Policy Statement

Tumour Marker Laboratory, PHSA Laboratories, Central Processing and Receiving, Lane Level.

The objective of the Tumour Marker Lab (TML) is to provide province-wide serum tumour marker service for oncologists and other physicians managing cancer patients. We have a number of services unique to the laboratory described in detail in the table below

While these assays have a variety of potential applications, in general, they are most valuable for patient monitoring and not applicable for diagnostic screening.

Overview of the clinical utility of serum tumour markers:

What are Serum Tumour Markers?

- Most are glycoproteins released by tumour cells into the blood stream or other body fluids
- One is a glycolipid (CA19-9) carrying the Lewis blood group determinant
- They are rarely tumour-specific or even organ specific
- They may be elevated in benign conditions

How Should Tumour Markers Be Used?

- They are not suitable for screening of asymptomatic patients
- They may be used as diagnostic ancillary tests in certain situations
- They are mainly used for the follow up and monitoring of patients post surgery or post chemotherapy/radiation therapy
- Any single value is less important than trends over time
- Levels may fluctuate and increase during therapy due to tumour cell death and may not reflect tumour burden

Should one use one Lab or Multiple Labs?

- Since longitudinal data is important, any given patient should be tested by the same lab each time because of methodological differences.
- For all types of tests that rely on immunoassays, there is no “gold standard”. Results vary by vendor due to analysis platform, calibrator standards, and reagents used.

Hours of service: **8am to 4:30pm, Monday to Friday**

Results and General inquiries: **1-877-747-2522**

For Clinical Consultation for Physicians only, contact:

For physician consultation only- please call VGH at 604-875-4111 and ask operator to page the Biochemist on call.

Turnaround times

A serum tumour marker test is rarely a stat test. As we run most assays on a daily basis, our turnaround time is generally the same working day or next working day. Because of our use of third party results distribution vendors that use batch printing or batch release of electronic results, those physicians who do not have access to the Cancer Agency Information System (CAIS) will not usually see the results until the next working day. Some assays (Chromogranin A, Beta 2 microglobulin) are of low volume usage and are only run once a week.

Measurement of uncertainty is available upon request

Tumour Marker Assays

	AFP	CA 125	CA 15-3	CA 19-9
Assay principle	**Direct Chemiluminometric Immunoassay**	**Direct Chemiluminometric Immunoassay**	**Direct Chemiluminometric Immunoassay**	**Direct Chemiluminometric Immunoassay**
Sample collection	**Serum sample**	**Serum sample**	**Serum sample**	**Serum sample**
Transport requirement	**4°C**	**4°C**	**4°C**	**4°C**
Turnaround Time	**48 hours**	**48 hours**	**48 hours**	**48 hours**
Reporting – Alert Limits	**≤8 µg/L**	**≤32 kU/L**	**≤32 kU/L**	**≤35 kU/L**
Assay Range	**1.3 – 1000 µg/mL (1.08 – 830 kU/L)**	**2 – 600 kU/L**	**0.50 – 200 kU/L**	**1.2 – 700 kU/L**

	CEA	ThCG	Beta 2 Microglobulin	SCC
Assay principle	**Direct Chemiluminometric Immunoassay**	**Please contact VGH**	**Chemiluminescent Immunoassay**	**Direct Chemiluminometric Immunoassay**
Sample collection	**Serum sample**		**Serum sample**	**Serum sample**
Transport requirement	**4°C**		**4°C**	**4°C**
Turnaround Time	**48 hours**		**7-10 days**	**72 hours**
Reporting – Alert Limits	**≤5 µg/L**		**<2.2 mg/L**	**≤ 70 µg/L**
Assay Range	**0.5 – 100 µg/L**		**Up to 20 mg/L**	**0 - 70 µg/L**

