**BIOMARKERS**

The term Biomarker was defined in 2001 at National Institute of Health. Biomarker is evaluated and measured as an indicator of physiological processes, as a response to pathogenic process or as a pharmacological response to a therapeutic intervention. The production of biomarker can be by diseased organ or by the body as a response to the disease. These Biomarkers play a pivotal role in various stages of patient management. Even before the diagnosis, biomarkers are used for screening and risk assessment. During the process of diagnosis, biomarker can be used for staging and grading of the diseases. They also help in selecting the mode of therapy. In the later stage, biomarker can be used to monitor the treatment, can help in guiding the physician to make any addition or deletion of drugs. They also help in monitoring the recurrence of the disease.

**There are different phases of evaluation of Biomarkers.**

Phase 1 – (pre-exploratory studies) the process involved in this phase are gene selection, gene expressions to distinguish abnormal (diseased tissues) and normal samples. The markers which are identified are prioritized based on their predictive value (diagnostic/prognostic/therapeutic). This would suggest their evolution into clinical use routinely. The specimens used during phase I should be ideally from a well-characterized cohorts, or from a trial with active follow ups.

Phase II – Is a phase characterized by establishing an assay methodology for the estimation of the biomarker. It could be RNA, DNA, proteins or a cell based techniques like ELISA, can use mass spectrometry etc. These assays are validated for reproducibility, sensitivity and specificity.

Phase III Clinically diagnosed cases are subjected for estimation of biomarker and are analyzed for sensitivity and specificity.

Phase IV This phase involves prospective cohort, on whom the evaluation of sensitivity and specificities of the test is being carried out. Unlike phase II a positive test result triggers a definitive diagnostic procedure at phase IV stage.

Phase V the overall benefits and risks of the newer diagnostic test are evaluated on the screened population in phase V

**Characteristics of an ideal Biomarker**

1. It should be safe and the measurement should be easy.
2. There should be a proven treatment methodology to modify the biomarker.
3. The follow up tests should be relatively of low cost
4. The biomarker used for assessment should be consistent across ethnic groups and across gender.

**Types of Biomarkers**

1. Diagnostic Biomarkers - These markers are used to confirm the presence of a disease or a medical condition
2. Monitoring Biomarker – These are used to assess the presence of the disease to know the extent of a disease, or to evaluate the response of an intervention.
3. Response Biomarker –These are used to assess the presence of the disease, to know the extent of a disease, or to evaluate the response of an intervention.
4. Predictive Biomarkers – These are used to identify the predictive nature or in other words, to identify the probability of development of a clinical event, after the exposure to an environmental agent
5. Prognostic Biomarker – These are used to measure the individuals risk to acquire a disease.
6. Safety Biomarker – These are used to predict toxic adverse events induced by medical intervention like drugs or exposure to environmental agents.

**Applications of Biomarkers**

1. They help in assigning predictability for certain diseases
2. They can be helpful in identifying precursors for advanced diseases such as blood disorders or cancers
3. They play an important role in the drug discovery and development process.

**USES OF BIOMARKERS**

Biomarkers can be used in assessment of the exposure (absorbed amount or internal dose) and effects of chemicals and the susceptibility of individuals. Biomarkers may be used to elucidate cause-effect and dose-effect relationships in health risk assessment. The measurement of Biomarkers provides the critical link between chemical exposure, internal dose and health impairment, and are of value in assessment of risk. There is a need to identify and validate those characteristic parameters for each organ system that are indicative of induced dysfunction, clinical toxicity or pathological change, also to establish the specificity and sensitivity of each biomarker and its method of measurement.

**Requirement of Biomarkers**

Some chronic diseases, which require the patient to take medicines for years, the diagnosis of such disease become important, especially when there are side effects associated with the treatment. In such case, biomarker play a very important role. Some diseases like Rheumatoid arthritis, Alzheimer’s diseases, usually begin with less symptoms, in such patients the biomarkers may help to know the probability of the patient developing the symptoms.

**Biomarkers in drug development**

Throughout the process of drug discovery and development, biomarkers are useful. The aim of drug development will be to produce an effective drug at lower cost. Eg- During the development of Gefitinib, epidermal growth factor receptor tyrosine kinase inhibitor (EGRF, TKI).

