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Technetium-99m-sestamibi scintigraphy in gynecological cancer imaging

J. Stelmachów, *M.D., Ph.D.*; **A. Timorek-Lemieszczuk,** *M.D.*

Chair and Department of Obstetrics, Gynecology and Oncology, II Faculty of Medicine. Medical University of Warsaw (Poland)

Summary

A new diagnostic method, technetium-99m-sestamibi scintigraphy, and its potential use in gynecological oncology is described. The biochemical mechanisms of uptake and retention of technetium-99m-sestamibi in neoplastic cells are presented and the grounds for the potential use of the tracer in predicting the response to first-line chemotherapy in ovarian cancer patients are discussed. Based on the available literature data and on our own studies, the sensitivity and specificity of technetium-99m-sestamibi scintigraphy in ovarian cancer diagnosis are assessed, and the current place of this method among other functional imaging methods applied in gynecological oncology is discussed. Technetium-99m-sestamibi scintigraphy seems to provide an attractive alternative method to the expensive PET imaging, and can be easily performed in most hospitals. However, further studies in a larger series of patients are necessary before this method is widely applied.

Key words: 99mTc-sestamibi scintigraphy; Ovarian cancer; Chemotherapy.

Hexakis (2-methoxyisobutylisonitrile) technetium-99m (99mTc-sestamibi) is a radiopharmaceutical, originally developed for myocardial perfusion imaging, as its myocardial accumulation depends on blood flow and relates to mibi cation retention by mitochondria [1-3]. The potential use of 99mTc-sestamibi in oncology was first proposed in 1989 by Hassan *et al*. [4]. The authors showed an abnormal, elevated uptake of the marker in ten out of 13 patients with lung cancer, with a sensitivity of 77%. These results, along with the subsequent data on bone cancer published by Caner *et al*. [5], inspired a series of studies on 99mTc-sestamibi application in cancer diagnosis.

O'Tauma *et al*. [6] have shown 99mTc-sestamibi to be a highly specific agent for detection of metabolic activity in childhood brain tumors. An intense uptake of the tracer was presented in tumor-containing areas compared to uninvolved brain. These findings "opened the doors" to the use of 99mTc-sestamibi for diagnostic purposes in patients with central nervous system tumors. Further studies on 99mTc-sestamibi proved its value for imaging primary and metastatic tumors of the thyroid gland, brain, bronchus and breast [7-11]. The physical properties of 99mTc-sestamibi, i.e., gamma-ray energy and radiation dose, enables its usage with the conventional gamma radiation camera systems.

^{18}Fluorodeoxyglucose (FDG) is a functional molecular imaging agent that detects the increased glucose metabolism of malignant tumors. FDG- position emission tomography (PET) imaging can reveal biochemical differences between normal and malignant tissues [12]. Hybrid PET-CT is a scanner that allows acquisition of spatially registered PET and CT data in one procedure, simultaneously providing anatomic and functional information and potentially improving both disease detection and its characterization.

Recently, PET-CT has emerged as a clinically important tool for evaluating patients with gynecologic malignancies, i.e., cervical, endometrial, and ovarian cancers [13-15], and the role of PET-CT in guiding therapeutic decisions is growing. PET-CT, with its functional imaging capabilities, offers the best available modality for detecting lymph nodes involved by cervical cancer [16]. The identification of metastatic lymph nodes with conventional CT and MRI is based on size, with short-axis diameter greater than 1 cm being a widely accepted criterion for the diagnosis of neoplastic involvement [17]. However, metastatic deposits in normal-sized lymph nodes and reactive lymph node enlargement do not allow us to reliably diagnose cancer infiltration either by CT or MRI, which can lead to false negatives and positives. Among contemporary imaging methods, functional imaging, besides morphological imaging, has emerged as a clinically important tool for diagnosing female genital tract diseases. There is a growing role of radioisotope-employing methods, especially in gynecological oncology. The development of specific new-generation tracers improves sensitivity and specificity of nuclear medicine techniques, which can be of a particular value when morphological imaging is ineffective or not conclusive.