	PSA	Free PSA	Testosterone	Chromogranin A
Assay principle	Direct Chemiluminometric Immunoassay	Direct Chemiluminometric Immunoassay	Direct Chemiluminometric Immunoassay	Enzyme Immunoassay (ELISA)
Sample collection	Serum sample	Serum sample	Serum sample	Serum sample
Transport requirement	4°C	4°C	4°C	4°C
Turnaround Time	48 hours STAT testing available with Medical consult	48 hours STAT testing available with Medical consult	48 hours STAT testing available with Medical consult	7-10 days
Reporting – Alert Limits	Age (years) / Upper Limit (µg/L) < 40 ≤ 2.0 40-49 ≤ 2.5 50-59 ≤ 3.5 60-69 ≤ 4.5 70-79 ≤ 6.5 ≥ 80 ≤ 7.2	Please refer to TML Program Guide to Services	Males Age (years) / Upper Limit (nmol/L) < 50 8.6-29 ≥ 50 6.7-26 Females Age (years) / Upper Limit (nmol/L) < 50 0.29-1.7 ≥ 50 0.10-1.4	<94 µg/L
Assay Range	0.01 - 100 µg/L	0.01 - 25 µg/L	0.24 – 52.0 nmol/L	Up to limit determined by test lot (typically 700 – 800 µg/L)

	Gastrin	**Soluble Interleukin 2**		
Assay principle	**Chemiluminescent Immunometric assay**	**Chemiluminescent Immunometric assay**		
Sample collection	**Serum sample**	**Serum sample**		
Transport requirement	**Frozen**	**4°C**		
Turnaround Time	**7-10 days**	**7-10 days** **STAT testing available with Medical consult**		
Reporting – Alert Limits	**< 115 pg/ml**	**≤19 yrs: No reference range** **>19 yrs: 241-846 U/mL**		
Assay Range	**5-1000 pg/ml**	**50 – 7500 U/mL**		

Carcinogenic Embryonic Antigen (CEA)

Because of a lack of specificity and sensitivity for early disease, CEA has no role in either screening or early diagnosis. In patients with colorectal cancer, CEA measurement before surgery is recommended by the ASCO because the result may complement pathologic staging and aid surgical treatment planning, and both the AJCC and College of American Pathologists (CAP) have proposed that CEA be included in the staging system for colorectal cancer.

Abnormal preoperative CEA values are associated with a higher risk of recurrence.

There is no evidence that patients benefit from adjuvant therapy solely on the basis of abnormal preoperative CEA concentrations.

The CEA assay is used primarily to monitor the treatment and possible recurrence of colorectal carcinoma. CEA may also be used for breast, lung, pancreatic and gastric malignancies.

Non-malignant elevations may be seen in benign polyps, colitis, cirrhosis, hepatitis, chronic lung disease and smokers. Values > 10-15 µg/L are rarely seen in benign conditions.

As with most tumour marker assays, it is the changing concentrations of CEA levels over a period of time that is used as a management aid. Where curative surgery is contemplated, pre-operative CEA levels may have prognostic significance. CEA is not recommended as a diagnostic screening test.

The CEA assay is a two-site sandwich immunoassay using direct chemiluminometric technology performed on the Siemens ADVIA Centaur. The assay standardization is traceable to an internal Siemens standard manufactured using highly purified material.

Serum is the recommended sample type.

Reference Range: < 5 µg/L

Interferences: Human anti-mouse antibodies (HAMA), heterophilic antibodies

Alpha-Fetoprotein (AFP)

Alpha-Fetoprotein levels are used primarily as markers in non-seminomatous testicular cancer and primary hepatocellular carcinomas. AFP may also be important in the diagnosis and staging of non-seminomatous tumours. Baseline levels are recommended as part of the initial work-up of patients suspected of having primary hepatocellular carcinoma or testicular cancer. Serial values may then be helpful for post treatment management. For non-seminomatous testicular carcinoma, AFP is usually used with ß-hCG.

AFP may also be elevated as a result of liver metastases arising from other primary cancers. Non-malignant elevations may be seen in hepatitis, cirrhosis and in normal pregnancy. Greater than normal pregnancy levels of AFP in maternal serum may be associated with fetal development problems such as neural tube defects.

