Clinical Utility of Histological and Radiological Evaluations of Tumor Necrosis for Predicting Prognosis in Pancreatic Cancer

Radiological tumor necrosis reflect prognosis

Masashi Kudo, MD¹,²,⁵, Tatsushi Kobayashi, MD³, Naoto Gotohda, MD, PhD², Masaru Konishi, MD², Shinichiro Takahashi, MD, PhD², Shin Kobayashi, MD, PhD², Motokazu Sugimoto, MD, PhD², Satoshi Okubo, MD, PhD², John Martin, PhD⁴, Horacio Cabral, PhD⁴, Genichiro Ishii, MD, PhD¹,⁵, Motohiro Kojima, MD, PhD¹*

¹ Division of Pathology, Exploratory Oncology Research & Clinical Trial Center, National Cancer Center, Kashiwa, Japan
² Department of Hepatobiliary and Pancreatic Surgery, National Cancer Center Hospital East, Kashiwa, Japan
³ Department of Diagnostic Radiology, National Cancer Center Hospital East, Kashiwa, Japan
⁴ Department of Bioengineering, Graduate School of Engineering, The University of Tokyo
Course of Advanced Clinical Research of Cancer, Juntendo University Graduate School of Medicine, Tokyo, Japan

• Corresponding author:
Motohiro Kojima, MD, PhD
Division of Pathology, Exploratory Oncology Research & Clinical Trial Center
National Cancer Center
6-5-1, Kashiwa-no-ha, Kashiwa, Chiba 277-8577, Japan
E-mail: mokojima@east.ncc.go.jp
Telephone: +81-4-7133-1111
Fax: +81-4-7131-4724

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Masashi Kudo designed the study and wrote the initial draft of the manuscript. Motohiro Kojima contributed to analysis and interpretation of data and assisted in the preparation of the manuscript. Other co-authors contributed to data collection and interpretation and critically reviewed the manuscript. All authors approved the final version of the manuscript and agree to be accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved.
Abstract:

Objectives:

Tumor necrosis is often found in pancreatic ductal adenocarcinoma (PDAC). Objective histological assessment and adequate radiological detection of necrosis could be used as biomarkers for therapeutic decision. However, standardized clinical utility of necrosis remains unknown. Here, we aimed to determine the prognostic potential of histological and radiological evaluations of necrosis.

Methods:

We investigated histological necrosis in 221 patients, who underwent surgery for PDAC, and classified its size as small (≤5 mm) or large (>5 mm). We also evaluated poorly enhanced areas on preoperative computed tomography to assess their ability for predicting histological necrosis and postoperative prognosis.

Results:

Tumor necrosis was found in 115 patients (52%), and was related to tumor area, lymph node metastasis, lympho-vascular invasion. Size of necrosis was significantly associated with tumor area, perimeter of necrosis, circularity of necrosis, number of ruptured cancer glands, and presence of collagen bundle ($P < 0.05$ for all). Both presence of necrosis and their size were strongly correlated to postoperative prognosis. Patients with
poorly enhanced areas showed worse prognosis ($P < 0.01$).

**Conclusion:**

Our findings underline the capacity of histological and radiological assessment of tumor necrosis for prognosis prediction in PDAC.

**Keywords:**

biomarker, CT, necrosis, pancreatic carcinoma, prognosis, pancreas
Abbreviations:

CI, confidence interval; CT, computed tomography; DSS, disease-specific survival; HTN, histological tumor necrosis; PDAC, pancreatic ductal adenocarcinoma; PEA, poorly enhanced areas; RFS, relapse-free survival; RR, risk ratio; TME, tumor microenvironment
Introduction

Pancreatic ductal adenocarcinoma (PDAC) is the third leading cause of cancer-related death and more than 53,000 new cases and over 41,000 deaths occur annually in the United States.\(^1\) Despite recent developments in diagnostic instruments and therapeutic regimens, the mortality rate has not shown any obvious improvement for decades. Thus, consecutive histological investigations and use of non-invasive imaging techniques are still important for understanding the biology and predicting the prognosis of PDAC.