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Krolicki *et al*. [18] were the first to apply 99mTc-sestamibi for initial diagnosis of pelvic masses. The increased uptake of the tracer was found in adnexal malignant tumors and abdominal metastases derived from these tumors. The authors have also described the method with its limitations related to the physiological uptake of the tracer by normal colon, liver, gallbladder, large abdominal vessels as well as by the pelvis in women of reproductive age. Initially, colonic uptake of the tracer was regarded to be an inevitable obstacle in pelvic lesion imaging. However, the imaging was found to be effective if performed shortly, i.e., 2-3 min after 99mTc-sestamibi injection; such procedure was found not to impair tumor uptake. In addition, intravenously injected glucagon was demonstrated to reduce background colon uptake and to improve the quality of images. Finally, a better differentiation between primary and metastatic tumors was possible if the tracer uptake in the pelvis and abdominal cavity was analyzed separately.

To evaluate the use of 99mTc-sestamibi in the diagnosis of pelvic masses and abdominal metastatic tumors further studies are necessary. We need to improve the methods used and to learn more about biochemical and biophysical properties of the tracer as well as the mechanisms of its uptake and release by malignant tumors.

Mechanisms of 99mTc-sestamibi cellular uptake

The 99mTc-sestamibi complex was described by Riche in the mid 1980s [19]. Stereochemical analysis has shown a stabile complex with a central technetium atom (Tc-99m), surrounded by six mibi ligands (2-methoxyl isobutyl isonitrile), with surface methoxyl groups being the only functional groups.

The mechanisms of 99mTc-sestamibi uptake and retention in a cell have been extensively studied by Piwnica-Worms *et al*. [20, 21] and Kronauge *et al*. [22], who examined novel agents for myocardial perfusion imaging. The neutral 99mTc-sestamibi complex was found to accumulate not exclusively in myocardial cells, and was shown to be stable in vivo. Further studies revealed biophysical grounds of the 99mTc-sestamibi complex retention in a cell [23]. The fundamental myocellular uptake mechanism of 99mTc-sestamibi was demonstrated to involve passive distribution across plasma and mitochondrial membranes. In this respect, the 99mTc-sestamibi complex behaves like lipophilic cationic traces that diffuse across the cellular membrane into the cell in response to transmembrane potential. The large negative transmembrane potentials, especially in mitochondria, are responsible for tracer retention within the intracellular structures [21]. Tracer retention follows the Nernst equation [24]. Biochemical and cellular pharmacological studies with the use of electron probe X-ray microanalysis (EPXMA) and conventional electron microscopy confirmed the accumulation of the lipophilic complex at the inner mitochondrial membrane. However, the majority of the complex remains unbound within the mitochondrial matrix.

The main qualities of the 99mTc-sestamibi as a radiopharmaceutical are as follows:

– high specific activity of 10^9 Ci/mol,
– rapid distribution kinetics,
– low level of non-specific binding to plasma proteins,
– short half-life of approximately 6.02 h [25].

In a subsequent study Piwnica-Worms *et al*. [26, 27] discovered an additional factor influencing intracellular accumulation of 99mTc-sestamibi, a trans-membrane P-glycoprotein (Pgp).

Pgp is a 170kD protein, first described by Ling and Juliano [28, 29]. Increased expression of Pgp, encoded by the *MDR1* gene, is associated with the development of multidrug resistance (MDR), thus it is linked to cancer treatment failure.

A classical experiment by Piwnica-Worms [26] has shown the potential use of the 99mTc-sestamibi complex to rapidly characterize Pgp expression in human tumors in vivo. Pgp was shown to actively transport the complex outside the cell. Studies on the cell lines with a different MDR1 gene expression confirmed this mechanism [30].

