and/or death. However, a recent study using methionine choline-deficient diet-fed mice demonstrated that an increase in serum ALT levels in NASH was attributable not to the enhancement of hepatocyte necrosis/apoptosis or hepatitis, but rather to the induction of hepatic ALT mRNA levels,[37] and a clear discrepancy between the changes in NAS and those in serum ALT levels has been observed in another clinical trial for NASH.[38] Our findings corroborate the idea that serum ALT levels are not always a reliable biomarker of NAFLD activity. Thus, we would like to propose determination of serum fragmented CK18 levels along with ALT levels for accurate assessment of therapeutic response in future NAFLD/NASH studies.

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In conclusion, this study established the usefulness of serum fragmented CK18 as a noninvasive, simple, and reliable biomarker that may complement the shortcomings of liver biopsy and serum ALT measurement. Although the number of patients in our longitudinal analysis was small, we believe that measurement of serum fragmented CK18 levels is a promising tool for monitoring disease status and assessing treatment response in patients with NAFLD. To confirm our observations, larger validation analyses and longitudinal prospective studies are needed.

## ACKNOWLEDGMENT

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TABLE 5. Characteristics of 20 Patients who Received a Follow-up Biopsy

| Characteristics | Values |
|---|---|
| Age (y) | 61 ± 15 |
| Female | 13 (65%) |
| NAS at first biopsy | 5.3 ± 1.6 |
| Interval between biopsies (y) | 2.9 ± 1.4 |
| Intervention | |
| Lifestyle correction | 8 (40%) |
| Treatment with highly purified EPA | 8 (40%) |
| Treatment with pioglitazone | 3 (15%) |
| Treatment with bezafibrate | 1 (5%) |
| Changes in NAS | |
| Improved | 13 (65%) |
| Unchanged | 0 (0%) |
| Deteriorated | 7 (35%) |

EPA indicates eicosapentaenoic acid; NAS, NAFLD activity score.

Platelet count, serum AST levels, and HOMA-IR values were also correlated with the stage of fibrosis, suggesting that these parameters may reflect the progression of fibrosis in NAFLD as well. Differently from a report by Diab et al,[25] fragmented CK18 levels did not show a meaningful association with the stage of fibrosis in the current analysis. This disagreement might be explained by the different proportion of patients with advanced fibrosis (3% in Diab et als' study vs. 27% in the present study).

It is noteworthy that fragmented CK18 levels correlated with NAS more strongly than ALT levels. More importantly, changes in NAS were mirrored by those in fragmented CK18 levels, but not by those in ALT levels. To our knowledge, this is the first trial to demonstrate fragmented CK18 as a potentially superior biomarker for evaluating NAS. Serum ALT levels are conventionally believed to be a surrogate biomarker of hepatocyte injury

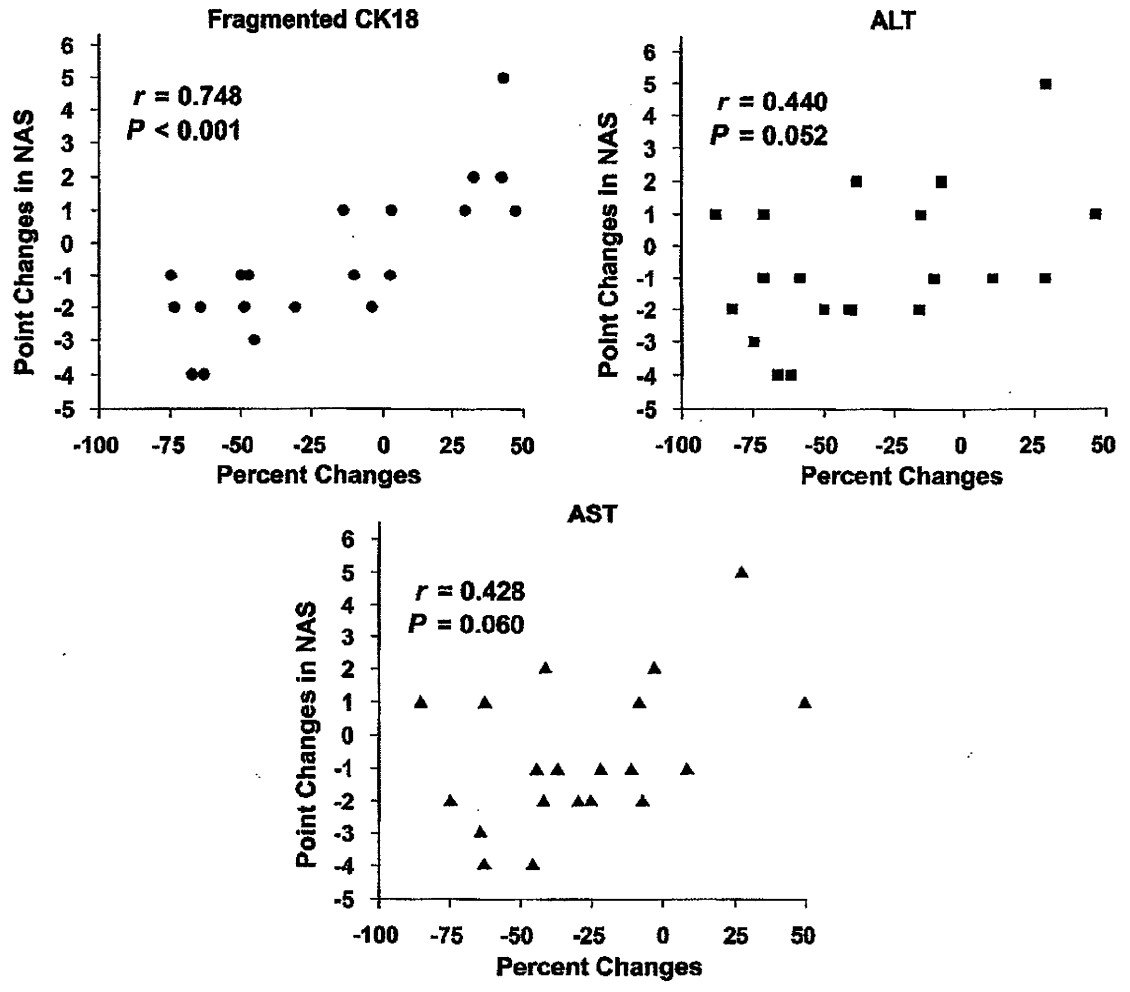


FIGURE 2. Correlations between changes in NAS and those in serum fragmented CK18, AST, and ALT levels in 20 NAFLD patients who received follow-up biopsies. Correlation coefficients were calculated using Spearman rank correlation analysis. ALT indicates alanine aminotransferase; AST, aspartate aminotransferase; NAFLD, nonalcoholic fatty liver disease; NAS, NAFLD histologic activity score.

and/or death. However, a recent study using methionine choline-deficient diet-fed mice demonstrated that an increase in serum ALT levels in NASH was attributable not to the enhancement of hepatocyte necrosis/apoptosis or hepatitis, but rather to the induction of hepatic ALT mRNA levels,[37] and a clear discrepancy between the changes in NAS and those in serum ALT levels has been observed in another clinical trial for NASH.[38] Our findings corroborate the idea that serum ALT levels are not always a reliable biomarker of NAFLD activity. Thus, we would like to propose determination of serum fragmented CK18 levels along with ALT levels for accurate assessment of therapeutic response in future NAFLD/NASH studies.

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In conclusion, this study established the usefulness of serum fragmented CK18 as a noninvasive, simple, and reliable biomarker that may complement the shortcomings of liver biopsy and serum ALT measurement. Although the number of patients in our longitudinal analysis was small, we believe that measurement of serum fragmented CK18 levels is a promising tool for monitoring disease status and assessing treatment response in patients with NAFLD. To confirm our observations, larger validation analyses and longitudinal prospective studies are needed.

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# Chronic Gastritis in the Setting of Autoimmune Pancreatitis

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**Abstract:** Autoimmune pancreatitis (AIP) is a recently recognized disease entity. In some patients, this disease is associated with other inflammatory diseases. In this study, we aimed to elucidate the pathologic characteristics of AIP-associated gastritis (AIP-G). We evaluated and compared the pathologic findings and immunohistochemical expressions of immunoglobulin G (IgG)4 and IgG in gastric biopsy specimens from 13 AIP-G patients with those from patients of 2 control groups. We divided the AIP-G patients who did not receive steroid therapy [AIP-G-ST(−)] into the following 2 groups: without *Helicobacter pylori* (HP) infection [AIP-G-HP(−)] and with HP infection [AIP-G-HP(+)]. The control groups comprised 19 patients who were diagnosed with chronic active gastritis associated with HP infection and 7 patients with nonsteroidal anti-inflammatory drug-induced gastritis. We classified the findings for the gastric mucosa into those for the upper and the lower lamina propria. The characteristic finding of AIP-G groups was diffusely lymphoplasmacytic infiltration in the lamina propria. The IgG4-positive plasma cell/IgG-positive plasma cell ratios (IgG4/IgG ratios) in both the upper and lower lamina propria in the AIP-G-ST(−) groups were predominantly higher than the corresponding values in the other groups. In the AIP-G-ST(−) groups, the IgG4/IgG ratio in the lower lamina propria was predominantly higher than that in the upper lamina propria, irrespective of the HP status. In conclusion, diffuse lymphoplasmacytic infiltration in the lamina propria and increased IgG4/IgG ratio in the gastric mucosa (notably in the lower lamina propria) may be the characteristic findings of AIP-G.

