
Ovarian Cancer: Biomarkers-Emerging Applications of Biomarkers in Healthcare

Biomarkers are small protein or peptide molecules which are part of cell receptors, cell signalling molecules involved in cell-cell signalling or cell protein kinase activation and transfer of signal to the nucleus with the regulation of gene transcription. Biomarkers have functional significance in the recognition of different stages of cancer, therapy for cancer, and identification of different types of cancer of different organs according to the biomarker present. It is also used for the diagnosis and analysis of mutations in various proteins and identification of single nucleotide polymorphism in genes, screening of diseases, classification of diseases in humans and development of target inhibitor molecules against the biomarkers [1][2]. Various other studies have indicated the presence of autoantibodies against proteins of the cells which act as biomarkers in different grades of cancer and autoimmune response[3].

Keywords: Biomarkers, ovarian cancer, breast cancer, prostate cancer, pharmacological markers, drug screening.

Introduction to applications of biomarkers

Potential applications of biomarkers include the analysis and screening of protein molecules, characterization, post-translational modifications like glycosylations, and prediction of structure of molecules. Other applications include the analysis and study of phosphorylation patterns and protein-protein interactions of protein biomarkers during activation of cell signalling mechanisms inside the cell. Biomarkers are used for the study of pharmacological inhibitors, study of mutations in biomarker proteins due to somatic and germline mutations in genes during transfer of characters and traits from one generation to another in humans. The other functions are analysis of subunit of protein biomarkers and their activation mechanism in cells during cancer, therapy models for identification and recognition of cancer of different organs, study of storage of biomarkers in various organelles as well the study of sorting and targeting of folded and misfolded protein biomarkers in endoplasmic reticulum, golgi apparatus along with their degradation by chaperone pathway[2]. Transcriptome-based genomic and proteomic techniques are used for the analysis and characterization of biomarkers involving digestion of peptide with enzymes like trypsin and study of peaks by mass spectroscopy, two-dimensional gel electrophoresis and isoelectric focusing[4].

Traditional applications of biomarkers

Applications of biomarkers involves the study of these molecules in various cancer diseases like hepatocellular cancer, prostate cancer, lung, liver, stomach, oral, nonsmall lung cancer, head and neck cancer, ovarian cancer and cervical cancer. Various techniques like peptide mass fingerprinting, surface-enhanced laser desorption studies, immunoaffinity metal chelations studies with nickel metal and biomarker protein molecules in columns, differential gel electrophoresis, cation exchange and anion exchange chromatography are essential for cancer studies. The other techniques are stable isotope labeling of amino acids in cell culture, multi-dimensional techniques, matrix-assisted laser desorption studies, mass spectroscopy and

capillary electrophoresis along with the study of post-translational modifications of amino acids in biomarker proteins are essential for the studies of proteins.[2,4](Fig no.1). Further microbial proteomics study is performed to study the pathogenicity clusters in bacteria, infection-causing proteins in bacteria, genome analysis of bacteria and protein toxins

Fig no.1—The applications include the techniques used in the identification and screening of biological biomarkers source-Mining the plasma proteome for cancer biomarkers [9].

causing disease in humans, antimicrobial proteins involved in killing other different microorganism population of bacteria in microbial consortium [4].In addition to this, miRNA profiling has been performed for the study of cancer biomarkers with the study of the regions of binding of protein biomarker miRNA along with target therapy for the analysis of inhibition of miRNA expressions and its downregulation and upregulation in various grades of cancer[5].

Emerging roles of biomarkers

The protein biomarkers are analysed in various cancerous organs for example glycoprotein CA-125, VEGF(vascular endothelial growth factor), transferrin, apolipoprotein A1 level and transthyretin levels, carcinogenic embryogenic antigen (CEA) in ovarian cancer, prostate-specific antigen, prostate acid phosphatases, and serum acid phosphatases in prostate cancer. Hepatocyte growth factor, survivin, alpha-fetoprotein, neuroserpin levels in hepatocellular carcinoma, atrial natriuretic factor in heart diseases, alpha-synuclein, PINK proteins(PARK), glial fibrillary acidic protein, galectin in Parkinson disease, chemokine ligand, tau proteins in Alzheimer's disease, estrogen receptor. Her-2 EGFR receptor in breast cancer, and KRAS mutations in colorectal carcinoma. [1,2,4].Various scientific committees have proposed the classification and nomenclature for protein biomarkers in organ cancer along with the significance and role of biomarkers[1].

