The Characteristics of Disseminated Tumor Cells in Pancreatic Cancer: A Black Box Needs to Be Explored

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Key Words
Pancreatic cancer · Disseminated tumor cells · Micrometastasis

Abstract
Despite recent advances in early diagnosis and surgical treatment, the clinical outcome of patients with pancreatic cancer has not been improved markedly. One of the reasons for the dismal outcome is early dissemination of tumor cells. Sensitive immunohistocytochemical and nucleic acid-based assays have detected disseminated tumor cells in the lymph nodes, bone marrow, peritoneal cavity or peripheral blood. Formation of the metastatic disease depends on the nature of the disseminated tumor cells. Standardization of protocols is mandatory to detect occult tumor cells in clinical practice. We present an overview of recent studies on the incidence, prognostic values and some characteristics of occult tumor cells disseminated in the secondary sites of patients with pancreatic cancer.

Introduction
Although pancreatic cancer is the thirteenth most common cancer in the world, its case fatality is ranked as the fifth in all cancer deaths [1]. Of those who had a chance to undergo curative surgical resection, the 5-year survival rate is still less than 10%. About 50% of this group of ‘lucky’ patients would die of local recurrence and/or distant metastasis shortly after operation [2]. Theoretically, the patients in R0 and N0 nodal status according to the International Union Against Cancer staging system for pancreatic cancer [3] should have a better outcome. However, the 5-year survival rate even in this subgroup of patients remains to be only 21% [4]. Meanwhile, the stage I and R0 patients have a poor 5-year survival rate of 17% [4]. Early metastatic recurrence after radical surgery indicates that disseminated tumor cells, undetectable by conventional pathological examinations, may have already been present at the time of surgery [5].

Further understanding of the characteristics and the fate of the occult tumor cells occurring in the secondary sites is crucial for designing treatment strategies and predicting the prognosis of the patients. The aim of this review is to summarize the recent achievements of researches concerning disseminated pancreatic cancer cells.

Technique Issues
Modern imaging techniques, e.g. spiral computed tomography and magnetic resonance imaging, have their own limitations in detecting lesions with a diameter of 3–5 mm, corresponding to about 10^8 cancer cells, and
cannot display a smaller disease in clinical practice [6]. In recent years, improved techniques for pathological examination using immunohistochemistry and polymerase chain reaction (PCR)-based techniques have been used to detect disseminated tumor cells in pancreatic cancer research.

**Hematoxylin-Eosin Staining of Serial Sectioning**

Compared with routine pathology, the serial sectioning technique has more accuracy in evaluation of the extent of lymph node metastasis [7]. According to one of the latest studies, 9 (0.9%) of 957 routine HE-negative nodes proved to be positive by serial sections in patients with pancreatic cancer, but this positive rate was lower than that of cytokeratin immunostaining [8]. The serial sectioning technique is too complicated and is inconvenient for clinical practice.

**Immunohistochemical Approaches**

Immunostaining techniques showed higher sensitivity to identify micrometastases from the peritoneal washings, lymph nodes, bone marrow and perineural plexus than conventional techniques [9, 10]. Immunohistochemical assays facilitated the identification of one disseminated cancer cell in a background of $10^5$–$10^6$ nonmalignant bone marrow cells [9]. One of the major advantages of immunohistochemistry is the possibility to assess the morphology of the positively stained cells under microscopy.

Cytokeratins are an essential family of components for both the normal and cancerous cells originating from the epithelium [11, 12]. Since pancreatic cancer originates from the pancreatic ductal epithelium, members of the cytokeratin family have been most commonly used as antigens for immunohistochemical detection of micrometastasis and isolated cells of pancreatic cancer.

Overestimation of immunostaining, partly being attributed to the different specificities of different antibodies, may cause frequent false-positive results and lead to different prognostic conclusions. Besides cancer cells, positive stains by cross-reactions have been reported in plasma cells, interstitial reticulum cells and mesothelial cells, depending on the different antibodies [13, 14]. Of the three well-known antibodies to cytokeratins, AE1/3, not CAM5/3 or PAM keratin, would not recognize plasma cells and interstitial reticulum cells. Benign mesothelial cells may embolize to regional lymph nodes in pleuritis or pericarditis, and could be easily distinguished from micrometastatic tumor cells. Since macrophages are often the cause for false-positive results in AE1/3 immunohistochemical staining, anti-CD68 antibody, which is a specific marker to the macrophages, has been used to differentiate disseminated cancer cells and the macrophages [14]. Anti-cytokeratin antibodies also have the possibility of cross-reaction with native bone marrow cells, but the prevalence is less than 5% [15].

