Serum tumor markers
Part I: Clinical Utility

By Daniel M. Hoefner, MT, PhD, DABCC, FACB

Tumor markers are chemicals found in the body (i.e., tissue and fluids) that have application in the management of patients who currently have, are suspected of having, or have had cancer. Tumor markers are usually constituents of healthy cells that are produced in greater abundance by cancerous cells; however, normal cells may also produce them in response to the malignancy. In addition, tumor growth may cause an obstruction and/or cellular breakdown of normal tissue — either of which could lead to increased levels of an analyte that could serve as a surrogate marker of malignancy. These tumor-related molecules may include enzymes, structural proteins, various cell-surface carbohydrate antigens, and receptors, as well as genes.

Many taxonomic approaches have been used to classify tumor markers. One method is to categorize them based on a molecular or functional descriptor (e.g., glycoproteins or mucins, oncofetal proteins, hormones, and so on). Some are integral tissue proteins (e.g., receptors) and are routinely measured by immunohistochemical staining of biopsy materials, while others are excreted in urine and measured therein. Another group of markers, loosely referred to as genetic or molecular tumor markers, consists of oncogenes, tumor suppressor genes, and receptors — molecules that are routinely detected by way of molecular testing, such as PCR or other amplification techniques. Serum tumor markers is a term commonly used to refer to molecules that can be detected in a blood sample by immunoneuocochemical methods; however, this term is somewhat of a misnomer, in that nearly all of the tumor markers can be detected in serum samples. This article is the first of a two-part review that will discuss the tests in highest usage (in this latter classification). These include α-fetoprotein (AFP), CA 125, CA 15-3, CA 27.29, CA 19-9, carcinoembryonic antigen (CEA), choriogonadotropin (hCG), PSA, and thyroglobulin. Part II will focus on practical considerations and limitations regarding their use.

Tumor markers in common use

In the clinical setting, there are many uses for these cancer-related tests; however, not all of the markers are appropriate for every purpose. Table 1 indicates their most common clinical utilities. As they are generally detectable in all healthy individuals, it is not their presence in serum, but their quantity, that makes tumor markers useful. Also, because noncancerous cells generally produce low levels of tumor-marker molecules, clinical specificity is usually low and due to the low prevalence of cancer, many false-positive results would occur if they were used to screen the mass population. Therefore, few serum tumor markers are recommended for general screening purposes. In cancer diagnosis, these analytes should never be used in vacuity. It is changes in their concentration, over months and years, which are used to assess disease progression and response to treatment, to monitor a patient for recurrence, or to indicate the aggressiveness of a malignancy. Unfortunately, early indications that are suggestive of cancer recurrence often have relatively little benefit on clinical outcome — as cancers come back, they are frequently more aggressive than the primary tumor. While tumor-marker levels that fall within the “normal” reference interval do not necessarily rule out that the cancer is still present, consecutively rising levels are a good indicator that the cancer has returned. In addition, when considered in light of the clinical picture, the markers can aid in the differential diagnosis and serve as an indicator of the primary site for malignancies that have metastasized.

Table 1. Common clinical uses of serum tumor markers

<table>
<thead>
<tr>
<th>Utility</th>
<th>Example</th>
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<tbody>
<tr>
<td>Diagnosis</td>
<td>While not used solely for this purpose, markers can aid in making a diagnosis and in locating the source of cancers that have metastasized.</td>
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<tr>
<td>Monitoring for recurrence</td>
<td>After a patient has been successfully treated, some markers are tested at regular intervals to indicate whether there has been a recurrence of the cancer.</td>
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<tr>
<td>Prognosis and staging</td>
<td>To aid in the estimation of tumor volume, as an indicator of disease progression and aggressiveness, or as an indication of metastatic involvement.</td>
</tr>
<tr>
<td>Detection of residual disease</td>
<td>After cancer surgery, testing can be used to indicate whether the entire tumor burden has been successfully removed.</td>
</tr>
<tr>
<td>Screening</td>
<td>Used to test patients without symptoms. With the exception of PSA, the screening population is usually reserved for those individuals at high risk for a given cancer (e.g., genetically-linked cancers).</td>
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<tr>
<td>Monitoring treatment</td>
<td>A means to assess the success of treatment by monitoring a patient’s response to various treatment regimens. In general, levels will drop if treatment is beneficial and will remain elevated or increase if it is ineffective.</td>
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</table>

The National Academy of Clinical Biochemistry tumor marker practice guidelines list approximately 150 analytes and methods that are currently under investigation for their utility as markers for various malignancies. At the time of this writing, all of the guidelines were in their draft stage, with six of the 17 documents still pending release. While the number of markers will likely continue to grow considerably, very few tests have received FDA approval for use as tumor markers. With the exception of hCG, the analytes presented in Parts I and II of this review have all been approved by the FDA for use as tumor markers, albeit the indications for their use in this capacity may be limited.

