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BIOSENSORS: AN EMERGING ANALYTICAL TOOL FOR THE EARLY DIAGNOSIS OF CANCER BIOMARKERS TO REDUCE THE MORTALITY RATE

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ABSTRACT

Cancer is the serious health problem characterized by the uncontrolled multiplication of cells. It is the second leading cause of mortality throughout the world. Effective and accurate early diagnosis is current need of hour to reduce the severity and mortality rate of cancer. In the past few years, biosensors have gained significant attraction in the detection of the emerging cancer biomarkers. These are the analytical devices that has vast potential to detect the biomolecules like protein, DNA, RNA and converting them into detectable electrical signal. Biosensor technology can be designed to provide reliable and accurate detection of cancer biomarkers, imaging study of cancer cells, and to detect the effectiveness of chemotherapeutic agents at various target sites. However, a major challenge in the development of biosensors is that cancer is highly complex set of diseases. In this review, we highlighted the challenges in the early detection of cancer, emerging use of biosensors as a diagnostic tool, and some future applications of biosensor technology.

KEYWORDS: Biosensors, biomarkers.

INTRODUCTION

In recent times, according to the report of the WHO Cancer is a leading cause of death globally, responsible for 71% of all deaths in 2016 and estimated near 18.1 million new cases and 9.6 million deaths in 2018. In 2018, the most frequently diagnosed cancer are over 200 different foams including was of the lung (11.6% of all cases), followed by female breast (11.6%) and colorectal cancer (10.2%). Lung cancer is also the leading cause of death from cancer (18.4% of all deaths), followed by colorectal (9.2%) and stomach cancers (8.2%). The 10 most common cancers are responsible for 60-70% of cancer incidence and mortality. Although the most common kind of cancer found in human is cervical cancer and breast cancer. The number of new cases and deaths continues to rise because of increasing life expectancy and epidemiology and demographic transitions.

Cancer is defined as uncontrolled and abnormal multiplication of cells due to environmental and genetic causes. Tumor formation occur due to uncontrolled division of cells. As the cancer progression occur in body, the tumor begins to spread in nearby organs and systems which lead to increase its severity. By considering this, early diagnosis of cancer is significantly important to reduce the severity and for the effective treatment.

The identification of clinical biomarkers plays important role in the diagnosis of cancer at early stage, and to detect the underlying pathophysiological process involved in the progression of disease. Biomarkers, chemical substances detected in blood, urine or body tissues of patients during the elevation of malignant tumors in body. Chemically, biomarkers can be enzymes or hormones, gene products, specific cells, which indicate the progression of the disease by detecting the changes in the expression of a protein. An ideal biomarker should have the following characteristics like high sensitivity and specificity, rapid quantitative estimation and rapid release in the blood.

A Biosensors can define as an analytical device that deals with the detection of biological parameters. In the past few years, biosensors have gained enormous attention in medicine and nanotechnology field due to strong potential to convert a physical quantity into readable signal. Emerging biosensor technology could be very effective for those cancers that responds ineffectively to treatments. The challenges of effective treatment and late diagnosis for such cancers can be

overcome by using the sensitive biosensor which enables to detect small quantity of samples with high sensitivity and selectivity. Biomarkers can be of various molecular origins, including DNA (i.e., specific mutation, translocation, amplification, and loss of heterozygosity), RNA, or protein (ie, hormone, antibody, oncogene, or tumor suppressor. Development of biomarkers play vital role in the early diagnosis of cancer.

Cancer Biomarkers

Cancer biomarkers are important indicators of tumor growth. They are used not only to diagnose and monitor disease, but also to provide a prognostic approach to treatment. The cancer biomarker is generally having high clinical sensitivity and specificity and they release quickly in blood to potentiate the early diagnosis, and also capable to remain for longer time in blood. There are 160 types of biomarkers are identified that are being used for detection of cancer. A partial list of tumor biomarkers is presented in Figure 1. Most of these biomarkers, sufficient sensitivity and specificity is yet to demonstrate for their translation into routine clinical use or for treatment monitoring. This is an area that biosensor technology can potentially improve upon.

