielsen 1979; Tsutsumi 1983). However, hardly any detailed studies have been made on the presence of Glic. cells in gastric tumors. More recently Shimamoto and Tahara (1982) have observed glicentin-producing carcinomas of the rat intestinal tract induced by 1,2-dimethylhydrazine. Further, in our previous study we have demonstrated production of gastrin, somatostatin and glucagon in carcinomas of the human stomach (Tahara et al. 1982a and b).

The purpose of the present study was to investigate the localization of Glic. cells in intestinal metaplasia, adenoma and carcinoma of the stomach and to clarify the significance of ectopic glicentin expression in gastric tumors.

Material and methods

Studies were made on 88 surgically resected stomachs composed of 49 cases of advanced gastric carcinoma including scirrhous argyrophil cell carcinoma, 35 cases of adenoma, and 4 cases of peptic ulcers. These cases were extracted from the files of the Department of Pathology, Hiroshima University School of Medicine and Kure Mutual Aid Hospital. For each case, 5–20 tissue sections including the main lesions were excised longitudinally after fixing in 10% formalin and embedding in paraffin. Two or three representative paraffin blocks were selected from each case for light microscopy and immunohistochemistry. Adjacent or serial sections cut 4 μm in thickness from these tissue blocks were stained with hematoxylin and eosin, periodic acid Schiff (PAS), and Grimelius silver nitrate technique for argyrophil reaction.

Histological classification of advanced gastric carcinoma was made according to the criteria of the Japanese Research Society for Gastric Cancer (1979). Argyrophil cell carcinoma of the stomach was diagnosed as carcinoma consisting predominantly of neoplastic argyrophil cells with a diffuse distribution throughout the tumor as described in detail by Tahara (1982). Histological definition and grading of gastric adenoma were made according the WHO classification and Nagayo's criteria (1983). In cases of tubular adenoma and peptic ulcers, the number of endocrine cells was counted in the area, which were divided into mucosal unit area of 5 mm in width × mucosal or tumor height.

Immunohistochemistry. A modification of the immunoglobulin enzyme bridge technique as described in detail by Tahara et al. (1982) was employed in light microscopy. Deparaffinized tissue sections 4 μm thickness were treated consecutively for 30 min with (1) specific rabbit antisera at room temperature, (2) goat anti-rabbit IgG antiserum, and (3) soluble peroxidase-antiperoxidase complex (rabbit, 1:60). Peroxidase was stained for 5–10 min, using 3, 3′-diaminobenzidine-tetrahydrochloride (DAB) (0.003% w/v) and hydrogen peroxide (0.001%) in 0.05 M Tris-HCl buffer (pH 7.6). The sections were counterstained with 3% methyl green or hematoxylin. In some tissue sections, the immunohistochemical procedure was performed after Grimelius technique for argyrophil reaction as double staining in order to confirm the morphological characteristics of endocrine cells.

Preparation and characterization of glicentin antiserum (R4804) and glucagon antiserum (OAL123) have been described previously (Yanaihara 1980). Briefly, glicentin antiserum has been shown to be specific for glicentin C-terminal fragment and demonstrated a specific reactivity to glicentin and proglucagon. Glucagon antiserum was prepared with glucagon C-terminal fragments (19–29) and showed a specific reaction to glucagon, but did not show a positive reaction to proglucagon or glicentin. Antigastrin serum was kindly supplied by Prof. Miyoshi, Department of Internal Medicine, Hiroshima University School of Medicine and was employed after dilution at 1:2,000. Antibody for somatostatin was purchased from Japan Immunoresearch Laboratories Co. Ltd, and lyophilized preparations were diluted 1:150.

