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Emphasizing the Role of Multi-omics Approach to Increase Survival Rate of Breast and Prostate Cancer Patients

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Abstract

The understanding of cancer biology has greatly advanced since the advent of genomics. A remarkable heterogeneity at the whole-genome (or omics) level exists amongst even histologically comparable cancers, demonstrating the enormous complexity of the cancer genome. A powerful resource that has the potential to translate high-throughput omics to better and quick overall survival is the massive accrual and public accessibility of multi-omics databases with accompanying clinical annotation, including tumor histology, patient response, and outcome. In this new era of high-throughput omics, this paper emphasizes the distinct benefits of a multidimensional approach to genomic analysis. It discusses the implications of translational omics research for the cancer population. Single-level data analysis of high-throughput technologies has constraints because it only displays a small window of cellular processes. Understanding the links across several cellular organization levels made possible by data integration across various platforms, including genomes, epigenomics, transcriptomics, proteomics, and metabolomics, is important. This review examines a few popular frameworks for integrating multi-omics data. It provides a general overview of multi-omics applications in tumor classification, prognosis, diagnostics, and the function of data integration in searching for novel biomarkers and treatment options.

Keywords: Biomarker Discovery, Cancer, Omics, Personalized Medicine, Risk Assessment

1. Introduction

The "Omics" technique is introduced as a promising biomarker sighting tool in the current biological period, focusing on many chemicals. The different tools that help to have a simple and systematic understanding of the extremely complex biological system include genomics (genome), proteomics (proteome), and metabolomics (metabolome). The "omics" approach for investigating complex biological systems differs greatly from the usual method. Theragnostic for precision medicine and advancements in cancer diagnostics would help with early detection, prognosis, and therapy of the disease and identify new molecular targets for drug development. Integration of regulatory layers may be beneficial for analyzing abnormal cellular processes that underline complex illnesses like cancer. A deeper knowledge of how genetic variations, environment, and interaction of the two affect complex biological systems is made possible by measuring biological samples on various omic scales. Using integrative models, multi-omics data analysis enhances the clustering of samples into physiologically significant groups, increases knowledge of prognostic and predictive phenotypes, analyses cellular responses to therapy, and supports translational research. This review highlights the benefits of such a comprehensive strategy while highlighting the innovative understanding that multi-omics has given to cancer modeling (Figure 1). The TWO common cancers diagnosed in females and males are Breast and Prostate cancer, respectively. An examination of cancer biomarkers, including somatic mutations, dysregulated expression, epigenetic control,

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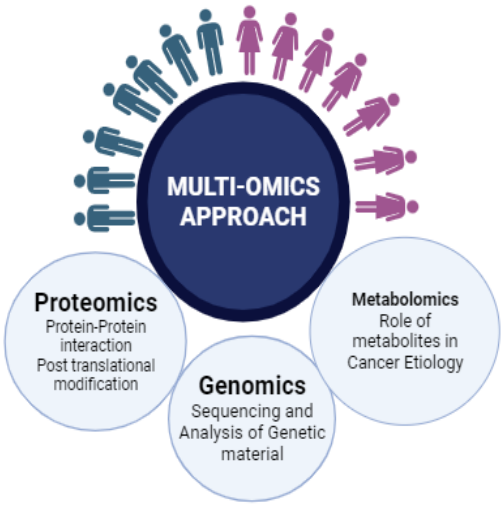

Figure 1. Multi-omics approach to increase cancer patient survival rate in breast and prostate cancer.

and genomic variants linked to significant tumors in the case of breast and prostate cancers, is attempted in this review.