**Key Words:** autoimmune pancreatitis, IgG4, gastritis

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The relationship between immunoglobulin G (IgG)4 and sclerosing disease with regard to autoimmune pancreatitis (AIP) was first reported in 2001.[6] AIP, also known as lymphoplasmacytic sclerosing pancreatitis, is a unique form of pancreatitis characterized by the following findings: (1) irregular narrowing of the main pancreatic duct because of fibrosis and lymphoplasmacytic infiltration and an increased number of IgG4-positive plasma cells and eosinophils in the pancreas; (2) hypergammaglobulinemia with a predominant increase in the IgG4 levels; and (3) alleviation of symptoms after steroid therapy.[9,14,32] AIP is now recognized as a distinct entity worldwide.[15,17,22]

AIP has also been reported to be associated with various diseases in organs other than those in the pancreaticobiliary system[2,10,17,25]; these diseases include chronic sialadenitis, retroperitoneal inflammation,[7,11,27] lymphadenopathy of the intra-abdominal, hilar, or mediastinal lymph nodes,[18] interstitial pneumonia,[8,24] pulmonary inflammatory pseudotumor,[28,30] hepatic inflammatory pseudotumor,[12,19,29] inflammatory abdominal aortic aneurysm,[13] prostatitis,[26] and tubulointerstitial nephritis.[23,24] Further, Kamisawa et al[10] have proposed that AIP is the pancreatic lesion involved in a new clinicopathologic entity, namely, IgG4-related sclerosing disease.

Many cases of chronic active gastritis associated with *Helicobacter pylori* (HP) infection (CAG) have been reported in Japan.[5] Few cases of gastritis not associated with HP infection have also been reported.[16] Recently, Shinji et al[21] found that the incidence of IgG4-positive plasma cells in gastric biopsy specimens from patients with AIP was significantly higher than that in specimens from patients with CAG. However, the distribution of IgG4-positive plasma cells and the characteristic histopathologic findings of AIP-associated gastritis (AIP-G) have not been reported. To elucidate the clinicopathologic characteristics of AIP-G, we evaluated the clinicopathologic findings of patients with AIP-G and compared the immunohistochemical expression of IgG4 in the AIP-G patients with that in the control groups, which included patients with abdominal distress.

## MATERIALS AND METHODS

### Patients and Materials

Between 2002 and 2009, we analyzed 13 AIP-G (Table 1) and 26 control patients at the Shinshu University

**TABLE 1.** Clinical Features of Autoimmune Pancreatitis-associated Gastritis

| Case No. | Age (y) | Sex | Pathologic Diagnosis | Biopsy Area | HP | Serum IgG4 (mg/dL)* | Endoscopic Finding | Symptom |
|---|---|---|---|---|---|---|---|---|
| 1 | 79 | M | CG | FA | Negative | 1950 | Redness | EG |
| 2 | 53 | F | CG | PA, FA | Negative | 227 | NP | NP |
| 3 | 59 | M | CG | PA, FA | Negative | 965 | Erosion | NP |
| 4 | 55 | M | CG | FA | Negative | 2970 | NP | NP |
| 5 | 76 | F | CG | PA | Negative | 420 | Erosion | NP |
| 6 | 56 | M | CAG | FA | Positive | 156 | NP | NP |
| 7 | 62 | M | CAG | PA, FA | Positive | 305 | Atrophy | NV |
| 8 | 60 | M | CAG | FA | Positive | 1240 | NP | NP |
| 9 | 62 | M | CAG | PA, FA | Positive | 825 | NP | NP |
| 10 | 78 | M | CAG | FA | Positive | 541 | NP | NP |
| 11 | 76 | M | CAG | PA, FA | Positive | 698 | Redness | NP |
| 12 | 56 | M | CAG | PA, FA | Positive | 265 | NP | EG |
| 13 | 67 | M | CAG | PA, FA | Positive | 500 | NP | EG |

*Normal value: IgG4, < 70 mg/dL (cut-off value, 135 mg/dL).
CAG indicates chronic active gastritis with HP infection; CG, chronic gastritis; EG, epigastralgia; F, female; FA, fundic area; HP, *Helicobacter pylori*; IgG, immunoglobulin G; M, male; NP, nothing particular; NV, nausea and vomiting; PA, pyloric area.

Hospital. Of the 13 API-G patients, 9 had no symptoms, 3 had epigastralgia (patients 1, 12, and 13), and 1 had nausea and vomiting (patient 7).

All the patients with AIP-G showed the following characteristic findings of AIP: (1) irregular narrowing of the main pancreatic duct, (2) sonolucent swelling of the pancreas, (3) hypergammaglobulinemia, and (4) high serum IgG4 level (cut-off value, 135 mg/dL). All the patients with AIP-G underwent enhanced abdominal computed tomography (CT) and Gallium (Ga)-67 citrate scintigraphy for the pancreatic lesions. One patient (patient 1) underwent pancreatoduodenectomy. The patient was diagnosed as having AIP, because of the presence of lymphoplasmacytic sclerosing pancreatitis without neutrophil infiltration and cancer. The other AIP-G patients did not undergo any operation for the pancreatic lesions and biopsy for suspected pancreatic cancer.

The histologic specimens obtained from the 13 AIP-G patients included biopsy specimens. The stomach biopsy specimens obtained from 7 patients (patients 2, 3, 7, 9, 11, 12, and 13) included specimens from both the fundic and pyloric areas, those from the other 5 patients (patients 1, 4, 6, 8, and 10) included specimens only from the fundic area, and those from the remaining 1 patient (patient 5) included specimens only from the pyloric area. In 4 patients (patients 2, 3, 11, and 13), the specimens were obtained from 6 biopsy points. HP infection was detected only in the specimens from 8 of the 13 patients (patients 6, 7, 8, 9, 10, 11, 12, and 13). Of the 8 AIP-G patients who had HP infection, 2 developed the infection after steroid therapy (patient 12 and 13). Patient 1 was receiving proton pump inhibitor therapy, although he was free of HP infection. First, we divided the AIP-G patients into the following 2 groups: steroid nontreated group [AIP-G-ST(−)] and steroid treated group [AIP-G-ST(+)]. We further divided the AIP-G-ST(−) patients into the following 2 groups: without HP infection [AIP-G-HP(−)] and with HP infection [AIP-G-HP(+)].

The control groups were divided into the following 2 groups: CAG and nonsteroidal anti-inflammatory drug (NSAID)-induced gastritis (NSAID-G). The CAG group comprised 19 patients who were clinically diagnosed with chronic active gastritis caused by HP infection. The NSAID-G group comprised 7 patients with no HP infection. None of the patients in the control groups showed the characteristic findings of AIP.

## Histopathology and Immunohistochemistry

All the specimens from the 39 patients were fixed in 20% formaldehyde and embedded in paraffin. Next, 4-μm-thick serial sections were cut from these blocks and stained with hematoxylin-eosin (HE). The sections were treated with rabbit monoclonal antibodies against human IgG (Dako, Glostrup, Denmark; dilution, 1:5000) and affinity-purified sheep polyclonal antibody against human IgG4 (The Binding Site, Birmingham, UK; dilution, 1:50) by using the avidin-biotin immunoperoxidase method. The number of immunohistochemically identified IgG4-positive plasma cells and IgG-positive plasma cells in 10 randomly selected 10000-$\mu m^2$-wide areas of the gastric mucosa specimens of the upper and lower lamina propria were counted. The fundic and pyloric mucosae were used for the assessments. The IgG4-positive plasma cell/IgG-positive plasma cell ratio (IgG4/IgG ratio) of the AIP-G group was compared with those of the control groups.

The number of mononuclear cells in the gastric mucosa in 10 randomly selected 10000-$\mu m^2$-wide areas of each specimen was also counted. We compared the number of mononuclear cells in the AIP-G group with those in the control group.

Mucosal neutrophilic inflammation, intestinal metaplasia, and glandular atrophy were separately graded as none (0), mild (1+), moderate (2+), or marked (3+), in accordance with the schematic diagrams provided in the Update Sydney System.[3] Eosinophil infiltration was assessed by 2 pathologists (T.U. and H.O.) and graded as

follows: absent or minimal (0), mild (1+), moderate (2+), and severe (3+).

The data are expressed as the median (25th to 75th percentile).

Comparisons were made using the Mann-Whitney *U* test. Differences were considered statistically significant when the *P* value was less than 0.05.

This study was approved by the Ethics Committee of Shinshu University, Japan.

## RESULTS

### Clinical Findings

Gastric biopsies revealed that, of the 13 patients with AIP-G, 5 had chronic gastritis and 8 had chronic active gastritis. An endoscopic examination revealed that 2 patients had redness, 2 patients had erosion, 1 patient had atrophy, and 8 patients had no abnormal findings. In patient 6, an enhanced abdominal CT image showed thickening of the stomach wall (Fig. 1A), and Ga-67 citrate scintigraphy revealed accumulation of radioactive material on the stomach wall, which was consistent with the findings of the CT analysis (Fig. 1B). Ga-67 citrate scintigraphy did not reveal such accumulations on the stomach walls of other AIP-G patients.

The serum IgG4 levels were not measured in the 26 patients in the control group because there was no evidence of AIP in these patients.

### Histologic Findings

We have summarized the histologic characteristics of the AIP-G patients in Tables 2 to 5. Some degree of diffuse lymphoplasmacytic inflammation was observed in the fundic and the pyloric mucosae of all the AIP-G patients (Figs. 2A, B).