Antirabbit IgG were prepared by MBL Company (Japan) and then were absorbed by

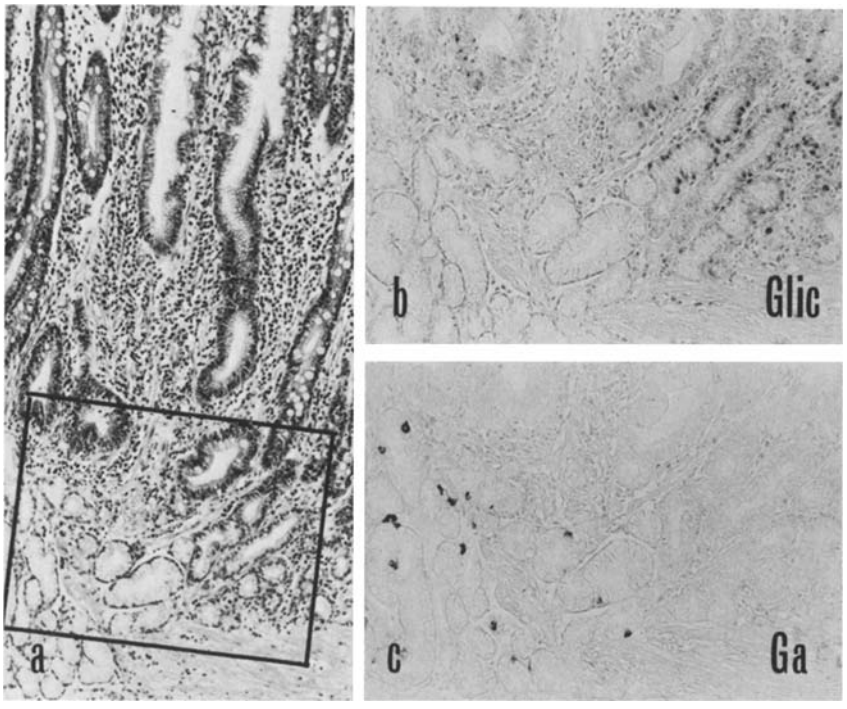

Fig. 1a–c. Serial section prepared from gastric mucosa with extensive intestinal metaplasia. **a** Hematoxylin and eosin (×80). **b** A higher magnification in square area of photomicrograph a. Glic. cells are found in deeper area of gastric mucosa. Immunostaining with anti-glicentin serum (×130). **c** A higher magnification in square of photomicrograph a. G cells are found in remained pyloric glands. Immunostaining with anti-gastrin serum (×130)

human immunogloblin without known cross-reactivities against other animal serum proteins. Peroxidase complex (rabbit) was obtained from Dakopatts A/C at a concentration of 1:60.

The specificity of the reaction was determined as described by Sternberger (1979): (1) Specific antisera to gastrin and somatostatin were absorbed at 4° C for 24 h with excess antigen; (2) non-immune rabbit serum as the first layer; (3) ommition of 3,3′-diaminobenzidin-tetrahydrochlorides or H_2O_2 from the incubation medium for the peroxidase reaction. Gastrin, glucagon and somatostatin were kindly provided by Japan Immunoresearch Laboratory Co., Ltd. Control reactions were invariably negative.

Appropriate positive control-slides were always stained as follows at the same time: Gastric antral mucosa for gastrin and somatostatin, pancreatic tissue for somatostatin, glucagon and glicentin, and ileum end for glicentin.

Results

1. Intestinal metaplasia

Glic. cells could not be found in the normal gastric mucosa idependently of the number of inflammatory cells in the lamina propria. They were first detected in the transitional area between metaplastic glands and pyloric or fundic glands (Fig. 1). These endocrine cells showed a columnar appearance with oval or elongated nuclei and strong immunoreactivity for antigli-

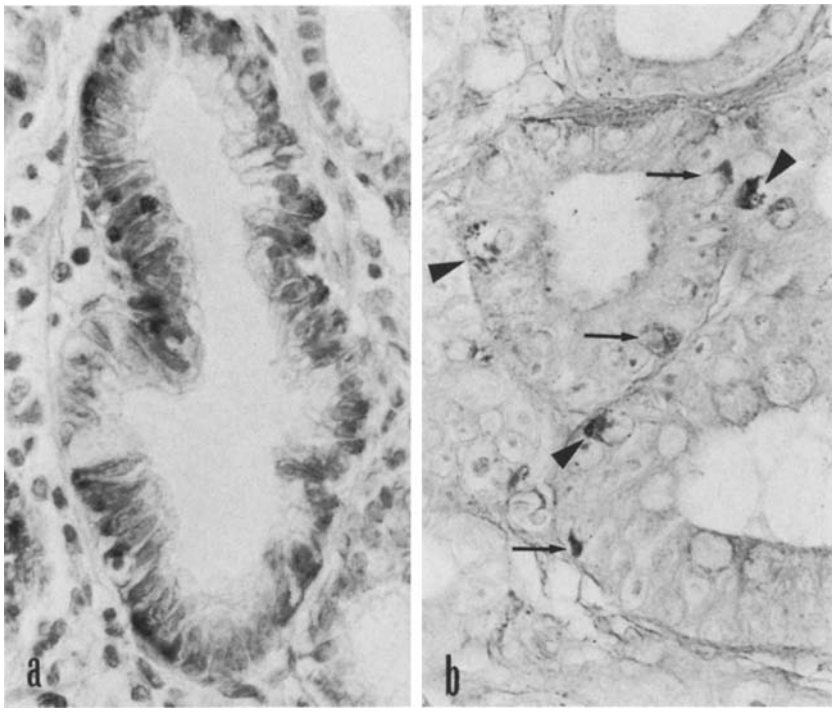

Fig. 2a, b. a A high magnification of Glic. cells showing a columnar appearance with oval or round nucleus. Immunostaining with anti-glicentin serum (×600). **b** Most of Glic. cells (*arrows*) show no argyrophil reaction. Grimelius positive argyrophil cells (triangles) do not coincide with the Glic. cells. Double staining with anti-glicentin serum and Grimelius argyrophil reaction (×600)

centin serum in the basal area (Fig. 2). They had obviously open-cell type character with a thin apex reaching the tubular lumen together with G and D cells in the antrum. Most of them showed no argyrophil reaction using Grimelius silver nitrate (Fig. 2).

