—Original—

# Evaluation of the Impact of Preoperative Values of Hyaluronic Acid and Type IV Collagen on the Outcome of Patients with Hepatocellular Carcinoma After Hepatectomy

Junji Ueda[1,2], Hiroshi Yoshida[1], Yasuhiro Mamada[1], Nobuhiko Taniai[1], Masato Yoshioka[1], Atsushi Hirakata[2], Youichi Kawano[1], Tetsuya Shimizu[1], Tomohiro Kanda[1], Hideyuki Takata[2] and Eiji Uchida[1]

[1]Department of Gastrointestinal Hepato-Biliary-Pancreatic Surgery, Nippon Medical School, Tokyo, Japan
[2]Department of Surgery, Nippon Medical School Tama Nagayama Hospital, Tokyo, Japan

**Background:** Recently, some reports have revealed a relationship between post-hepatectomy prognosis in hepatocellular carcinoma (HCC) and hepatic fibrosis markers. We evaluated the relationship between these markers of hepatic fibrosis, clinicopathological findings, and prognosis.

**Methods:** Three hundred and sixty patients underwent hepatectomy for HCC in the Nippon Medical School Hospital between 1993 and 2013. We divided these patients into two groups: normal serum hyaluronic acid (HA) levels and abnormal levels. We also divided patients into groups with normal serum type IV collagen levels and abnormal levels.

**Results:** The overall survival rate and recurrence-free survival rate of the normal group were significantly higher than those of the abnormal group. In the normal hyaluronic acid group, serum albumin and prothrombin time were significantly higher than in the abnormal group, and age, hepatitis C virus antibody (HCV)-Ab positivity, Child-Pugh grade B, liver cirrhosis, indocyanine green retention rate at 15 min (ICGR15), type IV collagen level, and type IV collagen 7s level were significantly lower than those in the abnormal group. In the normal type IV collagen group, HCV-Ab positivity, liver cirrhosis, ICGR15, HA level, and type IV collagen 7s level were significantly lower than those in the abnormal group, and the serum albumin level was significantly higher than that in the abnormal group. Multivariate analysis independently revealed the significant effect of serum type IV collagen on the overall survival rate as well as the significant effect of serum HA on the recurrence-free survival rate in patients who underwent hepatectomy for HCC.

**Conclusions:** Preoperative examinations of serum hyaluronic acid levels and type IV collagen levels are imperative for hepatic resection for HCC because these markers are significantly associated with liver function and prognosis.

(J Nippon Med Sch 2018; 85: 221–227)

**Key words:** hyaluronic acid, type IV collagen, hepatocellular carcinoma, liver cirrhosis

## Introduction

Hepatocellular carcinoma (HCC) is the fifth-most prevalent carcinoma in the world. HCC is the third-most common cause of cancer-related death worldwide[1,2]. From a clinical point of view, the critical problem is recurrence of HCC after treatment[3]. Despite improving techniques in non-surgical approaches, hepatectomy still represents a potentially curative treatment for HCC. Unfortunately, tumor recurrence is the major cause of worsening results in long-term survival, with an expected 5-year intrahepatic recurrence rate of up to 70 %[3]. Thus, the identification of new markers for recurrence and survival is important to predict the possible outcome of patients with HCC[4].

Correspondence to Junji Ueda, MD, PhD, Department of Surgery, Nippon Medical School Tama Nagayama Hospital, 1–7–1 Nagayama, Tama, Tokyo 206–8512, Japan
E-mail: junji0821@nms.ac.jp
Journal Website (http://www2.nms.ac.jp/jnms/)

Table 1 Clinical and pathological characteristics of the study

| | | |
|---|---|---|
| Age (yr) | | 66.7 (±9.63) |
| Sex | male/female | 263/97 |
| Infection | HBV/HCV/none | 70/224/77 |
| Child-Pugh | A (5)/A (6)/B (7)<br>B (8)/B (9)/C (10-) | 212/109/36<br>3/0/0 |
| ICG 15R (%) | | 18.2 (±12.3) |
| Tumor size (mm) | | 45.5 (±35.7) |
| Differentiation | wel, mod/por | 316/44 |
| Background | NL, CH/LC | 174/186 |
| pStage | I/II/III/IV | 48/129/104/74 |

(±standard deviation SD)