In the AIP-G-ST(−) groups, in the fundic mucosa, the number of mononuclear cell infiltrates in the upper lamina propria was not significantly different from that in the lower lamina propria. The number of mononuclear cell infiltrates in the upper lamina propria in the AIP-G-HP(+) group was significantly higher than those in the AIP-G-HP(−) and NSAID-G groups. However, the number of mononuclear cell infiltrates in the upper lamina propria in the AIP-G-HP(−) groups was significantly lower than that in the CAG group. The number of mononuclear cell infiltrates in the lower lamina propria in the AIP-G-ST(−) groups was significantly higher than those in the CAG and NSAID-G groups.

In the AIP-G-ST(−) groups, in the pyloric mucosa, the number of mononuclear cell infiltrates in the upper lamina propria was not significantly different from that in the lower lamina propria. The number of mononuclear cell infiltrates in the upper lamina propria in the AIP-G-HP(+) group was significantly higher than those in the AIP-G-HP(−) and NSAID-G groups. However, the number of mononuclear cell infiltrates in the upper lamina propria in the AIP-G-HP(−) group was significantly lower than that in the CAG group. The number of mononuclear cell infiltrates in the lower lamina propria in the AIP-G-HP(−) groups was higher than that in the NSAID-G group. Further, the number of mononuclear cell infiltrations in the lower lamina propria in the AIP-G-HP(+) group was significantly higher than that in the NSAID-G group.

In the AIP-G-ST(−) groups, the number of mononuclear cell infiltrates in the upper and lower lamina

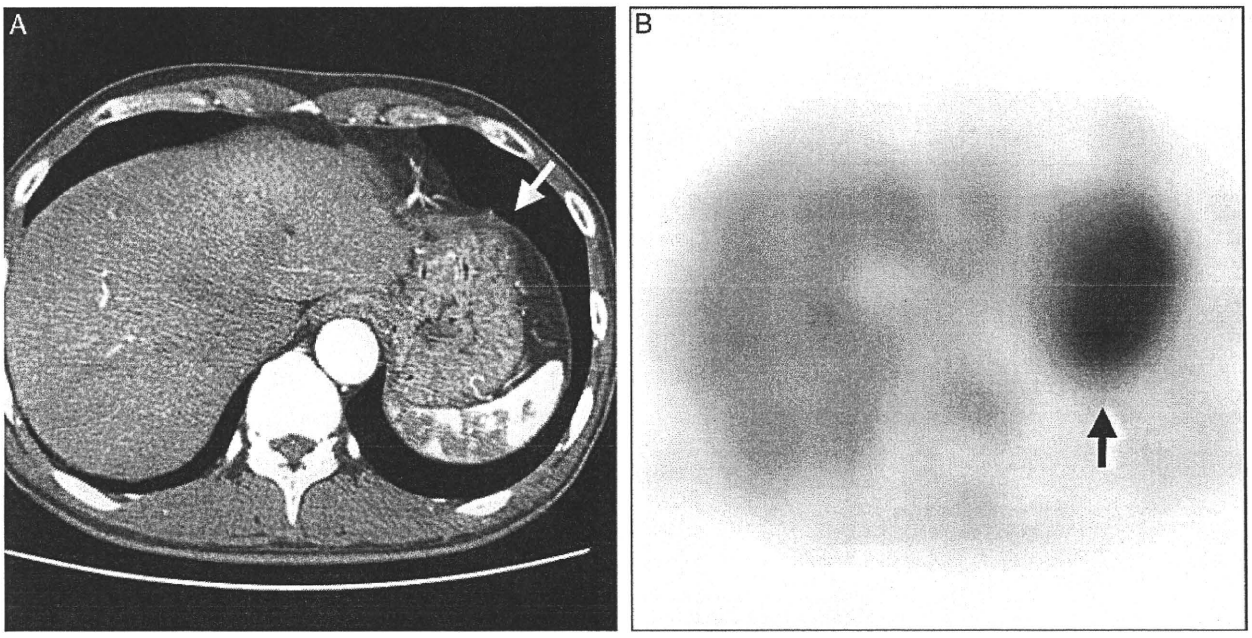


**FIGURE 1.** A, Enhanced abdominal CT scan shows thickening of the stomach wall (arrow). B, Gallium-67 citrate scintigraphy revealed accumulation of radioactive material on the stomach wall (arrow), which was consistent with the findings of the CT analysis. CT indicates computed tomography.

**TABLE 2.** Histologic Findings of the Fundic Mucosa From Patients With AIP-G-HP(−), AIP-G-HP(+), CAG, and NSAID-G

| | Mono | | | IgG4/IgG | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | Upper | Lower | *P* | Upper | Lower | *P* | Eosino | Neutro | IM | Atrophy |
| AIP-G-HP(−) (n = 4) | 14.50 (12.00-17.85) | 26.55 (11.20-39.30) | 0.3865 | 0.1 (0.05-0.27) | 0.76 (0.43-0.82) | 0.0433 | 2 (2-2) | 0 (0-0) | 0 (0-1.0) | 0.5 (0-1.5) |
| AIP-G-HP(+) (n = 6) | 38.75 (29.70-43.70) | 33.10 (28.50-37.50) | 0.4233 | 0.1 (0.03-0.19) | 0.34 (0.17-0.51) | 0.0547 | 2 (2-3) | 1 (1-2) | 0 (0-0) | 1 (1-1) |
| CAG (n = 19) | 29.20 (18.75-38.58) | 4.40 (3.13-5.35) | < 0.0001 | 0 (0-0.11) | 0 (0-0) | 0.4221 | 1 (1-1) | 2 (1-2) | 0 (0-0) | 1 (0.25-1) |
| NSAID-G (n = 7) | 6.50 (4.33-13.45) | 0.40 (0.23-2.45) | 0.0022 | 0 (0-0.01) | 0 (0-0) | 0.7494 | 0 (0-0) | 0 ( 0-0.75) | 0 (0-0) | 0 (0-0) |
| *P* | 0.019* | 0.0082† | | 0.0082† | 0.0140† | | 0.0082† | 0.0105* | NS | 0.0027‡ |
| | 0.0034‡ | 0.0027‡ | | 0.0184‡ | 0.010‡ | | 0.0034‡ | 0.0152‡ | | |
| | 0.0074§ | 0.0058§ | | | 0.0035§ | | 0.0231§ | 0.0021§ | | |
| | | 0.0003‖ | | | 0.0012‖ | | 0.0186‖ | | | |

Data are expressed as the median (25th to 75th percentile).
*AIP-G-HP(−) versus AIP-G-HP(+).
§AIP-G-HP(−) versus CAG.
†AIP-G-HP(−) versus NSAID-G.
‖AIP-G-HP(+) versus CAG.
‡AIP-G-HP(+) versus NSAID-G.
AIP-G indicates autoimmune pancreatitis-associated gastritis; AIP-G-HP(−), AIP-G without steroid therapy and HP infection; AIP-G-HP(+), AIP-G without steroid therapy and with HP infection; CAG, chronic active gastritis with HP infection; Eosino, eosinophil infiltration; HP, *Helicobacter pylori*; IgG, immunoglobulin G; IgG4/IgG, IgG4-positive plasma cells/IgG-positive plasma cells ratio; IM, intestinal metaplasia; Lower, in the lower lamina propria; Mono, number of mononuclear cells; Neutro, neutrophil infiltration; NS, not significant; NSAID-G, nonsteroidal anti-inflammatory drug-induced gastritis; Upper, in the upper lamina propria.

**TABLE 3.** Histologic Findings of Pyloric Mucosa From Patients With AIP-G-HP(−), AIP-G-HP(+), CAG, and NSAID-G

| | Mono | | | IgG4/IgG | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | Upper | Lower | *P* | Upper | Lower | *P* | Eosino | Neutro | IM | Atrophy |
| AIP-G-HP(−) (n = 3) | 20.10 (16.20-21.60) | 22.50 (20.80-34.50) | 0.5127 | 0.17 (0.10-0.25) | 0.37 (0.33-0.66) | 0.0495 | 2 (2-2.75) | 0 (0-0) | 1 (0.25-1) | 1 (1-1) |
| AIP-G-HP(+) (n = 3) | 46.30 (43.83-73.15) | 22.80 (21.30-38.95) | 0.1266 | 0.06 (0.04-0.12) | 0.22 (0.18-0.23) | 0.0495 | 2 (2-2) | 2 (2-2) | 0 (0-0.75) | 0 (0-0.75) |
| CAG (n = 19) | 45.00 (38.78-71.48) | 22.70 (15.15-29.85) | < 0.0001 | 0 (0-0.02) | 0 (0-0.03) | 0.9651 | 1 (1-1) | 2 (1-2) | 1 (0-2) | 2 (2-3) |
| NSAIDs-G (n = 7) | 6.50 (5.38-21.65) | 4.60 (2.58-15.10) | 0.2774 | 0 (0-0.02) | 0 (0-0) | 0.3711 | 0 (0-0) | 0 (0-0.0) | 0 (0-0.75) | 0 (0-1) |
| *P* | 0.0495* | 0.0167† | | 0.0167‡ | 0.0495* | | 0.0167‡ | 0.0495* | NS | 0.0495* |
| | 0.0167† | | | 0.0167† | 0.0304‡ | | 0.0167† | 0.0167† | | 0.0304† |
| | 0.0064§ | | | 0.0064§ | 0.0064§ | | 0.0147§ | 0.0064§ | | 0.0313§ |
| | | | | 0.0064‖ | 0.0129‖ | | 0.0217‖ | | | |

Data are expressed as the median (25th to 75th percentile).
*AIP-G-HP(−) versus AIP-G-HP(+).
§AIP-G-HP(−) versus CAG.
‡AIP-G-HP(−) versus NSAID-G.
‖AIP-G-HP(+) versus CAG.
†AIP-G-HP(+) versus NSAID-G.
AIP-G indicates autoimmune pancreatitis-associated gastritis; AIP-G-HP(−), AIP-G without steroid therapy and HP infection; AIP-G-HP(+), AIP-G without steroid therapy and with HP infection; CAG, chronic active gastritis with HP infection; Eosino, eosinophil infiltration; HP, *Helicobacter pylori*; IgG, immunoglobulin G; IgG4/IgG, IgG4-positive plasma cells/IgG-positive plasma cells ratio; IM, intestinal metaplasia; Lower, in the lower lamina propria; Mono, number of mononuclear cells; Neutro, neutrophil infiltration; NS, not significant; NSAID-G, nonsteroidal anti-inflammatory drug-induced gastritis; Upper, in the upper lamina propria.