In the contrast to the distribution of G and D cells in the gastric mucosa as described in detail elsewhere (Ito 1983), the number of Glic. cells increased with extension of intestinal metaplasia. In intestinal metaplasia frequently showing budding, a large number of Glic. cells were detected in the deeper zone of the gastric mucosa (Fig. 3a and b). Moreover, micronoduli of these cells were sometimes noted (Fig. 3c). However, in a single metaplastic gland showing complete intestinalization, they were present in a small numbers.

The correlation between average number of G cells and Glic. cells in intestinal metaplasia was examined in each unit area in the antrum of cases with peptic ulcer and gastric cancer. In the gastric mucosa, in which more than 60% of the glands were intestinalized, the number of Glic. cells became greater than that of G cells (Fig. 4). Glucagon-immunoreactive cells were also rarely noted in the intestinal metaplasia. Moreover, non-argentaffin argyrophil cells which did not show any immunoreactivity for gastrin increased in intestinal metaplasia.

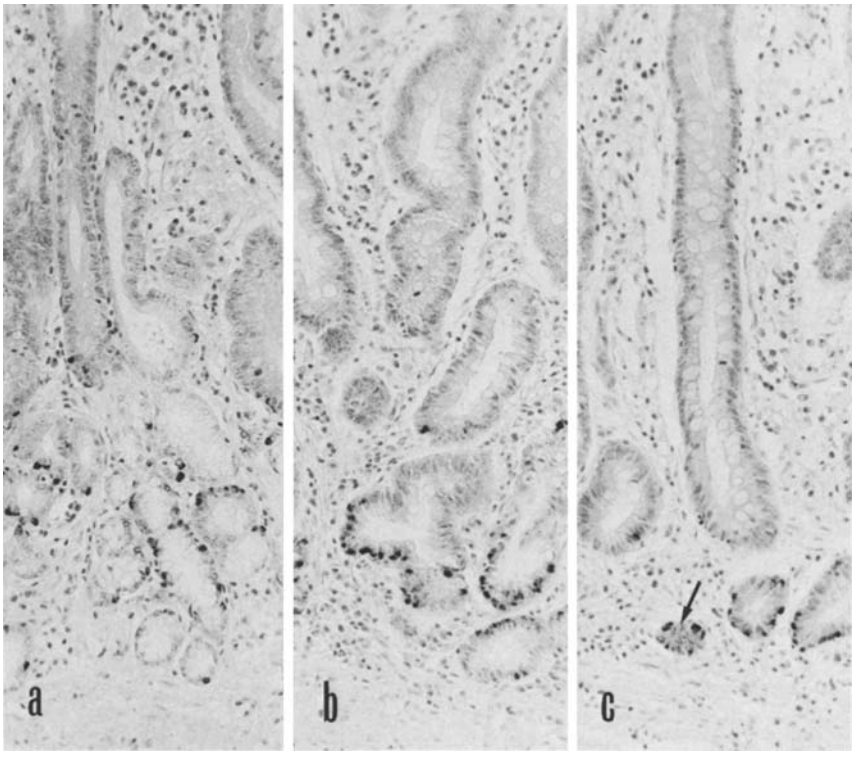

Fig. 3a–c. **a** and **b** A large number of Glic. cells are detected in the deeper zone of intestinal metaplasia frequently showing budding. Immunostaining with anti-glicentin serum (×160). **c** Only a few Glic. cells are found in a single metaplastic gland. Micronodule composed of Glic. cells (*arrow*) is also noted. Immunostaining with anti-glicentin serum (×160)

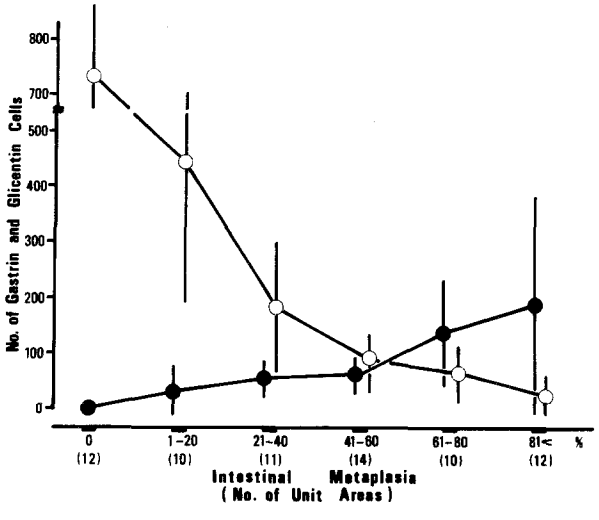

Fig. 4. Correlation between the number of gastrin and glicentin cells with intestinal metaplasia (○ = Gastrin cells; ● = Glicentin cells)

Table 1. Incidence of Glicentin, Gastrin and Somatostatin-Positive Cells in Adenoma and Carcinoma of the Stomach