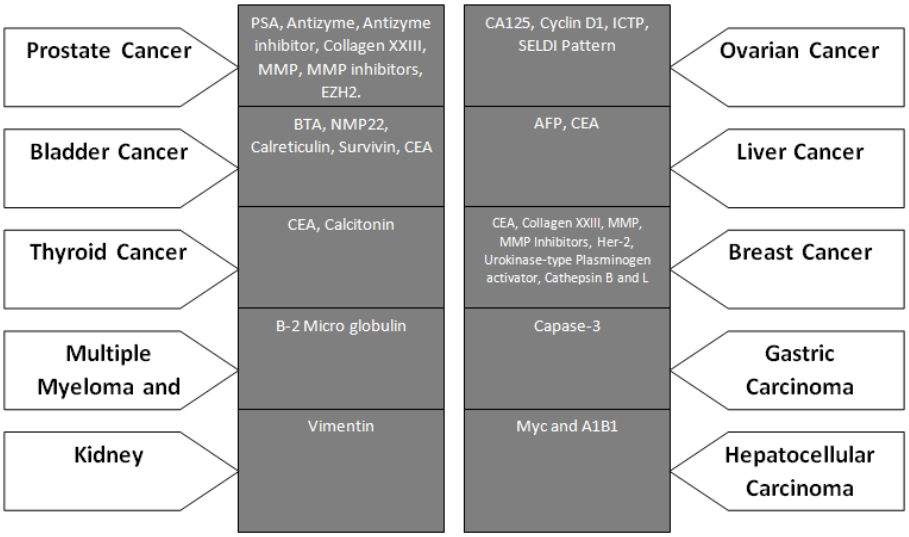


Figure 1: Various biomarkers related to specific cancer (Abbreviations) BTA: Bladder tumor antigen; CEA: Carcinoembryonic antigen; EZH: Enhancer of zeste homolog; ICTP: Carboxy terminal telopeptide of type I collagen; MMP: Matrix metalloproteinases; PAI: Plasminogen activatorinhibitor; PSA: Prostate-specific antigen; SELDI: Surface-enhanced laser desorption ionization.

Prostate-specific antigen (PSA)

Prostate -specific antigen is the first cancer biomarker to be identified and put into use for routine clinical application to diagnose and screening of prostate cancer. A normal PSA level in healthy men is considered to be 4.0 nanograms per milliliter (ng/mL) in blood, but this varies by age, for men in their 50s or younger men, a PSA level below 2.5 is considered as normal. In older men often have slightly higher PSA levels than younger men. A research study conducted by Smith *et.al* had demonstrated that roughly 30% of men with a PSA level between 4.1 and 9.9ng/mL had prostate cancer. In their study, Smith *et.al* demonstrated the screening test results and cancer detection rates based on a first-time screen and the pathologic features of a representative subset of screen detected tumors. Higher PSA level also indicate several other diseases like (hyperplasia, prostatitis, or smaller tumors). However, PSA level are not always purposeful for prostate cancer test. False PSA positive test are commonly, so while using of PSA for determination of prostate cancer certain factors like family history, age, ethnicity, digital rectal examination should be taken into consideration to nullify the low false examination.

Carcinoembryonic Antigen (CEA)

In 1965 gold and freedman first isolated CEA from human colorectal cancer (CRC). It is an oncofetal glycoprotein that normally expressed in low level by mucosal cell and high level of CEA got elevated in other cancers such as breast, liver, stomach, and pancreas. Several research studies have demonstrated that patients with high preoperative concentrations of CEA have more chances of cancer than those with low concentrations of the marker. The study had provided evidence that taking a reference value of 5 ng/ml of CEA, the sensitivity of CEA was at 37% only for patients with colorectal carcinoma at stage 1, 66.6% for patients at stage 2, and 75% for patients at stage 3. The specificities of the CEA for the cancers of the colon and rectum were at 76.98% with a reference value of 5 ng/ml and 86% with a reference value of 10ng/ml.