HBV: hepatitis B virus, HCV: hepatitis C virus, ICG: indocyanine green, wel: well differentiated hepatocellular carcinoma, mod: moderately differentiated hepatocellular carcinoma, por: poorly differentiated hepatocellular carcinoma, NL: normal liver, CH: chronic hepatitis, LC: liver cirrhosis, pStage: pathological stage, T-bil: total bilirubin, Alb: albumin, PT: prothrombin time, AFP: alfa-fetoprotein, AFP L3: Lectin-reactive alpha-fetoprotein, DCP: des-γ-carboxy prothrombin

Hyaluronic acid (HA) is one of the oldest serum parameters of hepatic fibrosis. HA is known as mucopolysaccharide. Several reports indicate that HA is a biomarker for fibrosis and cirrhosis in liver disease[5].

Type IV collagen codistributes with laminin in the basement membranes of vessels and bile ducts, and there is also "free" type IV collagen that is not associated with laminin and is distributed in the sinusoids as small, discrete, discontinuous deposits. During the transition from early to late fibrosis, trichrome (stains primarily collagen I) and collagen IV showed the steepest increase and appeared to be the most useful discriminators between the early and late stages of fibrosis[6].

The aim of this study was to evaluate the relationship between serum HA and type IV collagen levels, clinicopathological findings, and prognosis in patients who underwent hepatic resection for HCC.

## Materials and Methods

Consecutive patients who underwent hepatectomy for HCC between February 1993 and September 2013 at the Nippon Medical School Hospital were included in this study. This study was approved by the Human Ethics Review Committee of the Nippon Medical School.

Indications for hepatectomy were based on the results of preoperative and intraoperative imaging and liver function assessment.

Pathological diagnoses and clinicopathological factors were established using the general guidelines for primary liver cancer of the Liver Cancer Study Group of Japan[7].

All patients were followed up with examination of α-fetoprotein (AFP) and des-γ-carboxyprothrombin (DCP) levels, ultrasonography, dynamic computed tomography (CT), and magnetic resonance imaging (MRI). Recurrences were diagnosed as the appearance of a lesion that is typical of HCC, as confirmed by ultrasound imaging, CT, and MRI. If recurrence was detected, the patient received further treatment.

Serum HA levels and type IV collagen levels were measured within 4 weeks before surgery. We divided these patients into two groups: normal serum HA levels and abnormal levels. We also divided patients into groups with normal serum type IV collagen levels or with abnormal serum type IV levels. The normal HA level was under 50 ng/mL. The normal type IV collagen level was under 140 ng/mL.

Categorical variables were compared using the $\chi^2$ test and Student's t-test. Multiple logistic regression analyses were performed to confirm the relationship between variables, serum HA level, and type IV collagen level. Recurrence-free survival (RFS) and overall survival (OS) curves were determined with the Kaplan-Meier method. Multivariable Cox proportional hazards regression analyses were performed.

## Results

During the study, 360 consecutive patients underwent hepatectomy for HCC. The median age was 66.7 years, and 263 of the patients were men; 70 patients were hepatitis B surface antigen (HBs-Ag) positive, and 224 patients were hepatitis C virus antibody (HCV-Ab) positive. Child-Pugh grade A was present in 324 cases, and grade B was present in 36 cases. The mean tumor diameter was 45.5 mm. In the analysis of type IV collagen, 63 patients were excluded because their preoperative serum type IV collagen level had not been determined. The remaining 297 patients were analyzed (**Table 1**).

Regarding HA level, a comparison of the two groups is shown in **Table 2**. In the normal HA group, age, HCV-Ab positivity, Child-Pugh grade B, liver cirrhosis, indocyanine green retention rate at 15 min (ICGR15), type IV collagen level, and type IV collagen 7s level were signifi-