Besides antibodies to cytokeratin, Ber-EP4 in the lymph nodes, and carcinoembryonic antigen (CEA) and CA19-9 in the bone marrow and peritoneal washings were also used for investigation of disseminated pancreatic cancer cells [16, 17]. However, the diversity of antibodies used in the previous studies makes it impossible to compare the results reported. It is difficult to detect tumor cells in the body fluids such as peritoneal washings and peripheral blood by immunostaining because of the low incidence of disseminated cancer cells. Fortunately, application of enrichment protocols could increase the number of tumor cells isolated for detection. The immuno-magnetic cell separation system can separate tumor cells marked with magnetic beads via antibodies from unlabeled cells for detection, e.g. peripheral blood cells or bone marrow cells [18]. In density centrifugation methods, erythrocytes, platelets, and polymorphonuclear cells are separated in the pellet, and mononuclear cells, including tumor cells, gather in the so-called interphase. These interphase cells are used for the further evaluation of tumor cells by immunohistochemistry assay or RT-PCR [19].

**PCR-Based Assay**

In this set of techniques, Restriction Fragment Length Polymorphism is based on the findings of chromosomal abnormalities of cancer cells. Point mutations in some genes are consistently found in various types of malignancies. Since point mutation of the K-ras at codon 12 has been identified in 71–100% of pancreatic adenocarcinoma, it often serves as a marker of micrometastasis [20, 21]. Because K-ras mutation is present in chronic pancreatitis as well, either in pancreatic tissue or pancreatic secretions vary between 0 and 100%, the detection might be sometimes disturbed by false-positive results [22, 23]. For the benign pancreatic diseases, where the circulating cells with K-ras mutations originated is still unknown. Löhr et al. [24] figured out that the K-ras could be found in ductal hyperplasias and normal duct cells of pancreas, and also in heavy smoking people.

Since malignant cells often continue expressing markers that are characteristic of them or specific to the normal tissue from which the tumor originated, RT-PCR has become another main PCR strategy for the detection of dis-
seminated tumor cells in the lymph nodes, bone marrow, and peritoneal cavity washings in patients with pancreatic cancer. Theoretically, expression of tissue-specific mRNAs, such as for cytokeratins, CEA, etc., at a site where these transcripts are not normally present implies tumor spread.

RT-PCR has a high sensitivity, identifying one mRNA molecule from $10^6$ to $10^7$ normal cells. But the high false-positive rate is still a major problem. The following aspects could be the reasons. First, false positives come from the illegitimate transcription (i.e. transcription of any gene in any cell type) or processed pseudogenes – intronless, nontranscribed genes that are highly homologous to the exons of the transcribed genes [25, 26]. These processed pseudogenes may generate PCR fragments of the same size as the ones originating from the cDNA and bring the false-positive results. Meanwhile, some targeted genes can also be polymerized from nonmalignant cells. For example, CK-19 mRNA has been demonstrated in 20% of healthy volunteers by RT-PCR [27] and virtually, all of the other markers used (e.g. CEA and Muc1) have also proved to be nonspecific [28].

To resolve the problems in quantifying tumor burden, the real-time RT-PCR technique has been adopted recently as a choice for quantitating changes in gene expression [29]. However, its clinical value is still under discussion.

Detection of Disseminated Pancreatic Cancer Cells

The purpose of detection of disseminated cancer cells lies in the more precise staging of the malignancy and more exact prognostic evaluations of the patients with pancreatic cancer. During the last decade, several results and summaries have been reported about the detection and prognostic impact of disseminated pancreatic cancer cells [30]. In these studies, the diverse results should be attributed to the different techniques and divergent markers used.

The detection rate of disseminated cancer cells in the lymph nodes was reported to be 44–68% in pancreatic cancer (table 1). Hosch et al. [16] reported detection of disseminated cancer cells in the lymph nodes of patients with pancreatic cancer by using Ber-EP4 and described the prognostic relevance by multivariate analysis. In their study, however, the population of the lymph nodes examined was small and the specificity of Ber-EP4 to pancreatic cancer remained uncertain. Brown et al. [35] compared the immunohistochemical staining by AE1/3 and K-ras mutation using the PCR/Restriction Fragment Length Polymorphism technique to detect occult involvements of the lymph nodes. Of all the 30 patients, 14 were positive for disseminated cancer cells by cytokeratin staining, 19 by the PCR technique and 25 by a combination of the two techniques.

