

## Perspective

# Mesothelin: A New Target for Immunotherapy

Raffit Hassan, Tapan Bera, and Ira Pastan

Laboratory of Molecular Biology, National Cancer Institute, NIH, Bethesda, Maryland

### ABSTRACT

**Mesothelin is a differentiation antigen present on normal mesothelial cells and overexpressed in several human tumors, including mesothelioma and ovarian and pancreatic adenocarcinoma. The mesothelin gene encodes a precursor protein that is processed to yield the 40-kDa protein, mesothelin, attached to the cell membrane by a glycosylphosphatidyl inositol linkage and a 31-kDa shed fragment named megakaryocyte-potentiating factor. The biological function of mesothelin is not known. Mesothelin is a promising candidate for tumor-specific therapy, given its limited expression in normal tissues and high expression in several cancers. SS1(dsFv)PE38 is a recombinant anti-mesothelin immunotoxin that is undergoing clinical evaluation in patients with mesothelin-expressing tumors. There is evidence that mesothelin is an immunogenic protein and could be exploited as a therapeutic cancer vaccine. A soluble mesothelin variant has been identified and could be a useful tumor marker for malignant mesotheliomas.**

### INTRODUCTION

Mesothelin is a 40-kDa cell surface glycoprotein that is highly expressed in pancreatic cancers, ovarian cancers, mesotheliomas, and some other cancers (1). Mesothelin is not a cancer-specific antigen, but like CD20, it is a differentiation antigen that is present on normal cells and highly expressed in many cancers. Mesothelin is expressed on normal mesothelial cells lining the pleura, pericardium, and peritoneum. The limited distribution of mesothelin on normal tissues makes it a promising target for tumor-specific therapy. Also, because small amounts of mesothelin can be detected in the blood of some patients with mesothelin-positive cancers, measurement of mesothelin in the blood may be useful for the diagnosis and to follow the course of some of these patients (2). The mesothelin gene encodes a 69-kDa precursor protein that is processed to a 40-kDa membrane-bound protein termed mesothelin and a 31-kDa shed fragment called megakaryocyte-potentiating factor (MPF) that is released from the cell (Fig. 1). The mesothelin protein is the antigen recognized by the monoclonal antibody (Mab) K1, whereas MPF was isolated from the medium of a human pancreatic cancer cell line (3, 4).

### ISOLATION OF MAB K1 AND CLONING OF ITS 40-KDA ANTIGEN, MESOTHELIN

The Mab K1 was generated by immunization of mice with the human ovarian carcinoma cell line OVCAR-3 (3). The reactivity of Mab K1 against a variety of different human tumor cell lines was tested using immunofluorescence (3). It showed reactivity with several ovarian cancer cell lines, including OVCAR-2, OVCAR-3, OVCAR-5, A-1847, and SKOV3; cervical cancer cell lines HeLa and KB; and gastric cell lines AGS and HTB103. No reactivity was detected with breast, colon, or prostate cancer cell lines. Care must be taken with the use of Mab K1 because its reactivity is acid labile (5).

The reactivity of Mab K1 against normal human tissues was tested by immunohistochemistry using cryostat tissue sections (3). Most normal tissues showed no reactivity with Mab K1, with the exception of mesothelial cells that line the peritoneal, pleural, and pericardial cavities. There was also weak reactivity with the basal cells of the trachea and cells in the Fallopian tubes. A similar immunoreactivity was seen in cryostat tissue sections of cynomolgus monkeys, making monkeys a useful model for preclinical toxicology studies. Although mesothelin RNA has been found in extracts of normal lung, kidney, and liver, its presence is due to the fact that these tissues contain a mesothelial cell lining. Our data as well as data from other investigators indicate that there is no mesothelin expression in the parenchymal cells of these organs (3, 6).