Table 2 Comparison of the background characteristics of the two hyaluronic acid level groups

| Hyaluronic Acid | | Normal (n=58) | Abnormal (n=302) | *p*-value |
|---|---|---|---|---|
| Age (yr) | | 64.3 | 67.1 | 0.042 |
| Sex | male/female | 48/10 | 215/87 | 0.0975 |
| Tumor diameter (mm) | | 49.3 | 44.3 | 0.3399 |
| HBV | +/− | 16/42 | 54/246 | 0.1325 |
| HCV | +/− | 24/34 | 200/101 | 0.0005 |
| Child-Pugh | A/B, | 57/1 | 262/40 | 0.0399 |
| Differentiation | wel, mod/por | 43/7 | 223/37 | 0.999 |
| Background of liver | NL, CH/LC | 42/12 | 104/174 | 0.0001 |
| T-bil (mg/dL) | | 0.747 | 0.847 | 0.234 |
| Alb (g/dL) | | 4.12 | 3.75 | 0.001 |
| PT (%) | | 91.1 | 84.6 | 0.0034 |
| ICG15R (%) | | 10.9 | 19.6 | 0.0001 |
| AFP (ng/mL) | | 13,248 | 17,262 | 0.8993 |
| L3 (%) | | 9.09 | 17.6 | 0.114 |
| DCP (mAU/mL) | | 1,577 | 5,292 | 0.237 |
| Type IV collagen (ng/mL) | | 157 | 250 | 0.0001 |
| Type IV collagen 7s (ng/mL) | | 4.73 | 8.37 | 0.0308 |

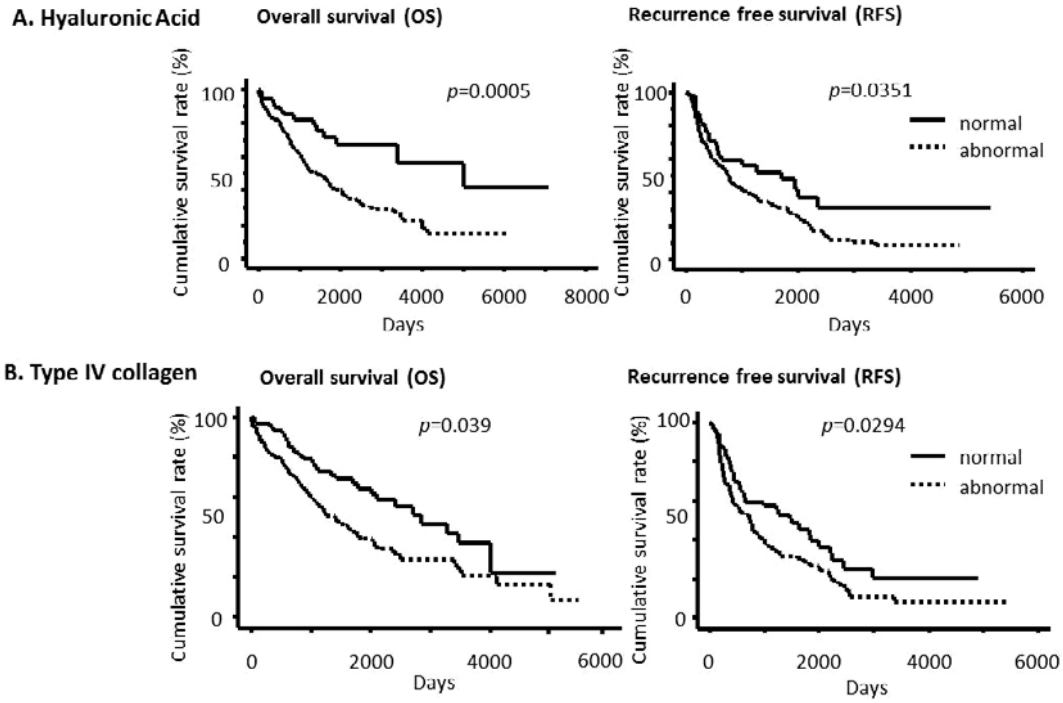


Fig. 1 **A**: A high serum hyaluronic acid (HA) concentration was associated with shorter overall survival (OS) (P=0.0005) and recurrence-free survival (RFS) (P=0.0351). In the normal HA group, OS and RFS were significantly longer than in the abnormal group. **B**: A high serum type IV collagen concentration was also associated with shorter OS (P=0.0039) and RFS (P=0.00294). In the normal type IV collagen group, OS and RFS were significantly longer than in the abnormal group.

cantly lower than in the abnormal group. Moreover, in the normal HA group, serum albumin level and prothrombin time were significantly higher than in the abnormal group. Anatomical resection of HCC was performed more often in the normal HA group than in the abnormal group. High serum HA concentrations were associated with shorter OS (P=0.0005) and RFS (P=0.0351). (**Fig. 1A**).