(New pharmacological drugs for screening of biomarker and drug inhibitor response in cell lines) Further pharmacological screening is performed for the efficiency, the affinity of binding, inhibition of biomarker protein in cells and drug-biomarker protein interactions in cell lines(Fig no.2). The drug-mediated targeting to the cancerous organ is performed with the help of liposomes, virus-mediated therapy, nanoparticles, micelles, dendrimers, photodynamic therapy, fluorescent labelling of biomarker protein molecules and visualisation by biomedical imaging[11] along with pharmacological docking, affinity studies, inhibition studies of drug and protein biomarkers like drug-EGFR(HER-2) mediated therapy[12] and drug interaction with mTOR signaling pathway proteins with rapamycin[13].The anti-EGFR receptor protein which is a biomarker binds to the inhibitor drug molecules and prevents the phosphorylation of the tyrosine amino acid on the EGFR receptor thus preventing the activation of cell signaling [12]. Major protein biomarkers used in drug metabolism include cytochrome P450 enzymes in liver[15]. Further pharmacodynamics and pharmacokinetics curves are plotted for the affinity of binding of drugs to various protein biomarkers in the cell lines under investigation. Additionally, the role and characterization of biomarkers include markers with mutations in proteins in cancerous cells, biomarker antigens for cancer detection, protein biomarkers in immunohistochemistry and fluorescent staining of cells and tissues in pathology. The other roles of biomarkers are radiolabeled protein biomarkers, quantum dot labeled biomarkers, biomedical imaging of fluorescent biomarkers in in-vivo studies of rats, circulating DNA found in tumour cancer cell lines and in blood of patients and pharmacodynamics and pharmacokinetics studies of drug-

protein biomarker interaction[14].

(New pharmacological drugs for screening of biomarker and drug inhibitor response in cell lines)

(Drug-docking studies of protein biomarker inhibitor response in vivo in humans and mice.)

(Binding affinity studies of drugs and proteins example for Alzheimer's disease)

(Clinical validation of inhibitor and drugs used for targeting cancer.)

Fig no.2—The screening of drug-biomarker protein interaction is confirmed along with mechanism of action of drugs, monitoring of effects of the interaction of drugs, diagnosis and treatment of cancers in patients [10].

No.

Role

Biomarker

Characteristics

Example disease

Author/year

1 Ovarian cancer

Carcinogenic embryonic antigen A and CA-125(carbohydrate antigen-125)

Present in ovarian cancer

cystadenocarcinoma

Sorenson et al.,2011, sreeja sarojini et al.,2012,Samimi et al.,2014.

2 Prostate cancer

Prostate specific antigen

Present in prostate cancer

Prostate adenocarcinoma

Chatterjee et al.,2012,Gu et al.,2014,Meng et al.,2013.

3 Hepatocellular carcinoma

Hepatocyte growth factor

Receptor on hepatic or liver cells

Hepatocellular carcinoma

Forner et al.,2012,Goodman et al.,2007,Hashem B et al.,2011.

4 Breast cancer

HER-2 EGFR receptor

Present of cell surface of mammary cells

Breast carcinoma

Yu et al.,2012,Tam et al.,2010, Makki et al.,2015.

5 Parkinson disease

Alpha –synuclein, glial fibrillary acidic protein

Aggregates of proteins in neurons

Neuronal disorder

Jankovic et al.,2014,Hauser et al.,2013,Clebak et al.,2013

6 Alzheimers disease

Tau proteins

Aggregates in neuronal cells

Neuronal disorder

Mattson et al.,2010,Palmer et al.,2011,Olokoba et al.,2011

Table-1 Shows various cancer diseases with biomarkers and their characteristic features.

Case studies of biomarkers

The study involves the sample analysis of urine of patients with cancer as compared to normal urine samples for the analysis of homovanillate, 4-hydroxy phenylacetate, the levels of metabolic products like 5-hydroxyindolyl acetate and urea formed in the body under two different prevalent conditions. Microarray analysis was performed along with statistical normal distribution analysis of the normal cell samples and cancer cell patient samples to study the levels of metabolic analytes. Further breast cancer adenocarcinoma cell samples were analysed for BRCA gene in patient samples. The analysis of urine samples was done by gas chromatography and mass spectroscopy after preparing the samples in buffer and digesting

them with beta-glucuronidase enzyme[6]. The P-value was calculated for the intensity of binding in microarray samples along with the study of the levels of tyrosine metabolism analytes in cancer samples and normal samples. The other study was conducted on ovarian samples of cancer from patients with the study of random intensity scatter distribution plots of the genes involved in ovarian cancer regulation[7].

A study of prostate cancer revealed the levels of sarcosine in cancer samples and normal samples along with the study of statistical parameters like mean, median, mode, and area under the curve for the intensity profile of the metabolite levels in samples under study[8]. Further high-performance liquid chromatography was done to purify the sarcosine in the samples under study and to analyse its levels under experimental observation.