2. Breast Cancer and the Need for Omics

Regardless of identical histological markers at diagnosis, breast cancer is known for its diverse clinical behaviors and patient outcomes. Breast cancer cells with varying degrees of treatment, resistance, and metastatic potential result from the heterogeneity and dynamics of clonal evolution. The presence or lack of estrogen, progesterone receptors, and human epidermal growth factor receptor 2 determines traditional therapy methods for specific individuals[1]. However, such clinical classifying techniques are useful for selecting targeted medicines and predicting short-term patient responses, but they cannot predict long-term survival. Cancer patients in rural locations are detected at late or advanced stages of the disease, with widespread metastases, indicating a need for more awareness, treatment, and early diagnosis. In recent years, India has been facing a difficult situation due to an increase of 13.5% in breast cancer incidence and a 10.6% increase in breast cancer mortality. In India, 162,468 women were newly diagnosed with breast cancer in 2018 alone. Breast cancer also accounted for 27.7% of all newly diagnosed malignancies in women. This indicates that, in India, one out of every four newly diagnosed cancers in women is breast cancer[2]. Breast cancer has risen to the top in all major urban registries due to increasing urbanization and westernization, as well as changing lifestyles and food habits. The lack of comprehensive breast cancer screening, disease detection at an advanced stage, and insufficient medical facilities are the leading causes of the reported increase in death rates. In India, one can see an increasing number of patients diagnosed with breast cancer in their early twenties. This is indeed an alarming situation in younger patients. There are a few comparable statistics for the survival rate of breast cancer in India. However, a rough estimate based on the Population-Based Cancer Registry (PBCR) and Hospital-Based Cancer Registry (HBCR) reports suggests that the 5-year survival rate for breast cancer patients in Indian women is less than 60%. Many cancer patients with HER2 positive and ER/PR negative or HER2/ER/PR negative subtypes have poor prognoses. Breast cancer is relatively common in the younger age group (25 to 49 years), accounting for around 37.7% of all cases, which is quite significant. Breast cancer peaks in the 50-69 age group (which accounts for approximately 46.5% of all cases) and then begins to decline in the 70-plus age group. It may not be decreasing, but rather it impacts life expectancy[3].

Breast cancer incidence and death metrics indicate an urgent need to develop robust knowledge-based prognostic systems capable of generating phenotypic estimates for an individual. To address this issue, personalized medicine seeks to provide the most effective treatment strategy based on the patient's medical history, genomic characteristics, and therapeutic response[4]. Current breast cancer therapies may be most effective if the tumor size decreases rather than increases in the patient. The assessment of tumor size from radiological imaging, the response of a tumor even to effective medication, and the high possibility that the measurement of tumor size will not change considerably in the two weeks between hospital visits are all subject to considerable errors in diagnosis. Tumors, however, can develop resistance to treatments. Over time, the likelihood of a therapy being effective decreases. The effectiveness decreases when the patient receives that therapy or, to a lesser extent, another therapy from the same line with similar functional mechanisms. In this context, current systems biology based on "omics" techniques has the potential to play a significant role in resolving these issues. Indeed, in the

age of precision medicine, "omics" methodologies and their application to the study of breast cancer could be viewed as a new biomarker discovery tool, resulting in novel biomarker molecules and molecular signatures with therapeutic use.

2.1 Genomics in Breast Cancer

The genomics of breast cancer greatly benefits from next-generation sequencing. Earlier, the breast cancer-causing genes BRACA 1 and BRACA 2 were identified. Many new oncogenes and tumor suppressor genes for breast cancer were discovered in the post-NGS era, and their interactions were also investigated. The genes are divided into several groups based on the alteration type (mutation/polymorphism) or cancer susceptibility (high/moderate/low penetrance). In the 1990s, researchers revealed that a woman's family history is the most accurate indicator of her risk of having Breast Cancer (BC). After intense research, two genes were eventually found. The first, BRCA1, was discovered in 1994, and the second, BRCA2, was linked to BC in women in 1995[6,7]. Notably, a meta-analysis revealed that at age 70 the average cumulative BC risks for carriers of the BRCA1 mutation were 57% and 49%, respectively[8]. Based on the multigene sequencing, additional genes were connected to the susceptibility of BC. BRCA1-Associated Ring Domain (BARD1), a direct companion of BRCA1, is anticipated to be a low-moderate penetrance BC risk gene[9]. Loss-of-Function (LoF) mutation of BRAD1 gene was present in 0.51 percent of BC patients.