The antigen recognized by Mab K1 was identified by expression cloning using cDNA prepared from a HeLa cell line. The gene was named mesothelin because immunohistochemical studies showed that the membrane-bound antigen it encodes is present on normal mesothelial cells (1). Alignment of the cDNA sequence with the human genome sequence shows that the mesothelin gene has 17 exons on human chromosome 16p13.3 (Fig. 2). The mesothelin cDNA is 2138-bp long, contains an open reading frame of 1884 bp, and encodes a 69-kDa protein. Two minor spliced forms of the major mesothelin transcript have been detected that encode two slightly altered proteins termed variant 1 and variant 2 (Fig. 2). Variant 1 has an insertion of 8 amino acids after glutamine 408. This variant was present in the original cDNA clone isolated from HeLa cells (1). Variant 2 retains the intron between exons 16 and 17 and probably leads to premature termination of the protein, resulting in its release from the cell. There are now over 60 mesothelin sequences in the expressed sequence tag database. Only two of these contain the insertion at position 408, and four others encode the prematurely terminated protein.

The major cell-associated glycoprotein recognized by Mab K1 has a molecular mass of ~40-kDa, although some full-length 71-kDa glycosylated protein has also been detected (1). The precursor protein contains 628 amino acids and has four potential *N*-linked glycosylation sites. A furin cleavage site, RPRFRR, is present at amino acids 288–293. Mesothelin is made as a 69-kDa polypeptide with a hydrophobic sequence at the carboxyl end that is removed and replaced by phosphatidylinositol. After glycosylation at one or more of its four putative

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**Requests for reprints:** Raffit Hassan, Laboratory of Molecular Biology, National Cancer Institute, 37 Convent Drive, Room 5116, Bethesda, MD 20892-4264. Phone: (301) 451-8742; Fax: (301) 402-9469; E-mail: hassanr@mail.nih.gov.

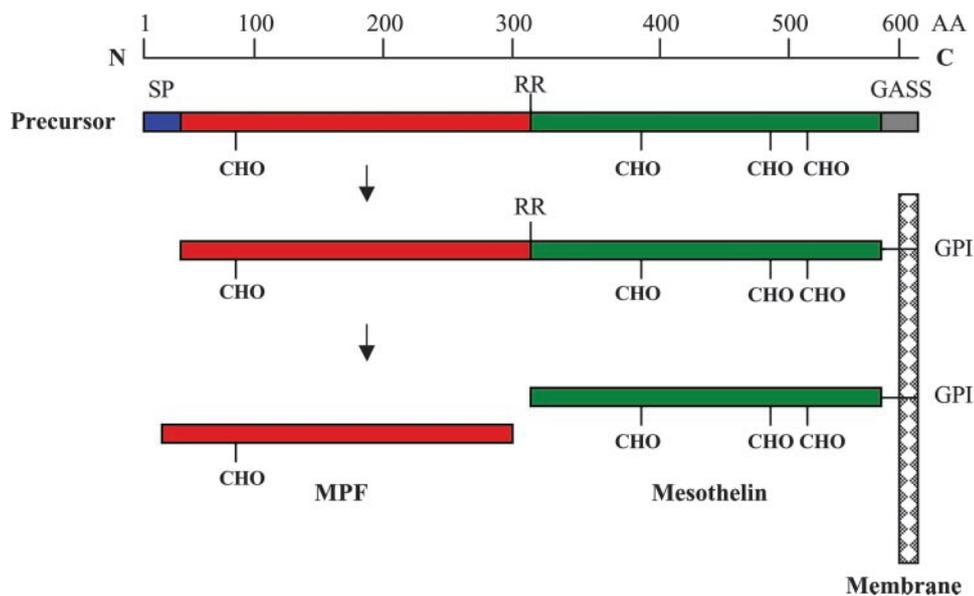


Fig. 1 Schematics showing maturation of mesothelin protein. Precursor protein for mesothelin is synthesized as a 622-amino acid polypeptide with a calculated molecular mass of 77 kDa. The potential signal peptide (SP) and the glycosylphosphatidylinositol anchor signal sequence (GASS) are predicted at the NH<sub>2</sub> terminus and the COOH terminus, respectively. The precursor protein has four predicted glycosylation sites (CHO) and a furin cleavage site (RR). Cleavage at the furin site generates membrane-bound mesothelin (green) and the secretory protein megakaryocyte-potentiating factor (red).

