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REVIEW ARTICLE

Biomarkers of cancer: A Comprehensive Review

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ABSTRACT
Cancer is one of the leading causes of death globally. Early detection with the help of biomarkers can be done and therapy can be planned. Biomarkers are substances that indicate the presence of cancerous cells. Different types of biomarkers can be antibodies, viral, mitochondrial, metabolic, and can be detected and used for diagnosis, prognosis and to evaluate the efficacy of the therapy. Identification of biomarkers has a major role in the diagnosis of disease and influences the treatment strategies in Cancer. Biomarkers can be used as targeted therapy and are one of the bases for precision medicine. As chemotherapy used in cancer treatment may affect normal cells in the body which may lead to various side effects. Using targeted therapy makes treatment less toxic and more tumor-specific. This article reviews various biomarkers of cancer and their application in diagnosis and as a therapeutic target.
Keywords: Cancer, Biomarkers, Targeted therapy

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INTRODUCTION

Cancer can be lookedas a disease process characterized by uncontrolled growth and spread of abnormal cells, has been the leading cause of death in many countries. It exerts a tremendous toll on society. The economic consequences of cancer are immense, in addition to the debilitating impact on patients and their families, both in terms of direct medical health facilities and in terms of the lack of human resources caused by early mortality. Cancer survival appears to be lower due most likely to a combination of a late diagnosis stage and insufficient access to prompt and effective care. Early and precise cancer detection is critical for clinical diagnosis, efficient control of toxicity and eventually successful cancer treatment.

A tumor marker is often referred to as a 'cancer biomarker' as a 'substance or action that can be objectively assessed and analyzed as an indication of normal biological processes, pathogenic processes or pharmacological responses to therapeutic interventions[1]. Cancer biomarkers are found in tumor tissues or serum and involve a wide range of molecules, including DNA, mRNA, enzymes, metabolites, transcription factors, and cell surface receptors[2].This characteristic/biomarker is either produced by the tumor or by the body in response to cancer. The cancer biomarker field's aim is to establish accurate, cost-effective, efficient cancer risk identification, early cancer detection and tumor classification strategies; so that the patient can receive the most appropriate therapy and monitor disease progression, regression and recurrence[3].

A characteristic could be measured by genetics, proteomics, cellular or molecular substances found in higher than normal amounts in the blood, urine or body tissues of a cancer patient. The biomarker is a quantitative and accurate variable or marker used to analyze the disease process or to determine whether or not a medication used in the treatment was successful[4].Based on their utility, biomarkers can be divided into the following categories (Figure 1) [5].

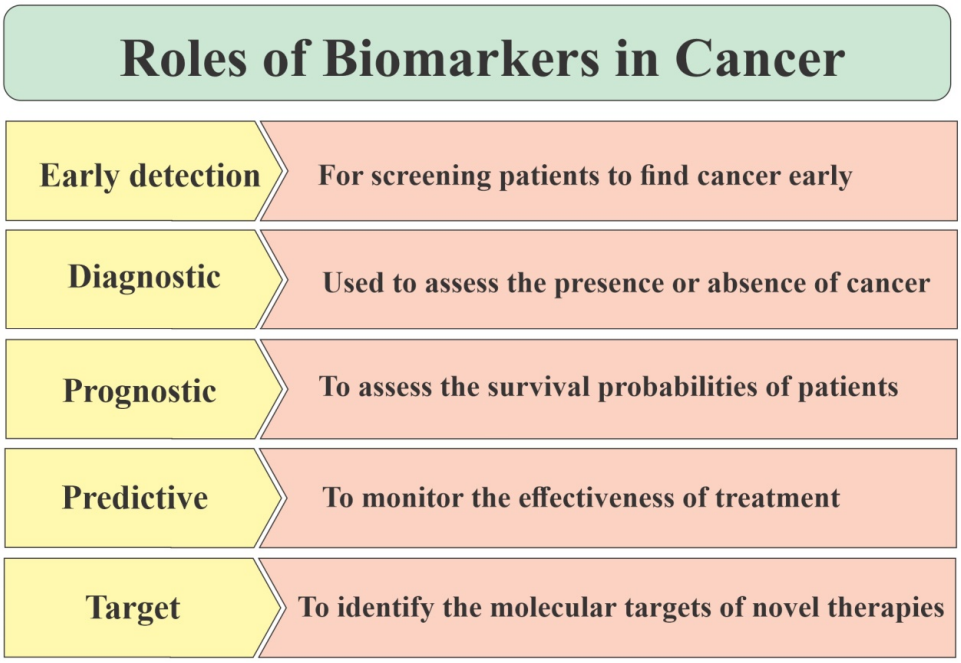

Figure 1 : Role of Biomarkers in Cancer

Cancer cells exhibit a wide spectrum of genetic alterations that include gene rearrangements, point mutations, and gene amplifications, leading to molecular pathway disruptions that regulate cell growth, survival, and metastasis. When such changes occur in most patients with a particular tumor type, they can be used as biomarkers for identification and development of targeted therapies, in addition to predicting responses to different therapies[6].

Genetics, genomics, proteomics, many noninvasive imaging techniques and other technologies allow measurement of several biomarkers. Currently, the protein targets and the pharmacologic consequences of drug administration results in advanced knowledge leading to a better understanding of the disease process that will facilitate development of disease specific drugs with minimal undesired systemic toxicity. Establishing biomarkers involves a thorough understanding of the molecular mechanisms and cellular processes underlying cancer initiation, with a specific emphasis on how minor changes can disrupt a number of cellular functions in only a few regulatory genes or proteins.