Histologic Type	Number of Cases	Peptide-Immunoreactivity			
		Glic.[b]	Gluc.[b]	Gast.[b]	Somato.[b]
Adenoma	35	27 (77.1%)	0 (0%)	5 (14.2%)	11 (31.4%)
Adenocarcinoma[a]					
pap., tub.	20	8* (40.4%)	2 (10.0%)	6 (30.0%)	1 (5.0%)
por., sig.	22	1* (4.5%)	1 (4.5%)	6 (27.3%)	3 (13.6%)
Scirrhous					
Argyrophil cell carcinoma	7	3 (42.9%)	3 (42.9%)	2 (28.9%)	3 (42.9%)

[a] According to the classification of Japanese Research Society for Gastric Cancer; pap = papillary adenocarcinoma, tub = tubular adenocarcinoma, por = poorly differentiated adenocarcinoma, sig = signet ring cell carcinoma.
[b] Glic. = Glicentin, Gast. = Gastrin, Somato. = Somatostatin.
* Significantly different from pap, tub and por, sig; $P < 0.025$

Table 2. Histological Grade and Number of Glicentin, Gastrin, Somatostatin-positive Cells in Adenoma of the Stomach

Histological Grade	Number of adenoma	Size of adenoma (mm)	Glicentin cells	Gastrin cells	Somatostatin cells
I Mild Dysplasia	17	7.7±3.0[b]	146.2±189.43[a]	0.6±0.8	26.4±68.3
II Moderate Dysplasia	9	13.6±5.9	16.0±25.9	1.0±3.0	3.3±6.2
III Severe Dysplasia	9	17.6±7.6	9.1±16.4	0.9±2.7	2.3±7.0

[a] Number of cells/5 mm length × tumor height
[b] Results are expressed as the mean ±SD

2. Gastric adenoma

Out of 35 cases of gastric adenoma, 24 cases were associated with gastric carcinoma and one case with gastric lymphoma, but they were localized in the mucosa distal from these main tumors. The adenoma had a longitudinal dimension ranging from 4 to 50 mm and most of them showed broad-based mucosal elevation with central erosion. Adenoma glands with various grades of cellular atypia occupied the upper portion of the mucosa and were frequently accompanied with cystic dilatation of the intact pyloric glands in the subjacent part of adenomas.

Immunohistochemically, Glic. cells were detected in 27 (77.1%) out of 35 adenomas (Table 1) and were located in the deeper area of the adenoma

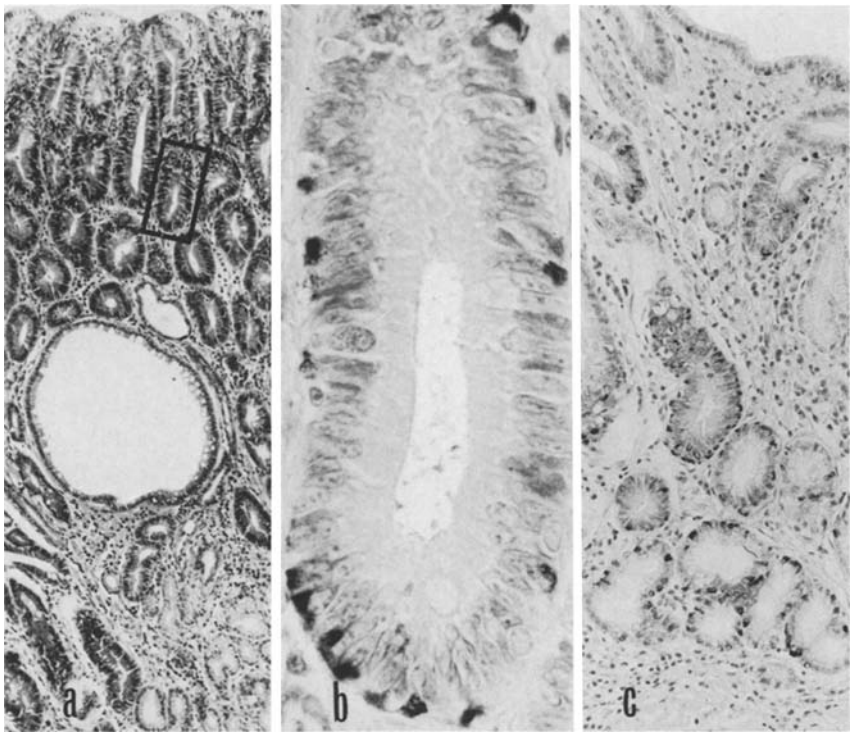

Fig. 5a–c. **a** Gastric adenoma with mild dysplasia is accompanied with cystic dilatation of the glands. Hematoxylin and eosin (×100). **b** A high magnification of the setting in square of photomicrograph a. Numerous Glic. cells are noted in the adenoma gland. Immunostaining with anti-glicentin serum (×400). **c** Hyperplasia of Glic. cells is found in subjacent mucosa of adenoma and in cystic dilated glands. Immunostaining with anti-glicentin serum (×260)