glycosylation sites, it is cleaved by furin to yield the 40-kDa fragment that was first found on the surface of OVCAR-3 cells and a smaller 32-kDa fragment that is released from the cell. As described above, this 32-kDa fragment is MPF, which was initially isolated from the medium of the human pancreatic cancer cell line HPY-5 (4).

**MPF**

MPF is a 32-kDa protein that stimulates the megakaryocyte colony-forming activity of murine interleukin-3 in mouse bone marrow cell culture (4). Yamaguchi *et al.* (4) examined 64 cell lines for their ability to produce megakaryocyte-potentiating activity. The pancreatic cancer cell line HPC-Y5 showed the

highest activity, and MPF was purified from the conditioned medium of these cells. On SDS-PAGE analysis, MPF was identified as a single band with a molecular mass of approximately 32-kDa. Glycopeptidase F digestion and amino sugar analysis demonstrated that MPF is a glycoprotein containing at least one N-linked sugar chain. Using a megakaryocyte colony-forming assay, the purified MPF potentiated megakaryocyte colony formation in the presence of interleukin-3. However, MPF alone did not have any intrinsic megakaryocyte colony-stimulating activity. Whether MPF has megakaryocyte-potentiating activity in humans is unknown, but it is of interest that patients with mesotheliomas often have elevated platelet counts.

Subsequently, the cDNA encoding human MPF was iso-

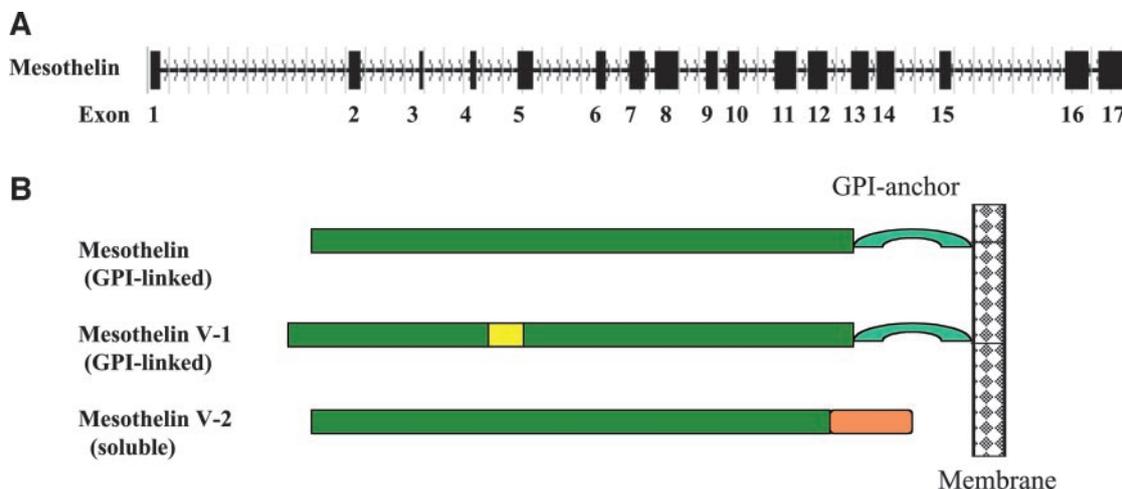


Fig. 2 Schematics describing the mesothelin gene and the proteins it encodes. A, genomic organization of mesothelin gene. There are 17 exons (filled boxes) in the mesothelin gene. B, drawing of three different forms of mesothelin protein described in the database. Mesothelin is the most common variant and is predicted to be membrane-bound; mesothelin V-1 has an additional 8 amino acids (yellow) and is also predicted to be bound to the membrane. Mesothelin V-2 has a modified COOH terminus (orange), lacks the glycosylphosphatidylinositol anchor signal sequence, and is predicted to be secreted.