Not only the ratio but also the sites of the lymph nodes involved are important for the outcomes of the patients with pancreatic cancer. In the report of Kanemitsu et al. [8], the intra-abdominal lymph nodes of the patients who underwent extended operation were analyzed by dividing them anatomically into three groups; N0, N1 and N2 based on the staging system proposed by the Japan Pancreas Association [36]. The survival rate of cytokeratin N0 and N1 cases was better than that of routine HE N0 and N1 patients. In the N2 group, the survival time with cytokeratin-positive N2 pancreatic cancer was not significantly longer than that with routine HE N2 [8]. So the authors would rather regard the pancreatic cancer with N2-positive nodes as a systemic disease, which could not be cured by extended surgical approaches. Our study [unpubl. data] showed that micrometastasis of the para-aortic lymph nodes has no prognostic value in patients who had undergone extensive surgery for pancreatic cancer.

Table 1. Frequency and prognostic value of disseminated tumor cells in lymph nodes in pancreatic cancer patients

<table>
<thead>
<tr>
<th>Markers/antibodies</th>
<th>Techniques</th>
<th>Detection rates</th>
<th>Prognostic value</th>
</tr>
</thead>
<tbody>
<tr>
<td>HE staining of serial sectioning</td>
<td>9/957 (0.9%)</td>
<td>–</td>
<td>8</td>
</tr>
<tr>
<td>Cytokeratin</td>
<td>IM</td>
<td>23/375 (2.4%)</td>
<td>–*</td>
</tr>
<tr>
<td>Ber-EP4</td>
<td>IM</td>
<td>16/37 (43.2%)</td>
<td>Yes</td>
</tr>
<tr>
<td>K-ras</td>
<td>PCR</td>
<td>4/6 (66.6%)</td>
<td>No</td>
</tr>
<tr>
<td>K-ras</td>
<td>PCR</td>
<td>8/13 (61.5%)</td>
<td>–</td>
</tr>
<tr>
<td>K-ras</td>
<td>PCR</td>
<td>17/25 (68%)</td>
<td>–</td>
</tr>
<tr>
<td>K-ras</td>
<td>PCR</td>
<td>8/13 (61.5%)</td>
<td>–</td>
</tr>
<tr>
<td>AE1/AE3</td>
<td>IM</td>
<td>14/30 (46.7%)</td>
<td>No</td>
</tr>
<tr>
<td>K-ras</td>
<td>PCR</td>
<td>19/30 (63.3%)</td>
<td>No</td>
</tr>
</tbody>
</table>

HE = Hematoxylin-eosin; IM = immunohistochemistry; PCR = polymerase chain reaction; – = not done by multivariate Cox regression analysis; Yes = Prognostic values were confirmed by multivariate Cox regression analysis; No = Prognostic values were not confirmed by multivariate Cox regression analysis.

* Detection rates were calculated by the number of lymph nodes involved.

Statistical significance was demonstrated only by univariate analysis.
The prevalence of the disseminated tumor cells in the bone marrow of patients with pancreatic cancer is about 24–58%. Up to now, to our knowledge, only one study with epithelial cytokeratin immunostaining mentioned the prognostic relevance confirmed by univariate analysis (table 2). Some studies reported disseminated tumor cells detected by immunohistochemical staining of CEA, CA19-9, and A45-B/B3 in the bone marrow [37–39] and one of them revealed a prognostic relevance by multivariate analysis [38]. Z’graggen et al. [41] detected isolated tumor cells in the bone marrow in more than 25% of patients with pancreatic cancer by using AE1/AE3 immunohistochemical assay with a specificity of 96% [41]. The assay using the antibody AE1/AE3 was approximately 3 times more sensitive than the antibodies for CK19, CK20, and CA19-9 [41].

K-ras, CEA, CK19, CK20 and chymotrypsinogen by PCR and AE1/AE3 by immunohistochemistry were used to detect dissemination of tumor cells in the peripheral blood (table 3). None of the studies could prove the prognostic significance by univariate and multivariate analyses. Bilchik et al. [45] reported a combination assay of different markers of MET, GalNac-T and β-hCG in the tumor biopsies, cultured tumor cells and blood of 33 patients with pancreatic cancer and healthy controls. In their study, the combination showed a benefit in terms of the sensitivity.