**TABLE 4.** Comparison of Histologic Features Between the Fundic and Pyloric Mucosa in AIP-G-HP(−) Patients

| | Fundic Mucosa (n = 4) | Pyloric Mucosa (n = 3) | *P* |
|---|---|---|---|
| Mono (upper) | 14.50 (12.00-17.85) | 20.10 (16.20-21.60) | 0.1573 |
| Mono (lower) | 26.55 (11.20-39.30) | 22.50 (20.80-34.50) | 0.7237 |
| IgG4/IgG (upper) | 0.1 (0.05-0.27) | 0.17 (0.10-0.25) | 0.4795 |
| IgG4/IgG (lower) | 0.76 (0.43-0.82) | 0.37 (0.33-0.66) | 0.4795 |
| Eosino | 2 (2-2) | 2 (2-2.75) | 0.4795 |
| Neutro | 0 (0-0) | 0 (0-0) | > 0.9999 |
| IM | 0 (0-1.0) | 1 (0.25-1) | 0.5959 |
| Atrophy | 0.5 (0-1.5) | 1 (1-1) | 0.5959 |

Data are expressed as the median (25th to 75th percentile).

AIP-G indicates autoimmune pancreatitis-associated gastritis; AIP-G-HP(−), AIP-G without steroid therapy and *Helicobacter pylori* infection; Eosino, eosinophil infiltration; IgG, immunoglobulin G; IgG4/IgG, IgG4-positive plasma cells/IgG-positive plasma cells ratio; IM, intestinal metaplasia; Lower, in the lower lamina propria; Mono, number of mononuclear cells; Neutro, neutrophil infiltration; Upper, in the upper lamina propria.

propria were not significantly different between the fundic and the pyloric mucosa. In the specimens obtained from 6 biopsy points in 3 patients of the AIP-G-ST(−) group, the distribution of the number of mononuclear cell infiltrates in every case was approximately uniform in the fundic and the pyloric mucosa.

In the AIP-G-ST(+) group, the number of mononuclear cell infiltrates in the fundic mucosa were 12.7 and 10.4 in the upper lamina propria, and 7.0 and 9.1 in the lower lamina propria; and that in the pyloric mucosa were 25.2 and 10.5 in the upper lamina propria, and 16.2 and 3.2 in the lower lamina propria. In the fundic and the pyloric mucosa, the number of mononuclear cell infiltrates in the upper lamina propria was higher than that in the lower lamina propria. In the specimens obtained from 6 biopsy points of 1 patient of the AIP-G-ST(+) group, the distribution of the number of mononuclear cell infiltrates was approximately uniform in the fundic and the pyloric mucosa.

All the AIP-G patients showed mild-to-severe eosinophil infiltration in the fundic mucosa (Fig. 2B). Among the 12 patients with AIP-G, 10 showed mild-to-moderate atrophy and 8 showed mild-to-moderate neutrophilic inflammation; however, none of the patients in the AIP-G-HP(−) group showed neutrophilic inflammation. Moderate intestinal metaplasia was observed in 1 patient in each of the AIP-G-HP(−), AIP-G-HP(+), and AIP-G-ST(+) groups. Patient 6 showed severe fibrosis in the lower lamina propria (Figs. 3A, B). Eosinophil infiltrates in the AIP-G-ST(−) groups were significantly higher than those in the CAG and NSAID-G groups. The number of neutrophil infiltrates in the AIP-G-HP(−) group was significantly lower than those in the AIP-G-HP(+) and CAG groups. The number of neutrophil infiltrates and the degree of atrophy in the AIP-G-HP(+) group were significantly higher than those in the NSAID-G group.

All the AIP-G patients showed moderate-to-severe eosinophil infiltration and mild-to-severe atrophy in the pyloric mucosa. Among the patients in the AIP-G-ST(−) groups, 3 showed mild metaplasia and moderate neutrophilic inflammation; however, none of the AIP-G-HP(−) group patients showed neutrophilic inflammation. Eosinophil infiltrates in the AIP-G-ST(−) groups were significantly higher than those in the CAG and NSAID-G groups. The number of neutrophil infiltrates in the AIP-G-HP(−) group was significantly lower than those in the AIP-G-HP(+) and CAG groups. The number of neutrophil infiltrates in the AIP-G-HP(+) group was significantly higher than that in the NSAID-G group. The degree of atrophy in the AIP-G-HP(−) group was significantly lower than those in the AIP-G-HP(+) and CAG groups. The degree of atrophy in the AIP-G-HP(+) group was significantly higher than that in the NSAID-G group.

In the AIP-G-ST(−) groups, there was no significant difference in the number of eosinophil and neutrophil infiltrates, intestinal metaplasia, and atrophy between the fundic and pyloric mucosa. In the specimen obtained from 6 biopsy points in 3 patients of the AIP-G-ST(−) group, the distribution of inflammation in every case was approximately uniform in the fundic and pyloric mucosa.

In the AIP-G-ST(+) group, the number of eosinophil and neutrophil infiltrates, intestinal metaplasia, and atrophy in the fundic mucosa were mild, moderate, none and moderate, and mild and moderate, respectively. In that group, the number of eosinophil and neutrophil

**TABLE 5.** Comparison of Histologic Features Between the Fundic and Pyloric Mucosa in AIP-G-HP(+) Patients

| | Fundic Mucosa (n = 6) | Pyloric Mucosa (n = 3) | *P* |
|---|---|---|---|
| Mono (upper) | 38.75 (29.70-43.70) | 46.30 (43.825-73.150) | 0.1213 |
| Mono (lower) | 33.10 (28.50-37.50) | 22.80 (21.30-38.95) | 0.6056 |
| IgG4/IgG (upper) | 0.1 (0.03-0.19) | 0.06 (0.04-0.12) | 0.7237 |
| IgG4/IgG (lower) | 0.34 (0.17-0.51) | 0.22 (0.18-0.23) | 0.8597 |
| Eosino | 2 (2-3) | 2 (2-2) | 0.6985 |
| Neutro | 1 (1-2) | 2 (2-2) | 0.1213 |
| IM | 0 (0-0) | 0 (0-0.75) | 0.7963 |
| Atrophy | 1 (1-1) | 2 (2-2.75) | 0.0389 |

Data are expressed as the median (25th to 75th percentile).

AIP-G indicates autoimmune pancreatitis-associated gastritis; AIP-G-HP(+), AIP-G without steroid therapy and with *Helicobacter pylori* infection; Eosino, eosinophil infiltration; IgG, immunoglobulin G; IgG4/IgG, IgG4-positive plasma cells/IgG-positive plasma cells ratio; IM, intestinal metaplasia; Lower, in the lower lamina propria; Mono, number of mononuclear cells; Neutro, neutrophil infiltration; Upper, in the upper lamina propria.