The AFP assay is a two-site sandwich immunoassay using direct chemiluminometric technology performed on the Siemens ADVIA Centaur. The assay standardization is traceable to World Health Organization (WHO) Reference Preparation for human AFP (72/225).

Serum is the recommended sample type.

Reference Range: ≤ 8 µg/L

Interferences: Human anti-mouse antibodies (HAMA), heterophilic antibodies, amniocentesis, hepatitis and cirrhosis.

Human Chorionic Gonadotrophin (beta subunit) β-Hcg

Testing has been referred to Vancouver General Hospital

Prostate Specific Antigen (PSA) – Total and Free

Total PSA (Prostate Specific Antigen) is used in the Tumour Marker Lab primarily in the management of prostate cancer. It is used as a diagnostic test, along with Free PSA and the Percent Free vs. Total PSA. It is of significant value in detecting metastatic or persistent disease following treatment. Persistent elevation following treatment or increase in post treatment levels is indicative of recurrent or residual disease.

PSA is of unknown value as a population screening test. Although there is good evidence that it increases the detection rate of early stage, clinically significant, prostate cancers, there is little evidence to date that such early detection leads to reduced mortality, the "gold standard" for evaluating screening tests. Fit men (men with at least 10 years life expectancy) between the ages of 50 and 70 should be made aware of the availability of PSA as a detection test for prostate cancer. They should be aware of the potential benefits and risks of early detection so they can make an informed decision as to whether to have the test performed.

Benign prostate hyperplasia (BPH) is a major problem that must be differentiated from prostate cancer. Options to help differentiate cancer include the rate of increase in PSA level (watchful waiting) over time, the free to total PSA ratio, PSA density (PSA level relative to prostate volume) and age adjusted PSA decision levels. Another clinical problem that can lead to very elevated PSA is prostatitis.

The PSA assay assay is is a two-site sandwich immunoassay using direct chemiluminometric technology performed on the Siemens ADVIA Centaur. The assay standardization is traceable to World Health Organization (WHO) Reference Standard for human PSA (96/670).

Serum and plasma (collected in Li-heparin or K_3-EDTA) are the recommended sample types.

Age related Reference Ranges for PSA-Total:

Age (Years)	PSA Upper Limit (µg/L)
< 40	≤ 2.0
40-49	≤ 2.5
50-59	≤ 3.5
60-69	≤ 4.5
70-79	≤ 6.5
≥ 80	≤ 7.2

FREE PSA

When total PSA is in the range of 4.0-10.0 µg/L, a free:total PSA ratio ≤ 0.10 indicates 49% to 65% risk of prostate cancer depending on age; a free:total PSA ratio > 0.25 indicates a 9% to 16% risk of prostate cancer, depending on age.

Free:Total PSA ratio	50-59 years	60-69 years	≥70 years
≤ 0.10	49.2%	57.5%	64.5%
0.11-0.18	26.9%	33.9%	40.8%
0.19-0.25	18.3%	23.9%	29.7%
> 0.25	9.1%	12.2%	15.8%

(Source: Mayo Clinic/Medical Laboratories)

Interferences: Human anti-mouse antibodies (HAMA), heterophilic antibodies, prostate manipulation including rectal examination, diagnostic intervention, biopsy, prostatectomy or prostate massage. It is recommended that PSA serum samples be taken prior to or 2-3 weeks after any of the above procedures.