Several histological findings, such as tumor size, lymph node metastasis, tumor grade, lymphatic invasion, venous invasion, and neural invasion, have been reported as biomarkers for predicting postoperative prognosis in patients with PDAC.\(^2-8\) However, most previously reported histological prognostic factors, such as tumor grade, vascular invasion, and neural invasion, have been difficult to detect non-invasively on preoperative imaging. Histological tumor necrosis (HTN), which is found in more than 60% of PDAC cases, has also been linked with a poor prognosis,\(^9,10\) and the poorly enhanced areas (PEAs) observed on computed tomography (CT) images could be indicators of HTN.\(^11\) Thus, if PEAs could predict the histological findings and postoperative prognosis early and noninvasively, the therapeutic strategy could be considered before surgery. Unfortunately, because HTN has a spectrum of origins and morphological changes within the varying tumor microenvironment (TME),\(^12\) such analysis could be difficult. In particular, since there are no uniformly applied diagnostic criteria of histological necrosis for PDAC, the morphological and histological details of necrosis in PDAC are still unclear.

Thus, the aim of this study was to investigate the morphological and histological events of tumor necrosis in PDAC and elucidate the clinical and histological findings.
associated with the postoperative prognosis. Based on these observations, the diagnostic
accuracy of PEAs for detecting HTN was then determined, with a particular focus on
validating the utility of PEAs as a prognostic factor.

Materials and Methods

Patients

From 2011 to 2017, a total of 281 patients underwent pancreatectomy for PDAC at the
National Cancer Hospital East, Japan. Of the 281 patients, 60 were excluded for the
following reasons: preoperative chemotherapy or chemoradiotherapy (n = 29); recurrent
PDAC in the remnant pancreas (n = 18); and inconsistent patient information (n = 13).
The remaining 221 patients who underwent pancreatectomy for PDAC were
investigated in this study. Single agent TS-1 was given as standard adjuvant
chemotherapy except for patients who received systemic chemotherapy due to early
recurrence or who refused standard adjuvant chemotherapy. This retrospective study
was approved by the National Cancer Ethical Review Board, and informed consent for
the use of medical records was obtained from each of the patients (reference 2017-328).

Histological analysis of surgical specimens

Surgical specimens of 5-mm thickness were cut for PDAC. Then, 4-µm-thick sections
were stained with hematoxylin-eosin. The hematoxylin-eosin slides were scanned using
the NanoZoomer 2.0 system (Hamamatsu Photonics, Hamamatsu, Japan), and
morphological and histological analysis was performed by two investigators (M. Kudo,
M. Kojima, one with more than 10 and one with more than 22 years of experience
examining pancreatic histology) without any radiological and clinical information. The
definition of necrosis was based on previous reports. In\textsuperscript{9,10} Intraluminal necrosis without extending to the stroma was not regarded as necrosis (Figure 1A). Both confluent cell death in an invasive area (visible at an objective lens magnification of x4) and smaller areas of necrosis (often accompanied by ruptured cancer glands) were regarded as necrosis. Many confluent cell deaths in an invasive area included ruptured cancer glands. On the other hand, all smaller areas of necrosis had ruptured cancer glands. Therefore, the size of necrosis was measured and classified into small (maximum diameter <5 mm, Figure 1B) and large necrosis (maximum diameter >5 mm, Figure 1C) to understand the size-dependent features of necrosis. Most areas of HTN showed a mixture of coagulation necrosis and liquefactive necrosis. Coagulation necrosis, which could be recognized as lesions with preserved cell outlines without nuclei, was easily regarded as necrosis. In contrast, liquefactive necrosis is difficult to distinguish from protein concretion or debris from a ruptured cancer gland. Therefore, histological necrosis was assessed only as lesions including preserved cell outlines without nuclei. The measurement of tumor area was based on previous reports.\textsuperscript{13} The circularity of necrosis was calculated as follows: $4 \times (\text{area of necrosis}) / (\text{perimeter of necrosis})^2$. The pathological stage of patients was defined according to the TNM stage in the 8\textsuperscript{th} Union for International Cancer Control staging system.