Additionally, BARD1-mutated BC patients had a significantly lower mean age at first diagnosis than the entire study cohort (48.6 years, range 17-92 years) (42.3 years, range 24-60 years)[10]. Similar to BRCA1, germline LoF mutations in BRIP1 (BRCA1 interacting protein C-terminal helicase 1), a low penetrance gene that interacts with BRCA1, are associated with a higher risk of BC development, particularly in people who are diagnosed with the disease at a young age[11]. SMAD4 is another gene that is inactive in BC patients. The Bone Morphogenetic Protein (BMP)/Transforming Growth Factor (TGF) signaling pathway uses SMAD4 as a shared signal transducer and transcription corepressor for the human Estrogen Receptor (ER). An area of the genome frequently deleted in BCs is 18q21, where SMAD4 is located[12]. The inactivation or inhibition of TGF-/SMAD4 signaling has been discovered to be crucial for BC development[13-15]. The Fanconi anemia-DNA repair pathway is disrupted by PALB2 (also known as FANCN) germline LoF mutations, which increase the likelihood of developing BC[16,17].

The pathogenic missense mutation L35Pa in PALB2 also interferes with the PALB2-BRCA1 interaction, which may fail BC suppression[18]. The other two genes recently utilized in screening BC susceptibility are RAD51C and RAD51D[19,20]. According to estimates, carriers of the RAD51C pathogenic variation and the RAD51D pathogenic variant produced a cumulative risk of 20% and 21%, respectively, of getting BC over the following 80 years. The BC risk for the RAD51C and RAD51D pathogenic mutations could range 44–46 % for carriers with two first-degree relatives diagnosed with BC[21]. Two more notable genes that are altered in BC are NBN and CDK12. Cyclin-Dependent Kinase 12 (CDK12) is a regulatory kinase with evolutionarily conserved functions in controlling transcription elongation. It is a low penetrance gene. In BCs, HER2 and CDK12 are typically co-amplified[22]. In 21% of primary, unselected BCs, CDK12 expression was found[23,24]. The Wnt/-catenin signaling pathway regulating gene APC (Adenomatous Polyposis Coli gene) is involved in preserving low levels of catenin in the cell. APC gene SNP rs2229992 was shown to be associated with high risk of breast carcinogenesis in a community of BC in eastern India[25].

2.2 Proteomics Approach for Breast Cancer

Proteomics is a thorough, high-throughput study of proteins that looks into how they are categorized, how much they are expressed, what they are made of, and what they do. Several studies on protein quantification have been done thus far, using strategies based on antibodies whose availability, amount, affinities, and specificities are all closely related. Proteomic tests have quickly advanced from total protein counts acknowledged and lists of proteins and peptides described in test samples. Breast cancer protein biomarkers may be produced by immune cells invading the tumor or stroma, tumor cells, or stromal cells. Due to variable degrees of incongruence between RNA and protein expression, functional biological characteristics are not fully captured in intrinsic gene expression profiles. The study of the "secretome" and cell markers has received considerable attention in recent years. The vast and intricate array of chemicals and proteins secreted by living cells and discharged

from their surfaces is called the "secretome." Since secretome proteins play a crucial role in cell signaling, communication, and migration, the study of tumor cell secretomes has come under increased scrutiny to identify and characterize diagnostic and prognostic markers and potential pharmacological and therapeutic targets[26]. In addition, many researchers have switched their attention from studying BC cell lines to Cancer Stem Cells (CSCs) in recent years. Nearly 65% of cases of cancer recurrence are known to involve CSCs in a major way[27,28]. Unlike cancer cells, CSCs are latent, making them immune to anti-cancer medications.

Additionally, after receiving anti-cancer medication, these cells are induced to proliferate quickly. Breast cancer patients who have Cathepsin D, a lysosomal protease gene (CTSD) expression upregulated, have a bad prognosis; moreover, the role of CTSD is completely unrevealed to date[29]. Patients inflicted with breast cancer with expression of greater levels of FABP7 (Fatty Acid Binding Protein 7) had worse survival rates and more instances of brain metastases as it plays a crucial role in proliferation and cell migration[30].