References:

1. Verification of Harmonization of Serum Total and Free Prostate-Specific Antigen (PSA) Measurements and Implications for Medical Decisions

Simona Ferraro,a,* Marco Bussetti,a Sara Rizzardi,b Federica Braga,a,c and Mauro Panteghinia
Clinical Chemistry 67:3 Cancer Diagnostics 543–553 (2021)

2. Serum Prostate-Specific Antigen Testing for Early Detection of Prostate Cancer: Managing the Gap between Clinical and Laboratory Practice

Simona Ferraro,* Marco Bussetti, and Mauro Panteghini
Clinical Chemistry 67:4 Mini-Review 602–609 (2021)

Cancer Antigen 15-3 (CA15-3)

The CA15-3 (Cancer Antigen 15-3) antigen is a highly polymorphic glycoprotein belonging to the mucin family and is the product of the MUC-1 gene. It is used primarily as a marker for cancer of the breast, but may also be elevated in cancers of the stomach, liver, pancreas, lung and ovary. As with other tumour markers, CA15-3 is most useful for serial monitoring.

Non-malignant elevations may be seen in hepatitis and cirrhosis.

The CA15-3 assay is a two-site sandwich immunoassay using direct chemiluminometric technology performed on the Siemens ADVIA Centaur. Currently, there is no reference standard for CA15-3. The assay standardization is traceable to an internal Siemens standard manufactured using highly purified material.

Serum is the recommended sample type.

Reference Range: ≤ 32 kU/L

Interferences: Human anti-mouse antibodies (HAMA), heterophilic antibodies and pregnancy.

Caveats:

Breast Cancer & CA 15-3 or CEA

- Low sensitivity in early-stage disease, lack of specificity, and controversy about whether the test benefits outcome
- Not recommended by any group for screening, diagnosis, or staging of breast cancer
- Major obstacles identified in relation to the use of CA15-3 as an indicator of asymptomatic recurrence include the low incidence of raised CA15-3 in early-stage disease, the lack of effective treatment options for recurrences detected, and the low efficiency of detection
- ASCO guidelines support use of CA15-3 to suggest treatment failure where disease is not readily measurable
- Use of CEA in breast cancer: CEA should only be measured if CA15-3 is not increased at presentation
- Mild to moderate elevations of CA 15-3 are seen in a variety of conditions, including liver and pancreatic cancer, cirrhosis, lung, pancreatic, ovarian, and colorectal cancers, benign breast disorders as well as in a certain percentage of apparently healthy individuals.
- The CA 15-3 elevations seen in non-cancerous conditions tend to be stable over time.

•Normal CA 15-3 levels do not ensure that a person does not have localized or metastatic breast cancer. It may be too soon in the disease for elevated levels of CA 15-3 to be detected or the person may be one of the 25% to 30% of individuals with advanced breast cancer whose tumors do not shed CA 15-3.

Cancer Antigen 125 (CA125)

The CA125 (Cancer Antigen 125) antigen is a 200 to 1000 kDa mucin-like glycoprotein primarily associated with cancer of the ovary. It is detectable in adult pleura, pericardium and peritoneum.

Elevated CA125 levels can also be seen in association with malignancies of breast, cervix, uterus, liver, pancreas, stomach, colorectum and lung.

Non-malignant elevations of CA 125 have been reported for ascites, cirrhosis, hepatitis, pancreatitis, fibroids, endometriosis, ovarian cysts, first trimester pregnancy and pelvic inflammatory disease.

CA125 is best used for patient monitoring. It is of only limited value as a diagnostic aid for intra-abdominal malignancy.

The CA125 assay is a two-site sandwich immunoassay using direct chemiluminometric technology performed on the Siemens ADVIA Centaur. Currently, there is no reference standard for CA125. The assay standardization is traceable to an internal Siemens standard manufactured using highly purified material.

Serum is the recommended sample type. Plasma should not be used because its performance has not been validated for this assay.

Reference Range: ≤ 32 kU/L

Interferences: Human anti-mouse antibodies (HAMA), heterophilic antibodies. Possible hook effect, leading to falsely lower values, may be seen in patients with very high CA125 levels.