In the early phase, cytology was used for the detection of disseminated tumor cells in the peritoneal cavity washings. The positive rate fluctuated from 10 to 39%, depending on the concentration of the tumor cells in the peritoneal cavity washings (table 4). CEA, CA19-9, 17-1-A, C54-0, Ra96, KL-1, B72.3 and Leu-M1 were used for im-

### Table 2. Frequency and prognostic value of disseminated tumor cells in bone marrow in pancreatic cancer patients

<table>
<thead>
<tr>
<th>Markers/antibodies</th>
<th>Techniques</th>
<th>Detection rates</th>
<th>Prognostic value</th>
<th>Ref.</th>
</tr>
</thead>
<tbody>
<tr>
<td>CK2, KL-1, A45-B/B3</td>
<td>IM</td>
<td>24/42 (57%)</td>
<td>–</td>
<td>37</td>
</tr>
<tr>
<td>CK2, KL-1, A45-B/B3</td>
<td>IM</td>
<td>25/48 (52%)</td>
<td>–</td>
<td>38</td>
</tr>
<tr>
<td></td>
<td></td>
<td>4/48 (8.3%)</td>
<td>–</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>16/48 (33.3%)</td>
<td>–</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>9/48 (18.6%)</td>
<td>–</td>
<td></td>
</tr>
<tr>
<td>CEA</td>
<td>PCR</td>
<td>2/3 (66%)</td>
<td>–</td>
<td>39</td>
</tr>
<tr>
<td>CEA, CA19-9, 17-1-A, C54-0, Ra96, KL-1</td>
<td>IM</td>
<td>15/26 (58%)</td>
<td>–</td>
<td>40</td>
</tr>
<tr>
<td>AE1/AE3</td>
<td>IM</td>
<td>13/54 (24%)</td>
<td>No</td>
<td>41</td>
</tr>
</tbody>
</table>

IM = Immunohistochemistry; PCR = polymerase chain reaction; – = not done by multivariate Cox regression analysis; Yes = prognostic values were confirmed by multivariate Cox regression analysis; No = prognostic values were not confirmed by multivariate Cox regression analysis.

* Statistical significance was demonstrated only by univariate analysis.

### Table 3. Frequency and prognostic value of disseminated tumor cells in peripheral blood in pancreatic cancer patients

<table>
<thead>
<tr>
<th>Markers/antibodies</th>
<th>Techniques</th>
<th>Detection rates</th>
<th>Prognostic value</th>
<th>Ref.</th>
</tr>
</thead>
<tbody>
<tr>
<td>K-ras</td>
<td>PCR</td>
<td>2/6 (33.3%)</td>
<td>–</td>
<td>42</td>
</tr>
<tr>
<td>CK20</td>
<td>PCR</td>
<td>22/28 (78%)</td>
<td>–</td>
<td>43</td>
</tr>
<tr>
<td>CEA</td>
<td>PCR</td>
<td>13/21 (61.9%)</td>
<td>–</td>
<td>44</td>
</tr>
<tr>
<td>MET, GalNac-T, β-hCG</td>
<td>PCR</td>
<td>8/16 (MET, 50%)</td>
<td>8/16 (GalNac-T, 50%)</td>
<td>45</td>
</tr>
<tr>
<td></td>
<td></td>
<td>7/16 (β-hCG, 3.8%)</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

IM = Immunohistochemistry; PCR = polymerase chain reaction; – = not done by multivariate Cox regression analysis.
munocytochemical examination of disseminated tumor cells from the peritoneal washings. No study demonstrated the significant prognostic value by multivariate analysis. The three reports using the PCR technique also showed no prognostic relevance of disseminated tumor cells in the peritoneal washings [49–51].

Perineural construction was regarded as a main pathway for pancreatic cancer infiltration in recent years. Suwa et al. [52] used both anti-cytokeratin19 antibody immunostaining and K-ras PCR detection for the dissemination of tumor cells in nerve plexus from 17 patients with resected pancreatic cancer. In their study, univariate analysis, rather than multivariate analysis, demonstrated a significant correlation between the prognosis and the positive neural margin.