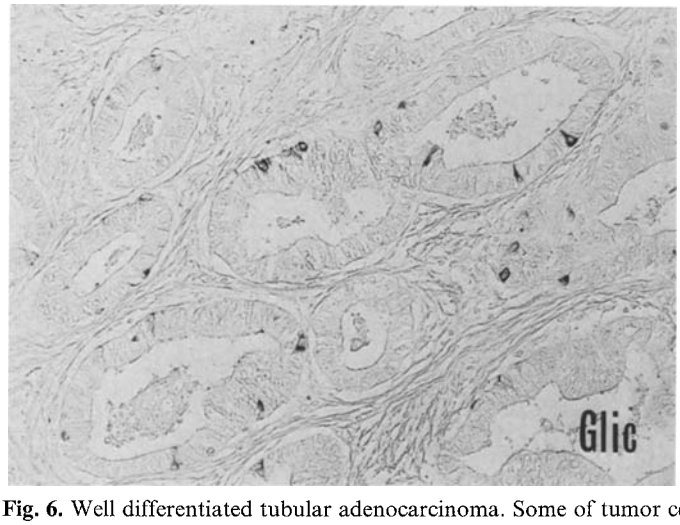

Fig. 6. Well differentiated tubular adenocarcinoma. Some of tumor cells show glicentin immunoreactivity. Immunostaining with anti-glicentin serum (×160)

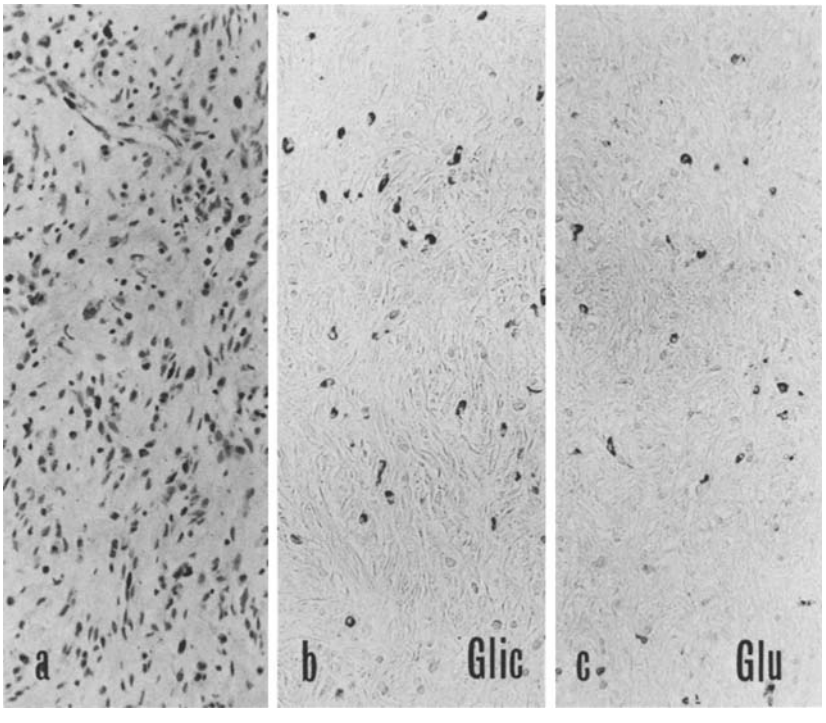

Fig. 7a–c. **a** Scirrhous argyrophil cell carcinoma with dense fibrosis. Hematoxylin and eosin (× 200). **b** Same tumor as in **a**. A good number of tumor cells show glicentin immunoreactivity. Immunostaining with anti-glicentin serum (× 200). **c** Glucagon-immunoreactive cells are also detected in the same area as in **b**. Immunostaining with anti-glucagon serum (× 200)

gland (Fig. 5a and b). Moreover, these endocrine cells were frequently found in the pyloric or cystic metaplastic glands, which were present under the adenoma (Fig. 5a and c).

The correlation between the grade of dysplasia and the number of Glic., G and D cells in adenoma was examined (Table 2). The grade of dysplasia was subdivided according to Nagayo's criteria (1983). Glic. cells were frequently found in adenoma with mild dysplasia and its number decreased with increase in grade of dysplasia. G cells and D cells were also found in adenoma, but they were remarkably lower in number when compared with Glic. cells. A cells were not observed in adenoma. Numerous non-argentaffin argyrophil cells without glicentin immunoreactivity were also present.

3. Gastric carcinomas

Of the 49 cases of advanced gastric carcinoma, 20 were well differentiated adenocarcinoma, 22 poorly differentiated adenocarcinoma and seven scirrhous argyrophil cell carcinoma.