Diagnostic and prognostic biomarkers are quantifiable traits that assist clinical oncologists in their first interaction with the patients suspected. These particularly aid in

(i) Identifying who is at risk,
(ii) Diagnose at an early stage,
(iii) Select the best treatment modality, and
(iv) Monitor response to treatment.

Such biomarkers occur in several different forms; typical biomarkers include those that can be tested using radiological techniques such as mammograms, etc., and circulating tumor-specific (related) antigens levels. For example, prostate-specific antigen (PSA).The plethora of potentially useful biomarkers for cancer has grown significantly to include DNA, RNA and protein sequence and expression levels, as well as metabolites[2].Advances in imaging techniques open the possibility that specific molecular biomarkers (e.g., those describing responses to therapy) can be tracked non-invasively in cancer patients.

Genetic and genome based approaches played an important role in cancer detection and prognosis. Changes in DNA content (hyper- and hypo-diploidy) due to genomic instability during dysregulated proliferation have been commonly used and in reality have been very effective in clearly dissecting subtle differences between various tumor stages as well as resolving similar tumor types, which were otherwise not easily discernable, but has its limitations.

TYPES OF BIOMARKERS

CELLS AS BIOMARKER

Cells begin to appear in the bloodstream in advanced tumor stages where they can be easily accessible.Cells tend to appear in the bloodstream in advanced tumor stages where they can be easily

tracked. Advanced clinical research in some malignancy has successfully used tumor and immune cells where it has acted as a strong prognostic biomarker, whereas its usage in other cancers is currently under review.

GENETIC BIOMARKERS

Cancer is a genetic disease facilitated by gene alterations, such as oncogenes and tumor suppressors, which regulate cell proliferation, survival and other homeostatic functions.Several non-random mutations, and translocations/ rearrangement within the regulatory region of the gene are also believed to be associated with different forms of malignancy. Such translocations aim for special clinical diagnosis as highly precise tumor markers.

Deletion of genomic material is important because there may be some tumor suppressor activity in the missing segment of DNA. Gene deletions are discovered through polymerase chain reaction (PCR) to various chromosomes and sites using microsatellite probes. Loss of heterozygosity can lead to microsatellite instability (MSI) as well as mutations within several proto-oncogenes. Although identification of microsatellite instability/alterations in pathological tissue samples involves a comparison with normal tissue, it is a valuable tool for early detection, prognosis and evaluation of chemotherapeutic drugs response occasionally at preneoplastic stage[7].

EPIGENETIC BIOMARKERS

Epigenetic modifications can occur directly through DNA methylation of genes and other proteins around which DNA is wound to form chromatin[8].Cytosine residue DNA methylation is the main human epigenetic modification that occurs in the context of 5'-CpG-3 dinucleotide [9]. It has become evident in recent years that epigenetic events are potentially responsible as genetic alterations for the initiation and progression of cancer, with hypo- and hyper-methylation of DNA facilitating the production of cancer.

Genomic Hypermethylation markers may be used for the detection of both primary and metastatic or recurrent cancer cases. Therefore, it was proposed, that changes in methylation patterns of gene groups in sputum samples could be an essential, non-invasive method for identifying smokers at risk of developing lung cancer. While, epigenetic work has resulted in improved survival of patients with certain types of lymphoma and leukemia by using drugs that modify DNA methylation and histone[10]**.**

CYTOGENETIC AND CYTOKINETIC MARKERS

Classical cancer markers are structural and numerical aberrations in chromosomes, as the link between chromosomal aberrations and neoplastic transformation which has been well known. Though deviations from the number of diploid chromosomes leading to both hyper- and hypo-diploidy as well as aneuploidy were noted in malignant tumors[11]. Somatic mutations are promising biomarkers for cancer risk as these can capture genetic events that are associated with malignant transformation[12].

Among other genome-based biomarkers, in some epithelial tumors, neoplasm recognition from the level of lesion-specific transcriptomes in the blood has been successfully employed[13]. Recently an innovative transcriptomes marker was produced to curtail the false positives in prostrate and other endocrine cancers[14]. Identification of S-phase cells and analysis of a number of other antigenic determinants of proliferation studied using a variety of cell biology techniques have also been used as complementary markers. Proteins encoded by the genes of minichromosome maintenance (MCM) were also proposed as useful proliferation markers; with high rates of gene expression suggesting poorprognosis[15]**.**

VIRAL BIOMARKERS

Among viral induced cancers, hepatocellular carcinoma (HCC) is one of the most common cancers worldwide and a leading cause of death in developing countriesdue to the endemic hepatitis B virus (HBV) infection.HBV can also promote carcinogenesis, in addition to immuno-inflammatory reactions, through genetic instability produced by its common integration into host DNA[16]. Various types of biomarkers were used to explain the etiology and development of HCC. Such markers involve analysis of viral DNA or proteins or antibodies that are generated against the viral proteins.

HPV viral load, a measure of the quantity of viral DNA in biopsy specimens, alone or in combination with well-characterized serological HPV assays, has been suggested to delineate the role of HPV in oral and oropharyngeal cases [17], while antibodies produced in HPV E6 and E7 subjects serve as markers of invasive HPV-associated malignancy [18]. The first human virus to become directly active in carcinogenesis was Epstein-Barr virus (EBV). Within B cells, the EBV genome is preserved either as a circular multicopyepisome in the host cell, or by incorporating the viral DNA into the host genome. Identification and quantitative analysis of plasma EBV DNA in patients with nasopharyngeal carcinoma and Hodgkin's lymphomaserves as a valuable molecular marker for diagnosis, tracking and prediction of relapse[19].