\textbf{Preoperative diagnostic imaging}

All CT imaging was performed using a multi-detector-row CT scanner at our institute (\textit{Aquilion ONE}, Canon Medical Systems Corporation, Ohtawara, Japan). All patients underwent precontrast and dynamic contrast-enhanced CT images. After precontrast CT scans, portal-dominant phase images of dynamic CT were obtained starting 60 seconds
after the beginning of the intravenous bolus injection (3 mL/s) of 100 mL of iodized contrast medium at 600 mL/kg. The scanning parameters were 2-mm slice thickness, tube voltage of 120 kV, and auto mA. The portal-dominant phase images were used in the analyses to detect HTN (Figure 1D). All magnetic resonance imaging studies were performed using one of the two 3.0-T scanners at our institute (Achieva or Ingenia, Philips Medical Systems, Amsterdam, The Netherlands), and T2-weighted images were used in the analyses (Figure 1E).

**Image analysis**

Poorly enhanced areas of PDAC were defined as obviously hypoattenuated regions showing Hounsfield unit values <45 located in tumor regions on the portal-dominant phase of dynamic CT imaging. Regions that were determined to be obvious dilatations of the distal pancreatic duct owing to obstruction by tumor invasion were excluded. Extreme hyperintensity regions on T2-weighted magnetic resonance imaging were evaluated to distinguish retention cysts from necrosis. A single investigator (M. Kudo), under supervision of an experienced radiologist (T. Kobayashi), calculated hounsfield unit values on the portal-dominant phase of dynamic CT imaging without any information about the histological data and reached a decision by consensus.

**Statistical analysis**

Categorical variables were evaluated using the chi-squared test and are presented as numbers and percentages, whereas continuous variables were evaluated using the Mann-Whitney U test and are presented as medians and range. The correlation between the maximum diameter of necrosis and the number of ruptured cancer glands was
assessed by Spearman’s rank correlation coefficient. Postoperative relapse-free survival (RFS) and disease-specific survival (DSS) rates were calculated by the Kaplan-Meier method, and these differences were compared by the log-rank test. Univariate and multivariate analyses for prognostic factors were performed using a Cox proportional hazards model. The factors that were found to be significant on univariate analyses were included in the multivariate analysis and are presented as the risk ratio (RR) and 95% confidence interval (CI). The cut-off value of PEA s was determined according to the areas under the receiver operating characteristic curves to assess the relationship between PEA s and survival. The correlation between histological necrosis and PEA s was analyzed by chi-squared tests, and the sensitivity, specificity, positive predictive value, and negative predictive value of PEA s were evaluated for accuracy. All P values were based on two-sided statistical tests, and the significance level was set at 0.05. Statistical analyses were performed using JMP (version 12.0.10; SAS Institute, Cary, NC).

Results

Patients’ characteristics

The clinical and histological characteristics of the patients are shown in Supplemental Table 1. The median age was 70 (range, 43-87) years, and 133 (60%) of the 221 patients were male. The tumor was located in the head of the pancreas in 149 patients (67%), the body or tail of the pancreas in 69 patients (31%), and the whole pancreas in 3 patients (1%). All patients were diagnosed with potentially resectable PDAC on preoperative CT. Histological tumor necrosis was found in 115 (52%) patients; 65 patients (29%) had small necrosis, and 50 patients (23%) had large necrosis.
Histological findings of necrosis

Correlations between histological findings and necrosis are shown in Table 1. Tumor area, lymph node metastasis, lymphatic invasion, and venous invasion were significantly associated with the presence of HTN. Large necrosis was associated with a large necrotic area, long perimeter of necrosis, lower circularity of necrosis, large number of ruptured cancer glands, and presence of collagen bundles. Debris and mucous substances from ruptured cancer glands were often mixed with necrosis, but collagen bundles were predominantly seen in large necrosis. Small necrosis was often accompanied by a single or few ruptured cancer glands with neutrophils, as reported previously. In large necrosis, dense collagen bundles were often found within necrotic areas, and some of these lesions formed a fibrotic focus, as described previously. Most areas of large necrosis also included scattered ruptured cancer glands, predominantly in the peripheral area of necrosis. There was a significant correlation between the size of necrosis and the number of ruptured cancer glands (Supplemental Figure 1; \( r = 0.744; P < 0.001 \)).