lated and found to encode a polypeptide consisting of 622 amino acids with a deduced molecular mass of 68 kDa, although the human MPF secreted by HPC-Y5 cells is only 32 kDa in size (7). The cDNA encoding MPF is identical to the mesothelin cDNA. Both the mesothelin and MPF cDNA encode the same precursor protein, which has a furin cleavage site. Cleavage by furin leads to a shed 32-kDa protein called MPF, and a 40-kDa fragment that remains attached to the cell membrane by a glycosylphosphatidyl inositol linkage is called mesothelin (Fig. 1). The gene encoding this precursor protein is referred to as Mesothelin, MPF, or Mesothelin/MPF gene.

### SOLUBLE MESOTHELIN

A soluble 42–45-kDa protein with an NH<sub>2</sub>-terminal amino acid sequence, identical to that of the membrane-bound portion of mesothelin, was identified using the murine Mab OV569, obtained by immunizing mice with malignant ascites of a patient with ovarian cancer (2). The antigen to which OV569 binds was also detected in the cell-free culture supernatant from antigen-positive carcinomas and in the cell-free malignant effusions of patients with mesothelin-positive carcinomas. The gene sequence and analysis of the expressed sequence tag database of mesothelin indicate that the major form of mesothelin is attached to the cell surface and is not soluble. The soluble form of mesothelin is likely due to an abnormal splicing event resulting in a frameshift mutation and premature termination at amino acid 600 deleting the amino acids at the COOH terminus that are responsible for its association with the cell membrane. Premature termination is probably due to lack of splicing of the intron between exons 16 and 17 (Fig. 2). Only 4 of the 60 expressed sequence tags in the database, which align to the 3' end of the mesothelin gene, could produce this soluble form of mesothelin, but it is also possible that soluble mesothelin is a proteolytically cleaved fragment of membrane-bound mesothelin.

To test whether soluble mesothelin-related proteins (SMR) could be valuable for diagnosis or follow-up of cancer patients, Scholler *et al.* (2) examined sera from 68 healthy donors, 3 patients with inflammatory disease, 1 patient with a benign tumor, and 105 patients with different tumors. Serum from all patients without cancer was negative for this soluble antigen. However, sera from 23 of 30 (77%) patients with ovarian cancer were antigen positive. This antigen was also detected in the serum of some patients with other tumors such as breast and lung cancer. When validated, this test may be useful for the diagnosis and follow-up of patients with mesothelin-expressing malignancies (see below). Because this soluble form of mesothelin is present in very small amounts in the blood, it should not interfere with antibody-based therapies that target the mesothelin antigen on cancer cells.

### BIOLOGICAL FUNCTION OF MESOTHELIN

To study the biological function of mesothelin *in vivo*, we generated mutant mice in which both copies of the mesothelin gene were inactivated (8). To our surprise, there were no detectable abnormalities in the mutant mice in terms of growth and reproduction as compared with their wild-type littermates. Also, there was no statistical difference in platelet counts between wild-type and mesothelin knockout mice. It was originally sug-

gested that mesothelin might have a role in adhesion because 3T3 cells transfected with a mesothelin expression vector were more difficult to remove from the culture dishes than nontransfected cells (1). The possibility that mesothelin may play a role in adhesion is supported by a recent study showing that mesothelin binds to CA125 and that this interaction mediates cell adhesion. Based on these findings, the authors (9) suggested that there may be an important role for CA125 and mesothelin in the metastatic spread of ovarian cancer.