Next, we analyzed type IV collagen levels. A comparison of the background characteristics of the two groups is shown in **Table 3**. In the normal type IV collagen group, HCV-Ab positivity, liver cirrhosis, ICGR15, HA

Table 3 Comparison of the background characteristics of the two type IV collagen level groups

| Type IV collagen | | Normal (n=68) | Abnormal (n=227) | *p*-value |
|---|---|---|---|---|
| Age (yr) | | 64.8 | 66.4 | 0.2659 |
| Sex | male/female | 51/17 | 165/62 | 0.9124 |
| Tumor diameter (mm) | | 43.0 | 48.1 | 0.2979 |
| HBV | +/− | 17/51 | 42/185 | 0.3022 |
| HCV | +/− | 33/35 | 153/74 | 0.0058 |
| Child-Pugh | A/B | 65/3 | 195/32 | 0.1796 |
| Differentiation | wel, mod/por | 59/9 | 201/26 | 0.6981 |
| Background of liver | NL, CH/LC | 49/19 | 99/128 | 0.0001 |
| T-bil (mg/dL) | | 0.756 | 0.873 | 0.234 |
| Alb (g/dL) | | 4.01 | 3.79 | 0.0012 |
| PT (%) | | 87.9 | 84.6 | 0.1681 |
| ICG15R (%) | | 14.6 | 19.0 | 0.0079 |
| AFP (ng/mL) | | 2,156 | 25,755 | 0.4827 |
| L3 (%) | | 13.5 | 18.3 | 0.2916 |
| DCP (mAU/mL) | | 2,178 | 5,347 | 0.323 |
| Hyaluronic acid (ng/mL) | | 94.3 | 266 | 0.0001 |
| Type IV collagen 7s (ng/mL) | | 4.96 | 7.75 | 0.0001 |

Table 4 Univariate analysis for overall survival rate

| Univariate analysis (OS) | *P* value | Hazard ratio | 95% CI |
|---|---|---|---|
| Age | 0.4196 | 1.008 | 0.989−1.027 |
| Tumor diameter (mm) | 0.0001 | 1.010 | 1.006−1.013 |
| Vascular invasion (−) | 0.0012 | 0.598 | 0.437−0.817 |
| Number of tumor | 0.9958 | 1.001 | 0.704−1.335 |
| Stage | 0.0085 | 1.523 | 1.317−1.860 |
| T-bil (mg/dL) | 0.7680 | 1.035 | 0.825−1.298 |
| Alb (g/dL) | 0.0001 | 0.489 | 0.360−0.666 |
| PT (%) | 0.5182 | 0.996 | 0.986−1.007 |
| ICG15R (%) | 0.0670 | 1.012 | 0.999−1.025 |
| AFP (ng/mL) | 0.0397 | 1.000 | 1.000−1.000 |
| L3 (%) | 0.0003 | 1.018 | 1.008−1.027 |
| DCP (mAU/mL) | 0.0001 | 1.000 | 1.000−1.000 |
| Hyaluronic acid (ng/mL) | 0.0001 | 1.001 | 1.000−1.001 |
| Type IV collagen (ng/mL) | 0.0001 | 1.002 | 1.001−1.002 |
| Type IV collagen 7s (ng/mL) | 0.7766 | 0.998 | 0.986−1.010 |

level, and type IV collagen 7s level were significantly lower than in the abnormal group. Moreover, in the normal type IV collagen group, the serum albumin level was significantly higher than in the abnormal group. A high serum type IV collagen concentration was also associated with shorter OS (P=0.0039) and RFS (P=0.00294) (**Fig. 1B**).

Factors that were associated with OS in univariate analysis were tumor diameter, vascular invasion, stage, serum albumin, serum AFP, serum AFP L3, serum DCP, serum HA, and serum type IV collagen (**Table 4**). These factors were included in the multivariate analysis. The multivariate analysis revealed that high preoperative serum type IV collagen levels independently predicted poor OS (**Table 5** hazard ratio (HR) 1.003, 95 percent confidence interval 1.000 to 1.006; P=0.0326). Factors associated with RFS in univariate analysis included tumor diameter, tumor number, stage, serum albumin, serum DCP, serum HA, and serum type IV collagen (**Table 6**). These factors were included in the multivariate analysis. The analysis revealed that high preoperative serum HA levels and tumor diameter independently predicted poor RFS (**Table 7**; HR 1.001, 95 % confidence interval 1.000 to 1.002; P=0.0121, HR 1.010, 95 % confidence interval 1.003 to 1.016; P=0.0035, respectively).

