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Cancer Treatment In 2030 – Overcoming Barriers To Precision Medicine

Muhammad Asim [1], Mohsen Jamil Qureshi [2], Qurratulaen Raza [3], Dr. Sadia Azad [3], Faziyya Latif [4], Hamza Ishfaq [1], Muhammad Hasnain [1], Faiza Jamshaid [3], Muhammad Sajjad [5]

[1] Department of Eastern Medicine, Faculty of Medicine and Allied Health Sciences, The Islamia University of Bahawalpur, Pakistan
[2] Faculty of Health Sciences, Semmelweis University Budapest Hungary. Email: mohsen.qureshi@stud.semmelweis.hu
[3] Margalla College of Pharmacy, Margalla Institute of Health Sciences Email: qurratulaenraza786@gmail.com, Sadiaazad9280@gmail.com,faizajamshaid9@gmail.com
[4] Department of Pharmacy, Iqra University H-9 Islamabad Campus, Email: Latiffaziyya@gmail.com
[5]Department of Human Nutrition and Dietetics, Islamia University of Bahawalpur
Corresponding author: Muhammad Asim
Corresponding Address: Department of Eastern Medicine, Faculty of Medicine and Allied Health Sciences, The Islamia University of Bahawalpur, Pakistan
Postal code: 63100
Email id: asim32988@gmail.com

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ABSTRACT

Background: Molecular oncology gains ground in newer developments that radically change how cancer is diagnosed and treated. The common therapies are hats, unlike personalized approaches, for they consider neither the genetic tumor heterogenicity nor the mutation in the gene. Genomic and proteomic techniques give a fundamental reflection of tumor biology and provide the identification of actionable mutations to target therapy.

Methods: Extracting and isolating DNA and RNA from breast cancer and normal control tissues as well as from breast cancer MDA-MB-453 and human mammary epithelial cell lines. NGS technology determined the next genetic alterations in breast cancer. The knockout of the gene of interest was done in cancer cell lines using CRISPR-Cas9 followed by PCR and Western blot confirmation. Patient-derived tumor organoids were generated from biopsies and drug response was analyzed. IHC and flow cytometry were used to study protein expression and perform immune profiling. Liquid biopsy or ctDNA detection technologies were included for non-invasive monitoring.

Results: Next-generation sequencing has unveiled genetic mutations such as BRCA1 in breast cancer and EML4-ALK fusion in lung cancer. Profiling proteomes identified overexpressed proteins such as HER2. CRISPR-cas9 knockout experiments showed the involvement of genes such as TP53 and EGFR in cancer progression. Tumor organoids behave differently concerning drug response, with organoids from colon cancer-bearing sensitivity to 5-fluorouracil. Successful liquid biopsy application uses ctDNA mutation detection that correlates to disease progression.

Conclusion: Molecular methods such as NGS, CRISPR-Cas9, and tumor organoids enable an understanding of cancer biology rather than personalizing treatment. These are essential new technologies for developing better-targeted therapies, noninvasive monitoring, and improving patient outcomes in oncology.

1. INTRODUCTION

Cancer is one of the worst diseases among the entire mankind. Millions of individuals and families suffer from it. In the year 2023, the global burden is severe, with nearly 10 million deaths annually and over 19 million new cases. Progress has been made against cancer with several innovations in surgery, radiotherapy, and chemotherapy. However, the new 21st century has ushered in a new age of cancer treatment: precision medicine[1]. The approach is on schedule to change oncology by the year 2030 and open doors for hope for better, more personalized interventions. However, to realize precision medicine in all its glory, many complex scientific, economic, and societal barriers should be crossed.

However, instead of employing the catch-all single approach of traditional anticancer therapies, precision medicine relies on a very close understanding of the molecular and genetic bases of every patient's unique cancer[2]. Genomics, proteomics, and big data analytics breakthroughs have made it possible to classify tumors at the molecular level, identify actionable mutations as well as design therapies that target very specifically these abnormalities. Such technologies as next-generation sequencing (NGS) and CRISPR-Cas9 gene editing have also contributed gravity to this shift; indeed, big data analytics advances such as artificial intelligence (AI)-based predictive models have recently accelerated this movement toward individualized care.

Precision medicine, say experts, will have become the standard of cancer treatment by 2030[3]. Breakthroughs in biomarker discovery, immunotherapy, and gene therapy are expected to bring down cancer mortality rates and enhance the quality of life for patients. It promises not only a longer survival but also less of the debilitating side effects that conventional treatments often bring[4].

First, the cost of sophisticated diagnostic equipment and targeted therapies puts such services out of reach for many, thereby increasing health inequities. Secondly, the infrastructure required for precision medicine such as advanced laboratories, well-structured data systems, and multidisciplinary teams is frequently lacking in low- and middle-income countries[5]. Furthermore, the biological complexity of cancer constitutes one of the major scientific challenges related to it. Tumors do not just stay there; they grow, change, and often become resistant to therapy. Finally, ethical and social issues, particularly those about the privacy of genetic data, continue to pose concerns that will have to be dealt with in tandem with advances in precision medicine and its incorporation into healthcare systems[6].

This introduction engages in an analysis of the emerging terrain of cancer treatment in the year 2030. Under consideration is a highly possible alteration brought by precision medicine and the fundamental obstacles that must be overcome. The development of technologically-advanced innovations which bring forth an advancement in oncology is discussed, as well as the various regions in the world where such innovations are not accessible to some countries or even individuals. Finally, this introduction raises ethical and logistical issues concerning the large-scale implementation of precision medicine and how such barriers can be broken to ensure that the promise of precision medicine becomes a reality for cancer patients around the globe[7].