2.3 Metabolomics for Breast Cancer

One of the newest members of the omics family, metabolomics, has advanced significantly over the past ten years, largely due to developments in mass spectrometry technology. Because metabolites serve as the link between genotype and phenotype, investigating the metabolome offers a considerable advantage by revealing the endpoints of biological events, as is now generally acknowledged. Metabolomics, in addition to genomes and proteomics, can show endpoint biomarkers for diagnosis or therapeutic response evaluation. Blood contains clinical biomarkers that are much easier to spot. Therefore, it is crucial to determine if the metabolic reaction seen in the blood is actually derived from tumor tissue or reflects a more general response of the body to the existence of a tumor[31,32]. Numerous studies have discovered indicators for the metabolism of fatty acids, amino acids, and glycolysis. Phe, Gluc, Pro, Lys, and N-acetyl-Cys levels were high in metastatic patients, in contrast to the low lipid levels in those patients. The by-products of -oxidation (Acac and 3-HB) and lipid breakdown (Gluc), as well as N-acetyl glycoproteins (NAC 1 and 2), Pyr, Glut, and mannose, have been detected in the blood of early and advanced BC patients. Lipids, 3-HB, Lact, His, Pro, and Phe are frequent marker metabolites[33]. As a result, several important routes for the early detection of BC have been identified, including the metabolism of taurine, hypotaurine, alanine, aspartate, and glutamate[34,35]. A dried blood spot method by Wang *et al.* showed better diagnostic sensitivity towards BC[36]. Another biological fluid that has been studied is saliva, and metabolites found in it, include 3- and 4-methyl-pentanoic acids, phenol, p-tert-butyl-phenol, acetic, propanoic, and benzoic acids, as well as 1,2-decane diol, 2-decanone, and decanal[37]. Due to their formation and accumulation, polyamines, particularly N-acetylated versions, are a different chemical class connected to tumor growth[38].

3. Prostate Cancer

Prostate cancer is the second most frequently diagnosed disease and the sixth most common cause of cancer mortality in males globally. In 2020, it was predicted that there would have been 1,414,259 new cases of prostate cancer and 375,304 deaths related to it[39]. Androgen Deprivation Therapy (ADT) and AR pharmaco-antagonists (androgen insensitivity syndrome) are two common treatments for PCa sensitive to aberrant alterations in AR. Point mutations and deletions are examples of changes in the AR genes. This insensitivity is caused by mutations in the AR receptor's second zinc-finger ligand-binding domain[40].

3.1 Genomics and Proteomics for Prostate Cancer

PTEN mutations and PCa aggressiveness are positively correlated, according to subsequent investigations. Recent research has demonstrated a beneficial correlation between PTEN loss and the de novo lipogenesis enzyme, Fatty Acid Synthetase (FASN) gene knockdown[41]. The amount of stromal micro-invasion decreased due to the downregulation of both genes. SREBP, a transcription factor that controls de novo lipogenesis and adipogenesis, was activated when PTEN was co-deleted with other genes, such as PML1[42]. Glycolysis and PCa cell glucose uptake are inhibited due to p53's suppression of the expression of glucose transporters. Through activating glutaminase 2 (GLS2) and regulating glutamine absorption, p53 expression supports OXPHOS. A study connecting p53 mutations in PCa cell lines with PCa primary human

metabolites in 10 out of 40 samples established p53 as a PCa tumor suppressor for the first time.