Table 5 Multivariate analysis for overall survival rate

| Multivariate analysis (OS) | *P* value | Hazard ratio | 95% CI |
|---|---|---|---|
| Tumor diameter (mm) | 0.2145 | 1.005 | 0.997–1.014 |
| Vascular invasion | 0.3361 | 1.581 | 0.622–4.018 |
| Stage | 0.3899 | 1.501 | 0.595–3.785 |
| Alb (g/dL) | 0.1644 | 0.630 | 0.328–1.209 |
| AFP (ng/mL) | 0.2723 | 1.000 | 1.000–1.000 |
| L3 (%) | 0.0730 | 1.012 | 0.999–1.025 |
| DCP (mAU/mL) | 0.0400 | 1.000 | 1.000–1.000 |
| Hyaluronic acid (ng/mL) | 0.9746 | 1.000 | 0.999–1.001 |
| Type IV collagen (ng/mL) | 0.0057 | 1.003 | 1.000–1.006 |

Table 6 Univariate analysis for recurrence-free survival rate

| Univariate analysis (RFS) | *P* value | Hazard ratio | 95% CI |
|---|---|---|---|
| Age | 0.8447 | 0.998 | 0.985–1.019 |
| Tumor diameter (mm) | 0.0001 | 1.009 | 1.004–1.012 |
| Vascular invasion (−) | 0.0130 | 0.654 | 0.978–1.403 |
| Number of tumor | 0.2933 | 0.827 | 1.082–1.743 |
| Stage | 0.3662 | 1.164 | 1.123–1.518 |
| T-bil (mg/dL) | 0.0736 | 1.221 | 0.981–1.520 |
| Alb (g/dL) | 0.0239 | 0.693 | 0.550–0.920 |
| PT (%) | 0.8279 | 0.999 | 1.000–1.000 |
| ICG15R (%) | 0.2234 | 1.009 | 0.998–1.022 |
| AFP (ng/mL) | 0.1077 | 1.000 | 1.000–1.000 |
| L3 (%) | 0.2650 | 1.006 | 0.994–1.014 |
| DCP (mAU/mL) | 0.0129 | 1.000 | 1.000–1.000 |
| Hyaluronic acid (ng/mL) | 0.0016 | 1.001 | 1.000–1.002 |
| Type IV collagen (ng/mL) | 0.0360 | 1.001 | 1.000–1.002 |
| Type IV collagen 7s (ng/mL) | 0.1233 | 1.044 | 0.987–1.102 |

Table 7 Multivariate analysis for recurrence-free survival rate

| Multivariate analysis (RFS) | *P* value | Hazard ratio | 95% CI |
|---|---|---|---|
| Tumor diameter (mm) | 0.0123 | 1.007 | 1.007–1.020 |
| Vascular invasion (−) | 0.3608 | 1.020 | 0.602–1.726 |
| Alb (g/dL) | 0.4849 | 0.630 | 0.387–1.026 |
| DCP (mAU/mL) | 0.1667 | 1.000 | 1.000–1.000 |
| Hyaluronic acid (ng/mL) | 0.0376 | 1.001 | 1.000–1.002 |
| Type IV collagen (ng/mL) | 0.3192 | 1.001 | 1.000–1.002 |

## Discussion

This study revealed that high preoperative serum HA levels and type IV collagen levels predict RFS and OS after hepatectomy for HCC. Specifically, a high HA level independently predicted RFS. Anatomical resection of HCC was performed more often in the normal HA group than in the abnormal group. Anatomical resection of HCC was introduced by Makuchi[8], who reported that this procedure improved the survival rate by reducing local recurrence. However, some recent reports did not show an improvement in recurrence-free survival following anatomical resection of HCC[9]. These findings are controversial. Prospective randomized control studies are required.

High type IV collagen levels independently predicted OS. These markers are very important for hepatic resection for HCC. Interestingly, the effects of tumor number, vascular invasion, stage, and some tumor markers were detected by univariate analysis, but these effects were not detected in multivariate analysis.

We will now review HA and type IV collagen and discuss the mechanism of how these factors affect OS and

RFS.