CANCER ANTIGENS (BIOMOLECULES) BASED BIOMARKERS

The cancer proteome includes information about probably any biological process occurring in cancer cells, cancer tissue microenvironment, and cancer cell-host interaction. Cancer cells release many proteins and other macromolecules through secretion into the extra-cellular fluid which can also serve as biomarkers.Some of these products will end up in the bloodstream and thus act as potential serum biomarkers.

Oral fluid includes proteomic signatures that can act as biomarkers for human illnesses including oral cancer. The most recent identification of five proteins in cancer patients' saliva was found to be useful oral cancer markers with 90% sensitivity and 83% oral squamous cell carcinoma specificity. Nonetheless, the validity of these possible biomarkers includes long-term research that include a significant number of oral cancer patients as well as subjects at high risk for developing oral cancer.

MITOCHONDRIAL BIOMARKERS

Mitochondria usually contains multiple haploid copies of their own genome (16.5 kb), including most transcription, translation, and protein components. At 1000-10,000 copies/cell, mtDNA is present and the vast majority of these copies are identical at birth (homoplasmic). Several mutations in mtDNA have recently been found, especially in the D-loop region, in breast, colon, oesophageal, endometrial, head and neck, liver , kidney, leukemia, lung, melanoma, dental, prostate, and thyroid cancer. Because of their clonal existence and large numbers of copies in cancer cells, mitochondrial mutations can provide an important molecular marker for the non-invasive cancer detection.

It may also be effective in the early detection, diagnosis, and prognosis of cancer outcome and/or in monitoring response to certain methodologies of prevention and intervention, as well as therapies[20].

METABOLIC BIOMARKER (GLUCOSE METABOLISM)

A cell bioenergetic index (BEC index) has been indicated that could be used for cancer classification and prognosis, in addition to predicting the therapy response[21].Positron emission tomography (PET), which enables non-invasive and quantitative study of different biological processes, uses a glucose analog (2-deoxy-D-glucose) labeled with Fluorine 18 positron emitter; FDG, which is partially metabolized and trapped as its phosphate (2-DG-6-P) in the tumor tissue, thus locating the tumor[22]. The use of glucose therefore tends to be an important metabolic marker for the diagnosis, prognosis and prediction of tumor response to a number of therapies[23].Various biomarkers in cancer are enlisted in (Figure 2) and various opportunities of identification(Figure 3)