The functional impact of p53 mutation, specifically deletion, on PCa progression was confirmed by subsequent p53 studies[43]. According to a recent study, the dietary component Phenethyl Isothiocyanate (PEITC) reduces PCa cell development by triggering apoptosis by saving mutant p53 in the VCaP and LAPC-4. Increased Serine One-Carbon Glycine synthesis (SOG), which is in charge of DNA methylation, is also connected to p53 loss[44]. It has been shown that c-MYC affects the expression of several enzymes involved in the glycolytic pathways, including HK2, PFK1, ENO1, LDHA, and GLUT1. Additionally, c-MYC controls GLS1 and the transporters it coordinates, promoting glutamine metabolism. The PI3K/AKT axis is activated by amplifying c-MYC. The relationship between c-MYC amplification and PI3K-associated dysregulation, including PTEN and all AKT homologs, has been shown in localized and metastatic PCa[45]. In all PCa cell types, c-MYC and AKT1 activities promote increased metabolites linked to lipogenic and glycolytic processes. According to a recent study, there is a favorable correlation between c-MYC expression and AR activity. However, due to both proteins sharing identical enhancer binding sites in a different study, overexpression of c-MYC had a negative impact on AR activity and transcription in PCa cell lines. In advanced PCa, c-MYC exhibited an inverse correlation with the AR target genes KLK3 (PSA) and GNMT[46].

3.2 Metabolomics for Prostate Cancer

Metabolomics is a new and promising approach for PCa biomarker research that has just emerged. Numerous multivariate biomarker sets have been investigated for various use cases. Production of citrate, PSA, and polyamines like spermine, the main constituents of prostatic fluid, is one of the primary tasks of prostate cells. Therefore, prostate cells have a unique metabolic profile because they produce these and other chemicals. Compared to other organs, prostate cells produce a large amount of citrate. Citrate oxidation is one of the most prominent changes because cancer cells cannot collect zinc. Since zinc levels are low in cancer cells, m-aconitase is no longer blocked and can catalyze citrate oxidation. More effective energy production arises from converting citrate build-up in healthy prostate cells to oxidized citrate in malignant prostate cells. This likely occurs before the histological detection of malignant cells and is an early stage in the development of malignancy[47].

In a recent study, it has been shown that Androgen Receptor (AR) signaling brings about increased levels of Glucose-6-Phosphate Dehydrogenase (G6PD), NADPH, and ribose synthesis in hormone-sensitive PCa cells and Castrate-Resistant PCa (CRPC) cells. G6PD is a key enzyme for the pentose phosphate pathway[47]. The overexpression of G6PD is stopped by rapamycin, which inhibits the mammalian target of rapamycin. As a result, these investigations demonstrated a connection between the mammalian target of rapamycin and the activation of G6PD via AR. These findings suggested that the pentose phosphate pathway is crucial for growth[48].

Increased lipid production is necessary for cell division and intercellular communication. Because it is a precursor for lipogenesis and cholesterogenesis and can be formed by the cytosolic transformation of citrate, acetyl-CoA also plays a significant role in this metabolic change. Sterol regulatory element-binding protein-1 boosted lipogenesis, NADPH oxidase expression, reactive oxygen species generation, and PCa cell proliferation, migration, and invasion. Choline and creatine levels are elevated in PCa cells due to increased membrane production for cell proliferation[49].

The interactions between the tumor and the stroma are significant in PCa progression. The reverse Warburg effect depends on the Myo-fibroblastic microenvironment, created when cancer cells interact with "cancer-associated fibroblasts." The Warburg effect, generated by epithelial cancer cells, causes cancer-associated fibroblasts in the Myo-fibroblastic microenvironment to release lactate and pyruvate. The PCa cells absorb the lactate and pyruvate and utilize them for the Krebs cycle, anabolic metabolism, and cell growth[50].

Sarcosine (N-methylglycine), one of the most critical metabolomics investigations, was found to be a potential PCa biomarker in urine[51]. Sarcosine was found to be considerably raised during PCa progression to metastasis and was either undetectable or present in insignificant amounts in the urine of healthy individuals. Sarcosine is an intermediary product in the synthesis and breakdown of glycine[52,53].

In addition to changes in sarcosine levels, metabolomics analyses of serum and plasma from PCa patients also showed changes in fatty acids, amino acids, lyso-phospholipids, bile acids, and metabolites connected to the route for the manufacture of steroid hormones.