Several studies have generated data indicating that activation of specific signaling pathways that are important in cancer can lead to an increase in mesothelin expression. Yamashita *et al.* (10) reported that in the Eker rat carcinoma cell line, there is increased expression of the *Erc* gene, the rat homolog of the human mesothelin gene. Eker rats develop hereditary renal carcinomas due to mutations of the tumor suppressor gene *Tsc2*.  $\beta$ -Catenin levels are also elevated in Eker rat tumors (11). The mesothelin gene is also differentially regulated by members of the Wnt signal transduction pathway (12). Using C57MG mouse mammary epithelial cells, mesothelin was up-regulated by Wnt-1. Interestingly, tumors with constitutive activation of the Wnt signaling pathway, such as ovarian and pancreatic cancers, have high mesothelin expression. Additional studies are needed to fully define mesothelin function as well as the role of mesothelin in carcinogenesis.

### MESOTHELIN EXPRESSION IN HUMAN MALIGNANCIES

Mesothelin gene expression in different human cancers has been studied using serial analysis of gene expression (SAGE) tag analysis. The presence and frequency of a specific tag for a particular gene are indicative of the frequency of expression of that gene in different tissues or cancers (13). The presence of mesothelin protein has been studied by immunohistochemistry using anti-mesothelin Mabs K1 and 5B2. The commercially available anti-mesothelin Mab 5B2 (Novocastra, Newcastle-on-Tyne, United Kingdom) is an IgG1 murine Mab that was obtained by immunizing mice with a recombinant prokaryotic fusion protein corresponding to approximately 100 amino acids that are present in the membrane-bound form of the mesothelin molecule.

The only normal tissues that show strong mesothelin immunoreactivity are the mesothelial cells of pleura, pericardium, and peritoneum. On immunohistochemistry, a strong staining of a single layer of mesothelial cells is seen (3). Mesothelin-positive cancers exhibit a diffuse membranous staining of the tumor cells, usually of similar intensity to that seen in normal mesothelial cells. The percentage of tumor cells that are mesothelin positive varies with specimen quality, with better results seen with frozen tissue, especially when using Mab K1.

**Mesothelin Expression in Ovarian Cancer.** The immunohistochemical localization of Mab K1 in ovarian cancer was initially tested using cryostat tissue sections (3). Ten of the 15 (66%) nonmucinous ovarian cancers examined were mesothelin positive, whereas none of the 4 mucinous ovarian cancers were positive. A study performed on paraffin-embedded tissue sections using the anti-mesothelin antibody 5B2 also showed mesothelin immunopositivity in the majority of cases of nonmuci-

nous ovarian cancer (6). Other studies have noted high mesothelin expression at both the mRNA and protein levels in samples from serous ovarian carcinoma (14). There are many tags for the mesothelin gene present in nine SAGE libraries made from ovarian cancers, supporting the fact that mesothelin is overexpressed in ovarian cancer.

**Mesothelin Expression in Mesotheliomas.** Using cryostat sections obtained from patients with pleural mesothelioma, Mab K1 reactivity was observed in all 15 cases of epithelial mesotheliomas tested, but in none of the 4 cases of sarcomatous mesothelioma tested (15). Similar results, showing mesothelin positivity in epithelial mesotheliomas, were seen in a recent study (16) evaluating mesothelin expression in paraffin-embedded tissue sections of mesotheliomas, using the anti-mesothelin antibody 5B2. SAGE tag analysis shows that there are 55 tags for mesothelin cDNA in the single library made from a peritoneal mesothelioma.

**Mesothelin Expression in Pancreatic Cancer.** In the study reported by Argani *et al.* (17), the tag for mesothelin was consistently present in pancreatic cancer samples, but not in normal pancreas. Mesothelin mRNA was confirmed to be present in the cancer cells by *in situ* hybridization in 4 of 4 resected primary pancreatic adenocarcinomas and by immunohistochemistry using the 5B2 antibody in all 60 resected primary pancreatic adenocarcinomas. Labeling was intense ( $\geq 3+$ ) in 54 of the 60 samples tested. No mesothelin reactivity was noted in the adjacent normal pancreas.