There is increasing interest in the use of 99mTc-sestamibi scintigraphy in oncology, both for diagnostic and predictive purposes. In our preliminary studies, we have examined the employment of 99mTc-sestamibi scintigraphy in patients with gynecological tumors. To date, most patients included in the study presented benign and malignant ovarian tumors, others were diagnosed with endometrial, uterine and vulvar carcinomas. Before scintigraphic imaging, all patients had been clinically examined, and abdominal and pelvic ultrasound scanning along with color Doppler scanning had been performed. At the beginning, we were afraid that physiological colon uptake of the tracer would make the imaging of gynecological lesions impossible, but scanning proved to be successful if performed within a few minutes after 99mTc-sestamibi injection. Physiological uptake of the tracer by the liver, gallbladder and small intestine is an obstacle in interpreting abdominal images, and further studies and experience are necessary to improve preoperative diagnosis. Tracer uptake was assessed separately in pelvic and abdominal areas. Out of 70 patients with ovarian tumors 30 were diagnosed by histopathological methods with ovarian cancer as FIGO Stages IB to IV. 99mTc-sestamibi scintigraphy was characterized by 70% sensitivity at 70% specificity, lower to that of color Doppler ultrasound (sensitivity 93%, specificity 81%). However, when abdominal metastases were diagnosed, 99mTc-sestamibi scintigraphy was found more sensitive at a comparable specificity.

Piwnica-Worms *et al*. [27] in the studies on cancer cell lines have shown that 99m-Tc sestamibi is a substrate for Pgp and can be used for cellular Pgp detection [31-33]. 99m-Tc sestamibi accumulation was found to diminish with

the increase of *MDR1* expression in breast cancer cell lines [34, 35]. Thus, 99m-Tc-sestamibi uptake may provide information about the Pgp status of the cells. These discoveries opened up extensive studies on 99m-Tc-sestamibi scintigraphy application for predicting response to chemotherapy in patients with breast and lung carcinomas, malignant melanoma, sarcomas and lymphomas [36-42]. The 99m-Tc sestaMIBI complex application in gynecological oncology was first examined in our pilot study [43] on 12 patients. We have demonstrated that the tracer accumulation assessment in patients undergoing chemotherapy may be useful for monitoring treatment response. We have also addressed the question of the value of 99m-Tc-sestamibi scintigraphy for monitoring the response to first-line chemotherapy in ovarian cancer patients. The 5-year survival rate for patients with clinically advanced ovarian cancer patients remains poor in spite of considerable efforts to improve the effects of treatment. The majority of cases are diagnosed at a late stage because there are no reliable screening techniques and early-stage ovarian cancer is generally asymptomatic. The essential ovarian cancer treatment is optimal surgery followed by chemotherapy.

We have examined 25 patients with epithelial ovarian cancer FIGO Stages IIb to IV, following primary surgical treatment. All patients were scheduled for scintigraphy after surgery and after three and six courses of paclitaxel/platin chemotherapy, which is now the "gold standard" of the first-line chemical treatment of ovarian cancer patients. Following the sixth course of treatment, the response to treatment was assessed by physical examination, ultrasound and CT imaging, and CA 125 level measurement.

Before treatment, 18 patients (72%) presented 99m-Tc-sestamibi uptake. After treatment, 13 women had a complete clinical remission, ten patients presented disease progression, and the remaining two had a partial clinical remission. In patients with complete clinical remission, there was no uptake of the tracer, in patients with partial remission the tracer uptake remained unchanged, while in 80% of those with disease progression there was a high level of tracer accumulation.

We have found a strong correlation between the scintigraphic and the standard estimation of response to chemotherapy in ovarian carcinoma patients. Only one patient with FIGO Stage IIIc ovarian carcinoma and progression of disease after completion of chemotherapy, and with a high level of 99m-Tc-sestamibi uptake before treatment, presented no tracer uptake after treatment. This is in accordance with the results of Goldstein [44] and Bourhis *et al*. [45], who have shown low levels or the lack of P-glycoprotein expression in ovarian cancer at the time of diagnosis. The lack of false-positive results in our study points to the high specificity of 99m Tc sestamibi scintigraphy in estimating the response to treatment of patients with ovarian cancer. False-negatives were shown in two patients. Similar results were presented by Marshall *et al*. [46] and Dunnwald *et al*. [47] in breast cancer patients.

In summary, 99m-Tc-sestamibi scintigraphy in ovarian cancer patients seems to have a clinical value both for the preoperative diagnosis and for monitoring of treatment response. Further studies in a larger series of patients are necessary to confirm the utility of this method.

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Address reprint requests to:
J. STELMACHÓW, Ph.D., M.D.
Department of Obstetrics,
Gynecology and Oncology
II Faculty of Medicine
Medical University of Warsaw (Poland)
e-mail: klingi@amwaw.edu.pl