

## **BIOMARKERS**

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## **I. BIOMARKERS**

Biomarker is anything that can be measured as an indicator of a biological process. 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 biomarker can be produced by the body as a response to a disease. These biomarkers play a pivotal role in various stages of patient management. Even before the diagnosis, biomarkers are used for risk assessment and also for screening. During the process of diagnosis, biomarker can be used for staging and grading of the disease. 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.

### **1. 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 differentiate between abnormal and normal samples. The biomarkers 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 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 can 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 measuring the levels 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

### **2. Characteristics of an Ideal Biomarker**

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

### **3. Types of Biomarkers**

- Diagnostic Biomarkers : These markers are used to confirm the presence of a disease or a medical condition
- 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.

- **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.
- **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
- **Prognostic Biomarker:** These are used to measure the individuals risk to acquire a disease.
- **Safety Biomarker:** These are used to predict toxic adverse events induced by medical intervention like drugs or exposure to environmental agents.

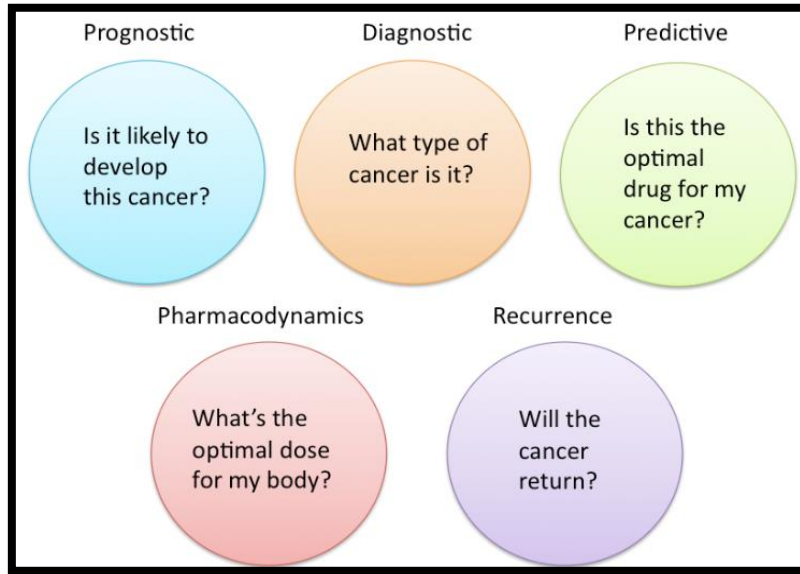
#### 4. Applications of Biomarkers

- They help in assigning predictability for certain diseases
- They can be helpful in identifying precursors for advanced diseases such as blood disorders or cancers
- They play a pivotal role in drug discovery and development process.

## II. 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.

1. **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. 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.
2. **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 (EGFR, TKI).
  - **Biomarkers of cancer:** One of the important use of Biomarkers is in the diagnosis and management of cancer. The questions that can be answered by the biomarkers can be described from Fig 1



**Figure 1: Questions That Can Be Answered By Cancer Biomarkers**

**Table 1: Biomarkers Used In Various Types Of Cancer**

| Type of Cancer                   | Biomarker   |
|----------------------------------|---|
| Breast                           | ER/PR (estrogen receptor/progesteron receptor) <sup>(1,2)</sup>             |
|                                  | Human epidermal growth factor receptor (HER-2/neu) <sup>(1,2)</sup>         |
| Colorectal                       | Epidermal Growth Factor Receptor (EGFR) <sup>(1,2)</sup>                    |
|                                  | KRAS <sup>(1,3)</sup>   |
|                                  | UGT1A1 <sup>(1,3)</sup>   |
| Gastric                          | HER-2/neu <sup>(1)</sup>  |
| Gastro Intestinal Stromal Tumors | c-KIT <sup>(1,4)</sup>  |
| Leukemia/lymphoma                | CD20 <sup>(2,5)</sup>   |
|                                  | CD30 <sup>(1,6)</sup>   |
|                                  | FIP1L1-PDGFRalpha <sup>(1,7)</sup>  |
|                                  | Platelet derived growth factorDGFR <sup>(1,8)</sup>                         |
|                                  | Philadelphia chromosome (BCR/ABL) <sup>(1,9,10)</sup>                       |
|                                  | PML/RAR-alpha <sup>(1,11)</sup>   |
|                                  | TPMT <sup>(1,12)</sup>  |
|                                  | UGT1A1 <sup>(1,13)</sup>  |
| Lung                             | Echinoderm microtubule associated protein-like 4EML4/ALK <sup>(14,15)</sup> |
|                                  | EGFR <sup>(1,2)</sup>   |
|                                  | KRAS <sup>(1,2)</sup>   |
| Melanoma                         | BRAF <sup>(1, 15)</sup>   |
| Pancreas                         | Elevated levels of leucine, isoleucine and valine <sup>(16)</sup>           |
| Ovaries                          | CA-125 <sup>(17)</sup>  |