Other studies have also confirmed mesothelin expression in the majority of pancreaticobiliary tumors (6, 18). Currently there are 19 tags for mesothelin in four different SAGE libraries made from pancreatic cancers.

**Mesothelin Expression in Other Tumors.** Mesothelin immunoreactivity was noted in the majority of frozen tissue sections from squamous cell carcinomas of the cervix, head and neck, vulva, lung, and esophagus (19). However, this immunoreactivity in squamous cell carcinomas was much less when formalin-fixed, paraffin-embedded tissue sections were examined (6, 20). Other tumors that show some mesothelin expression include lung adenocarcinomas, endometrial carcinoma, biphasic synovial sarcomas, and desmoplastic small round cell tumors (6, 20).

Very little or no expression of mesothelin is seen in breast adenocarcinomas, carcinomas of thyroid, renal cell carcinoma, transitional cell carcinoma of the bladder, melanomas, and hepatomas (6). Mesothelin expression by immunohistochemistry is also infrequently seen in tumors of the gastrointestinal tract. In the case of gastric adenocarcinoma, mesothelin expression is seen in 14–29% of cases (3, 6). This low frequency of mesothelin positivity in gastric tumors by immunohistochemistry is somewhat surprising because SAGE tag analysis shows that mesothelin is highly expressed in many gastric cancer specimens. There are currently 151 SAGE tags for the mesothelin gene in six SAGE tag libraries made from gastric cancer tissues.

## MESOTHELIN AS A TARGET FOR TUMOR-SPECIFIC IMMUNOTHERAPY

Given the high mesothelin expression in several human malignancies and its limited expression in normal tissues, mesothelin is a good target for tumor-specific antibody-based ther-

apies. In addition, biodistribution studies of the anti-mesothelin Mab K1 in nude mice show that it localizes to mesothelin-expressing tumors and could therefore be useful for imaging or immunotherapy of such tumors (5).

**Development of the Anti-Mesothelin Immunotoxin, SS1(dsFv)PE38 (SS1P).** Because the murine anti-mesothelin Mab K1 by itself is not cytotoxic toward mesothelin-positive cells, we decided to develop anti-mesothelin immunotoxins using the potent bacterial toxin *Pseudomonas* exotoxin A (PE). This toxin consists of three functional domains: (a) a cell-binding domain that causes binding to a cell surface protein; (b) a translocation domain that brings the active fragment of the toxin to the cytosol; and (c) an ADP-ribosylation domain that inhibits protein synthesis leading to cell death (21). PE38 is a truncated 38-kDa form of PE that is not toxic to target cells because the cell binding domain is deleted (22). Conjugation of PE38 to an antibody results in targeting of the toxin to the antigen recognized by the antibody and produces cell death.

The first anti-mesothelin immunotoxin consisted of Mab K1 chemically linked to a modified form of PE38 (23). Although it had antitumor activity, it had drawbacks for clinical use including its low affinity and large size, which limits tumor penetration. Therefore, efforts were directed toward developing a molecule with favorable clinical properties leading to the development of SS1(dsFv)PE38 (SS1P). SS(scFv) is an anti-mesothelin single chain Fv (scFv) that was obtained from the splenic mRNA of mice using antibody phage display (24). Using “hot spot” mutagenesis, the affinity of SS(scFv) was improved over 15-fold to yield SS1(scFv) (25). Because immunotoxins containing a scFv are often unstable at 37°C, we have developed a method of stabilizing the Fv by replacing the peptide linker connecting the light and heavy chains with a disulfide bond (26). The SS1scFv was converted to a disulfide stabilized Fv (dsFv) and used to construct the recombinant immunotoxin SS1(dsFv)PE38. This immunotoxin has significant antitumor activity *in vitro* and *in vivo* against mesothelin-positive tumor cells and was chosen for evaluation in clinical trials and given the name SS1P.