Prognostic significances according to the size of necrosis

The prognostic significance according to the size of necrosis is shown in Figure 2. The median RFS and DSS in the 221 patients were 17.1 and 55.6 months, respectively. Patients with necrosis showed a poor clinical prognosis, both RFS and DSS, and size-dependent deterioration of the clinical prognosis was also seen (Figure 2A, B). Next, the clinical prognosis was analyzed in pathological T2 (primary tumor size >2 cm but not >4 cm) patients to reduce data confounding, and similar results to those including all
patients were obtained (Figure 2C, D).

Risk analysis of prognostic factors associated with DSS in patients with PDAC

Univariate and multivariate risk analyses of prognostic factors associated with DSS are shown in Table 2. On multivariate risk analyses, Cox proportional hazards models identified 3 independent prognostic factors: lymph node metastasis (RR, 2.34; 95% CI, 1.33-4.35; $P = 0.003$); venous invasion (RR, 5.34; 95% CI, 1.17-95.65; $P = 0.026$); and histological necrosis (RR, 3.16; 95% CI, 1.91-5.44; $P < 0.001$).

Correlation between PEAs on preoperative CT and histological necrosis

To detect histological necrosis before surgery, the correlation of PEAs on the portal-dominant phase of dynamic CT and histological necrosis was analyzed (Table 3). There was a significant correlation between PEA-positive and the presence of histological necrosis ($P < 0.001$). The sensitivity, specificity, positive predictive value, and negative predictive value of PEAs for detecting histological necrosis were 65.2%, 78.3%, 76.5%, and 67.5%, respectively. Furthermore, there was a significant correlation between PEAs and size of necrosis ($P < 0.001$). Based on the histological analysis, the causes of false-positive PEAs were thought to be large neoplastic glands in 14 patients (14%), dilated pancreatic ducts in 3 patients (3%), and steatosis in 1 patient (1%) (Supplemental Table 2).

Prognostic significance according to PEAs

Kaplan-Meier estimates of RFS and DSS according to positive or negative for PEAs are shown in Figure 3. The 3-year RFS was significantly higher in the PEA-negative group
than in the PEA-positive group (48.0% vs. 19.7%, *P* < 0.001). The 5-year DSS was also significantly higher in the PEA-negative group than in the PEA-positive group (64.5% vs. 25.2%, *P* < 0.001). Kaplan-Meier curves of RFS and DSS were significantly different between the two groups.

Discussion

Based on a comprehensive analysis of the morphological, histological, and radiological findings of PDAC, it was possible to correlate histological and radiological observations of necrosis and size-dependent histological alterations in PDAC as a prognostic factor. The clinical utility of PEAs, which represent histological necrosis, as a prognostic factor was also shown. The present findings provide a clear approach for translating histological or radiological observations into predicting patient survival.

Based on the present findings, size-dependent histological alteration of necrosis in PDAC is important (Figure 4). Small necrosis was accompanied by ruptured cancer glands and often showed debris and mucous substances extravasating from the ruptured cancer glands. These observations suggest that necrosis in PDAC is initiated and formed in close association with ruptured cancer glands. Interspersed debris and mucous substances from the ruptured cancer glands can trigger necrosis formation in PDAC. Next, the number of ruptured cancer glands was strongly correlated with the maximum diameter of necrosis (Supplemental Figure 1), and large necrosis was thought to be formed by the fusion of areas of small necrosis. During this step, the outline of necrosis became irregular, which was reflected by lower circularity of areas of large necrosis, and collagen bundle formation was also accelerated during its growth. Histological tumor necrosis was associated with vascular invasion and lymph node metastasis (Table
1. Previously, the physiological abnormality of hypoxia was found to correlate with tumor necrosis, lymphatic invasion, and other characteristics of this biological progression. Recently, the addition of a TME re-engineering therapy, which aims to modulate the TME to alleviate hypoxia, when combined with chemoradiotherapy, increased downstaging rates for resection compared to historical studies in locally advanced PDAC and is now being considered to alleviate immunosuppression. Additionally, low pH- or hypoxia-targeting strategies of TME re-engineering therapies could further improve the selectivity of TME re-engineering therapies to tissues prone to necrosis. Thus, in addition to the potential for radiological detection of PEAs for predicting patients’ prognosis, it might also enable selection of patients with unresectable PDAC, in which histological confirmation is impossible, for hypoxia-alleviating TME re-engineering therapy. In addition, several studies have reported that neoadjuvant therapy improved the survival of patients with potentially resectable PDAC. However, predictors of therapeutic effect and prognosis are still elusive for patients who receive neoadjuvant therapy. Therefore, PEAs may also become a novel biomarker for predicting the effectiveness of neoadjuvant therapy in PDAC, and distinct therapeutic strategies can be considered based on PEAs in the future. Therefore, various studies focused on histological and radiological necrosis as a method to provide basic information for medical innovation in PDAC patients are needed.