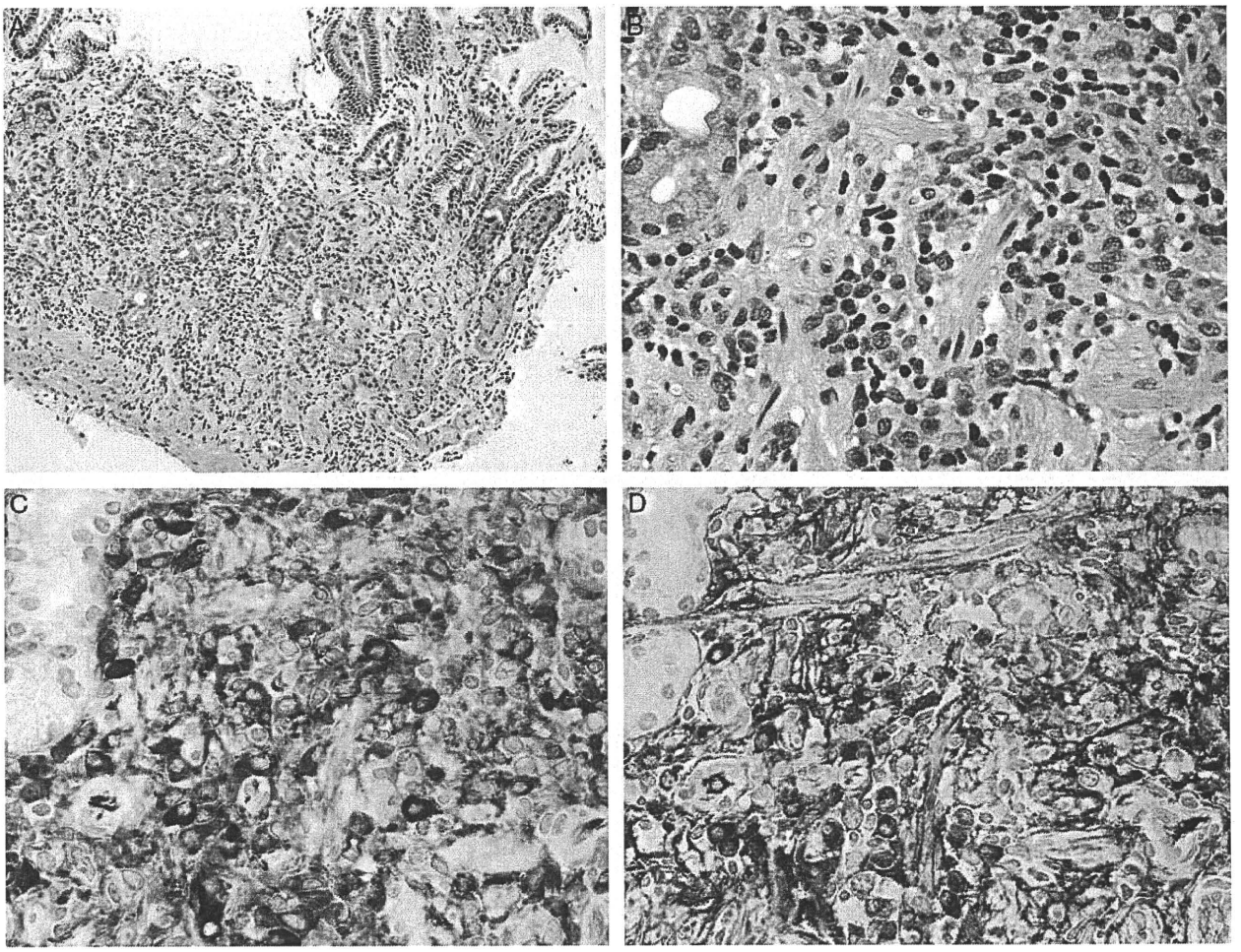


**FIGURE 2.** Stomach biopsy specimen of a patient with autoimmune pancreatitis-associated gastritis without steroid therapy and *Helicobacter pylori* infection. A, Inflammatory cell infiltration showing diffuse distribution in the fundic mucosa (HE staining, original magnification, 25 ×). B, Various degrees of lymphoplasmacytic and scattered eosinophil infiltration are found (HE staining, original magnification, 100 ×). Infiltration of (C) IgG-positive (immunostaining for IgG, original magnification, 100 ×) and (D) IgG4-positive plasma cells can be noted (immunostaining for IgG4, original magnification, 100 ×). HE indicates hematoxylin-eosin; IgG, immunoglobulin G.

infiltrates, intestinal metaplasia, and atrophy in the pyloric mucosa were mild and moderate, moderate, none and moderate, and mild and moderate, respectively. In the specimens obtained from 6 biopsy points of 1 patient of the AIP-G-ST(+) group, the distribution of inflammation was approximately uniform in the fundic and pyloric mucosa.

## Immunohistochemical Findings

In the fundic mucosa of the AIP-G-HP(−) group, the IgG4/IgG ratio in the lower lamina propria was significantly higher than that in the upper lamina propria (Figs. 2C, D). In the fundic mucosa of the AIP-G-HP(+) group, the IgG4/IgG ratio in the lower lamina propria was higher than that in the upper lamina propria (Figs. 3C, D); however, the difference was not statistically significant. The IgG4/IgG ratios for the upper and lower lamina propria were not significantly different between the AIP-G-ST(−) groups. The IgG4/IgG ratios in the upper lamina propria in the AIP-G-ST(−) groups were significantly higher than those in the NSAID-G group. The IgG4/IgG ratios in the lower lamina propria in the AIP-G-ST(−) groups were significantly higher than those in the CAG and NSAID-G groups.

In the pyloric mucosa, in the AIP-G-ST(−) groups, the IgG4/IgG ratio in the lower lamina propria was significantly higher than that in the upper lamina propria. There was no significant difference in the IgG4/IgG ratios in the lower and upper lamina propria between the AIP-G-ST(−) groups. The IgG4/IgG ratios in the upper and lower lamina propria in the AIP-G-HP(−) groups were significantly higher than those in the NSAID-G group. The IgG4/IgG ratio in the upper lamina propria in the AIP-G-HP(+) group was significantly higher than that in the NSAID-G group. In the AIP-G-HP(+) group, the IgG4/IgG ratio in the lower lamina propria was higher than that in the NSAID-G group; however, the difference was not statistically significant. The IgG4/IgG ratios in

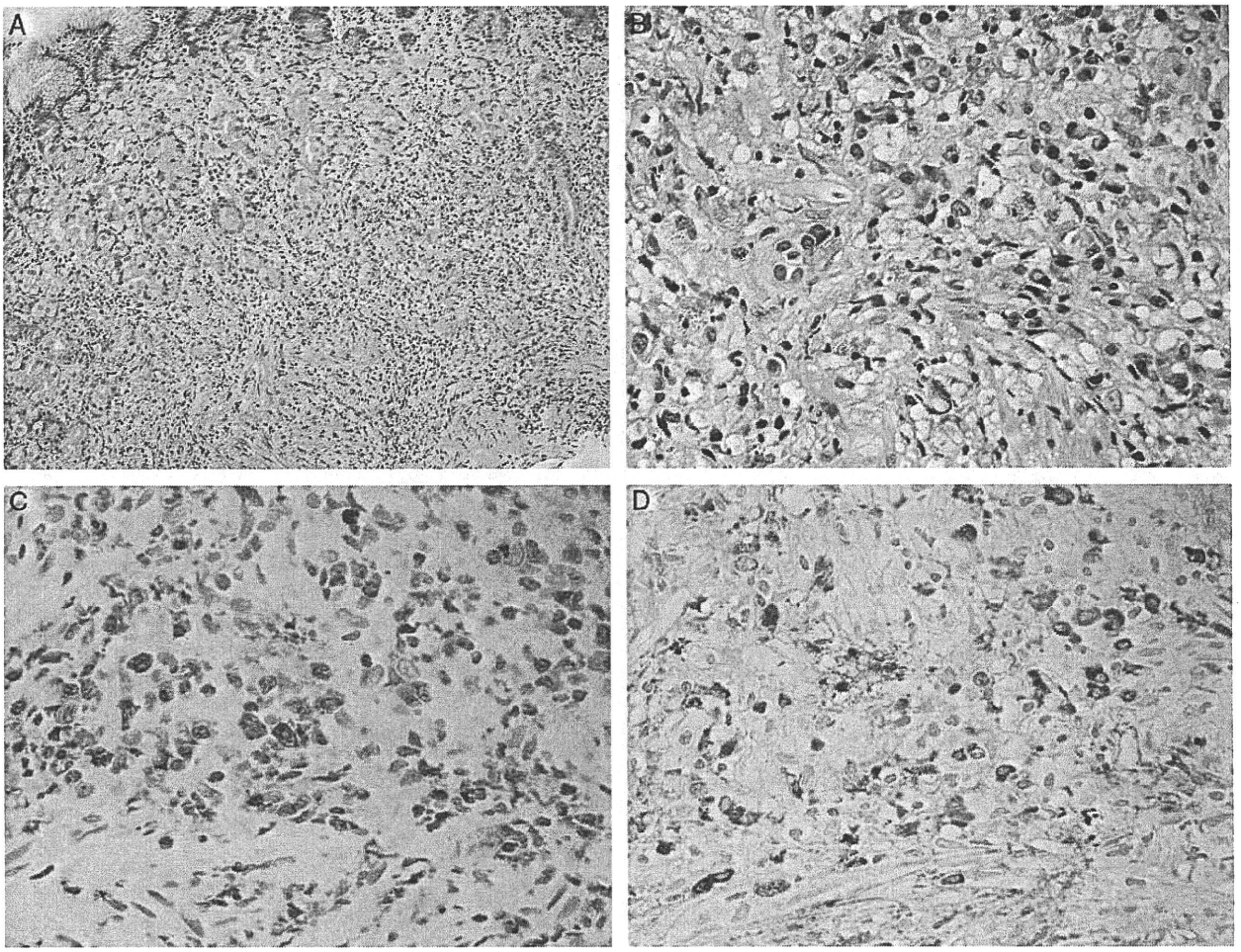


**FIGURE 3.** Stomach biopsy specimen of a patient with autoimmune pancreatitis-associated gastritis without steroid therapy and with *Helicobacter pylori* infection. A, Diffuse inflammatory cell infiltration is noted in the fundic mucosa (HE staining, original magnification, 25 ×). B, Various degrees of lymphoplasmacytic and scattered eosinophil infiltration are found. Fibrosis is also noted (HE staining, original magnification, 100 ×). Infiltration of (C) IgG-positive (immunostaining for IgG, original magnification, 100 ×) and (D) IgG4-positive plasma cells can be noted (immunostaining for IgG4, original magnification, 100 ×). HE indicates hematoxylin-eosin; IgG, immunoglobulin G.

the upper and lower lamina propria in the AIP-G-ST(−) groups were significantly higher than those in the CAG group.

In the AIP-G-ST(−) groups, the IgG4/IgG ratio in the upper and lower lamina propria were not significantly different between the fundic and the pyloric mucosa. In the specimens obtained from 6 biopsy points in the 3 patients of the AIP-G-ST(−) group, the distribution of the IgG4/IgG ratio in every case was approximately uniform in the fundic and the pyloric mucosa.