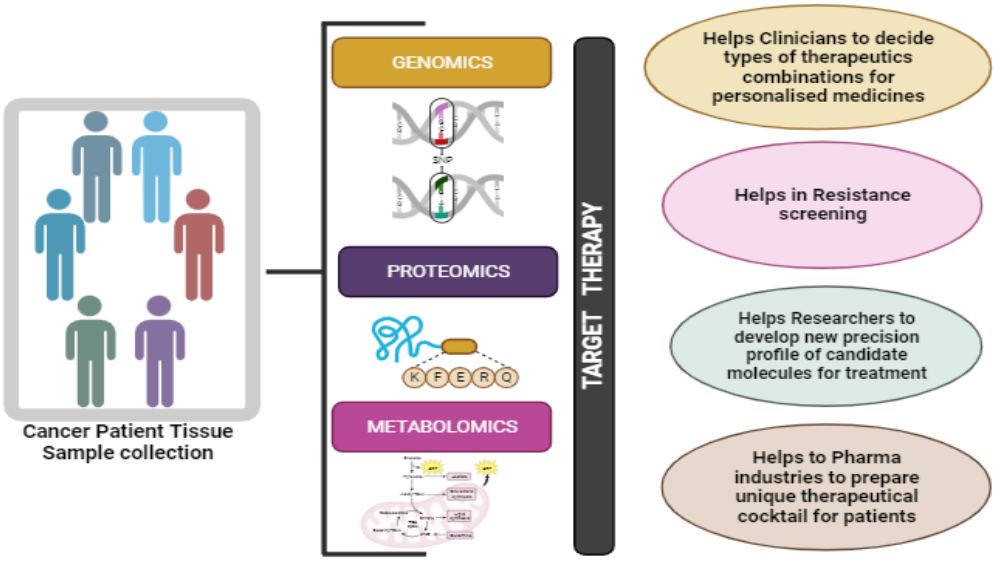

Figure 2. Schematic of possible approach explaining diagram to predict future precision medicine.

Variations in lipid -oxidation, which are required to produce the energy for aberrant cell proliferation, are connected to changes in fatty acid composition. Increased glucose levels in serum samples at the time of PCa diagnosis were linked to an increased risk of recurrences following treatment with radical prostatectomy or radiation therapy. Alterations in energy metabolism are also common[54,55].

To find changes in cancer cell metabolism and non-invasive biomarkers for PCa detection, PCa metabolomics investigations may also be conducted on seminal and prostatic fluids. When comparing PCa groups to controls, prostatic and seminal fluid analyses using different analytical methods (fluorescence technique and NMR) found decreased levels of zinc and citrate. The citrate-level testing is more effective at detecting PCa than Prostate Specific Antigen (PSA) tests[56]. Additionally, the efficacy of citrate analysis in semen is equivalent to that of citrate analysis in prostatic secretion for detecting PCa[57,58].

Studies on metabolomics in cell lines can also be utilized to assess the changes brought on by pharmacological treatment. Changes in choline and energy metabolism seem to result from PCa cell therapy[59]. Due to the increased pyruvate uptake into mitochondria, pyruvate dehydrogenase kinase is inhibited by Dichloroacetate (DCA), which has the potential to reverse the Warburg effect. In contrast to poorly metastatic cells, which showed no changes in lactate/metabolite ratios following treatment, highly metastatic cells displayed significantly reduced levels of lactate/metabolite ratios [Lac/Cr, Lac/Cho, Lac/Al, and Lac/(Cho + Cr + Al)]. These results imply that cells with high levels of metastatic behavior are more dependent on lactate generation[60].

4. Concluding Remarks

A systemic approach, integrating multi-omics studies, is essential to understand cancer biology and investigate its molecular pathogenesis. Through multi-omics data analysis, some common molecular characteristics can be identified across multiple tumor types. This allows us to differentiate between patient subgroups and identify cancer subtypes based on their molecular characteristics. With multi-omics, we can systematically summarize biological interactions from an individual cell or tissue to a patient with a primary tumor and possible metastases, embracing different layers of quantitative information, which help clinicians, pharmaceutical companies, and researchers in the direction of future precision medicine (Figure 2). Moreover, such integration enables the identification of the molecular characteristics of tumors at various levels, from genes to proteins, as well as the different stages of cancer.

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