HA is a high molecular weight ($10^6$–$10^7$ Da) glycosaminoglycan polymer that is composed of repeating disaccharides: β1, 3 N-acetyl glucosaminyl-β1, 4 glucuronide[10]. Serum HA levels indicate the severity of underlying chronic liver disease[11–13]. Serum HA levels reflect liver fibrosis more closely than ICG R15[14]. Moreover, because portal pressure is correlated with hepatic fibrosis[15], serum HA may be a biochemical marker of portal pressure and a predictor of postoperative hepatic failure[14]. Serum HA concentration reflects the function of hepatic sinusoids and is a sensitive marker of liver endothelial cell dysfunction after liver ischemia[16]. Some reports have revealed that HA is a useful marker for the prediction of post-hepatectomy hepatic functional reserve[15,17,18]. HA is involved in tumorigenesis[10]. HA receptors such as CD44 (cluster of differentiation 44) are implicated in the cell signaling cascades associated with cancer initiation and progression[19]. The CD44 receptor is a widely distributed transmembrane glycoprotein that plays a critical role in malignant cell activities[10]. CD44 has some isoform types. HA/CD44 and HA/CD44v interactions regulate stem cell migration and homing[20]. CD44s have been implicated in promoting epithelial-mesenchymal transition (EMT)[21]. These mechanisms may contribute to shortening the RFS at a high HA level. Type IV collagen is common in a variety of tumors[22–26]. A positive correlation between type IV collagen expression and tumor metastasis has been reported in several studies[28]. Native type IV collagen induced an EMT-like process, increasing matrix metalloproteinase (MMP)-2, focal adhesion kinase (FAK) and nuclear factor kappa-light-chain-enhancer of activated B cells (NF-κB) activation, cell migration, and invasion in MCF10A human mammary epithelial cells[29]. In experiments, shRNA targeting type IV collagen gene-transfected cells formed significantly fewer and smaller lung nodules[29].

## Conclusion

Preoperative examination of serum hyaluronic acid levels and type IV collagen levels are imperative for the hepatic resection of hepatocellular carcinoma because these markers are significantly associated with liver function and prognosis.

**Conflict of Interest:** None.

## References

1. El-Serag HB: Hepatocellular carcinoma. N Engl J Med 2011; 365: 1118–1127.
2. Forner A, Llovet JM, Bruix J: Hepatocellular carcinoma. Lancet 2012; 379: 1245–1255.
3. Colecchia A, Schiumerini R, Cucchetti A, Cescon M, Taddia M, Marasco G, Festi D: Prognostic factors for hepatocellular carcinoma recurrence. World J Gastroenterol 2014; 20: 5935–5950.
4. Mima K, Beppu T, Ishiko T, Chikamoto A, Nakagawa S, Hayashi H, Watanabe M, Sakamaki K, Baba H: Preoperative serum hyaluronic acid level as a prognostic factor in patients undergoing hepatic resection for hepatocellular carcinoma. Br J Surg 2014; 101: 269–276.
5. Rostami S, Parsian H: Hyaluronic Acid: From Biochemical Characteristics to its Clinical Translation in Assessment of Liver Fibrosis. Hepat Mon 2013; 13: e13787.
6. Chen W, Rock JB, Yearsley MM, Ferrell LD, Frankel WL: Different collagen types show distinct rates of increase from early to late stages of hepatitis C-related liver fibrosis. Hum Pathol 2014; 45: 160–165.
7. Liver Cancer Study Group of Japan: The general rules for the clinical and pathological study of primary liver cancer. Jpn J Surg 1989; 19: 98–129.
8. Makuuchi M: Surgical treatment for HCC—Special reference to anatomical Resection. Int J Surg 2013; 11 [Suppl 1]: S47–S49.
9. Koo Jeong Kang, Keun Soo Ahn: Anatomical resection of hepatocellular carcinoma: A critical review of the procedure and its benefits on survival. World J Gastorentrol 2017; 23: 1139–1146.
10. Arpicco S, Milla P, Stella B, Dosio F: Hyaluronic acid conjugates as vectors for the active targeting of drugs genes and nanocomposites in cancer treatment. Molecules 2014; 19: 3193–3230.
11. Murawaki Y, Ikuta Y, Koda M, Nishimura Y, Kawasaki H: Clinical significance of serum hyaluronan in patients with chronic viral liver disease. J Gastroenterol Hepatol 1996; 11: 459–465.
12. Engstrom-Laurent A, Loof L, Nyberg A, Schroder T: Increased serum levels of hyaluronate in liver disease. Hepatology 1985; 5: 638–642.
13. van Leeuwen DJ, Howe SC, Scheuer PJ, Sherlock S: Portal hypertension in chronic hepatitis: relationship to morphological changes. Gut 1990; 31: 339–343.
14. Yachida S, Wakabayashi H, Okano K, Suzuki Y: Prediction of posthepatectomy hepatic functional reserve by serum hyaluronate. Br J Surg 2009; 96: 501–508.
15. Krogsgaard K, Gluud C, Henriksen JH, Christoffersen P: Correlation between liver morphology and portal pressure in alcoholic liver disease. Hepatology 1984; 4: 699–703.
16. Idobe Y, Murawaki Y, Ikuta Y, Koda M, Kawasaki H: Post-prandial serum hyaluronan concentration in patients with chronic liver disease. Intern Med 1998; 37: 568–575.
17. Nanashima A, Tobinaga S, Abo T, Nonaka T, Takeshita H, Hidaka S, Sawai T, Nagayasu T: Reducing the incidence of post-hepatectomy hepatic complications by preoperatively applying parameters predictive of liver function. J Hepatobiliary Pancreat Sci 2010; 17: 871–878.
18. Nanashima A, Abo T, Arai J, Matsumoto H, Kudo T, Nagayasu T: Functional liver reserve parameters predictive for posthepatectomy complications. J Surg Res 2013; 185: 127–135.
19. Ghosh SC, Neslihan Alpay S, Klostergaard J: CD44: a validated target for improved delivery of cancer therapeutics. Expert Opin Ther Targets 2012; 16: 635–650.