**Biomarkers of cancer:** One of the important use of Biomarkers is in the management of cancer. The questions that can be answered by the biomarkers can been described from Fig 1

Fig 1: QUESTIONS THAT CAN BE ANSWERED BY CANCER BIOMARKERS

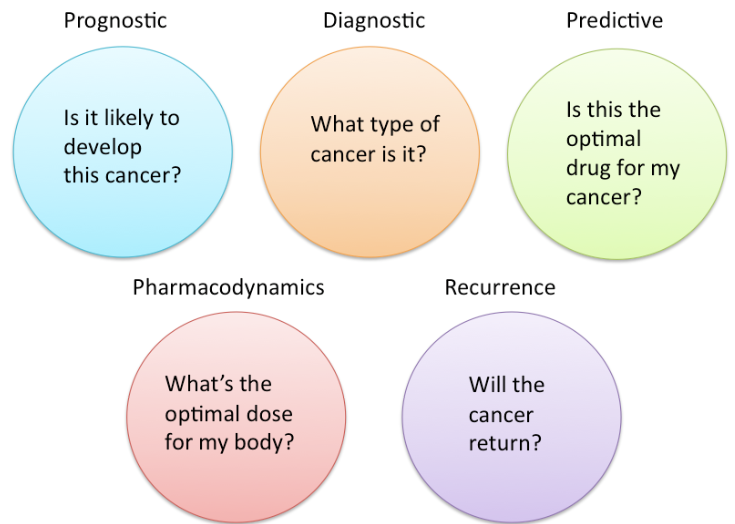


Table 1: BIOMARKERS USED IN VARIOUS TYPES OF CANCER

|  |  |
| --- | --- |
| Type of Cancer | Biomarker |
| Breast | [ER](https://en.wikipedia.org/wiki/Estrogen_receptor)/[PR](https://en.wikipedia.org/wiki/Progesterone_receptor) (estrogen receptor/progesteron receptor)(1,2) |
|  | Human epidermal growth factor receptor ([HER-2/neu](https://en.wikipedia.org/wiki/HER2))(1,2) |
| Colorectal | Epidermal Growth Factor Receptor (EGFR)(1,2) |
|  | [KRAS](https://en.wikipedia.org/wiki/KRAS)(1,3) |
|  | [UGT1A1](https://en.wikipedia.org/wiki/UGT1A1)(1,3) |
| Gastric | HER-2/neu(1) |
| Gastro Intestinal Stromal Tumors | [c-KIT](https://en.wikipedia.org/wiki/C-KIT)(1,4) |
| Leukemia/lymphoma | [CD20](https://en.wikipedia.org/wiki/CD20)(2,5) |
|  | [CD30](https://en.wikipedia.org/wiki/CD30)(1,6) |
|  | [FIP1L1](https://en.wikipedia.org/wiki/FIP1L1)-[PDGFRalpha](https://en.wikipedia.org/wiki/PDGFRA)(1,7) |
|  | [Platelet derived growth factor DGFR](https://en.wikipedia.org/wiki/PDGFR)(1,8) |
|  | [Philadelphia chromosome](https://en.wikipedia.org/wiki/Philadelphia_chromosome) ([BCR](https://en.wikipedia.org/wiki/BCR_(gene))/[ABL](https://en.wikipedia.org/wiki/ABL_(gene))) (1,9,10) |
|  | [PML](https://en.wikipedia.org/wiki/Promyelocytic_leukemia_protein)/[RAR-alpha](https://en.wikipedia.org/wiki/RAR-alpha)(1,11) |
|  | [TPMT](https://en.wikipedia.org/wiki/TPMT)(1,12) |
|  | UGT1A1(1,13) |
| Lung | Echinoderm microtubule associated protein-like 4[EML4](https://en.wikipedia.org/wiki/EML4)/[ALK](https://en.wikipedia.org/wiki/Anaplastic_lymphoma_kinase)(14,15) |
|  | EGFR (1,2) |
|  | KRAS (1,2) |
| Melanoma | [BRAF](https://en.wikipedia.org/wiki/BRAF_(gene))(1, 15) |
| Pancreas | Elevated levels of [leucine](https://en.wikipedia.org/wiki/Leucine), [isoleucine](https://en.wikipedia.org/wiki/Isoleucine) and [valine](https://en.wikipedia.org/wiki/Valine)(16) |
| Ovaries | [CA-125](https://en.wikipedia.org/wiki/CA-125)(17) |