Besides the clear observations of histological and radiological necrosis, and their correlation with prognosis, the limitations of the present study are also worth noting. First, this was a retrospective, single-institute study. However, the sample size was large enough to perform a reliable analysis. Second, the accuracy of PEAs for detecting necrosis was not high enough to definitively determine preoperative therapy. However,
the present study showed similar diagnostic accuracy to a previous report. Further investigation is needed to establish more precise approaches for the detection of histological necrosis before surgery. Third, there was a large number of patients who underwent chemotherapy or chemoradiotherapy before surgery. Because neoadjuvant therapy might have an impact on HTN and PEAs, its effect was excluded from the present analysis. The impact of chemotherapy or chemoradiotherapy on tumor necrosis would be the subject of future studies.

In conclusion, the prognosis of PDAC is associated with histological necrosis, which is thought to be initiated by ruptured cancer glands. PEAs on preoperative CT could consistently detect histological necrosis, and their presence was a prognostic factor for resectable PDAC. The clinical utility of histological necrosis and PEAs should be further investigated in the future.

Acknowledgments:

This study protocol conformed to the ethical guidelines of the 1975 Declaration of Helsinki and was approved by the institutional review board of the National Cancer Center, and informed consent was obtained from all patients. The authors would like to thank all participating patients and their families who made this study possible. The datasets generated and/or analyzed during the present study are available from the corresponding author upon reasonable request.
References


5. Shubert CR, Bergquist JR, Groeschl RT, et al. Overall survival is increased among stage III pancreatic adenocarcinoma patients receiving neoadjuvant chemotherapy compared to surgery first and adjuvant chemotherapy: An


Figure legends

FIGURE 1 The definition of the size of necrosis in pancreatic cancer according to maximum diameter and diagnostic images of a necrotic portion. The regions of tumor and necrosis are encircled by blue and red lines, respectively. Tumor necrosis is absent or limited in a cancer gland (A). Necrosis with maximum diameter less than 5 mm is defined as small necrosis (B). Necrosis with maximum diameter greater than 5 mm is defined as large necrosis (C). Portal-dominant phase dynamic CT imaging shows a PEA (arrow) (D), and the signal intensity of the same area is slightly high on T2-weighted MRI (arrow) (E). The same area of the resected specimen shows a necrotic portion (arrow) (F). Yellow and blue arrow heads show the portal vein and duodenum, respectively.

FIGURE 2 Kaplan-Meier estimates of relapse-free survival (A) and disease-specific survival (B) of the 239 patients with pancreatic cancer according to the size of necrosis. Kaplan-Meier estimates of relapse-free survival (C) and disease-specific survival (D) of Union for International Cancer Control pathological T2 with pancreatic cancer according to the size of necrosis. All Kaplan-Meier curves are significantly different among the three groups according to the size of necrosis.