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Tests in highest usage

Figure 1 indicates the relative testing volumes for our laboratory, based on the first two quarters of 2005. Of all analytes tested at our facilities, these tests represent the tumor markers of highest volume. Calcitonin (not discussed in this article) would be next on the list with only ~15% of the volume of CA 15-3; test volumes for all other individual serum-based tumor markers were less than 0.1% of that for total PSA. Since requests do not indicate the intended utility, volumes for tumor marker use are not easily estimated for hCG; however, due to its utility in cancer assessment, it is likely that it is on the “Top 10” list of serum tumor markers, based on volumes.

Table 2 is a summary of estimated hospital test volumes for 2004 based on data supplied by the Voice of the Clinical Lab, a sister publication of the Market Monitor (Information Dynamics; West Chester, PA). AFP and hCG are not included, as the usage of these two tests is not confined to cancer testing; thyroglobulin data were not collected. The column, Average Annual Volume per Hospital, was calculated as the Total Annual Hospital Volume/5,134 x Percent of Hospitals Performing Test. For projection purposes, 5,134 is the estimated number of acute care hospitals in the United States.

It is interesting to note that, while the number of CA 27.29 tests performed is about fourfold higher than CA 15-3 in our reference lab (Figure 1), CA 15-3 is offered ~2½ times more hospital-based laboratories than CA 27.29. Based on the average annual volume per hospital (Table 2), one can speculate that larger hospitals or those that do the majority of testing for a hospital consortium are likely to be the facilities to offer testing services for analytes such as CA 19-9 and CA 27.29, since the average annual volume for these analytes is much higher than for the other tumor markers. For CA 27.29, this is likely due to only one vendor supplying an automated method. Therefore, if smaller hospitals do not use that vendor for their immunoassay testing, requests for CA 27.29 from those locations are probably sent to a reference laboratory. This may also explain why reference laboratories perform significantly more CA 27.29 testing than CA 15-3.

Because PSA is recommended for routine screening of men over the age of 50, the testing volumes are significantly higher than the other markers (Figure 1, Table 2). No other cancer-related marker has been approved for general screening purposes.

Clinical practice guidelines

Table 3 summarizes two of the most comprehensive cancer guidelines available — by the European Group on Tumour Markers (EGTM) and the National Academy of Clinical Biochemistry (NACB) — as well as a few cancer-specific recommendations. The proposals put forth for tumor-marker use are not always clear-cut in every instance. For some tumor markers, there are caveats associated with specific recommendations (e.g., “if no other test is available, this marker can be used to monitor treatment response...”). For further clarification, please refer to the cited materials referenced in Table 3. Since hCG has not been FDA-approved for use as a tumor marker, it has not been included in this table. Although the evidence is not sufficient for practice guidelines to suggest routine use of most tumor markers for the detection of residual disease following treatment, several markers are employed in certain situations for this purpose.

Summary of clinical utilities

α-Fetoprotein (AFP). In the normal fetus, circulating AFP is present in significant levels; however, after birth, the concentration declines to adult levels by ~18 months. Due to leakage across the placenta, this analyte is elevated in maternal serum (gestational age-dependent) to levels well above the reference interval for nonpregnant individuals. It is routinely measured in maternal serum as a marker of open neural tube defects and trisomy disorders of the fetus. For oncological purposes, this marker is used primarily for the detection of liver cancer and germ cell malignancies of the testis and ovary. It is also frequently used in tandem with hCG for monitoring patients with nonsemionomatous germ cell tumors. While in the United States, this marker is not used routinely to screen for cancers, it has been shown to have utility for this purpose in areas of the world with a high incidence of hepatocellular carcinoma (e.g., Africa and parts of Asia).

Cancer antigen 125 (CA 125). CA 125 is primarily used for ovarian and endometrial cancers. Unfortunately, this marker is not sensitive enough for use in detecting very early stages of these cancers and is ineffective as a screening tool in asymptomatic women. It is, however, elevated in ~50% of patients with stage I ovarian cancer, increasing to above 90% in advanced stages of disease. Its greatest utility is to serve as a means to assess cancer therapy and to monitor patients for recurrence. Following treatment, there is an association between significantly elevated levels and a bleak prognosis. In women with an ambiguous pelvic mass, it can also aid in the differentiation between malignant and benign tumors. CA 125
for routine screening of ovarian cancer is not recommended by any medical organization. A recent report by Smith, et al, however, suggests that early diagnosis of ovarian cancer may be significantly delayed in some women because physicians do not order the appropriate testing, such as CA 125, early enough in patients with unexplained “target symptoms.”

**Cancer antigen 15-3 and cancer antigen 27.29 (CA 15-3, CA 27.29).** These two markers are listed together because they actually measure different epitopes of the same antigen. Their primary use is for monitoring therapy in patients with breast cancers — although they may be elevated in other cancers as well. Their principal utility is in monitoring therapy; however, they also have value for evaluating patients for breast-cancer recurrence, assessing prognosis, and checking for residual disease following surgery. While overall volumes for CA 15-3 testing are slightly higher than CA 27.29, this appears to be a function of the number of vendors that market an immunoassay for the following surgery. While overall volumes for CA 15-3 testing are slightly higher than CA 27.29, this appears to be a function of the number of vendors that market an immunoassay for the former test, as neither analyte appears to hold an advantage over the other for use as a marker of malignancy.