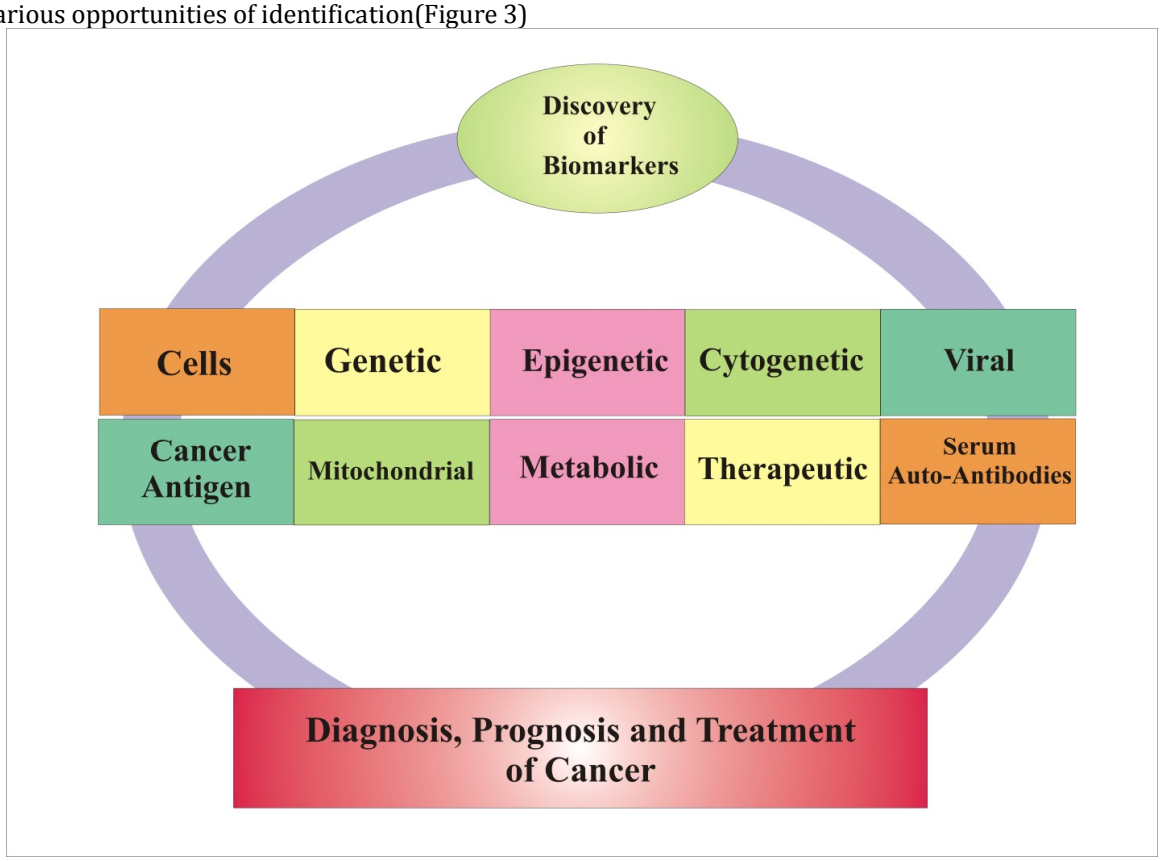

Figure 2 : Biomarkers in Cancer

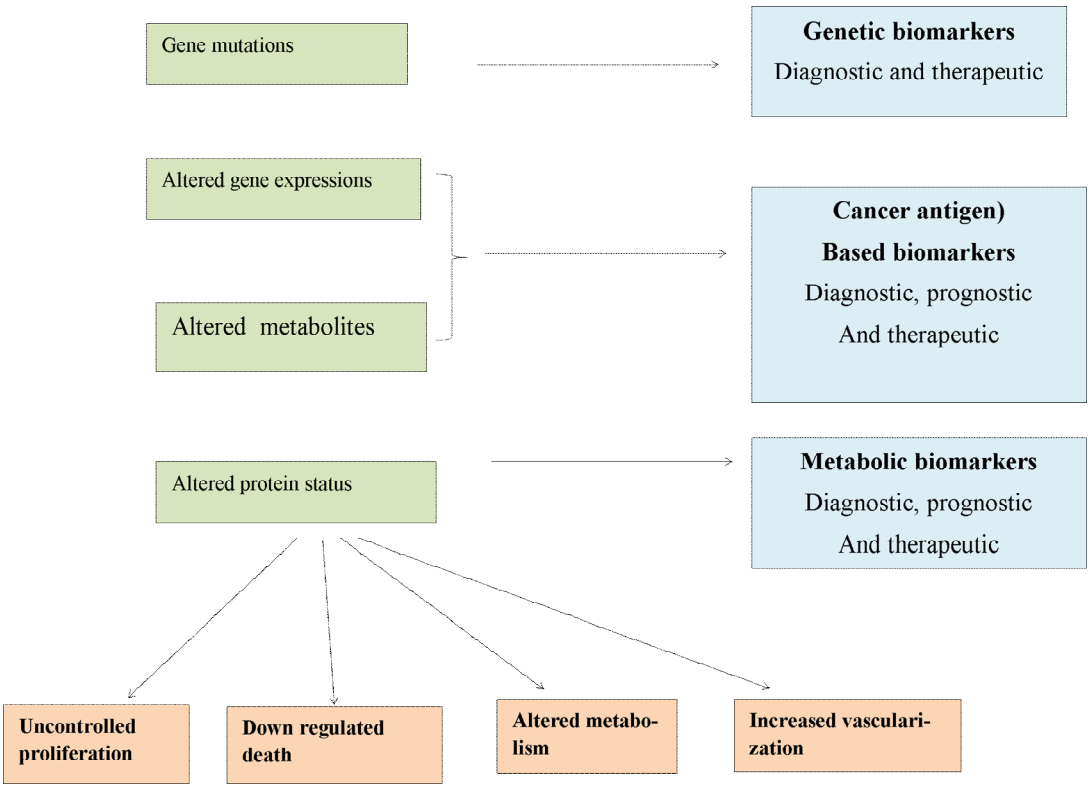

Figure 3. The process of carcinogenesis, showing opportunities of identifying biomarkers.

THERAPEUTIC BIOMARKERS

In principle, "Targeted therapies" show more selectivity for tumor cells in theory, and indeed many such therapies have already shown promise in the clinic. These include small molecule drugs that inhibit the activity of protein tyrosine kinases and neutralizing antibodies that inhibit trans-membrane signaling receptors. Other therapeutic approaches include drugs that block molecular activity in the host microenvironment supporting the growth of tumours. To date, many of these therapies have only conferred modest benefits on patient survival, but refining of the way these drugs are used (e.g., as combination therapies and with biomarker-guided patient selection) is expected to improve therapy efficacy.

MAMMALIAN TARGET OF RAPAMYCIN (mTOR)

This is an evolutionarily preserved serine-threonine protein kinase which belongs to the family of kinase-related PIKK [phosphoinositide 3-kinase (PI3 K)] and plays an important role in regulating cell growth and proliferation. Upon activation, mTOR increases the phosphorylation rates of its downstream targets including p70S6 K and 4EBP1, leading to increased translation speeds, ribosome biogenesis, and actin cytoskeleton reorganization and autophagy inhibition[24].Activation of this pathway leads to cell growth and proliferation dysregulation and can be verified by biomarkers such as PTEN mRNA loss or protein production in tumor tissue. Biochemical rapamycin inhibition of mTORs can be assessed by biomarkers such as the ample amount of the phosphorylated form of the ribosomal protein S6, and its therapeutic effects on tumor cells can be assessed by the proliferation marker Ki-67[25].

TELOMERASE

Telomerase belongs to a class of enzymes known as reverse transcriptases which use RNA as a template to create DNA and contains both components of RNA and proteins. The enzyme ensures telomer maintenance and so protects the cell against degradation and death[26]. Because telomerase is present in approximately 90% of human cancers and is responsible for the irreversible growth of cancer cells[27], it has been a target for anticancer therapy that cuts off telomerase and thus prevents the growth of tumours. Most human tumors not only express telomerase but also have very short telomeres which are interesting. Telomerase is one of the best markers for human cancer, linked only to malignant tumors and not to the benign lesions that make it an ideal diagnostic marker for chemotherapy[28].Within normal cells, telomerase is sequestered far from the chromosomes in a region of the nucleus called the nucleolus. The enzyme is released during cell division only when required, and then returns quickly to the nucleus afterwards. However, in cancer cells, telomerase is present in the cell, suggesting a deficiency of the

telomerase-shuttling mechanism. Identifying and controlling the proteins usually involved in the movement of telomerase may be useful targets for anti-telomerase therapies[29].

p53

The p53 gene is one of the tumour suppressor genes that normally prevent uncontrolled multiplication of abnormal cells and experimental findings from the last two decades[30]. Upon stimulation, p53 activates molecular processes that delays the cell cycle progression of proliferating cells and simultaneously stimulating DNA repair processes[31]. On the other hand, higher level of damage has been found to activate p53 mediated cell death (typically apoptosis), a mechanism that is purported to be responsible for the prevention of carcinogenesis.