Carbohydrate Antigen 19-9 (CA19-9)

CA19-9 (Carbohydrate Antigen 19-9) is present in the fetal epithelium of the stomach, intestine, liver and pancreas. In adults traces can be found in the pancreas, liver, gallbladder and lung. Its primary use is as a marker for cancer of the pancreas. It is also elevated in cancers of the liver, lung, breast, uterus and ovary (mucinous). CA 19-9 is high in normal mucinous secretions such as saliva, bile and semen.

CA19-9 may also be elevated in non-malignant liver diseases such as cirrhosis and hepatitis. Other less frequent non-malignant disease elevations include pancreatitis, autoimmune diseases, cystic fibrosis and disorders of the kidney, lung and the GI tract.

The CA19-9 assay is a two-site sandwich immunoassay using direct chemiluminometric technology performed on the Siemens ADVIA Centaur. Currently, there is no reference standard for CA19-9. The assay standardization is traceable to an internal Siemens standard manufactured using highly purified material.

Serum is the recommended sample type. Plasma should not be used because its performance has not been validated for this assay.

Reference Range: ≤ 35 kU/L

Interferences: Human anti-mouse antibodies (HAMA), heterophilic antibodies. Please note that patients with the Lewis negative blood group phenotype may not express CA19-9 antigen.

Caveats

- Several benign diseases including chronic and acute pancreatitis, liver cirrhosis, cholangitis and obstructive jaundice may give rise to elevated CA 19-9 levels.
- In patients with cholangitis, levels of CA 19-9 in excess of 1000 kU/L have been found. These elevated levels may return to normal after treatment of cholangitis and appropriate decompression of the common bile duct.
- In patients presenting with obstructive jaundice, levels of CA 19-9 should therefore be measured following biliary decompression.
- CA 19-9 can be increased in multiple types of adenocarcinoma, especially in advanced gastrointestinal cancers:
    - 67% of patients with bile duct cancer,
    - 41% of patients with gastric cancer,
    - 34% of patients with colorectal cancer,
    - 22% of patients with esophageal cancer
    - 49% of patients with hepatocellular carcinoma
- CA 19-9 is not expressed in subjects with Lewis A negative genotype.
- The molecules on which the CA 19-9 epitope is found is a sialylated Lewis A blood group antigen.
- Subjects who are genotypically Lewis a- b- therefore cannot synthesize the CA 19-9 epitope.
- Approximately 5%–10% of the Caucasian population are believed to have this genotype.
- Little information is available on the prevalence of the Lewis a- b- genotype in non-Caucasian populations.

Testosterone

Testosterone is a male sex hormone secreted by the testes. It gradually increases from childhood through puberty until its adult level. In females testosterone is mainly produced by peripheral conversion of prehormones and remains low. Testosterone monitoring is used clinically to diagnose and differentiate endocrine disorders in both males and females.

In the oncology setting, anti-testosterone (anti-androgen) therapy is used as a form of hormonal treatment in men with prostate cancer. The goal is to produce chemical castration by eliminating the testosterone stimulation of the prostate cancer cells. Serum testosterone levels are measured to determine the success of such treatment.

The Testosterone assay is a competitive immunoassay using direct chemiluminescent technology performed on the Siemens ADVIA Centaur. This assay has been standardized using Isotope Dilution-Gas Chromatography/Mass Spectrometry (ID-GC/MS).

Serum and plasma (other than sodium citrate anticoagulated plasma) are the recommended sample types.

Reference Ranges:	Males < 50 years:	8.6-29 nmol/L
	Males ≥ 50 years:	6.7-26 nmol/L
	Females < 50 years:	0.29-1.7 nmol/L
	Females ≥ 50 years:	0.10-1.4 nmol/L

Interferences: Human anti-mouse antibodies (HAMA), heterophilic antibodies

ß$_2$-Microglobulin (B2M)

Useful For

Prognosis assessment of multiple myeloma

Clinical Information

Beta-2-microglobulin (beta-2-M) is a small membrane protein (11,800 Dalton) associated with the heavy chains of class I major histocompatibility complex proteins and is, therefore, on the surface of all nucleated cells. The small size allows beta-2-M to pass through the glomerular membrane, but it is almost completely reabsorbed in the proximal tubules.