FIGURE 3 Kaplan-Meier estimates of relapse-free survival (A) and disease-specific survival (B) of the 221 patients according to the presence of PEAs are significantly different between the two groups. PEA, poorly enhanced area
FIGURE 4 Schematic of size-dependent histological alterations of necrosis in pancreatic ductal adenocarcinoma. First, the cancer cell necrosis occurs in the cancer glands. Second, debris and mucous substance are extravasated out of the ruptured cancer glands. The necrosis is accompanied by lymphatic invasion and venous invasion, but collagen bundles are rarely seen in small necrosis. Third, the areas of necrosis become conglutinated and grow into an irregular shape. Large necrosis is accompanied by many ruptured cancer glands and collagen bundles. Finally, lymph node and liver metastases develop.

Supplemental Figure 1 Correlation between maximum diameter of necrosis and number of ruptured cancer gland. There was a significant correlation between the maximum diameter of necrosis and number of ruptured cancer gland. Spearman’s rank correlation coefficient and P value were calculated ($r=0.744$, $P<0.001$).
List of Supplemental Information:

Supplemental Table 1 Clinical and histological characteristics

Supplemental Figure 1 Correlation between the maximum diameter of necrosis and number of ruptured cancer glands

Supplemental Table 2 Histological reasons for false-positive PEAs
FIGURE 1

No necrosis

Small necrosis

Large necrosis

Portal-phase CT imaging

T2-weighted MRI

Resected specimen
FIGURE 2

**A**

Recurrence rate (%)

No. at risk:
- Absent: 106, 78, 55, 36, 27, 21
- Small: 65, 31, 17, 11, 6, 3
- Large: 50, 16, 5, 3, 2, 2

Time (months)

**B**

Survival rate (%)

No. at risk:
- Absent: 106, 94, 71, 48, 33, 25
- Small: 65, 52, 30, 22, 11, 4
- Large: 50, 29, 15, 8, 4, 3

**C**

Recurrence rate (%)

No. at risk:
- Absent: 52, 39, 27, 17, 14, 12
- Small: 43, 21, 11, 8, 3, 2
- Large: 35, 10, 4, 3, 2, 2

Time (months)

**D**

Survival rate (%)

No. at risk:
- Absent: 52, 47, 36, 26, 17, 13
- Small: 43, 34, 19, 15, 7, 3
- Large: 35, 19, 7, 4, 2, 2

**P**
- Absent vs. Small: $P < 0.001$
- Small vs. Large: $P = 0.002$
- Absent vs. Large: $P = 0.005$
- Absent vs. Small: $P = 0.012$
- Small vs. Large: $P = 0.013$
FIGURE 3

PEA (−)  PEA (+)  \[ P < 0.001 \]

Recurrence rate (%)

No. at risk:
PEAs (−)  123  81  55  37  28  19
PEAs (+)  98  44  21  12  7  6

Survival rate (%)

No. at risk:
PEAs (−)  123  104  76  55  36  23
PEAs (+)  98  72  39  22  11  8
Maximum diameter of necrosis

Number of ruptured cancer glands

$r = 0.744$

$P < 0.001$

Supplementary Figure 1
<table>
<thead>
<tr>
<th>Presence of necrosis</th>
<th>Size of necrosis</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Tumor area (mm²)</strong></td>
<td><strong>Small necrosis</strong></td>
</tr>
<tr>
<td>(n = 106)</td>
<td>(n = 65)</td>
</tr>
<tr>
<td>(73 (10-277)</td>
<td>106 (26-315)</td>
</tr>
<tr>
<td>(140 (26-363))</td>
<td>173 (64-363)</td>
</tr>
<tr>
<td><strong>P</strong></td>
<td>&lt;0.001*</td>
</tr>
</tbody>
</table>

**Histological findings of necrosis**

- **Necrotic area (mm²)**
  - Absent: 14.8 (0.6-182.7)
  - Present: 5.7 (0.6-27.7)
  - P: <0.001*

- **Necrotic area / tumor area (%)**
  - Absent: 13.8 (0.5-81.8)
  - Present: 6.0 (0.5-27.0)
  - P: <0.001*

- **Perimeter (mm)**
  - Absent: 13.0 (2.8-65.4)
  - Present: 8.3 (2.8-15.0)
  - P: <0.001*

- **Circularity**
  - Absent: 0.66 (0.30-0.93)
  - Present: 0.71 (0.33-0.93)
  - P: <0.001*