Although p53 is not a typical cancer-specific antigen, its central role in the control of cell growth and apoptosis and frequent mutations in tumours make p53 a unique target for cancer therapy. Radiation and most of the other anticancer drugs cause serious harm to cancer cell DNA and trigger the p53 action leading to apoptosis. Investigations in several types of cancer have shown that the p53 gene is a potentially useful biomarker for predicting prognosis and patient's response to therapy to various type of cancer [32]**.**

TYROSINE KINASE

Tyrosine kinases are a class of enzymes that control multiple cellular processes by primarily acting as essential transducers of extracellular signals that influence various functions such as cell growth, differentiation, migration, and apoptosis that contribute to the production and progression of tumours. Many human tumors show aberrant activation of tyrosine kinases caused by genetic changes that may be associated with malignant transformation[33].In a variety of tumors, the erbB or HER family of transmembrane tyrosine kinase receptors, in particular receptors erbB1 (or EGFR) and erbB2 (or Her2 / neu), were identified as an important therapeutic target.

It has been quite promising to target protein tyrosine kinases as a therapeutic technique, and the findings of recent clinical trials are also very encouraging. Current approaches include blocking kinase-substrate interaction, inhibiting the binding site of the enzyme adenosine triphosphate (ATP), and blocking receptors of the extracellular tyrosine kinase on tumor cells. Several tyrosine kinase inhibitors (TKIs) have already been approved as anti-cancer agents (i.e., gefitinib and trastuzumab).

HISTONE DEACETYLASES (HDACs)

Protein acetylation orchestrates the dynamic interplay between different processes, such as DNA repair, cell cycle arrest and apoptosis, which determines the cellular response to radiation and various chemotherapeutic drugs. This acetylation is catalyzed by histone acetylases (HATs) Chromatin remodeling during the regulation of gene expression is orchestrated by a concerted action of HATs and HDACs that condense and decondense the structure of chromatin by acetylating and deacetylating histones and other proteins of the nuclear receptors. Furthermore, HDACs appear to be closely linked to oncogenesis by regulating the expression of certain tumor suppressor genes which result in excessive proliferation and tumourogenesis[34].

HDAC have recently been among some of the attractive targets for cancer therapeutics, and HDAC inhibitors with diversified structures have indeed shown promising anti-tumour activity both in vitro and in vivo[35].Many of the HDAC inhibitors in a number of haematological malignancies and solid tumors are currently under clinical investigation[36]. It appears therefore, that HDAC inhibitors with pleiotropic actions in modulating multiple genes, signaling pathways and biological features of malignancy are useful in the treatment of cancers[37].

PIN1

In human breast cancer cell lines and tissues, overexpression of Pin1 has been reported and its expression closely correlates with cycline D1 (important cycline needed for cell proliferation) in tumors[38].Pin1 overexpression enhances transformed new/ Ras-transformed phenotypes of mammary epithelial cells and is involved in mitotic regulation[39]. In comparison, Pin1 inhibition suppresses the transformed phenotypes caused by Neu- and Ras, or induces tumor cells into mitotic arrest and apoptosis[40]. Pin 1 inhibition by different means, such as mutations, deletions or antisense expression, induces mitotic arrest and apoptosis in the tumor cell lines[41]. It seems that Pin1 can be used as a diagnostic marker for cancer detection or to stage the disease, although it also seems to be a desirable target for diagnosis and therapy in only certain types of cancers[42].

Serum Auto-Antibodies as Biomarkers

The main medical focus has been on early cancer detection that allows curative care to be administered before cancer progresses to late (and most often incurable) stages. Accordingly, serum biomarkers are highly sought after before cancer begins[43]. Autoantibodies that target specific tumor-associated antigens (TAAs) are one potential group of serum biomarkers. Since the first serological identifications of

tumor antigens from the sera of patients with melanoma[44], the number of reports of TAAs and autoantibodies in cancer patients has increased[45]. The immune response to TAAs functions in eliminating precancerous lesions during early carcinogenesis events[46]. The development of autoantibodies as a result of cancer immunosurveillance has therefore been found to precede clinical symptoms of tumorigenesis manifestations by several months to years[47]. Thus these serological biomarkers will act as early reporters in tumorigenesis for aberrant cellular processes[48].

PRODUCTION OF AUTOANTIBODIES

Robert W. Baldwin was the first person to establish an immune response to solid tumors[49]. Immunosurveillance to cancer cells is caused to cause destruction of antigen-specific tumors[49, 50]. The autologous proteins of tumor cells, usually referred to as TAAs, are believed to be altered in such a way as to immunogenize these proteins[51]. These selfproteins could be overexpressed, mutated, misfolded, or aberrantly degraded such that autoreactive immune responses in cancer patients are induced.

Immune system can perceive TAAs that have undergone post-translation modifications (PTMs) as foreign substance.The presence of PTMs (e.g. glycosylation, phosphorylation, oxidation and proteolytic cleavage) could induce an immune response by generating a neo-epitope or by enhancing self-epitope presentation and affinity to the major histocompatibility complex or the T-cell receptor. The immune response against these immunogenic TAA epitopes triggers autoantibodies to be developed as serological biomarkers for cancers.

Although some of the immune responses in cancer patients identify neo-antigens present only in tumors, most tumor-associated autoantibodies are directed toward aberrantly expressed self-antigens (e.g. HER2, p53, and ras). The immunogenicity of p53 was believed to be caused by its overexpression, missense point mutation and aggregation of cancer cells in the cytosol and nucleus[52]. They appears to increase the antigenic load and prime antibody production when proteins are overexpressed in cancer patients.