Characteristics of Disseminated Tumor Cells in Pancreatic Cancer

The different conclusions regarding the prognostic value of disseminated tumor cells in the peripheral blood, bone marrow, lymph nodes and other secondary sites may contribute to the confusion of the different concepts and poor understanding of the characteristics of the disseminated tumor cells, besides the different sensitivity and specificity of the various detecting techniques.

To discuss the characteristics and the fates of disseminated tumor cells, it is necessary to clarify the various concepts that have been used for the description of the tumor cells escaping from the primary site. The abuse of terminology often causes controversial conclusions in the prognostic value of micrometastasis.

The following two concepts are most easily misunderstood: isolated tumor cells and micrometastasis. Compared to isolated tumor cells pathologically, micrometastasis usually occurs as a small group of tumor cells, which are smaller than 0.2 cm in greatest dimension, in contact with vessel or lymph sinus wall, invading or penetrating the vessel and lymph sinus wall, and further evoking extravascular stromal reaction and proliferating outside the vessel and lymph sinus after penetration. On the other hand the isolated tumor cells mean single tumor cells or small clusters in circulation or disseminated in secondary sites [53]. For the convenience of discussion, we termed the isolated tumor cell and micrometastasis together as disseminated tumor cells.

The formation of a metastasis is a complex process. Before forming a secondary tumor, cancer cells in the primary site would pass through a series of discrete steps, involving a number of possible pathways (fig. 1) [54]. The molecular mechanisms involved in the process from disseminated tumor cells to metastatic disease have not been completely understood but those associated with cell-cell and cell-matrix adhesion regulated by abundant adhesive molecules, such as intercellular adhesion molecules-1, vascular cell adhesion molecule [55, 56] and integrin [57], with the degradation of extracellular matrix regulated by the members of the matrix metalloproteases [58], urokinase plasminogen activator and tissue plasminogen activator [59], as well as with the initiation and maintenance of early growth at the new site have generally been accepted as critical for pancreatic cancer.

Although metastasis is the foremost cause of death for cancerous diseases, it is fortunately an extremely inefficient process [60], with few of the many cells shed from a primary tumor successfully forming secondary tumors. In recent years some studies tried to understand what happens to cancer cells lost between their entry into the circulation and the formation of metastases.

Some malignant cell disappearance may occur in the circulation due to hemodynamic forces or the immune system. Only a very small percentage of circulating tumor cells (0.05%) survived and initiated metastatic foci [61]. Numerous experimental data indicated that the isolated tumor cells in the peripheral blood would apoptosis rapidly, unless they were able to adhere to the neighboring cells or the extracellular matrix of the distant sites. This phenomenon could be explained by the conception of Anoikis, originating from ‘homelessness’ in Greek. Its essence is that cell anchorage can regulate several apoptosis-suppressing pathways by many kinds of adhesive mol-

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<tr>
<td>CEA, CA19-9,17-1-A, C54-0, Ra96, KL-1</td>
<td>PCR</td>
<td>24/62 (39%)</td>
<td>–*</td>
<td>49</td>
</tr>
<tr>
<td>K-ras</td>
<td>PCR</td>
<td>2/20 (10%)</td>
<td>–</td>
<td>50</td>
</tr>
<tr>
<td>CEA, Ca19-9</td>
<td>PCR</td>
<td>4/20 (20%)</td>
<td>–</td>
<td>51</td>
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PCR = Polymerase chain reaction; – = not done by multivariate Cox regression analysis.
* Statistical significance was demonstrated only by univariate analysis.
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ecules expressed on the cell membrane and extracellular matrix. The process of this detachment-induced apoptosis, depending on different cells, has been related to an abundance of adhesive molecules, the members of integrin and cadherin families, endogenous galactoside-binding lectin, as well as a frightening number of complex signal transduction molecules, i.e. MAP/ERK kinase kinase-1, caspase, cytochrome c, Fas ligand, and ras-extracellular signal-regulated kinase [62, 63]. Duxbury et al. [64] demonstrated recently that different pancreatic cancer cell lines showed variation in their anoikis resistance, being associated with the level of focal adhesion kinase (FAK) expression, which can transduct intracellular messages that are accompanied with growth factor signaling and cell-extracellular matrix interactions. Meanwhile, FAK gene silencing by RNA interference promoted in vitro anoikis and inhibited metastasis of pancreatic adenocarcinoma cells in vivo.

