IMMUNOFLUORESCENT STAINING OF THE GASTRIC
MUCOSA-SPECIFIC EPITHELIAL GLYCOPROTEIN IN
METASTASIZED GASTRIC CANCER CELLS ; A CASE
REPORT OF KRUKENBERG TUMOR

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Previous immunofluorescent studies (1) revealed that the gastric mucosa-specific epithelial glycoprotein distributed in mucoid cells and secreted mucus in the normal gastric mucosa as well as even in the diffusely and invasively proliferated gastric cancer cells in the mucosal, submucosal and muscular layer.

Krukenberg tumor, which metastasizes frequently not only to the ovary but also to other various organs, is generally characterized by mucin-producing epithelial cells having a signet ring form. Its primary tumor is recognized to be most often in the gastrointestinal tract (2-5).

The present investigation dealt with the immunofluorescent staining of such cancer cells which metastasized to different organs apart from the primary gastric lesion.

CASE REPORT

Clinical course :

Y. T., a 35 year old married woman, was admitted to Kurume Daiichi Hospital, Kurume city, on January 13, 1970, with the chief complaint of epigastric pain and abdominal inflation.

She was poorly nourished and appeared to be anemic. Palpable epigastric mass was recognized. Roentgen examination of the stomach (Fig. 1) revealed a scirrhus-type carcinoma with an absence of regular peristalsis and a deformation of gastric contour and moreover with an infiltration into the duodenum region. There were no palpable masses for liver, spleen, kidney and others. There were no gynecological complaints.

The ascites was bloody and showed a Class V in a Papanicolaou stain. And therefore the peritoneal implant was estimated to be widespread. On the diagnosis of inoperable gastric carcinoma, the patient was continued to receive only a conservative treatment.

Fig. 1 Stomach X-ray

Fig. 2 Chest X-ray

A few months following admission, the patient noticed an increasing impairment of appetite, epigastric pain and abdominal inflation. With an extreme elevation of serum alkali phosphatase value, there was the onset of jaundice. In this period, chest X-ray (Fig. 2) demonstrated the acute diffuse miliary lesions in the lung. Regurgitation of food took place and increasing dysphagia developed. Diarrhoea was severe.

On May 9, the patient was dead.

Pathological findings :

I. Macroscopic observations

Stomach and adjacent organs : More than half of the stomach wall was

occupied with tumor mass extending from pylorus up to corpus ventriculi (Fig. 3). And also there was the infiltration to duodenum and pancreas. Vater papille of the duodenum was invaded with cancer cells, consisting a cause of obstructive jaundice. Omentum and lymph nodes adjacent to the stomach were intensely metastasized.

Fig. 3 Stomach and adjacent organs

Fig. 4 Right ovary

Ovary : Right ovary (Fig. 4) was enlarged up to 8 cm in diameter, being solid in consistency with a cystic, hemorrhagic or gelatinous area. The external surface was smooth with firm capsules and with no tendency to become adherent to the surrounding structures. Left ovary was also enlarged to the hen's egg size.

Rectum : It was adherent to the uterine mass posteriorly. The rectal wall was tough and its mucosa was edematous and rugosed.

Lung and liver : On their cut surfaces, a large number of metastasized small tumor masses was recognized in the lung and a few in the liver.

II. Microscopic observations

Stomach, HE stain (Fig. 5-a) and PAS-alcian blue stain (Fig. 5-b), showing the characteristic signet ring cells proliferated diffusely from the mucosa through the muscular layer. They were stainable with PAS or alcian blue and partially with the both. And the mucoid liquefaction was remarkable. According to "The general rules for the gastric cancer study in surgery and pathology" (6), it was classified to be adenocarcinoma of tubular and mucocellulo-nodular type.

Ovary (Fig. 5-c), showing clearly the large polyhedral and rounded cells with mucoid contents pushing the nuclei to one side, giving the signet ring appearance as well as an accumulation of mucoid ground substances.

Rectum (Fig. 5-d), liver (Fig. 5-e) and lung (Fig. 5-f), showing the metastasized and scattered signet ring carcinomatous cells. The mucoid ground substances were also recognizable.

Fig. 5-a Stomach (H. E.) *Fig. 5-b* Stomach (PAS-alcian blue) *Fig. 5-c* Ovary (H. E.) *Fig. 5-d* Rectum (H. E.) *Fig. 5-e* Liver (H. E.) *Fig. 5-f* Lung (H. E.)

From the above-described pathological observations, it was diagnosed to be Krukenberg tumor with a primary gastric cancer.

Immunofluorescent studies :

Rabbit specific antisera against the epithelial glycoprotein (blood-group active) of human gastric mucosa were prepared and labeled with fluorescein isothiocyanate as described in the preceding paper (1). Before use, the FITC-

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labeled antisera were absorbed with lyophilized human serum, liver powder and also with the ethylene glycol extract of normal intestinal mucosa.

Immunofluorescent tissue sections of the primary gastric cancer and various metastasized organs, as shown in color photos, demonstrated an intensely fluoresced cancer cells especially of signet ring form, correlating well with PAS staining.

COLOR PHOTOS

1. Stomach, cancer cells in mucosal (above left) and submucosal (below right) layer.
2. Stomach, cancer cells in muscular (below left) and submucosal (above right) layer.
3. Ovary, metastasized cancer cells.
4. Rectum, metastasized cancer cells (above left) and apparently normal rectal mucosa (below right).
5. Lymph node in a neighbourhood of the stomach, metastasized cancer cells.
6. Omentum, metastasized cancer cells.
7. Liver, metastasized cancer cells (above right) and apparently normal liver structure (bottom left).
8. Lung, metastasized cancer cells.

DISCUSSION

In the study of Krukenberg tumor there had been two pathological questions that attract attention; First, is this tumor primary in the ovary or is it secondary to some gastrointestinal carcinoma? Second, how can one explain the combination of malignant connective tissue and epithelial areas? A number of pathologists (2-5) who had been consulted regarding this question were unable to give the definite decision in these respects.

The attempt of this experiment was to offer a precise procedure determining the primary lesion in such a widely metastasized cancer from the immunochemical standpoint. This idea was based on the persistency of the same or cross-reacted antigenic epithelial glycoprotein in the undifferentiated gastric cancer cells, as demonstrated in a previous paper (1). And also the characteristic signet ring cells possessing a high content of mucoid materials was considered to give a favorable example for such a mucoid staining.

The results revealed that the cancer cells in the stomach as well as other different organs including omentum, ovary, rectum, lung and liver were intensely stainable with the FITC-labeled antibody, though the normal areas of the latter organs were scarcely fluoresced. It might, therefore, be deduced that such metastasized cancer cells in various organs apart from the primary gastric lesion persisted in producing or secreting the epithelial glycoprotein specific

for the gastric mucosa.

Such a fluorescent antibody technic might give one of the interesting procedures for determining the primary lesion in some kinds of cancerous metastasis.

SUMMARY

Immunofluorescent staining of the gastric mucosa-specific epithelial glycoprotein was carried out on the metastasized cancer cells in a case of Krukenberg tumor with a primary gastric cancer.

The cancer cells, especially of signet ring form, in the stomach as well as other metastasized organs including omentum, ovary, rectum, lung and liver were found to be intensely stainable with the FITC-labeled antibody, though the normal areas of the latter organs were scarcely fluoresced. It might, therefore, be deduced that such metastasized cancer cells in various organs apart from the primary gastric lesion persisted in producing or secreting the epithelial glycoprotein specific for the gastric mucosa.

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