**Biomarkers of cardiac diseases.**

|  |  |  |  |
| --- | --- | --- | --- |
| **Test** | [**Sensitivity and specificity**](https://en.wikipedia.org/wiki/Sensitivity_and_specificity) | **Approximate peak** | **Description** |
| [Troponin test](https://en.wikipedia.org/wiki/Troponin_test) | Troponin is the most sensitive and specific test for [myocardial](https://en.wikipedia.org/wiki/Myocardium) damage. The reason being it has increased specificity compared with CK-MB, Troponin C, Cardiac troponin I, and Cardiac troponin T are the three components of Troponin. Troponin I especially has a high affinity for myocardial injury. | 12 hours | Troponin is released from the cytosolic pool of the myocytes during Myocardial infarction. Troponin T and I, are specific to myocardium. Troponin elevation can also be seen in heart failure, acute infarction, severe pulmonary embolism causing acute right heart overload, myocarditis. Troponins can also calculate infarct size but the peak must be measured in the 3rd day. After myocyte injury, troponin is released in 2–4 hours and persists for up to 7 days.  Normal value are - Troponin I <0.3 ng/ml and Troponin T <0.2 ng/ml |
| [Creatine Kinase (CK-MB) test](https://en.wikipedia.org/wiki/CPK-MB_test) | It is relatively specific only when skeletal muscle damage is ruled out | 10–24 hours | CK-MB an isoform of [creatine kinase](https://en.wikipedia.org/wiki/Creatine_kinase" \o "Creatine kinase) is expressed in heart muscle. It is located in the cytoplasm. CK-MB facilitates movement of high energy phosphates into and out of mitochondria. Since it stays for a short duration, it cannot be used for late diagnosis of acute MI but can be used to suggest infarct extension if levels rise again. This is usually back to normal within 2–3 days. Normal range - 2-6 ng/ml |
| [Lactate dehydrogenase](https://en.wikipedia.org/wiki/Lactate_dehydrogenase) (LDH) | LDH is not as specific as troponin. | 72 hours | Lactate dehydrogenase is involved in the conversion of [pyruvate](https://en.wikipedia.org/wiki/Pyruvic_acid) to [lactate](https://en.wikipedia.org/wiki/Lactic_acid). The isozyme found in heart muscle is LDH-1. The isoenzyme predominantly found in blood serum is LDH-2. A high LDH-1 level to LDH-2 suggests MI. his is usually back to normal 10–14 days. |
| [Aspartate transaminase](https://en.wikipedia.org/wiki/Aspartate_transaminase) (AST) |  |  | This was used earlier. It is not specific for heart damage |
| [Myoglobin](https://en.wikipedia.org/wiki/Myoglobin) (Mb) | low specificity for [myocardial infarction](https://en.wikipedia.org/wiki/Myocardial_infarction) | 2 hours | Myoglobin is not commonly used. Myoglobin is the primary oxygen transporter to the muscles. The levels increase when muscle tissue is damaged but it lacks specificity. The only advantage of Myoglobin is it responds rapidly (18) rising and falling earlier than CK-MB or troponin. It also has been used in assessment of reperfusion after [thrombolysis](https://en.wikipedia.org/wiki/Thrombolysis)(19) |
| [Ischemia-modified albumin](https://en.wikipedia.org/wiki/Ischemia-modified_albumin) (IMA) | low specificity |  | IMA can be detected by albumin cobalt binding (ACB) test. Myocardial ischemia alters the N-terminus of albumin which leads to reduction in the ability of cobalt to bind to albumin. IMA measures ischemia in the blood vessels and thus the results in minutes unlike the traditional markers of necrosis that take hours. |
| [Pro-brain natriuretic peptide](https://en.wikipedia.org/wiki/Pro-brain_natriuretic_peptide) |  |  | Increased values of BNP are seen in patients with heart failure. It has been an approved marker for acute congestive heart failure. |
| [Glycogen phosphorylase isoenzyme BB](https://en.wikipedia.org/wiki/Glycogen_phosphorylase_isoenzyme_BB) |  | 7 hours | one of the three isoforms of [glycogen phosphorylase](https://en.wikipedia.org/wiki/Glycogen_phosphorylase) is the Glycogen phosphorylase isoenzyme BB (GPBB). This isoform exists in cardiac and brain tissue. Because of the blood–brain barrier, GP-BB can be used as a specific marker to heart muscle. GP-BB is one of the "new cardiac markers" that can lead to early diagnosis in acute coronary syndrome. During ischemia, GP-BB is converted into a soluble form and is released into the blood. A quick rise in blood levels can be seen in unstable angina and myocardial infarction. GP-BB is found to be elevated 1–3 hours after process of ischemia. |

Disadvantages of Biomarkers

1. Most of the biomarkers cannot be used as surrogate endpoints to assess the clinical outcomes.
2. There is lot of difficulty associated with validation of methods used to measure the biomarkers. They also require validation at different levels
3. It is difficult to measure the success of a therapeutic intervention, using biomarkers alone.

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