Cancer antigen 125 (CA 125)

Cancer antigen 125 is also known as mucin 16 or MUC16. It is only biomarker which is suggested for diagnosis and management of ovarian cancer. This monoclonal antibody was firstly isolated by Robert Bast *et.al* in 1981. CA 125 level is also associated with

several malignant condition including epithelial ovarian carcinoma, endometrial carcinoma, pancreatic carcinoma, breast cancer, lymphoma, lung cancer, colorectal cancer. Some nonpathological conditions like menstruation and pregnancy also increase CA 125 level. The reference range of CA 125 is 0-35 units/ml (0-35ku/L) it determines from the distribution of values in healthy 4 individuals to include 99% of the normal population. A serial determination of CA 125 biomarker provides more potency in determining response treatment.

Enhancer of Zeste Homolog 2

Enhancer of zeste homolog 2 (EZH2) is a histone-lysine N methyl transferase enzyme encoded by EZH2 gene, which participates in histone methylation and transcriptional repression. EZH2 is not present in healthy adults, it is only found in actively dividing cells. Because of this characteristic of EZH2 its overexpression can be used as a diagnostic marker of cancer and some neurodegenerative disorders. EZH2 help cancerous cells divide and proliferate. Its present in large amount in healthy cells associated with wide range of cancer including breast cancer, prostate cancer, bladder cancer, uterine and renal cancer as well as in melanoma and lymphoma.

Carboxy-terminal Telopeptide of type 1 Collagen (ICTP)

The serum concentration of trivalent cross-linked carboxyterminal telopeptide of type I collagen (ICTP), reflects changes in the extracellular matrix of the ovarian tumor and the surrounding tissues during cytotoxic chemotherapy and follow-up. The amino terminal propertied of type III procollagen (PIIINP), an indicator of type III collagen metabolism, is also a useful complement to CA 125 in monitoring the clinical changes of ovarian cancer. The clinical use of preoperative serum ICTP level determination gives specific information about the epithelial ovarian cancer and the consequent prospects of survival independently of conventional tumor markers.

Matrix metalloproteinase

It is the more promising application in both diagnosis and prognostic, MMP is also used to detect the tumour, also it primary and adjuvant therapy for breast cancer. MMP is useful in the prediction of therapeutic efficiency.

Ling cancer: MMP-1 over expression has been reported in lung cells, the DNA variant of MMP-1 are linked to lung cancer.

Ovarian cancer: -MMP-2-9 are the most studies biomarkers for ovarian cancer, ovarian cancer progression has also been reported for MMP-2-9, a recent study which shows more than 90% of clear-cell carcinomas expressed moderate to MMP-2.

Plasminogen activator inhibitor (PAI)

Plasminogen activator inhibitor-1 (PAI-1) also known as endothelial plasminogen activator inhibitor or serpin E1 is a protein that in humans is encoded by the SERPINE1 gene. The deregulation of PAI1 expression has been involved in cardiovascular diseases, obesity, metabolic syndrome and various types of cancer. A Patients with acute leukaemia, breast cancer or hepatocarcinomatous show an increase in the plasma levels of PAI1, which are also associated with histological grade of endometrial cancer. The PAI1 expression is also correlated with poor outcome in several other cancer subtypes, such as node-negative breast cancer and ovarian serous carcinoma. PAI1 has a role in cancer development, especially in breast cancer where it has been validated clinically, but the specific functions and roles of PAI1 depend on the type of cancer.

Prostate-specific antigen (PSA)

Prostate-specific antigen (PSA), also known as gamma seminoprotein or kallikrein-3 (KLK3), is a glycoprotein enzyme encoded in humans by the KLK3 gene. PSA is normally present in the blood at very low levels. The reference range of less than 4ng/mL. A patient with PSA level between 4 to 10 ng/mL have 25% chance of having prostate cancer and if PSA level 10 ng/mL and above there is 50% chance that person has a prostate cancer. However, other pathological conditions in the prostate, such as benign prostatic hyperplasia (BPH) or prostatitis can elevate the serum PSA levels, resulting in a "false positive" PSA test. PSA concentration in blood has been heavily explored for detection of prostate cancer, as well as treatment response and progression free survival monitoring thereof.