<table>
<thead>
<tr>
<th>Table 3. Summary of clinical guidelines for the use of tumor markers in selected cancers</th>
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<tbody>
<tr>
<td>Marker (cancer type)</td>
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<tr>
<td>-----------------</td>
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<tr>
<td>AFP (germ cell tumors)</td>
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<tr>
<td>AFP (liver cancer)</td>
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<tr>
<td>CA 125 (ovarian)</td>
</tr>
<tr>
<td>CA 15-3 / CA 27.29 (breast)</td>
</tr>
<tr>
<td>CA 19-9 (pancreatic)</td>
</tr>
<tr>
<td>PSA, total (prostate)</td>
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<tr>
<td>Thyroglobulin (thyroid)</td>
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</tbody>
</table>

**Abbreviations used**

- D: Diagnostic aid
- M: Monitoring for recurrence
- P: Prognosis and staging
- R: Detection of residual disease
- S: General screening
- T: Monitoring therapy
- Calendar year
- Other recommendations

**Human chorionic gonadotropin (hCG, βhCG).** The primary utility of hCG is for determining pregnancy status. While this analyte has not been FDA-approved for use as a cancer marker, the literature supports its frequent use for this purpose; in addition, it has been used in this capacity for longer than any of the other listed analytes. Generally, elevated levels are seen in patients who have trophoblastic tumors and testicular cancer. In combination with AFP, this marker is frequently used to monitor patients with nonseminomatous germ cell tumors. hCG is also commonly used to monitor patients with gestational trophoblastic disease (e.g., molar pregnancy). With the high analytical specificity of hCG assays currently in use, there is insignificant interference from hormones that share the common heterodimer α subunit (e.g., LH, FSH, and TSH) — a phenomenon that was a concern in the past. Some tumors, however, produce primarily the β subunit, and assays that measure total βhCG (intact and free) may correlate better with tumor burden than assays that just measure intact hCG. 7

**Prostate-specific antigen (PSA).** This is one of the few organ-specific markers; unfortunately, it is not cancer-specific. Many have considered this to be a promising and near-ideal marker for prostate cancer. While studies that were performed 15 or more years ago suggested much potential, that was probably in large part due to the higher numbers of men with undiagnosed advanced prostate cancer. Now that the marker has been in use for many years, analyses by receiver-operator characteristic curves indicate that, due to its lack of specificity at levels that are required for sensitivity, it is mediocre in its utility. Many attempts have been made to enhance the effectiveness of this analyte, including the assessment of increasing in levels over time (PSA velocity), estimating the amount of PSA per unit volume of prostate tissue (PSA density), and the use of age-specific reference intervals. The primary utilization of PSA is for screening patients for prostate cancer; recommendations suggest that this be done in conjunction with digital rectal examination of the prostate gland.

The use of free PSA is advocated in patients with total PSA levels between 4 ng/mL and 10 ng/mL. In this group, the specificity for testing can be increased by reporting the percent free PSA value (free PSA / total PSA) — values >25% suggest that biopsy be performed. 8 Patients with free PSA >25% should continue to be monitored with annual digital rectal examination and PSA.

**Thyroglobulin (Tg).** This is another tumor marker that is organ-specific; its use is primarily for patients who have differentiated thyroid cancer. Before surgery, it is important to establish the thyroglobulin concentration, which is used for two purposes — to assess whether the tumor is producing thyroglobulin and to use the value as a reference to judge the effectiveness of surgery and radioablation therapy at eradicating the malignancy. If the tumor does not release a significant amount of thyroglobulin into the circulation, it will not be an effective tool for evaluating the therapy. In patients who have undergone successful total thyroidectomy and/or radioablation, thyroglobulin levels should be virtually undetectable; and as long as there is no residual or recurrent disease, they should remain that way.

In conjunction with measuring Tg, it is critical to measure antithyroglobulin antibody levels in all samples. These antibodies are not necessarily the result of, or indicative of,
pathology. If they are present in the patient’s serum, however, the thyroglobulin results could be rendered invalid since the patient’s anti-Tg antibodies may interfere with the Tg assay. This important factor should be taken into consideration as the clinician evaluates the patient’s status.

Summary

Most tumor markers in current use are glycoproteins that are measured by routine immunoassay techniques. The primary utility of serum tumor markers is for evaluating the effectiveness of therapy in advanced stages of cancer; in addition, they are used for monitoring “cured” patients for cancer recurrence. Regrettably, this latter use has not led to a significant improvement in patient outcomes when a recurrence is detected early.

Sensitivity and/or specificity is frequently lacking for most tumor marker tests and their utility as screening tools to detect early cancer is extremely limited. For the most part, decisions based on the concentration of these cancer-associated molecules should always be made in light of the entire clinical picture. A monumental goal in cancer research is to find markers that are significantly more sensitive and specific for early cancer detection, as well as the other uses described herein.

In Part II of this series, practical considerations and limitations regarding laboratory testing of tumor markers will be addressed.

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References