Conclusion

The study helps in the analysis of various grades of cancer affecting different organs by the study of particular expressions of protein biomarkers in various organs of the body. Various methods like immunohistochemistry and fluorescence in situ hybridization were performed to analyze the location and levels of biomarkers in cell and tissues under study [1]. There is a range of protein biomarkers used for the study of cancers in different organs according to the role of these protein biomarkers in cell signaling, kinase phosphorylation, and transcriptional regulation in tumor and cancerous cells as compared to normal cells[2]. The applications involve the use of various techniques like mass spectroscopy for the study of biomarker proteins and their amino acid characterization in the sequence along with post-translational modification[2,3]. The autoimmune antibodies formed in the body against self-altered proteins of the body also act as protein biomarkers in various autoimmune diseases[3]. The particular protein markers specific to organs and tissues were studied in samples under experimental analysis[1,4].miRNA levels were analyzed in various cancer samples along with the study of its regulation in the genome[5]. Further microarray analysis was performed to study the levels of breast cancer genes like BRCA[6], ovarian cancer genes[7] and antigen prostate-specific antigen in cancer samples and normal samples as well as sarcosine metabolite samples with statistical analysis[8].

References

1. Cancer biomarkers--N. Lynn Henryra, Daniel F. Hayes b-Molecular Oncology, 6 (2012) 140e1 4 6.
2. Proteomics and Human Diseases --Rosa Lippolis and Maria De Angelis-Journal of Proteomics and Bioinformatics., Lippolis and De Angelis, J Proteomics Bioinform 2016, 9:3.
3. Serologic Autoantibodies as Diagnostic Cancer Biomarkers—A Review--Pauline Zaenker and Melanie R. Ziman., Cancer epidemiology, biomarkers and prevention., September 20, 2013;
4. The cancer secretome: a reservoir of biomarkers--Hua Xue, Bingjian Lu and Maode Lai., Journal of translational medicine., 2008, 6:52.
5. MicroRNAs in cancer: Biomarkers, functions and therapy--Hayes J, Peruzzi P, Lawler S., Trends in Molecular Medicine, vol. 20, issue 8 (2014) pp. 460-469.
6. Combining tissue transcriptomics and urine metabolomics for breast cancer biomarker identification--Hojung Nam¹, Bong Chul Chung², Younghoon Kim¹, KiYoung Lee³ and

Doheon Lee¹—Bioinformatics, Vol. 25 no. 23 2009, pages 3151–3157.

7. Study design and data analysis considerations for the discovery of prognostic molecular biomarkers: a case study of progression free survival in advanced serous ovarian cancer, Li-Xuan Qin and Douglas A. Levine², Qin and Levine BMC Medical Genomics (2016) 9:27.
8. A case control study of sarcosine as an early prostate cancer detection biomarker- Donna P. Ankerst^{1,2,3*}, Michael Liss², David Zapata², Josef Hoefler¹, Ian M. Thompson² and Robin J. Leach^{2,4.}, Ankerst et al. BMC Urology (2015) 15:99.
9. Mining the plasma proteome for cancer biomarkers--Samir M. Hanash¹, Sharon J. Pittner¹ & Vitor M. Faca¹, Nature., 452, 571-579 (3 April 2008).
10. Biomarkers for Alzheimer's disease: academic, industry and regulatory perspectives- Harald Hampel, Richard Frank, Karl Broich, Stefan J. Teipel, Russell G. Katz, John Hardy, Karl Herholz, Arun L. W. Bokde, Frank Jessen, Yvonne C. Hoessler, Wendy R. Sanghai, Henrik Zetterberg, Janet Woodcock & Kaj Blennow.
11. Drug targeting to tumors: Principles, pitfalls and (pre-) clinical progress, Twan Lammers, Fabian Kiessling, Wim E Hennink, Gert Storm, Journal of Controlled Release, Volume 161, Issue 2, 20 July 2012, Pages 175-187.
12. HER2 therapy. Small molecule HER-2 tyrosine kinase inhibitors--Spector N, Xia W, El-Hariry I, Yarden Y, Bacus S--Breast Cancer Research, vol. 9, issue 2 (2007) p. 205.
13. Targeting the mTOR Signaling Network for Cancer Therapy Funda Meric-Bernstam Ana Maria Gonzalez-Angulo, Clinical Oncology, 27, no.13 (May 2009), 2278-2287.
14. Pharmacological Biomarkers in the Development of Stratified Cancer Medicine- William Douglas Figg Sr., Pharm.D., MBA, David R. Newell., Clin Cancer Res. 2014 May 15; 20(10): 2525–2529.
15. Pharmacogenomic biomarkersxAna Maria Gonzalez-Angulo-Kirkwood SC1, Hockett RD Jr., Dis Markers. 2002;18(2):63-71. Search for articles by this author