**Activity of SS1P against Mesothelin-Expressing Tumors in Nude Mice.** The antitumor activity of SS1P was evaluated using both a tumor xenograft and metastatic model. A431-K5 (human epidermoid carcinoma cell line expressing mesothelin by transfection) cells were established as s.c. tumors in athymic nude mice. Animals with growing tumors received three i.v. injections of SS1P on days 5, 7, and 9 after the injection of tumor cells. Mice treated with 4, 6, or 8  $\mu\text{g}/\text{dose}$  of SS1P had complete tumor regression.<sup>1</sup> The activity of SS1P in inhibiting lung metastases was also evaluated. Mice received i.v. injection with either mesothelin-positive tumor cell line NCI-H226 (derived from a pleural mesothelioma) or mesothelin-negative lung adenocarcinoma cell line PC14PE6. When SS1P was administered to these nude mice, SS1P selectively inhibited the growth of lung metastases produced by the mesothelin-producing NCI-H226 cells (27). These data indicate that SS1P can be active in a metastatic setting.

<sup>1</sup> Unpublished data.

**Activity of SS1P against Mesothelin-Expressing Human Tumor Cells.** The cytotoxic activity of SS1P against tumor cells obtained from patients was evaluated *in vitro*. The activity of SS1P against human ovarian cancer cells obtained from patients undergoing cytoreductive surgery was evaluated using a three-dimensional *in vitro* culture system. Using tumor cells obtained from five patients with ovarian cancer and one with cervical squamous cell carcinoma and apoptosis as an end point, we showed that tumors expressing mesothelin showed a significant dose-dependent sensitivity to SS1P with cytotoxic activity observed at concentrations as low as 1 ng/ml. No antitumor activity was seen at 100 ng/ml in tumors that did not express mesothelin (28). Similarly, the activity of SS1P against tumor cell lines established from ascites of patients with peritoneal mesotheliomas was evaluated using a cell proliferation assay. Seven tumor cell lines were established from the ascites of patients with peritoneal mesothelioma. Six of the seven cell lines expressed mesothelin, whereas one cell line did not. Cell lines that expressed mesothelin were very sensitive to SS1P, with  $IC_{50}$  values ranging between 0.08 and 3.9 ng/ml. The peritoneal mesothelioma cell line that did not express mesothelin was resistant to SS1P with an  $IC_{50}$  of  $>100$  ng/ml.<sup>2</sup>

#### CLINICAL TRIALS OF SS1(DSFV)-PE38 (SS1P)

Based on the preclinical studies showing antitumor activity of SS1P against mesothelin-expressing tumors, as well as toxicology studies in monkeys, SS1P has been approved for investigational use in patients with mesothelin-positive tumors. Two institutional review board-approved Phase I studies of SS1P are currently ongoing (29, 30). One study involves giving SS1P as an i.v. bolus injection every other day for three or six doses. The other involves administration of SS1P as a continuous i.v. infusion over 10 days. Both studies are open to accrual, and dose escalation is ongoing.

#### MESOTHELIN AS A THERAPEUTIC CANCER VACCINE

There is experimental evidence to suggest that mesothelin is a strongly immunogenic protein. In a Phase I clinical trial of patients with pancreatic cancer who were vaccinated with irradiated, granulocyte macrophage colony-stimulating factor-secreting pancreatic tumor cell lines, a dose-dependent systemic antitumor immunity against autologous tumors was seen (31). Some patients treated in this study had clinical benefits that may have led to an improvement in progression-free and overall survival. All patients who had this benefit had a strong anti-mesothelin T-cell immune response as measured by the ELISPOT assay. Based on these results, preclinical studies to develop a therapeutic mesothelin vaccine for the treatment of mesothelin-expressing cancers are ongoing.<sup>3</sup>

<sup>2</sup> L. Quian, C. F. Verschraegen, J. Mendoza, and R. Hassan: Cytotoxic activity of the recombinant anti-mesothelin immunotoxin, SS1(dsFv) PE38, towards tumor cell lines established from ascites of patients with peritoneal mesotheliomas. *Anticancer Research*, in press.