In the AIP-G-ST(+) group, the IgG4/IgG ratio in the fundic mucosa were 0.00 and 0.00 in the upper lamina propria and 0.21 and 0.00 in the lower lamina propria. In the AIP-G-ST(+) group, the IgG4/IgG ratio in the pyloric mucosa were 0.00 and 0.00 in the upper lamina propria and 0.00 and 0.00 in the lower lamina propria. In the fundic and the pyloric mucosa, the IgG4/IgG ratios in patients with AIP-G-ST(+) group were more discreet than those in the AIP-G-ST(−) group. In the specimens obtained from 6 biopsy points in 1 patient of the AIP-G-ST(+) group, the distribution of the IgG4/IgG ratio was approximately uniform in the fundic and the pyloric mucosa.

## DISCUSSION

Characterization of variant forms of gastritis other than CAG, such as AIP-G, is important. This study revealed that AIP-G patients had lymphoplasmacytic and eosinophilic infiltration accompanied by the presence of many IgG4-positive plasma cells in the stroma and high serum IgG4 concentrations. The result of this study suggested that AIP-G had characteristics of IgG4-related disease and might be one of the IgG4-related sclerosing diseases.

In this study, although 1 patient with AIP-G-HP(−) had epigastralgia, he did not have HP infection. AIP-G-HP(−) patients did not have ulcers, but 2 of them

had erosion and 1 of them had redness. This finding contradicted the findings of Chang et al,[1] who reported that ulcer formation was a characteristic of AIP-G. This contradiction might be explained by the fact that most of the reported cases in that study had HP infection,[1,21] which may be associated with ulcer formation.

AIP-G has received worldwide recognition since it was first reported by Shinji et al[21] in 2004. Although many of their cases were accompanied with HP infection, they reported their pathologic findings as characteristic of AIP-G. They also reported that the inflammatory scores for AIP-G and CAG were not significantly different; however, in our study, eosinophil infiltration in the AIP-G patients was significantly higher than that in the CAG patients. Eosinophil infiltration is one of the most characteristic findings of IgG4-related sclerosing disease and is important in the diagnosis of AIP-G. No neutrophilic infiltration may also be a characteristic finding that distinguishes AIP-G as an IgG4-related sclerosing disease, provided HP infection is absent. Interestingly, the mononuclear cell density in the upper and lower lamina propria in the AIP-G-ST(−) groups was almost identical, but that in the upper lamina propria was higher than the corresponding value in the control groups. Diffuse mononuclear cell infiltration in the lamina propria might be another characteristic finding of AIP-G. The AIP-G-ST(−) groups had no difference in the degree of inflammation between the pyloric and the fundic mucosa, although inflammation is known to be significantly higher in the pyloric mucosa than in the fundic mucosa in CAG.[4]

Interestingly, much IgG4-positive plasma cells were observed a lot in the lower lamina propria than the upper propria in the AIP-G-ST(−) groups, irrespective of the HP status. In 1 patient with AIP-G-HP(+), diffuse fibrosis was observed in the lower lamina propria. These pathologic findings resembled those of IgG4-related sclerosing disease. The presence of gastric wall thickening and the accumulation of radioactive material on the stomach wall, which was revealed by Ga-67 citrate scintigraphy, indicated that there may be a marked inflammation in the deep gastric wall. This might explain the absence of significant changes on the gastric surface mucosa during the endoscopic examinations of AIP-G patients. Further studies must be conducted to elucidate the clinicopathologic characteristics of AIP-G.

Many studies suggest that not only the number of IgG4-positive plasma cells but also the IgG4/IgG ratio is important for the diagnosis of extrapancreatic IgG4-related sclerosing disease.[30,31] An IgG4/IgG ratio of greater than 40% to 50% usually seems to be highly suggestive of IgG4-related disease. In this study, the IgG4/IgG ratio in the lower lamina propria of fundic mucosa was more than 50%. Sepehr et al[20] reported that the cut-off value for the IgG4/IgG ratio in the Vater papilla of an AIP patient was 0.1. Although the IgG4/IgG ratio in almost all the AIP-G-HP(+) patients was more than 0.1, we could not distinguish whether this inflammation was caused by HP infection or AIP-G. However, the IgG4/IgG ratio in the AIP-G-HP(−) patients suggests that AIP-G could have caused the stomach inflammation, even in the absence of HP infection. It is also important to note that patients without HP infection may have gastritis resembling pancreatic inflammation observed in AIP.

Steroid therapy is effective against IgG4-related sclerosing disease,[28] as the IgG4/IgG ratios in the AIP-G-ST(+) group were predominantly lower than those in the AIP-G-ST(−) groups. AIP-G may also be effectively treated using steroid therapy. But the symptoms in the AIP-G-ST(+) group did not alleviate after steroid therapy; this could be because some clinical findings associated with HP infection persisted.

In summary, AIP-G is also one of the IgG4-related sclerosing diseases, and the IgG4/IgG ratio is a useful marker for further research on IgG4-related sclerosing diseases.

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# B型肝炎再活性化の病態と対策

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総　説

# B型肝炎再活性化の病態と対策

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**要旨**：B型肝炎再活性化が注目されるようになった背景には，医療の進歩にともない免疫を強く抑制する治療が一般化したことが挙げられる．非活動性キャリアからの再活性化は以前より知られている病態であり，その防止には核酸アナログ薬の予防投与が行われている．既往感染者からの再活性化はde novo B型肝炎とも呼ばれ，劇症化しやすく死亡率も高い．対策として，核酸アナログ薬の予防投与や血中HBV DNAの定期検査が検討されている．後者では，できるだけ早期にウイルスの再増殖を検出し，肝炎発症前に治療を開始する．両法ともde novo B型肝炎の防止に有用であるが，一般化するためには費用対効果などの問題を明らかにする必要がある．

**索引用語**：De novo B型肝炎，再活性化，HBV cccDNA，免疫抑制，核酸アナログ薬

## はじめに

近年，B型肝炎再活性化の問題が広く注目を集めるようになった．この背景には，医療の進歩にともない，より強力に免疫を抑制する治療薬が悪性腫瘍，膠原病，移植などの領域で広く用いられるようになったことがある．一方，B型肝炎ウイルス（HBV）感染のインパクトは強く，世界では22億人（人口の1/3）がHBVに感染したことがあり，この内3億5千万人がキャリアであること，また，日本では2600万人（人口の1/5）がHBVに感染したことがあり，この内140万人はキャリアであることが推定されている[1)~3)]．再活性化の問題はキャリアのみならず既往感染者にも及ぶため，その影響範囲は広い．

臨床的観点から，B型肝炎の再活性化は大きく2つに分けることができる．1つは非活動性キャリアからの再活性化であり，もう1つは既往感染者からの再活性化である．前者の病態は以前より知られており，キャリアからの再活性化に対しては予防策が実践されている．これに対し後者は，最近ようやく一般に認識されるようになった病態であり，これに対する対策はいまだ十分とはいえない．HBV既往感染者における再活性化はde novo B型肝炎とも呼ばれており，劇症化しやすく死亡率も高いことから，その対策の確立が急がれている[4)]．本稿ではB型肝炎の再活性化について，その病態や対策などについて述べる．

## I　再活性化の病態とHBV cccDNA

HBV cccDNA（covalently closed circular DNA）はHBVの複製においてその起点となる分子であり，B型肝炎の再活性化を考える上で極めて重要な因子である[2)5)~8)]．HBV粒子がレセプターを介して肝細胞内に侵入すると，その遺伝子である不完全二重鎖のHBV DNAが完全二重鎖に修復されcccDNAとなり核内にプールされる．このHBV cccDNAはHBV複製のすべての起点であり，ここから主に2系統のRNAが転写される（Figure 1）．2.1と2.4kbのmRNAからはHBs抗

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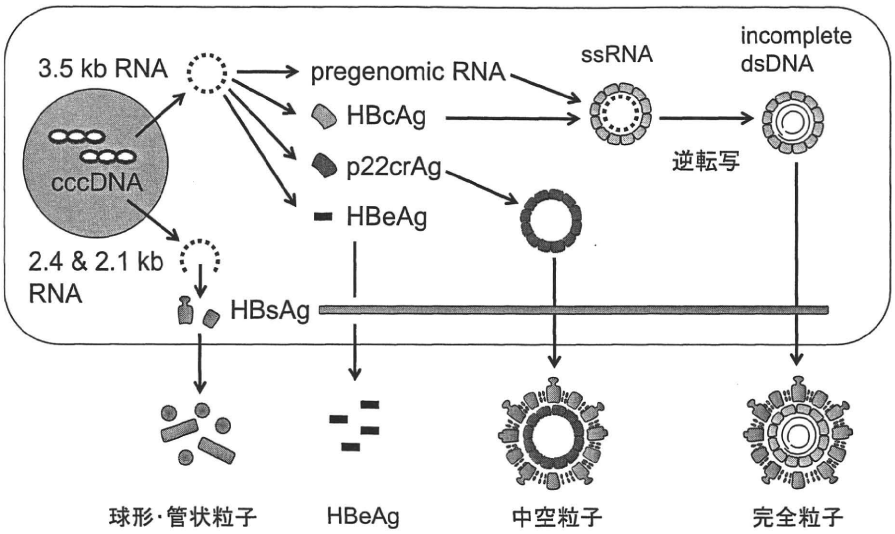


**Figure 1.** HBV cccDNA（covalently closed circular DNA）を起点としたウイルスの複製.

**Table 1.** 肝細胞内 HBV cccDNA の特徴

| |
|---|
| 1. 安定した分子で破壊されにくい. |
| 2. 肝細胞核内にプールを作る(5～50コピー/細胞). |
| 3. HBV 複製の起点である. |
| 4. HBV 増殖の活動性と相関する. |

cccDNA：covalently closed circular DNA.