This chemotactic activity of tissue-specific TAAs may alert the immune system to danger signals from damaged tissues and promotes tissue repair. TAAs that interact with juvenile dendritic cells are immunogenic because they are liable to be sequestered and then addressed to the cellular immune system in an aberrant way.

The developed sera autoantibodies that target these TAAs could serve as early molecular signatures for cancer patients' diagnoses and prognoses. Additionally, most autoantibodies found in cancer patients' sera target cellular proteins with modifications, aberrant localization or expression associated with carcinogenic processes such as progression of the cell cycle, signal transduction, proliferation, and apoptosis[53]. The detection and functional characterisation of these immunological 'reporters' or 'sentinels' for tumorigenesis-related cellular pathways will help identify early molecular cancer events.

The immune response to TAAs occurs during tumorigenesis at an early stage, as shown by the discovery of high autoantibodies titers in patients with early stage cancer[54]. It has also been shown that the immune response to TAAs associated with malignant transformation progression[55]. Thus autoantibodies production can be identified before any other biomarkers or phenotypic aberrations are found, making these autoantibodies invaluable as biomarkers for early detection of cancer[56].

Additionally, autoantibodies have different characteristics which allow them to be valuable biomarkers of early cancer[57]. First, autoantibodies can be found in the asymptomatic cancer stage and can be identified in some cases as early as 5 years before the onset of the disease[58]. Second, autoantibodies against TAAs are present in cancer patients' sera, where they are readily available for screening. Third, autoantibodies are largely stable and remain in the serum for a fairly long time, since they are usually not subject to the proteolysis forms found in other polypeptides. The autoantibodies' durability and stability give them an advantage over other biomarkers, including the TAAs themselves, which are transiently secreted and can be easily degraded or cleansed. In addition, the autoantibodies are present at significantly higher concentrations than their respective TAAs; in response to a single autoantigen many autoantibodies are amplified by the immune system.Thus, autoantibodies may be more readily detectable than their respective TAAs. Lastly, sample collection is simplified as a result of the long half-life (7 days) of the autoantibodies, which minimizes hourly fluctuations

Nonetheless, autoantibodies do have their limitations. A single autoantibody test lacks the sensitivity and specificity required to screen and diagnose the cancer. Typically, autoantibodies against a particular TAA are found in only 10–30% of patients[57]. The reason for this decreased sensitivity lies in the heterogeneous nature of cancer, whereby different proteins in patients with the same type of cancer are aberrantly processed or regulated. Therefore no protein is known to be widely disrupted or immunogenic in a particular type of cancer. Many TAAs may also be unspecific, as they exist in cancer as well as in other diseases, particularly those with an autoimmune background such as systemic lupus erythematosus,

Sjogren's (sicca) syndrome, rheumatoid arthritis, type 1 DM and autoimmune thyroid disorder[59]. In addition, autoantibodies can be found in normal individuals in certain circumstances.

A panel of TAAs can solve this problem by allowing simultaneous detection of multiple autoantibodies. For example, patients with 10 different types of cancer and autoimmune diseases have been shown to differ from normal subjects in a panel of two TAAs (Koc and p62)[60]. Using a panel of seven TAAs (IMP1,p53, cyclin B, p62, c-myc, Koc and survivin), Koziol et al. is able to classify normal individuals and differentiate between patients with breast, bowel, stomach, liver , lung or prostate cancers, with sensitivities ranging from 77% to 92%, and between 85% and 91%. Zhang et al. analyzed 527 sera of six different types of cancer [breast, lung, prostate, stomach, colorectal, and hepatocellular carcinoma (HCC)] and showed that the successive addition of antigen to the same panel of seven TAAs improved immunoreactivity in cancer patients to 44–68 percent, but did not increase immunoreactivity in safe individuals[61, 62].

METHODS FOR IDENTIFYING AUTOANTIBODIES

Initial TAA research focused on a couple of antigens at a time, using techniques such as 1D SDS/PAGE or ELISA. Improvements in technologies such as proteomics platforms have allowed a panel of TAAs to be generated that exhibits better diagnostic value than a single TAA marker. In this area of research, five main techniques may be used, including serological screening of cDNA expression libraries, phage display libraries, protein microarrays, 2D western blots and 2D immunoaffinity chromatography (Figure 4) [61].