The incidence of tumor cells which showed immunoreactivity for glicen-

Table 3. Pathological and Immunohistochemical Findings of 7 cases with Scirrhous Argyrophil cell Carcinoma of The Stomach

Patients			Site of Tumor	Gross findings (Borrmann's classification)	Immunoreactivity[b]			
No.	Age	Sex			Glic.[c]	Gluc.	Gast.	Somato.
1.	29	F	M[d]	III	−	−	+++	−
2.	26	M	M	III	−	−	−	−
3.	29	M	C[e]	III	+++	+++	++	+
4.	25	F	M	III	−	−	−	++
5.	29	M	C	III	++	+	−	−
6.	71	M	M	IV	+	+	−	−
7.	76	F	C	IV	−	−	−	++

[a] According to the classification of Japanese Research Society for Gastric Cancer
[b] These reactions are graded +, ++ and +++ on the basis of the frequency of staining of individual cells
[c] Glic. = Glicentin, Gluc. = Glucagon, Gast. = Gastrin, Somato. = Somatostatin.
[d] Middle third of stomach corresponds to intermediate zone and lower fundus
[e] Upper third of stomach corresponds to cardia and upper fundus

tin, glucagon, gastrin and somatostatin is shown in Table 1. Glicentin was detected in eight (40.0%) of the 20 cases of well differentiated adenocarcinoma and in one (4.5%) of the 22 cases of poorly differentiated adenocarcinoma with a significant difference ($P < 0.025$) in the incidence of glicentin between well differentiated type and poorly differentiated type of gastric carcinoma. Immunoreactive gastrin and somatostatin were also detected in 12 (28.6%) and four (9.5%) cases, respectively, but there was no difference in incidence between the two histological types. Furthermore, immunoreactive glucagon was found in tumor cells of 3 cases, which were not necessarily associated with glicentin positive cases. All of these peptide-positive cells were scattered or restricted to a small area of the tumor (Fig. 6).

Among the seven cases of scirrhous argyrophil cell carcinoma which had numerous argyrophil cells with extensive fibrosis, three had various numbers of Glic. cells. In addition, these three cases showed glucagon-immunoreactivity in the same area of the tumor (Fig. 7). Furthermore, one of them having a good number of glicentin- and glucagon positive cells had gastrin and somatostatin at the same time (Table 3). The remaining two cases which showed no immunoreactivity for glicentin had gastrin or somatostatin-reactive cells. No overt endocrine syndrome could not be observed in these scirrhous argyrophil cell carcinoma cases. Macroscopically, all of these tumors showed ulcerative tumor, Borrmann's type three or four carcinoma and were observed in the intermediate zone or fundus. The results of the immunohistochemical analysis are summarized in Table 3.

Discussion

The physiological functions of glicentin have not been adequately elucidated, but there is no doubt that glicentin has a trophic action on intestinal epithelia

(Bloom 1982). This is supported by the clinical study that serum enteroglucagon level is significantly higher in coeliac disease or acute tropical sprue than in normal cases (Besterman 1982). Furthermore, Moody (1983) has reported that glicentin also has an inhibitory effect on gastric acid secretion.

Meanwhile, glicentin producing cells which correspond to L cells according to the Lausanne classification (Solcia 1978) are localized in the ileum, but cannot be detected in the normal stomach (Bloom 1978). Thus, they might be regarded to be intestinal type endocrine cells. However, in the earlier stage of human fetuses, many Glic. cells occur in the gastric mucosa (Iwanaga et al. 1983). In this study, Glic. cells were first detected in the transitional area between intestinal metaplastic glands and pyloric or fundic glands. With the extension of intestinalization of the gastric mucosa, Glic. cells increased and frequently showed hyperplasia in the deeper zone of the gastric mucosa to exceed G cells in number. However, in a completely intestinalized single gland, only a few Glic. cells could be seen. These findings suggest that glicentin might play an important role in the development of intestinal metaplasia. Glic. cells might promote more or less the intestinalization of gastric mucosa by inhibiting gastric acidity and its trophic action via paracrine secretion.

Intestinal metaplasia has been subdivided into two types, complete and incomplete (Kawachi 1974; Matsukura 1980). The former is characterized by total expression of intestinal marker enzymes without sulfomucin-containing goblet cells and the presence of Paneth cells. The latter, incomplete type, which was considered to have a closer relation to gastric cancer than complete type, is characterized by partial expression of intestinal marker enzyme with sulfomucin in goblet cells and the absence of Paneth cells. The distinction of these two types of intestinal metaplasia, however, could not be clarified in this study, because Glic. cells are found in gastric mucosa in or near intestinal metaplasia with both types.

In view of the histological features, the close relation between intestinal metaplasia and gastric adenoma has been discussed (Nakamura 1966; Nagayo 1971; Sugano 1979). Furthermore, it is well known that intestinal metaplasia is frequently complicated by dysplasia of all grades, which is observed in adenomas, a precancerous lesion in the stomach (Morson 1980; Grundmann 1983). An adenoma is composed of several types of epithelial cells, such as absorptive cells, goblet cells and also argentaffin cells (Watanabe 1972). However, few reports on the presence of other kinds of endocrine cells in gastric adenoma have been made. Here we found 1) numerous Glic. cells were located in the deeper zone in both adenoma and metaplasia; notably in adenoma with mild dysplasia, 2) gastric type endocrine cells, G and D cells decreased dramatically, indeed they were almost absent. Because of these similarities in the distribution of endocrine cells between gastric adenoma and intestinal metaplasia, it is suggested that gastric adenoma may develop in gastric mucosa in the process of intestinal metaplasia. Cystic dilatation of glands in the subjacent part of adenoma is one of the characteristic appearances of gastric adenoma. We have observed scattered Glic. cells in the epithelium of cystic dilated glands. These findings show

that dilated cystic glands are not retention cysts but are newly formed and are one of the components of gastric adenoma, as reported by Nagayo (1971).