### 3. Biomarkers of cardiac diseases.

| Biomarker                    | Sensitivity and specificity   | Approximate peak | Description  |
|------------------------------|---|------------------|--|
| Troponin I                   | Troponin I is one of the specific marker for diagnosis of myocardial infarction. It is a more specific marker than CK-MB <sup>(12)</sup> . The three components of Troponin are Troponin C, Troponin I and Troponin T | 18-24 hours      | Troponin I even though is a specific marker for myocardial infarction, it is a non enzymatic marker unlike Creatinine kinase (CK-MB). It starts rising 4-10 hours after myocardial infarction, reaches its peak level at 18-24 hours and comes back to normal level by 8-14 days. The normal value of Troponin I is 0 – 0.04 ng/mL.  |
| Creatine Kinase (CK-MB) test | It is relatively less specific marker when compared to Troponin I, it is an enzymatic marker unlike Troponin I.   | 18–24 hours      | Creatine kinase has several isoenzyme forms, CK-MB, CK-BB and CK-MM. among these CK-MB is cardiac specific. It is less specific than Troponin I, But CK-MB has a very good role in assessment of reperfusion. This enzyme starts to rise by 3-6 hours, reaches its peak by 18 – 24 hours and comes back to normal by 36 to 72 hours. The normal range of creatinine kinase is 15 – 100 U/L for males and 10 – 80 U/L in females. n |
| Lactate dehydrogenase (LDH)  | LDH is an enzymatic marker. LDH is less specific cardiac marker compared to Troponin I  | 72 hours         | Lactate dehydrogenase is the enzyme required for conversion of pyruvate to lactate. The isozyme found in heart muscle is LDH-1. Normally the levels of LDH-2 is more than LDH-1 which is called flipped pattern. An increased LDH-1/LDH-2 ratio indicates the possibility of Myocardial infarction.  |

|                                     |   |         |  |
|-------------------------------------|---|---------|--|
| Aspartate transaminase (AST)        |   |         | This was used earlier. It is a non-specific marker for detecting heart damage  |
| Myoglobin                           | This biomarker has low specificity for detection of myocardial infarction | 2 hours | Myoglobin is one of the not- commonly used marker. The function of Myoglobin is to transport oxygen to the muscles. The increase in the levels of moglobin is seen when muscle tissue is damaged, but it lacks specificity. One of the important advantage of Moglobin is its rapid response. <sup>18)</sup> The rise in CK-MB is seen before any other markers raise. It also has been used in assessment of reperfusion after thrombolysis <sup>(19)</sup> |
| Ischemia-modified albumin (IMA)     | Less specific marker  |         | This biomarker can be measured by albumin cobalt binding (ACB) test. When the myocardial cells undergo ischemia, there is alteration in the N-terminus of albumin. This reduces the affinity of cobalt to albumin. IMA measures ischemia in the blood vessels. Hence it takes very minimal time to obtain the report unlike the other cardiac markers which take hours to obtain the report.   |
| Pro-brain natriuretic peptide       |   |         | This marker has a role in detection as well as prediction of heart failure. Hence can be used for early detection of structural heart disease.   |
| Glycogen phosphorylase isoenzyme BB |   | 7 hours | Glycogen Phosphorlase-BB is one of the "new cardiac markers"used in early diagnosis of acute coronary syndrome. Ischemia leads to  |

|  |  |  |   |
|--|--|--|---|
|  |  |  | conversion of GP-BB into a soluble form, which 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 for upto 3 hours after process of ischemia. |
|--|--|--|---|

### Disadvantages of Biomarkers

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

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