Furthermore, the implantation of tumor cells appears inefficient in the formation of the pathological metastasis. Freeman et al. [65] and Rehders et al. [66] found that the great majority of circulating tumor cells was noncycling or remained in G0 phase of cell cycle, while only the minority of them had the ability to form micrometastasis. One research reported that although more than 80% of injected melanoma tumor cells can successfully extravasate as solitary cells in the liver, only 5% of the extravasated cells underwent either proliferation or apoptosis, while 95% of them were dormant. Very few of these nondormant cells formed colonies in the liver. One in forty had formed micrometastasis by day 3, while only 1% of the micrometastasis developed into macroscopic metastasis by day 13 [67]. The isolated tumor cells entering in the bone marrow may remain in a dormant state for a long period until they are eradicated by immune surveillance or until they acquire a blood supply to permit growth [68]. It is unclear what exact factors determine the duration from the dissemination of tumor cells to the appearance of clinical metastasis [69]. The biological significance of these disseminated cells still remains to be determined.

To grow up to a size larger than 1–2 cm, micrometastatic tumor cells need the ability of angiogenesis [70]. For pancreatic cancer, many factors are involved in these processes; vascular endothelial growth factor, acidic and basic fibroblast growth factors 1 and 2, integrin and transmembrane tyrosine kinase receptors expressed on endothelial cells and pancreatic cancer cells [71, 72].

Since a great amount of details remain unknown, the processes of tumor cell dissemination and the formation of secondary disease are a black box for us that needs to be explored.

Fig. 1. To form the secondary tumor growth, the disseminated tumor cells would pass through a series of steps. Since little is known about this complicated process, we would like to call it ‘black box’.


Conclusions

Despite the recent progress of diagnostic and therapeutic modalities, the clinical outcome of pancreatic cancer remains dismal, partly due to dissemination of tumor cells in early phase. Detection of disseminated tumor cells by immunocytohistochemical and molecular methods seems to be a promising approach to arrive at a more precise staging, which may enable us to identify putative candidates at high risk of recurrence. Standardization of detection methods and elucidation of the characteristics of the disseminated cancer cells are crucial for future researches on pancreatic cancer.

References


Reduction of morbidity and mortality due to invasive ductal adenocarcinoma of the pancreas will require progress in any or all of the following areas: (1) prevention, (2) early detection, and (3) treatment. The current inefficiency in early diagnosis underlies the low rates of both operability and cure among patients with carcinoma of the pancreas. The review of disseminated pancreatic cancer cells by Su et al. focuses on important issues that must be addressed to improve treatment of this disease when it is discovered at the clinical stages when 90–95% of new patients are now diagnosed.

These issues include detection of disseminated tumor cells in lymph nodes, blood, and other metastatic sites, and recognition of traits of these cells that correlate with prognosis and provide targets for treatment. Analysis of these ‘traits’ will take investigators into the realms of genomics and proteomics [1, 2]. The cost and complexity of these approaches, and even those of multiple immunohistochemical studies of individual carcinomas, require that the value of the assays be validated in the coinage of prediction of prognosis and directing therapy.

As is noted in the Tables provided by the authors, simple detection of disseminated tumor cells in lymph nodes, blood, and other metastatic sites, and recognition of traits of these cells that correlate with prognosis and provide targets for treatment. Analysis of these ‘traits’ will take investigators into the realms of genomics and proteomics [1, 2]. The cost and complexity of these approaches, and even those of multiple immunohistochemical studies of individual carcinomas, require that the value of the assays be validated in the coinage of prediction of prognosis and directing therapy.

As is noted in the Tables provided by the authors, simple detection of disseminated tumor cells by a variety of molecular and immunostaining approaches has usually failed to correlate with prognosis in a meaningful way. In general these are retrospective studies that have not been linked to the evaluation of specific therapies other than surgical resection. Thus, the review by Su et al. serves to point directions for future research. But, because of limitations in our current level of understanding, the authors do not provide guidelines for management of individual new patients or for design of the clinical trials that will be required to meet the goals they identify.

This review references many relevant studies on the dissemination of cancer cells beyond the primary site based on both pancreatic adenocarcinoma and, to a lesser extent, neoplasms that arise in other organs. There is scant attention to highly relevant studies of this problem in model systems that do not involve pancreatic neoplasms [3]. Thus, in depth consideration of this problem will require the reader to extend consideration to papers from Ward et al. [3], Fidler [4], Ouatas et al. [5] and Oft et al. [6] that are not referenced in the review.

References