Surface Enhanced laser desorption ionization (SELDI):

Surface-enhanced laser desorption/ionization (SELDI) is a soft ionization method in mass spectrometry (MS) used for the analysis of protein mixtures. The SELDI method in conjunction with mass spectrometry (SELDI-MS) use for the diagnosis, prognosis and therapeutic monitoring of cancer and offers an attractive approach to cancer biomarker discovery from tissues and biological fluids. The use SELDI Compared to some other more traditional proteomic platforms, such as 2D polyacrylamide gel electrophoresis, it has a high-throughput capability and can resolve low-mass proteins.

Biosensors

Biosensors have immense potential to come up with advancement of new molecular diagnostic techniques for patients suffering with cancerous diseases. Diagnosing a cancer through cross sectional imaging (CT scan) and biopsy is costly and mostly uncomfortable approach for patients and yield considerable false-negative rates and a limited potential for early diagnosis of disease. The recognition of clinical biomarkers plays a vital role in the early detection of a cancer, design of individual therapies, and to identify underlying processes involved

in the disease. Biomarkers are chemical substances related with the elevation of malignant tumours which are found in blood, urine, or body tissues.

Various cancer markers are being broadly utilized for recognizing cancer, but carcinoembryonic antigen (CEA) is known to be a tumour marker related with colon, lung, ovarian and breast cancer that are dependable for more than half of all cancer passing each year. There are a number of potential preferences of biomarkers over other strategies of cancer determination, particularly decreased test time, comfort, tall affectability and selectivity, uncomplicatedness, miniaturized and flexibility. Biosensor-based diagnostics can help cancer screening and progress the rates of prior conclusion and orderly progressed figure. Biosensors have potential for multi-target examinations, computerization, and taken a toll viable testing.

Types of biosensor

Electrochemical biosensors

Electrochemical biosensors are the foremost common sort of biosensor utilize nowadays due to their transportability, taken a toll sustainability, little measure, and ease of utilize. Electrochemical biosensors can be used at domestic or within the doctor's office as point-of-care (POC) gadgets. The glucose sensor, which has been used for the blood glucose readings, is an electrochemical biosensor. Potentiometric and amperometry biosensors are the two most common sorts of electrochemical biosensors. Potentiometric biosensors use ion-selective anodes to identify an electrical reaction within the molecular acknowledgment element. Not however in clinical use, but appearing incredible guarantee within the zone of cancer discovery, may be a light-addressable potentiometric sensor (LAPS) that's coupled to a phage acknowledgment element. The phage-LAPS was able to distinguish the cancer biomarker hPRL-3 and cancer cells (MDA/MB231 breast cancer cell line) with tall affectability. This modern potentiometric-based biosensor has been proposed for utilize within the clinical discovery of cancer and anticancer sedate assessment.

Amperometric transducers degree current that's delivered when a potential is set between two terminals. Oxidation or decrease responses deliver a current, which can at that point be measured.

Optical biosensors

Optical biosensors are light-based sensors that degree changes in particular wavelengths of light. The transducer can be glow, fluorescence, colorimetric, or interferometry based. Optical transducers change over changes in wavelengths or SPR in reaction to analyst's acknowledgment into an electrical/digital readout. Photonic gem biosensors are a recently rising course of biosensors that utilize an optical transducer. The photonic gem sensor is planned to capture light from exceptionally little ranges or volumes, permitting for more noteworthy affectability of estimation, and after that transmit that light into a tall electromagnetic field for show. By measuring the light reflected by the precious stone, this procedure can distinguish when and where cells or particles tie to or are expelled from the gem surface. Utilizing this sort of biosensor, Chan and colleagues were able to screen changes in expansion and apoptosis of breast cancer cells uncovered to doxorubicin and decide the IC50 of doxorubicin. This sort of biosensor innovation would be valuable for pre-treatment screening of compelling measurements in arrange to adjust viability of treatment and poisonous quality.