Since the report of Hamperl (1972), it has been well known that argentaffin or argyrophil cells are observed in ordinary gastric carcinoma except for classical carcinoid (Azzopardi 1963; Kubo 1972; Watanabe 1974; Tahara 1975). More recently, we have demonstrated a synchronous production of gut peptides, amine and also glycoproteins such as CEA, HCG and lysozyme in scirrhous argyrophil cell carcinoma (Tahara 1982a and b). The relationship between endocrine cells in the gastrointestinal tract and tumors has also been reported by Tahara in 1975 as follows; (1) classical carcinoid of endocrine cell origin, (2) endocrine cell carcinoma often showing poorly differentiated adenocarcinoma, and (3) endocrine cell clones with scattered appearance in the tumor. The advanced gastric cancers in the present study correspond to (3) and are considered to be attributable to differentiation of the tumor. Glicentin immunoreactivities in tumor cells were found more frequently in well differentiated adenocarcinomas than poorly differentiated adenocarcinomas. Tumor cells of well differentiated adenocarcinoma of the stomach are well known to have many features of intestinal metaplasia and therefore, glicentin-positive cells in well differentiated adenocarcinoma can be considered to be an expression of intestinal endocrine cells.

On the other hand, scirrhous argyrophil cell carcinoma in the present report corresponds to (2) and is assumed to originate from totipotent stem cells of endodermal origin as described by Tahara (1975) and Shimamoto et al. (1983). In the present study, three out of seven cases of scirrhous argyrophil cell carcinoma showed synchronous production such peptides as glicentin, glucagon, and gastrin or somatostatin. It is of interest that glicentin and glucagon were detected synchronously in all three cases. Glicentin-immunoreactivity in these tumors might be proglucagon. These tumors might have undevelopment of the enzyme-system in converting glicentin into glucagon, as happens in human fetuses. Therefore, production of glicentin or proglucagon and glucagon in the tumor may be an expression of a fetal marker. In fact, Glic. cells and A cells are present in the human fetal gastric body (Ito 1981; Iwanaga et al. 1983).

The role of immunoreactive glicentin in gastric carcinoma on its tumor growth, however, could not be elucidated in this study. Furthermore, it must be considered that immunoreactive glicentin may not be identical to biologically active glicentin, especially in carcinoma. Nevertheless, glicentin or Glic. cells seemed to have an intimate relationship with intestinal metaplasia, gastric adenoma, well differentiated adenocarcinoma and also scirrhous argyrophil cell carcinoma of the stomach, as mentioned above. Clinical attention should be paid to the serum glicentin level in patients with these gastric diseases. It may serve as a useful marker for intestinal metaplasia, adenoma and carcinoma of the stomach.

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References

Azzopardi JC, Pollock DJ (1963) Argentaffin and argyrophil cells in gastric carcinoma. J Pathol Bacteriol 86:443–451

Besterman HS, Adrian TE, Mallinson CN, Christofides ND, Sarson DL, Pera A, Lombardo L, Modinglianin R, Bloom SR (1982) Gut hormone release after intestinal resection. Gut 23:854–861

Bloom SR, Polak JM (1978) Gut hormone overview. In: Bloom SR (ed) Gut hormones. Churchill Livingstone. pp 3–18

Bloom SR, Polak JM (1981) Enteroglucagon and the gut hormone profile of intestinal adaptation. In: Robinson JWL, Dowling RH, Riecken EO (eds) Mechanisms of intestinal adaptation. MTP press: Lancaster, Boston, The Hague. pp 189–199

Bordi C, Pavazzola M (1979) Endocrine cells in the intestinal metaplasia of gastric mucosa. Am J Pathol 96:391–398

Grundmann E (1983) Classification and clinical consequence of precancerous lesions in the digestive and respiratory tracts. Acta Pathol Jpn 33:195–217

Hamperl H (1927) Ueber die „gelben (chromaffinen)" Zellen in gesunden und kranken Magen-Darmschlauch. Virchow Arch Pathol Anat 226:509–548

Ito H, Tahara E (1983) Immunohistochemical study on G and D cells in the human resected stomach with peptic ulcer diseases. In: Miyoshi A (ed) Gut peptides and ulcer. Biochemical Research Foundation, Tokyo. pp 180–187

Ito S, Iwanaga T, Kusumoto Y, Sudo M, Suzuki T, Shibata A (1979) Is glucagon present in the human gastric fundus? Horm Meta Res 282:260–266

Iwanaga T, Ito S, Fujita T, Yanaihara N (1983) Brain-gut peptides and their precursors: Big gastrin, glicentin, proinsulin and somatostatin. An immunohistochemical study. In: Miyoshi (ed) Gut peptides and ulcer. Biochemical Research Foundation, Tokyo. pp 93–101