Serum beta-2-M levels are elevated in diseases associated with increased cell turnover. Levels are also elevated in several benign conditions such as chronic inflammation, liver disease, renal dysfunction, some acute viral infections, and a number of malignancies, especially hematologic malignancies associated with the B-lymphocyte lineage.

In multiple myeloma, beta-2-M is a powerful prognostic factor and values <4 mg/L are considered a good prognostic factor.

Reference Ranges: < 2.2 mg/L

Chromogranin A (CgA)

Chromogranin A is a protein, located in neuroendocrine cells, which is co-secreted with a wide variety of peptide hormones and neurotransmitters. Tumours with neuroendocrine properties typically secrete large quantities of CgA into the circulation. The assay is used primarily in the diagnosis and monitoring of patients with carcinoid tumours, islet cell tumours, pheochromocytoma, neuroblastoma, and other tumours of neuroendocrine origin. It may also be used for small-cell lung cancer and prostate cancer, in men with normal PSA levels.

Non-malignant elevations may be seen in kidney, liver or heart failure, It may also be elevated in patients experiencing significant stress because CgA is co-released with catecholamines.

CgA is measured using the Cisbio ELISA double antibody sandwich assay. Due to a lack of a recognized reference standard for CgA, correlation is poor between different methods.

Serum (preferred) and plasma are the recommended sample types.

Testing is performed weekly.

Reference Range: <94 µg/L

Caveats

Causes of Elevations of Serum Chromogranin A (CGA) Concentration Unrelated To Carcinoids or Other Neuroendocrine Tumors

Proton Pump Inhibitor (PPI) Drugs:

> Proton pump inhibitors (eg, omeprazole; PPI), which are used in the treatment of esophageal and gastroduodenal ulcer disease and dyspepsia, will result in significant elevations of serum CGA levels, often to many times above the normal range.. PPI should therefore be discontinued for at least 2 weeks before CGA measurements, because the biological effects of PPI persist for a significant time period after the drugs are discontinued.
>
> Atrophic gastritis and pernicious anemia also lead to false elevations in serum CGA levels, by the same mechanism as PPI; lack of feedback inhibition of gastrin production due to gastric achlorhydria.

Impaired Hepatic or Renal Function:

> CGA and its peptide fragments are cleared by a combination of hepatic metabolism and renal excretion. The effects of hepatic failure are relatively minor in the absence of hepatocellular carcinoma or fulminant liver failure. However, even modest renal impairment can elevate serum CGA, and end-stage renal failure is associated with elevations similar to those observed in patients on PPI

Non-neuroendocrine Tumors:

> Various non-neuroendocrine tumors might be associated with elevations, usually modest, in serum CGA concentrations. One example is testicular cancer.

Interferences: Human anti-mouse antibodies (HAMA), heterophilic antibodies

Reference:

https://www.ncbi.nlm.nih.gov/pmc/articles/PMC4754364/

Squamous Cell Carcinoma (SCC)

Squamous Cell Carcinoma Associated Antigen is a tumour marker purified from human squamous cell carcinoma tissue of the uterine cavity. It is primarily used as a marker for squamous cell carcinoma of the cervix and uterus. However, it is also seen in cancers of the anus, vagina, vulva, lung, esophagus, skin, head and neck. SCC is primarily used as a monitor of response to therapy.

SCC is a chemiluminescent magnetic microparticle immunoassay (CMIA) performed on the Abbott ARCHITECT platform. This assay has been standardized using an Abbott internal reference manufactured from stock SCC diluted with borate buffer with protein (bovine) stabilizer.

Serum (preferred) or EDTA- or heparin-anticoagulated plasma are the recommended sample types.

Reference Range: < 1.5 µg/L

Interferences: Human anti-mouse antibodies (HAMA), saliva, other body fluids. SCC samples are very easily contaminated (eg. sneezing) and must be handled with care.