Mass-based biosensors

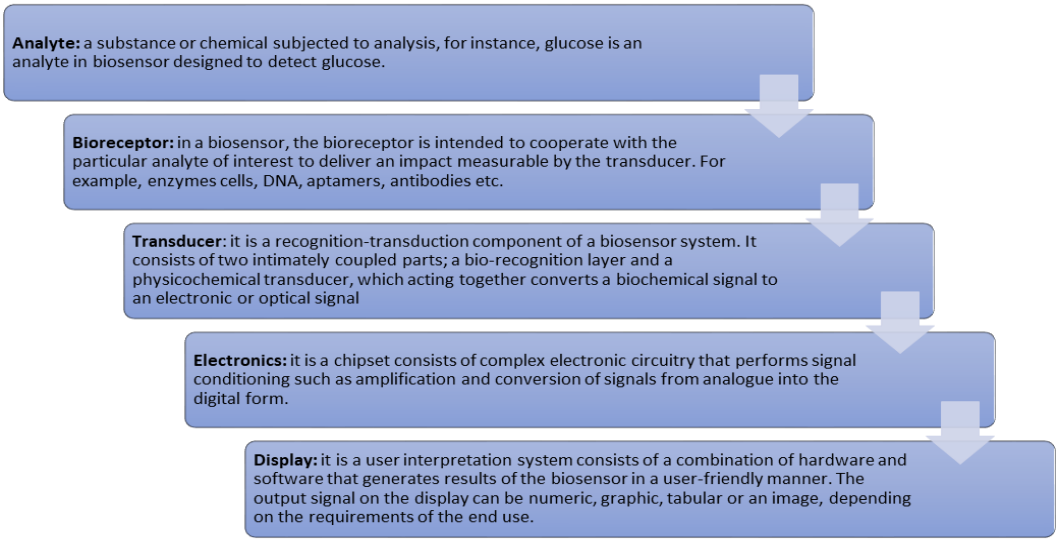
Piezoelectric and acoustic wave biosensors make up the lesson of mass-based biosensors. In terms of cancer location, piezoelectric biosensors are more commonly utilized. Piezoelectric sensors are based on changes within the mass of quartz gems when potential vitality is connected to them. This alter in mass creates a recurrence, which can be changed over into a flag. Immunosensors and microcantilever sensors that utilize piezoelectric technology have demonstrated valuable within the distinguishing proof of cancer biomarkers. A study about this conducted by Dell'Atti and colleagues outlined the utilize of a piezoelectric biosensor coupled with PCR intensification to distinguish point changes within the human p53 quality, which underlie in portion nearly all sorts of cancer. Since transformations of p53 are basic to cancer advancement and the viability of cancer medicines, there has been significant exertion within the improvement of speedy, reasonable, and viable ways to identify p53 changes.

Calorimetric biosensors

Calorimetric biosensors are less common than other biosensors for cancer diagnostics, but the presentation of nanotechnology to the field of biosensors has expanded the extend of applications for these sorts of biosensors. Calorimetric biosensors degree exothermic responses. Numerous enzymatic responses produce warm, and changes in warm can be utilized to degree analyte concentration. The response is checked by measuring enthalpy changes, which by implication give data almost substrate concentration. Calorimetric biosensors are not commonly utilized for the determination and forecast of cancer, but a few cancer-detecting capabilities have been illustrated. A later report by Mixture and colleagues illustrated the utilize of an aptamer-based gold nanoparticle calorimetric biosensor for the discovery of cancer. The analysts were able, utilizing the gold nanoparticles, to distinguish two diverse cell sorts, intense leukaemia cells and Burritt's lymphoma cells. This report outlines the achievability of combining aptamer-based acknowledgment components with a calorimetric transducer to identify cancer cells and possibly segregate between typical and cancer cells.

Components of Biosensor

A typical biosensor consists of following components.