Japanese Research Society for Gastric Cancer (1979) The general roles for gastric cancer study in surgery and pathology. Kanahara Shuppan Tokyo

Kawachi T, Kogure K et al. (1974) Studies of intestinal metaplasia in the gastric mucosa by detection of disaccharidases with "Tes-Tape" J Natl Cancer Inst. 53:19–30

Kubo T, Watanabe H (1971) Neoplastic argentaffin cells in gastric and intestinal carcinomas. Cancer 27:447–454

Matsukura N, Suzuki K et al (1980) Distribution of marker enzymes and mucin in intestinal metaplasia in human stomach and relation of complete and incomplete types of intestinal metaplasia to minute gastric carcinomas. J Natl Cancer Inst 65:231–240

Moody AJ, Thim L, Kirkrgaard (1983) Glicentin: Its structure and possible function as an enterogastrone. In: Miyoshi A (ed) Gut peptides and ulcer. Bioch Res Found, Tokyo. pp 20–26

Morson BC, Sobin LH, Grundmann E et al. (1980) Precancerous conditions and epithelial dysplasia in the stomach. J Clin Pathol 33:711–721

Nakamura K, Sugano H, Takagi K, Fuchigami A (1966) Histopathological study on early carcinoma of the stomach: Criteria for diagnosis of atypical epithelium.GANN 57:613–620

Nagayo T (1971) Histological diagnosis of biopsied gastric mucosa with special reference to that of borderline lesions. Gann Monograph Canc Res 11:245–256

Nagayo T (1983) Precancerous changes of the stomach from the aspect of dysplasia of gastric mucosa – Histological study. In: Sherlock P, Morson BC, Barbara L, Veronesi U (eds) Precancerous lesions of the gastrointestinal tract. Raven Press, New York. pp 115–126

Nielsen HO, Teglbjaerg PS, Hage E (1979) Gastrin and enteroglucagon in human antrum, with special reference to intestinal metaplasia. Scand J Gastroenterol [Suppl] 14:101–103

Shimamoto F, Tahara E, Yanaihara N (1983) Gut endocrine cells in rat intestinal tract carcinoma induced by 1,2-dimethylhydrazine. J Cancer Res Clin Oncol 105:221–230

Solcia E, Polak JM, Pearse AGE, Forssman WG, Larsson LI, Sundler F, Lechago J, Grimelius

L, Fujita T, Creutzfeldt W, Gepts W, Falkmer S, Lefrance G, Heitz PH, Hage E, Buchan AMJ, Bloom SR, Grossman MI (1978) Lausanne 1977 classification of gastroenteropancreatic endocrine cells. In: Bloom SR (ed) Gut hormones. Churchill, Livingstone, Edinburgh. pp 40–48

Sternberger LA (1979) Immunochemistry. Wiely, New York, Chichester, Brisbane, Tronto

Sugano H, Nakamura K, Takagi K (1971) An atypical epithelium of the stomach: A clinicopathological entity. Gann Monograph on Canc Res 11:257–269

Sundby F, Markussen J, Moody AJ (1976) Purification and characterization of a protein from porcine gut with glucagon-like immunoreactivity. Horm Metab Res 8:366–371

Tahara E, Haizuka S, Kodama T, Yamada A (1975) The relationship of gastrointestinal endocrine cells to gastric epithelial changes with special reference to gastric cancer. Acta Path Jpn 25:161–177

Tahara E, Ito H, Shimamoto T, Sumiyoshi H, Kajihara H, Yamamoto M, (1982a) Argyrophil cells in early gastric carcinoma: An immunohistochemical and ultrastructural study. J Cancer Res Clin Oncol 103:187–202

Tahara E, Ito H, Nakagami K, Shimamoto F, Yamamoto M, Sumii K (1982b) Scirrhous argyrophil cell carcinoma of the stomach with multiple production of peptide hormones, amine, CEA, lysozyme and HCG. Cancer 49:1904–1915

Thim L, Moody AJ (1981) The primary structure of porcine glicentin (proglucagon). Reg Pept 2:139–150

Tsutsumi Y, Osamura RY, Nagura H, Watanabe K, Yanaihara N (1983) Immunohistochemical studies on gastrointestinal hormones in the intestinal metaplasia of the stomach. In: Miyoshi A (ed) Gut peptides and ulcer. Biochemical Research Foundation, Tokyo pp 171–179

WHO International Reference Centre for the Histological Classification of Gastro-oesophageal Tumors (1977) World Health Organization, Geneva Watanabe H (1972) Argentaffin cells in adenoma of the stomach. Cancer 30:1267–1274

Watanabe H (1974) Argentaffin cells in the non-neoplastic mucosa, adenoma and carcinoma of the stomach. Jap J Cancer Clin 20:519–535

Yanaihara N (1980) Immunochemical application of synthetic peptides to studies of the prohormone-hormone system. Biomed Res 1:105–116

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