- **Number of areas of necrosis**
  - Absent: 3 (1-22)
  - Present: 2 (1-22)
  - P: 0.104*

- **Number of ruptured cancer glands**
  - Absent: 6 (0-36)
  - Present: 3 (0-17)
  - P: <0.001*

- **Neutrophil infiltration**
  - Absent: 80 (70)
  - Present: 35 (30)
  - P: 0.929†

- **Collagen bundle**
  - Absent: 69 (60)
  - Present: 46 (40)
  - P: <0.001†

- **Lymph node metastasis**
  - Absent: 51 (48)
  - Present: 55 (52)
  - P: 0.224†

- **Lymphatic invasion**
  - Absent: 34 (32)
  - Present: 72 (68)
  - P: 0.400†

- **Venous invasion**
  - Absent: 13 (12)
  - Present: 93 (88)
  - P: 0.074†

- **Neural invasion**
  - Absent: 6 (6)
  - Present: 100 (94)
  - P: 0.124†

Data expressed as n (%) or median (range). There were significant differences in terms of the tumor area, lymph node metastasis, lymphatic invasion, and venous invasion according to the presence of necrosis. Necrotic area, necrotic area / tumor area, perimeter, and number of ruptured cancer forming tubules were significantly increased in areas of large necrosis compared to areas of small necrosis. However, circularity of the necrosis was significantly decreased in areas of large necrosis compared to areas of small necrosis.

*Mann-Whitney U-tests or †chi-squared tests.
<table>
<thead>
<tr>
<th>Variable</th>
<th>Median DSS (months)</th>
<th>Univariate analysis*</th>
<th>Multivariate analysis*</th>
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<tr>
<td></td>
<td></td>
<td>RR (95% CI)</td>
<td>P</td>
</tr>
<tr>
<td>Age (y)</td>
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<tr>
<td>≥75 (n = 57)</td>
<td>56.4</td>
<td>1.30 (0.76-2.13)</td>
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<tr>
<td>&lt;75 (n = 164)</td>
<td>43.4</td>
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<td>Sex</td>
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<tr>
<td>Male (n = 133)</td>
<td>54.7</td>
<td>0.97 (0.62-1.54)</td>
<td>0.886</td>
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<tr>
<td>Female (n = 88)</td>
<td>61.9</td>
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<td>Body mass index (kg/m^2)</td>
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<td>≥25 (n = 33)</td>
<td>56.4</td>
<td>1.08 (0.57-1.89)</td>
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<td>&lt;25 (n = 188)</td>
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<td>≥37 (n = 146)</td>
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<td>2.23 (1.34-3.93)</td>
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<td>Lymph node metastasis</td>
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<tr>
<td>Absence (n = 78)</td>
<td>-</td>
<td>3.47 (2.06-6.23)</td>
<td>&lt;0.001</td>
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<tr>
<td>Presence (n = 143)</td>
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<td>55.6</td>
<td>0.98 (0.41-1.97)</td>
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<td>3 (n = 22)</td>
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<td>Lymphatic invasion</td>
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<tr>
<td>Absence (n = 54)</td>
<td>-</td>
<td>2.59 (1.43-5.18)</td>
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<tr>
<td>Presence (n = 167)</td>
<td>43</td>
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<td>Venous invasion</td>
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<tr>
<td>Absence (n = 17)</td>
<td>-</td>
<td>9.93 (2.21-175.01)</td>
<td>&lt;0.001</td>
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<tr>
<td>Presence (n = 204)</td>
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<tr>
<td>Neural invasion</td>
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<tr>
<td>Absence (n = 9)</td>
<td>75.2</td>
<td>2.77 (0.87-16.87)</td>
<td>0.092</td>
</tr>
<tr>
<td>Presence (n = 212)</td>
<td>54.7</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Histological necrosis</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Absence (n = 106)</td>
<td>-</td>
<td>4.12 (2.54-6.97)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Presence (n = 115)</td>
<td>30.5</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

On multivariate analysis, lymph node metastasis, venous invasion, and histological necrosis were considered independent good prognostic factors.