Working Mechanism of Biosensor

Biosensors are operated based upon the principal of signal transduction. The Transduction measures This interaction and outputs a signal. The detection of clinical biomarkers plays a crucial role in the early detection of a cancer, design of individual therapies, and to identify underlying processes involved in the disease. The Bio transducer is the Recognition transduction component which consist of two parts {A} Bio-Recognition layer and {B} Physiochemical Transducer. Recognition element detect a signal from environment in the form of analysis of data. The combination unit of biosensor (sensitive elements and recognition transducer) will convert the biological response into electrical signal. Depending on the type of enzyme. The transducer converts the biological signal into electrical output. The output of transducer will be either current or voltage depending upon the type og recognition element (enzymes), if the output is current than it must be converted into equivalent voltage using an OP-Amp based current to voltage converter. The voltage output signal is generally very low amplitude and superimposed on a high frequency noise signal. So, the single should be amplified (Op-Amp based amplifier and pass through low pass RC filter. The amplifying and filtering process of the signal is the job of a Signal Processing Unit or a Signal Conditioning Unit. The output of the signal processing unit is an analog signal that is equivalent to the biological quantity being measured. The analog signal can be displayed directly on an LCD display but usually, this analog signal is passed to a Microcontroller, where the analog signal is converted into digital signal, since it is easy to analyse, process or store a digital signal. The Basic Work of the component of biosensor is to produce the Electric signal by detecting the group of analytes. Based on the highly selectively recognition of antibodies for antigens, Immunosensors have been manufactured and are widely applied in monitoring diseases related to proteins. This gesticulation can obtain from the changes of proton concentration, uptake or release of gases like oxygen and Ammonia, Light emission. Other mechanism comes out by the action of sensing molecules. This gesticulation also converted by the transducer among considerably responses. Biological and Electrical gesticulation are controlled again by amplification or processing. While the gesticulation might be running in principle, tool can be configured to yield one measurement to get specific market need.

Future Application of Biosensor

Advancements in the biosensor technology in the last few years put major impact in the Research and Development Sector. The large growth in the field of molecular biology has helped in the making of highly advanced Biosensors. The bio-components of the biosensor can be developed on the basis of molecular level which are more rapid in detection and cheaper to produce. This interest has brought about a more prominent prerequisite of the powerful and ideal way for the advancement of the biosensors to give as the front finish of estimation and control frameworks. Future bearings in biosensors innovation will address the expanded scope of analytes, totally incorporated frameworks having different periods of test dealing with, microfluidics, location and show conceivably telemetry, re-established accentuation on entire cell and tissue biosensor. A few biosensor applications for cancer diagnostics are depicted. Biosensors and micro-array chips that are based on location of hybridization/interaction in brief strands of nucleic acids propose a working framework for applications such as screening of genomes, discovery of pathogenic living beings, and compelling look in compound libraries to identify and study potential restorative operators. Early-stage lung cancer, one of the foremost common and forceful cancers, slaughters around 1.4 million individuals around the world each year – so the interest of modern strategies to precisely identify it remains a worldwide challenge. Presently, a profoundly sensitive graphene biosensor has appeared potential in electronic nose gadgets, which analyze the components of vapor blends such as breath. Multi-layered graphene can identify biomarkers specifically and delicately, giving

trust that a cheap, reusable, and exact breath test for early-stage lung cancer might gotten to be a reality.

As of late, uses of nanomaterials in biosensors give novel open doors for building up another age of biosensor innovation. Nanomaterials can improve mechanical, electrochemical, optical and attractive properties of biosensors.

CONCLUSION

In vitro molecular biosensors are nowadays playing an important role in biomedical diagnosis as well as a wide range of other areas such as point-of-care monitoring of treatment and disease progression, environmental monitoring, food control, drug discovery, forensics and biomedical research. The trends in biosensor technology over the past 30 years have taken this equipment from simple and cheap components to the integration of several sensor systems into one unit including multiple analytes, making these systems smaller and tailored for mass production. The vision for the biosensor industry is to create microscale technology that will be suitable for performing sample preparation, analysis and diagnosis all with one chip.

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