原が翻訳される．一方，3.5kb の mRNA からは HBc 抗原，p22cr 抗原，HBe 抗原が翻訳されるとともに，この 3.5kb の RNA 自身が pregenomic RNA となる．HBe 抗原は分泌蛋白として血中に分泌され，p22cr 抗原は遺伝子を持たない中空粒子を作る[9]．HBc 抗原は pregenomic RNA と一緒にヌクレオキャプシドを形成する．形成されたヌクレオキャプシド内の pregenomic RNA は DNA に逆転写され，さらに不完全2本鎖の DNA 遺伝子となる．この逆転写が完了すると HBs 抗原を含む脂質二重膜のエンベロープをかぶり完全ウイルス粒子として肝細胞から血中へ分泌される．

Table 1 に HBV cccDNA の特徴を整理した．HBV cccDNA は非常に安定した分子で DNAase などにより破壊されにくい．HBV が感染すると1肝細胞当たり 5～50 コピーの核内プールが作られる．また，HBV 複製の起点であり，これがあれば HBV の複製を完了させることが可能である．さらに，cccDNA 量は HBV の活動性と相関することが知られている[10]．これらの性質から，複製がなくなっても HBV は cccDNA の形で肝細胞核内に長期間潜伏感染し，免疫抑制にともないここから HBV の複製が再開される[11]．

HBV キャリアの自然経過は，免疫寛容期，HBe 抗原陽性または陰性の慢性肝炎期，非活動性キャリア期，回復期などの病期に分類される．この中で，非活動性キャリア期では HBV に対する宿主の免疫が優位になり HBV の複製は大きく低下する．さらに，一部では HBs 抗原までもが陰性化し回復期となる．これらの病期では肝炎は治癒し肝発癌率も低下するので予後は良い[12]．しかし，宿主の免疫能が何らかの原因で抑制されると，肝細胞の核内の HBV cccDNA を起点として HBV の複製が活性化し B 型肝炎が再燃する．HBV の既往感染でも同様で，年余にわたり肝細胞核内に HBV cccDNA が潜伏感染しているので免疫抑制により B 型肝炎が再活性化する．

## II 再活性化の多様性

B 型肝炎再活性化対策の難しさは，その病態が一様でないことや関連する診療科や免疫抑制薬が多岐にわたる点にある．まず，免疫抑制療法前の

Table 2. B型肝炎再活性化と関連する免疫抑制療法とその特徴

| 免疫抑制療法 | 特徴 |
|---|---|
| グルココルチコイド | 多くの診療科が関与し，使用頻度が高い．HBV特異的な再活性化促進作用がある． |
| 化学療法 | 多くの診療科で行われ，使用頻度の高い治療である． |
| リツキシマブ | 本薬を加えた化学療法では再活性化が高頻度である． |
| TNFα阻害薬 | 最近，使用頻度が高くなり，再活性化が報告されるようになった．グルココルチコイドとの併用が多く，長期に使用される傾向にある． |
| 造血幹細胞移植 | 免疫担当細胞の組み換えがおこる．長期に免疫抑制を行う必要がある． |
| 肝移植 | ドナーのHBVが原因となる．長期に免疫抑制を行う必要がある． |
| 新しい治療法 | 問題となる新しい治療法が今後増えることが予測される． |

HBVの感染状態では，活動性キャリア，非活動性キャリア，既往感染（回復期）の場合に分けて考える必要がある．再活性化と呼ばれるのは非活動性キャリアと既往感染の場合のみであるが，活動性キャリアにおいても免疫抑制がかかると肝炎がさらに活動性となるので，再活性化と同様に何らかの対策が必要である．また，既往感染者はHBVキャリアとは認められていないため，対策にも自ずと制限が生ずる．

医学の進歩にともない化学療法や免疫抑制療法も多様化し，さらにより高度になっている．B型肝炎の再活性化の頻度は治療法や治療薬により異なるので，対策もそれぞれについて検討する必要がある．B型肝炎の再活性化と関連する免疫抑制療法とその特徴をTable 2にまとめた．

グルココルチコイドによるB型肝炎の再活性化については以前より知られており，注意が喚起されていた．この薬物の特徴は，多くの診療科で使用しその使用頻度も高い点である．また，HBV遺伝子のS領域にはグルココルチコイド感受性領域が存在することが報告されている[13)~15)]．グルココルチコイドを投与すると細胞質内にあるグルココルチコイド受容体が核内に移動し，この受容体とHBV遺伝子の感受性領域が結合することによりHBVの複製が活性化される．このように，グルココルチコイドはHBV特異的にその増殖を活性化するので，その使用量が少ない場合でも注意が必要である．

近年リツキシマブが悪性リンパ腫などの治療で広く用いられるようになり，通常の化学療法と比較して再活性化の頻度が明らかに高くなった．TNFα（tumor necrosis factor α）阻害薬は慢性関節リウマチやクローン病などで最近広く使用されるようになり，長期使用によるB型肝炎再活性化の危険性が指摘されている．移植診療一般に，強力で長期の免疫抑制を必要とするためB型肝炎再活性化の危険性は低くない．この中で，造血幹細胞移植は免疫担当細胞の組み換えがおこるため，HBVの複製を抑制する力はより弱くなる可能性がある．肝移植については，ドナーのHBVが原因となるため，宿主の免疫がこれを効率よく抑制できない可能性がある．

### III 非活動性キャリアにおける再活性化

HBVキャリアの85～90%は自然経過でウイルスの増殖が低下し非活動性キャリアとなる．非活動性キャリアでは肝炎はないので臨床的には安定した状態となる．しかし，非活動性キャリアになっても，化学療法薬や免疫抑制薬を使用することによりHBVに対する免疫監視能が低下し再増殖がおこる[16)17)]．この再増殖を確認するためのマーカーとしては血中HBV DNA量の測定が有用である．一般に，非活動性キャリアでは血中のHBV DNAは4.0log copy/ml未満に抑制されている．しかし，免疫が抑制されるとDNA量は4.0log copy/ml以上に増加し，しばしば7.0～8.0log copy/mlまたはこれ以上の高ウイルス量となる．ウイルスの増殖が盛んになった状態で免疫抑制薬や化学療法薬を中止すると，これまで抑制されて

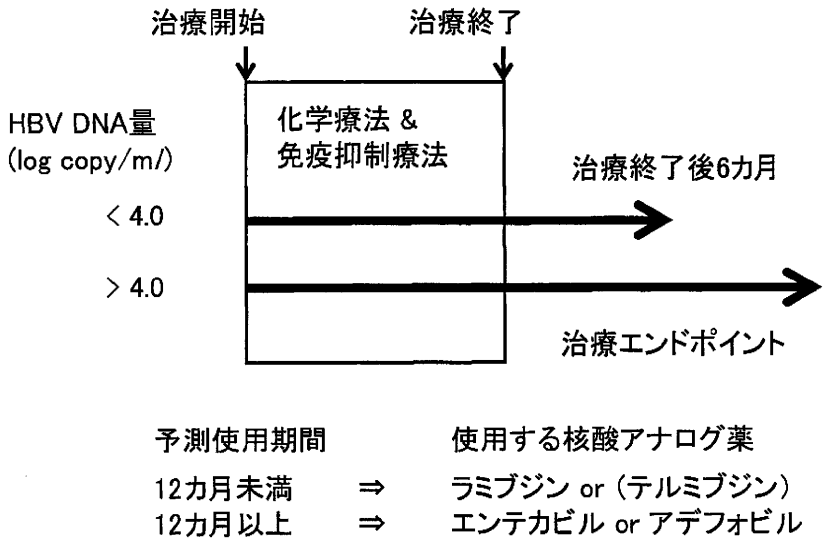


**Figure 2.** 非活動性キャリアにおけるB型肝炎再活性化の対策（文献18から改変）.

いた免疫力が回復し強いウイルス排除反応，すなわち肝炎の再活性化がおこる．再活性化したB型肝炎は重症化・劇症化し致死的になることもまれではない．このため，キャリアで化学療法薬や免疫抑制薬を使用する場合は再活性化に対する注意が必要であり，最近では核酸アナログ薬による予防が積極的に行われるようになっている．

B型肝炎再活性化の頻度はHBVの感染状態と免疫抑制の程度により異なる．HBVキャリアでは，免疫抑制の程度が比較的弱くても20～50%に再活性化がおこり，造血幹細胞移植や肝移植では50%を超える再活性化がおこりさらに危険性が高い．悪性腫瘍の化学療法にともなう再活性化の報告には悪性リンパ腫，乳癌，多発性骨髄腫，肺癌，膵癌などの報告があるが，報告が多いのは悪性リンパ腫と乳癌である．膠原病などの免疫抑制療法にともなう再活性化ではステロイドによるものが多い，最近はTNFα阻害薬などの分子標的薬による再活性化の報告が増加している[17]．