CI, confidence interval; DSS, disease-specific survival; RR, risk ratio

*Cox-regression proportional hazards model.
**TABLE 3** Correlation Between PEAs and Histological Necrosis

<table>
<thead>
<tr>
<th>Presence of Necrosis, n (%)</th>
<th>Size of Necrosis, n (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Absence</td>
<td>Presence</td>
</tr>
<tr>
<td>(n = 106)</td>
<td>(n = 115)</td>
</tr>
<tr>
<td>PEAs (+)</td>
<td>23 (22)</td>
</tr>
<tr>
<td>PEAs (-)</td>
<td>83 (78)</td>
</tr>
</tbody>
</table>

There were significant correlations between PEA-positive and histological necrosis.

PEAs, poorly enhanced areas

*Chi-squared tests.
<table>
<thead>
<tr>
<th>Characteristics</th>
<th>n = 221</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age, median (range), y</td>
<td>70 (43-87)</td>
</tr>
<tr>
<td>Sex, male, n (%)</td>
<td>133 (60)</td>
</tr>
<tr>
<td>Tumor location, n (%)</td>
<td></td>
</tr>
<tr>
<td>Head</td>
<td>149 (67)</td>
</tr>
<tr>
<td>Body and tail</td>
<td>69 (31)</td>
</tr>
<tr>
<td>Whole pancreas</td>
<td>3 (1)</td>
</tr>
<tr>
<td>Tumor size, n (%)</td>
<td></td>
</tr>
<tr>
<td>&lt;20mm</td>
<td>61 (28)</td>
</tr>
<tr>
<td>20-40mm</td>
<td>137 (62)</td>
</tr>
<tr>
<td>≥40mm</td>
<td>23 (10)</td>
</tr>
<tr>
<td>Lymph node metastasis, n (%)</td>
<td></td>
</tr>
<tr>
<td>Absence</td>
<td>78 (35)</td>
</tr>
<tr>
<td>Presence</td>
<td>143 (65)</td>
</tr>
<tr>
<td>Tumor grade, n (%)</td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>36 (16)</td>
</tr>
<tr>
<td>2</td>
<td>163 (74)</td>
</tr>
<tr>
<td>3</td>
<td>22 (10)</td>
</tr>
<tr>
<td>Lymphatic invasion, n (%)</td>
<td></td>
</tr>
<tr>
<td>Absence</td>
<td>54 (24)</td>
</tr>
<tr>
<td>Presence</td>
<td>167 (76)</td>
</tr>
<tr>
<td>Venous invasion, n (%)</td>
<td></td>
</tr>
<tr>
<td>Absence</td>
<td>17 (8)</td>
</tr>
<tr>
<td>Presence</td>
<td>204 (92)</td>
</tr>
<tr>
<td>Neural invasion, n (%)</td>
<td></td>
</tr>
<tr>
<td>Absence</td>
<td>9 (4)</td>
</tr>
<tr>
<td>Presence</td>
<td>212 (96)</td>
</tr>
<tr>
<td>Size of necrosis, n (%)</td>
<td></td>
</tr>
<tr>
<td>Absence</td>
<td>106 (48)</td>
</tr>
<tr>
<td>Presence</td>
<td>65 (29)</td>
</tr>
</tbody>
</table>
### Supplemental TABLE 2 Histological Reasons for False-Positive PEAs

<table>
<thead>
<tr>
<th>Histological findings account for false positive of PEAs</th>
<th>Large neoplastic gland</th>
<th>Dilated pancreatic duct</th>
<th>Steatosis</th>
<th>Unclear</th>
</tr>
</thead>
<tbody>
<tr>
<td>PEAs positive (n = 98)</td>
<td>76 (78)</td>
<td>14 (14)</td>
<td>3 (3)</td>
<td>1 (1)</td>
</tr>
</tbody>
</table>

Data expresses as n (%). The causes of false positive of PEAs were thought to be large neoplastic gland in 14 patients (14%), dilated pancreatic duct in 3 patients (3%), and steatosis in a patient (1%).

PEAs, poorly enhanced areas