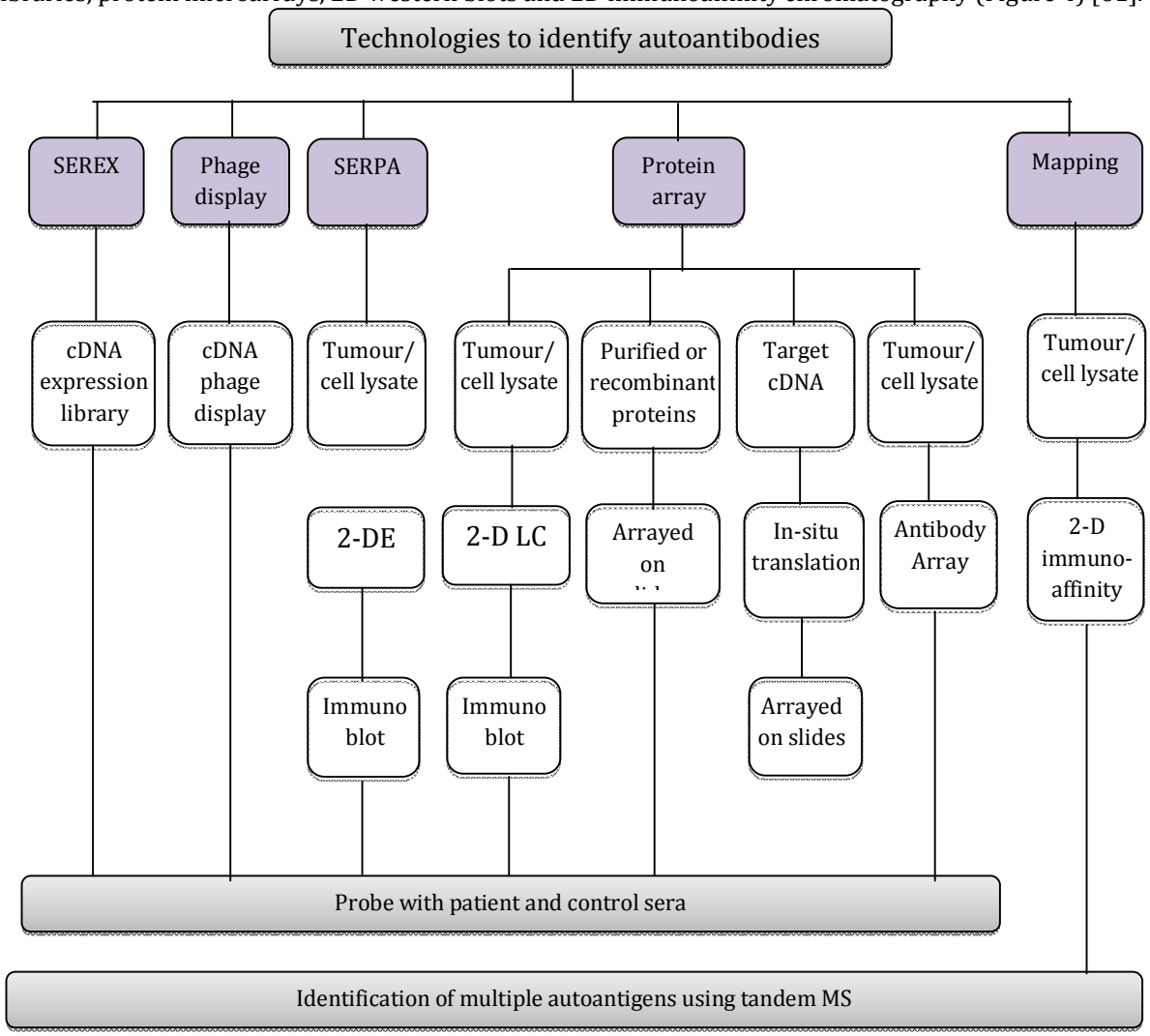

Figure 4 . Overview of 5 different approaches that enable identification of multiple autoantibodies 2-DE, 2-Dimentional Electrophoresis; 2-DLC, 2-Dimentional Liquid Chromatography; cDNA, complementary Deoxyribonucleic acid; SEREX. Serological analysis of recombinant tumor cDNA expression libraries.

CANCER-ASSOCIATED AUTOANTIBODIES

In recent years, the hunt for specific autoantibodies has stepped up, as demonstrated by a search on PubMed for 'autoantibodies and cancer.' Many cancers such as HCC and in the lung, colorectal, breast, stomach, prostate and pancreatic cancershave been found in autoantibodies and TAAs. Oncoproteins are among the growing list of TAAs found in cancers (e.g. HER-2/Neu, ras and c-MYC), tumor suppressor proteins (e.g. p53), survival proteins (e.g. survivin), cell cycle regulatory proteins (e.g. cyclin B1), mitosis-associated proteins (e.g. centromere protein F), mRNA-binding proteins (e.g. p62, IMP1, &Koc), differentiation and CTAs (e.g. tyrosinase and NY-ESO-1)[62].

3. COMBINATION OF BIOMARKERS IN CANCER DETECTION, DIAGNOSIS AND PROGNOSIS

While several potential biomarkers are known using high-throughput technologies, it remains to be determined their clinical application. SELDI – TOF was used by many investigators to test a serum protein pattern as a biomarker for ovarian, breast, and prostate cancer and lung cancer[63]. So a panel of biomarkers may provide more valuable knowledge and boost individual biomarkers statistical performance. In a five-center case-control study, Chan and colleaguesanalyzed the serum proteome of 153 patients with invasive epithelial ovarian cancer as an example of the usefulness of combining three serum biomarkers with CA125 for increased sensitivity and specificity (the best known biomarker for ovarian cancer and approved for ovarian cancer monitoring), 42 Ovarian tumor patients, 166 pelvic tumor patients and 142 healthy women[64]. The authors identified three biomarkers for early detection: apolipoprotein, a truncated form of transthyretin, and an interα-trypsin-inhibitor cleavage fragment.When these biomarkers were combined with CA125, the sensitivity was 74% for early stage ovarian cancer, which is significantly higher than CA125 alone (65%) at a matched specificity of 97%.

Prostate cancer, for example, is caused by the particular stimulation of cell growth by the androgen hormone and its ablation is one of the therapeutic strategies. The more aggressive type of prostate cancer, however, is usually due to androgen-independent pathways[65]. PSA is regulated by androgen, and its levels in prostate cancer are high. Nonetheless, detecting the androgen-independent prostate cancer, or when androgen levels are reduced by chemotherapy or castration, may not be a successful biomarker, making it an unattractive marker for prostate cancer to advance.Using prostate biomarkers that monitor these different pathways would not only improve the current low sensitivity of PSA but also improve the measures used to follow androgen-independent, aggressive cancer of the prostate. In summary, a panel of biomarkers identified by the use of multiple, high-performance platforms enables multivariate and simultaneous analyzes, thus allowing better and more efficient methodology[5].