## IV 非活動性キャリアにおける再活性化対策

アメリカ肝臓学会が2007年に発表したB型慢性肝炎のガイドラインの中に，化学療法・免疫抑制療法にともなうB型肝炎再活性化対策がある(Figure 2)[18]．対策としては核酸アナログ薬の予防投与が基本であり，化学療法・免疫抑制療法開始前より投与を開始する．核酸アナログ薬は化学療法・免疫抑制療法中は継続して投与する必要があり，治療が終了してもすぐには中止しない．中止時期は治療開始前のウイルス量により異なっており，血中HBV DNA量が4.0log copy/ml以下であれば化学療法・免疫抑制療法終了後6カ月で核酸アナログ薬を中止する．一方，ウイルス量が4.0log copy/ml以上であれば通常の慢性B型肝炎の治療に準じて治療を継続する必要がある．使用薬物は，予防投与期間が12カ月以内と予測される場合はラミブジンまたはテルビブジン（日本では未承認）が，またこの期間が12カ月以上と予測される場合は耐性株が出現しにくいエンテカビルまたはアデフォビルが推奨されている．日本でのB型慢性肝炎治療のガイドラインではエンテカビルが第一選択であり，再活性化の予防についてもエンテカビルが選択される傾向にある．インターフェロンは骨髄抑制の副作用があるため使用しない．

Hsuら[19]は非ホジキンリンパ腫の患者を対象として，ラミブジンを予防的に使用した場合と肝炎発症後の治療に使用した場合を前向きに比較検討した．この結果，ラミブジンの予防投与はB型

肝炎の再活性化の頻度や重症度を有意に低下させることを報告した．Saabら[20]はDecision Analysis Modelを用いて，HBVキャリアの悪性リンパ腫の患者に対してラミブジンを予防的に内服させることは費用対効果が高いことを報告した．さらに，ラミブジン投与群では原疾患による死亡数も減少することが予測されている．これは，ラミブジン投与によりB型肝炎の再活性化がおこらなければ，抗腫瘍薬などの休薬による原疾患加療の中断がないためと考えられている．このように，HBVキャリアにおける再活性化対策として核酸アナログ薬の予防投与が有用であり，これらの成績を基に対策が立てられた．予防投与の期間は，化学療法終了後2カ月までででは不十分であり，基本的に6カ月までの投与が推奨されている．

## V De novo B型肝炎

HBs抗原陰性でHBc抗体and/or HBs抗体陽性者，すなわちHBVの既往感染者は本邦の人口の約1/5を占める．以前は，既往感染状態になるとHBVは宿主から完全に排除されていると考えられていた．しかし，その後の検討で，HBV遺伝子がcccDNAの形で核内に残存しごく低レベルで複製していることが明らかになった．HBVに感染すると，非特異的な自然免疫に引き続きウイルス特異的な免疫反応がおこる．この時，細胞障害性T細胞（CTL）による細胞性免疫がウイルス排除に重要な役割を果たしている[21]．この細胞性免疫反応は急性肝炎治癒後，年単位で持続する．これは，肝細胞内で感染性のあるウイルスの産生が持続していることを示すが，CTLの働きにより血中へのウイルスの放出は極めて少量に抑えられている．これがHBs抗原陰性で既往感染とされる場合の感染状態である．

宿主の免疫能が低下し，特異的CTLがウイルスの増殖をコントロールできない状態になるとHBVの増殖が盛んになりB型肝炎が再燃する．この状態がde novo B型肝炎である．化学療法後のde novo B型肝炎の報告は1975年のWandsら[22]によるものが最初と思われる．しかし，その当時はHBVの潜伏感染に関する知識に乏しく，その臨床的意義は不明であった．1991年，Lokら[23]は化学療法を受けた悪性リンパ腫患者についてB型肝炎の再活性化を前向きに調査した．この結果，HBVキャリアでの再活性化は高頻度であったが，de novo B型肝炎の発症は2%程度とまれであることを報告した．これに対し，血液疾患にともない同種骨髄移植を施行した症例ではde novo B型肝炎の発症は14～50%と比較的高率であることが報告されている．この両者の頻度の差は，おそらく免疫抑制の程度が骨髄移植では通常の化学療法よりも強いためと考えられる．

1998年，Uemotoら[24]は生体肝移植症例におけるde novo B型肝炎を検討し，ドナーがHBVの既往感染の場合，移植後94%（15/16）のレシピエントにde novo B型肝炎を発症したことを報告した．さらに，この報告ではドナー肝のHBVとレシピエントから分離されたHBVの塩基配列が一致することを確認し，レシピエントのHBVがドナー由来であることを証明した．その後，肝移植後のde novo B型肝炎に関する報告は多く[25)~33)]，その特徴をまとめると以下のようになる．発症時期は移植後しばらくして経過してからであり，中央値は12カ月と長い．ドナーが既往感染者の場合の感染率は高く，33～96%と報告されている．肝炎を発症するとHBVは高率に持続感染化する．肝炎の重症度は症例により大きく異なり予測は困難である．レシピエントがHBs抗体陽性でも感染は成立する．しかし，HBIG（hepatitis B immunoglobulin）またはHBワクチンでHBs抗体価を保つことにより予防可能である．

近年，de novo B型肝炎が肝臓の専門家だけでなく血液の専門家の間でも注目を浴びるようになった．その理由は，悪性リンパ腫の治療に抗CD20抗体であるリツキシマブが使用されるようになったためである．リツキシマブの使用で悪性リンパ腫の治療成績は大きく改善したがB型肝炎再活性化の頻度も高くなった．リツキシマブの使用によってde novo B型肝炎を発症した症例が2001年にDerviteら[34]により初めて報告された．すなわち，HBs抗原陰性，HBs抗体陽性の患者がリツキシマブを含む化学療法施行後にHBs抗原とHBV DNAが陽性となり劇症肝不全をおこ

して死亡したという症例である．その後もリツキシマブ使用と関連したde novo B型肝炎例が報告され注目を集めた．

香港のHuiら[35]は，悪性リンパ腫の治療を受けた244名を対象に前向きコホートでde novo B型肝炎，劇症肝不全の発症率およびそれらの危険因子を検討した．彼らの報告では244例中8例（3.3%）でde novo B型肝炎を発症しており，その内3例で劇症肝不全を発症し1例が死亡した．De novo B型肝炎発症の危険因子はリツキシマブとステロイドの併用であった．さらに，de novo B型肝炎の発症は劇症肝不全を引きおこす唯一の危険因子であった．香港のYeoら[36]も104例の悪性リンパ腫症例を対象にde novo B型肝炎の発症を検討した．HBs抗原陰性でHBc抗体陽性の既往感染者が104例中46例あり，この46例中R-CHOP療法を受けた21例では4例（24%）でde novo B型肝炎を発症したのに対し，CHOP療法を受けた25例ではde novo B型肝炎の発症はなかった（P=0.015）．これらの成績から，リツキシマブの使用が有意にde novo B型肝炎の発症に関連していることは明らかである．

近年，白血病などの治療に骨髄幹細胞移植が広く用いられるようになり，これにともなうde novo B型肝炎の報告も増えている[37]~[45]．骨髄幹細胞移植での特徴は，悪性リンパ腫のR-CHOP療法と比較するとde novo B型肝炎の発症時期が遅い症例が多いことである．すなわち，R-CHOP療法後は中央値で3カ月（範囲0~9カ月）であるのに対し，骨髄幹細胞移植後は中央値で16カ月(範囲3~25カ月)と長い傾向にある．これは，骨髄幹細胞移植では長期に免疫抑制薬を使用することが主な要因と考えられる．

インフリキシマブやエタネルセプトなどのTNFα阻害薬は，近年，関節リウマチやクローン病の治療に広く用いられるようになった．TNFα阻害薬もHBVの増殖を活性化することが知られており，非活動性キャリアからの再活性化に加えde novo B型肝炎の報告もみられる[46]~[51]．特徴としては，病歴が長く，本薬に加えグルココルチコイドなどの再活性化を促進する薬物も併用されていることが多い．短期での発症は少ないことが予測され[52]，長期に使用する場合は注意が必要である．

### VI 本法でのde novo B型肝炎の実態調査

本邦ではde novo B型肝炎のまとまった疫学的なデータがなかったため，平成17年度の厚生労働省研究班（熊田博光班長）で全国調査を行った[53]．この調査では，5年間に新たにHBs抗原陽性になった患者として，B型急性肝炎1184例とde novo B型肝炎55例（4.4%）が登録された．劇症化率はde novo肝炎で27%，急性肝炎で7%であり，de novo肝炎で有意に高い傾向がみられた．また，劇症化例の死亡率はde novo肝炎で100%，急性肝炎で44%であり，これもde novo肝炎で有意に高い傾向がみられた．すなわち，これらの成績は，一旦de novo B型肝炎を発症して劇症化すると救命は非常に困難であることを示す．さらに，肝炎発症後に核酸アナログ薬を投与しても劇症化が回避できないことがわかり，早期の核酸アナログ薬投与の重要性が示された．

De novo B型肝炎が重症化しやすい要因はいくつか考えられる．第一は，通常のB型急性肝炎に比較して年齢が高いことである（中央値：63歳 vs. 33歳，P<0.001）．約30歳の差があり，明らかにde novo肝炎患者で不利である．第二は基礎疾患の有無であり，de novo肝炎患者の多くは背景に重篤な疾患を持ち，肝炎発症前から全身状態が不良である．第三は，すでに数々の治療が行われており，薬物性肝障害などの他の肝障害をおこす要因があることから，肝炎を発症してもde novo B型肝炎と認識されずに対応が遅れることが多いことが挙げられる．

### VII De novo B型肝炎の対策

Table 3に，現在実施または検討されているde novo B型肝炎対策を整理した．HBIGやHBワクチンによるHBs抗体価の維持は移植後のde novo B型肝炎の予防に用いられている．具体的には肝移植時からHBIGを投与しHBs抗体価を維持する方法である[24]．通常，その後の経過でHBワクチンを併用しHBs抗体価を維持する．信州大学での検討では，90%以上の症例で最終的に