<sup>3</sup> Dr. Elizabeth Jaffe, personal communication.

#### SOLUBLE MESOTHELIN AS A DIAGNOSTIC TUMOR MARKER

The small amount of mesothelin shed into the serum could make it a valuable diagnostic tool in cancers that express mesothelin. Using a double determinant (sandwich) ELISA, the serum concentrations of SMR were assayed in samples obtained from 44 patients with mesothelioma, 160 patients with other inflammatory or malignant lung and pleural diseases, and 68 matched healthy controls, of whom 40 had asbestos exposure (32). Thirty-seven of the 44 patients (84%) with mesothelioma had elevated SMR, in contrast to only 3 of 160 patients (2%) with other diseases of the lung and pleura. In patients with mesothelioma, SMR correlated with the stage and tumor burden, with elevated SMR levels in patients with advanced stage and increased disease burden. No correlation between SMR and platelet counts in patients with mesothelioma was noted. In the healthy control group of 68 patients, 28 patients who had no asbestos exposure had no elevation in SMR, whereas 7 of the 40 patients with exposure to asbestos had increased serum SMR. Within 1–5 years, three of these seven patients developed mesothelioma, and one developed lung carcinoma. This study suggests that serum SMR could be a helpful tumor marker for the diagnosis of mesothelioma. It could also potentially be helpful in screening asbestos-exposed individuals for early evidence of developing mesothelioma.

The value of soluble mesothelin for the diagnosis of ovarian cancer is also being studied. It appears that soluble mesothelin by itself lacks sensitivity and specificity as a tumor marker in ovarian cancer but may complement CA125 for the detection of ovarian cancer (33). Additional studies of the utility of mesothelin, in combination with other ovarian tumor markers, are needed to fully define its role as a screening test for ovarian cancer.

#### MESOTHELIN IMMUNOHISTOCHEMISTRY FOR TUMOR DIAGNOSIS

Initial studies using cryostat sections of human tumors suggested that Mab K1 could help distinguish epithelial mesothelioma from lung adenocarcinoma because lung adenocarcinomas were uniformly mesothelin negative (15). However, two recent studies using the anti-mesothelin Mab 5B2 showed mesothelin positivity in 41–53% of lung adenocarcinomas (6, 20). Despite differences in the pattern of mesothelin staining in epithelial mesotheliomas (in which the staining is membranous) and lung adenocarcinomas (in which the staining is predominantly cytoplasmic), it is unlikely that mesothelin immunostaining will help the pathologist to distinguish between these two tumors. However, because mesothelin immunostaining is a very sensitive positive marker for epithelial mesothelioma, a negative mesothelin immunostain suggests a diagnosis other than mesothelioma (16).

Another tumor type in which mesothelin immunostaining may be of value to the pathologist is pancreatic adenocarcinoma because mesothelin expression is noted only in the cancerous tissue and not in normal pancreas or other benign histologies, such as chronic pancreatitis (6, 17, 18). This is especially important in the interpretation of pancreatic fine-needle aspiration specimens, a procedure now commonly used for initial

pathological diagnosis, given the cytologic overlap between malignant and reactive processes (34).

## CONCLUSION

Mesothelin is a differentiation antigen that is highly expressed in several malignancies and is being evaluated as a target for antibody- and vaccine-based therapies of cancer. Also, soluble mesothelin could be valuable as a tumor marker for diagnosis and follow-up of patients with mesothelin-expressing malignancies. In addition, immunohistochemistry studies evaluating mesothelin expression show it to be valuable to the pathologist for tumor diagnosis.

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