PROTEOMICS IN THE BIOMARKER DISCOVERY

Recently there is development of new technologies that have set the pace for biomarker discovery and provide methodologies which have been used to identify novel biomarkers for the early detection of cancer and premalignancy and some of these biomarkers are currently validated on tumor sets assays. Cancer proteomics includes the detection and quantitative study of differentially expressed protein compared to healthy tissue equivalents at various stages of disease, from preneoplastic to neoplastic. Proteomics supplements genomic methods in cancer research. Research indicate up to six distinct forms of protein per gene in humans [6], and understanding their functional role at non-diseased and multiple stages of disease development will provide insights into the nature of prevention , treatment and therapeutic strategies. Protein expression and function are regulated by transcription and by post-transcription and translational events. Identifying and recognizing these shifts are the principles that underlie the proteomics of cancer.

FACTORS AFFECTING PERFORMANCE OF A BIOMARKER

A response to biomarkers refers to the percentage of cases (individuals with documented disease) that screen for the biomarker as positive.Specificity refers to the proportion of control subjects who test negative for the biomarker (individuals without disease). Ideal biomarkers would have a specificity and sensitivity rating of 100 per cent, i.e. everyone with cancer should have a positive biomarker test, and everyone without cancer would have a negative test. The lower the sensitivity, the more people with cancer won't be detected; the lower the precision, the more people without cancer will test positive. It is the lack of sensitivity and/or specificity which causes the discarding of many potential biomarkers. Additionally, specifications for a test's output features differ with the intended use.

Biochemical tests used in cancer screening and diagnosis differ not only in precision, reliability and validity but also in performance, e.g. positive predictive value (PPV), sensitivity, accuracy and negative predictive value (NPV). PPV talks of the number of people that have a positive result who do have the disease, while NPV is the percentage of people that do not have the disease with a negative test. PPV offers details on the likelihood that the disease is present if the test is positive.Consequently, it is clear that a statistical and inferential framework for assessing candidate markers in confirmatory clinical trials

is needed in cancer screening. Replacing an established technology is not a prudent decision, unless a biomarker offers sufficient knowledge about disease screening.
Other than these, there are various other factors) that can impact a biomarker or biomarker-based assay performance are as follows:

- Experimental designing
- Quality and source of cancer specimen
- Progressive biological heterogeneity
- Pre-analytical factors such as age, sex, dietary status, smoke exposure, use of tobacco, geographical and environmental factors.
- Analytical factors like errors during sample collection and processing, dilution errors, purity of reference standards, cross contamination of selected biomarker, fluctuations in temperature and instrument performance, contamination in purity of chemicals, and calculation errors.
- Social and economic issues[66].

Biomarkers can also aid in redefining the diseases and their treatments by changing the focus of current approaches to a more rational objective molecular basis, based on symptoms and morphology.

ISSUES AFFECTING MOLECULAR DETECTION, SCREENING AND TREATMENT

Biomarker development for the screening, detection, and treatment of cancer involves both biological and economic challenges. Most of the diagnosis approaches used to date classify fully formed cancer, not pre-malignant or early (intermediate stage) lesions that can be respected and cured. Although a screening test may detect cancer at the preclinical level, micro metastasis may not be identified and therefore the advantage of early detection and treatment may be limited[67]. Another problem is that in many organs, for example, prostate or colon, preneoplastic lesions are much more common than aggressive cancers and only 10% or less develop into a malignant tumor[68]. However, there is evidence that a subset of prostatic intraepithelial neoplasia develops into particularly aggressive phenotypes. This raises the issue whether any screening method should focus solely on early lesions, or whether it should also analyze the tumors behavior. To our minds, the discovery of serum, genetic or other tumor biomarkers should enable detection of the subset of cancer that is likely to lead to clinically important cancer, such as in prostate cancer[5, 43].

DISCUSSION AND CONCLUSION

Cancer biomarkers are important indicators of tumor growth. They are not only used to diagnose and control illness but also to provide a prognostic treatment strategy. Carcinogenesis and advancement of the tumour are complex and progressive processes associated with a variety of genetic and epigenetic alterations, some of which can also be observed in plasma and serum. Although there are cancer protein blood biomarkers some of them have been approved by the American Society of Clinical Oncology, but their number and clinical use are limited.
In these review we tried to cover various types of biomarkers which involves Cell, Blood (Plasma, Serum); Genetic, Epigenetic, Cynogenetic, Viral, Mitochondrial, Metabolic, Cancer Antigen and Sarum Antibody Biomarkers and also summarized various methods or techniques for their detection. It is possible to detect metabolomics in biological samples, which might provide an appropriate level to study cancer phenotype, also researcher said Tumor-specific circulating miRNAs and ctDNA analysis may provide a viable option for the early cancer diagnosis and prognosis, which further act as promising markers for various tumor entities which have already been identified as non-invasive biomarkers.
Biomarkers of DNA differ in several ways from the general biomarkers. However, improvements in technology for detection and diagnosis are rapidly overcoming some of the issues of analytical sensitivity, and it is likely that mutation and methylation analysis of these markers will improve specificity for the diagnosis of cancer. Though isolation, quantification and normalization strategies have to be standardized before any of these novel biomarkers are made applicable for clinical routine.
Clinical application and detection type plays a major role in reshaping life science industry and thereby influencing the treatment of many diseases particularly cancer. The above mentioned panel of biomarkers helps in diagnosis and facilitating therapy for each of those cancer type. Also, the upcoming genomic and proteomic technologies are quite promising in identifying new biomarkers, which can significantly enhance the efficacy of cancer management which helps to identify and understand the signaling pathways of specific targets for developing newer drugs and therapeutic strategies.

CONFLICT OF INTEREST

The authors declare no conflict of interest, financial or otherwise.

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