20. Williams K, Motiani K, Giridhar PV, Kasper S: CD44 integrates signaling in normal stem cell, cancer stem cell and (pre)metastatic niches. Exp Biol Med (Maywood) 2013; 238: 324–338.
21. Brown RL, Reinke LM, Damerow MS, Perez D, Chodosh LA, Yang J, Cheng C: CD44 splice isoform switching in human and mouse epithelium is essential for epithelial-mesenchymal transition and breast cancer progression. J Clin Invest 2011; 121: 1064–1074.
22. Kainz C, Kohlberger P, Tempfer C, Sliutz G, Gitsch G, Reinthaller A, Breitenecker G: Prognostic value of CD44 splice variants in human stage III cervical cancer. Eur J Cancer 1995; 31A: 1706–1709.
23. Zhu GG, Risteli L, Makinen M, Risteli J, Kauppila A, Stenback F: Immunohistochemical study of type I collagen and type I pN-collagen in benign and malignant ovarian neoplasms. Cancer 1995; 75: 1010–1017.
24. Kauppila S, Stenback F, Risteli J, Jukkola A, Risteli L: Aberrant type I and type III collagen gene expression in human breast cancer in vivo. J Pathol 1998; 186: 262–268.
25. Nasser NJ: Heparanase involvement in physiology and disease. Cell Mol Life Sci 2008; 65: 1706–1715.
26. Huijbers IJ, Iravani M, Popov S, Robertson D, Al-Sarraj S, Jones C, Isacke CM: A role for fibrillar collagen deposition and the collagen internalization receptor endo180 in glioma invasion. PLoS One 2010; 5: e9808.
27. Burnier JV, Wang N, Michel RP, Hassanain M, Li S, Lu Y, Metrakos P, Antecka E, Burnier MN, Ponton A, Gallinger S, Brodt P: Type IV collagen-initiated signals provide survival and growth cues required for liver metastasis. Oncogene 2011; 30: 3766–3783.
28. Espinosa Neira R, Salazar EP: Native type IV collagen induces an epithelial to mesenchymal transition-like process in mammary epithelial cells MCF10A. Int J Biochem Cell Biol 2012; 44: 2194–2203.
29. Chen SY, Lin JS, Yang BC: Modulation of tumor cell stiffness and migration by type IV collagen through direct activation of integrin signaling pathway. Arch Biochem Biophys 2014; 555–556C: 1–8.

(Received, December 26, 2017)
(Accepted, March 6, 2018)