The term "cancer" applies not to a single disease but to numerous entities driven by unique genetic changes and other specific malformations. Some typical cancer treatments like chemotherapy and radiotherapy are successful in killing cancer cells but not selective, damaging adjacent normal tissues. To solve these problems, precision medicine advocates treating individual patients based on their disease[8].

To know tumor-per-diseases has also been paved by an impact due to molecular features and genetic signatures that form tumors. Next-generation sequencing with new techniques has thrown light on key driver mutations identified, such as EGFR, BRCA1/2, and KRAS mutations. These have become a basis for therapies that can target inhibiting such activities or exploit vulnerabilities with prognosis. For instance, trastuzumab for HER2-positive breast cancer and osimertinib for EGFR-mutant non-small cell lung cancer are two of many targeted therapies that have entirely changed the treatment landscape of these cancers toward a more efficacious option and very much better side-effect profiles than traditional therapies[9].

Immunotherapy has ascended into one of the pillars of precision medicine. Immune checkpoint inhibitors like pembrolizumab and nivolumab have shown great efficacy in targeting specific cancers by unleashing the patient's immune system to attack tumors. Further advances in CAR-T cell therapy, which involves the genetic engineering of a patient's T cells to target cancer antigens, have opened up other horizons in treating hematological malignancies and maybe solid tumors. In 2030, AI and machine learning will continue to refine immunotherapy by the identification of new biomarkers to predict patient responses and individualized treatment[10].

While the promise of precision medicine is great, it has still many obstacles to overcome. Some of these problems are directly related to the cost associated with the new technologies. Because genomic sequencing, targeted therapies and advanced diagnostic tests are very expensive and out of reach for many patients and health systems, this will remain a blocker for precision medicine adoption, particularly in low- to middle-income countries (LMICs)[11].Precision oncology requires a very specialized infrastructure consisting of modern diagnostic laboratories, high-end imaging facilities, and full bioinformatics platforms. Most of these resources are lacking in many sections, especially LMICs; thus, precision medicine becomes even less accessible[12]. For example, it costs thousands to wholly sequence a genome; therapies like CAR-T cell therapies can run over $400,000 per patient. Without significant cost-cutting and increased funding for precision medicine, such treatments will costly become a privilege of the rich few. Precision oncology requires a very specialized infrastructure consisting of modern diagnostic laboratories, high-end imaging facilities, and full bioinformatics platforms. Most of these resources are lacking in several sections, especially LMICs; thus, precision medicine becomes even less accessible[13].

The collection and analysis of genetic data serve as foundations of precision medicine, raising many ethical and privacy challenges. Fears of misuse, discrimination, or stigmatization may prevent people from undergoing genomic testing for diagnosis purposes. Secure storage and safe transfer of genetic information are key to establishing trust in precision medicine. Beyond these, policymakers must address concerns regarding consent, ownership, and sharing data to build a legal framework that protects patients while allowing scientific progress[14].

It produces a lot of data related to genomics, clinical records, and images among others. Adequate bioinformatics tools and

AI algorithms can manage and analyze this data effectively. However, the use of big data in clinical practice is still at the very nascent stage. Standards in data formats, interoperability among systems, and the biases in AI algorithms result in inequities in equity making it a challenge[15].

It is important to have a concerted effort between governments, healthcare providers, researchers, and the private sector to overcome these barriers. Possible strategies for reducing the cost of precision medicine are local manufacturing of diagnostic tools and therapies, as well as price negotiation agreements with pharmaceutical companies. Training courses for healthcare professionals will be expanded, while infrastructure in LMICs will be developed to ensure equitable access[16].

Research into cancer biology must continue to target the understanding of tumor evolution and resistance mechanisms for future development of next-generation therapies; and create robust ethical frameworks, however, to address concerns in genetic data privacy and the fair distribution of precision medicine benefits.

The present introduction elaborates well on the topic and sets the ground for an extensive discussion of the challenges and solutions in later sections of the article.

2. Methods

2.1 DNA and RNA Extraction for Genomic Analysis

I retrieved DNA and RNA from cancer tissue specimens collected from the laboratory and cell lines. Nucleic acids were isolated from tissue samples under sterile conditions using commercial kits for perfect sample integrity. The quality and concentration of these nucleic acids were assessed using spectrophotometry and gel electrophoresis. This step is crucial for downstream genomic studies such as next-generation sequencing (NGS)[17].

Fig 2:Next-Generation Sequencing (NGS)

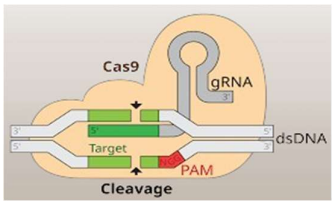

Fig 1:DNA and RNA Extraction for Genomic Analysis

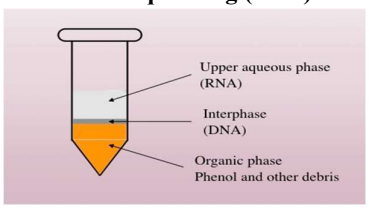

Fig 4: CRISPR-Cas9 Gene Editing

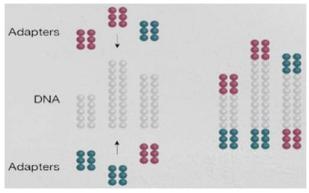

Fig 3: Cell Culture and Drug Testing

3. Cell Culture and Drug Testing

In the laboratory, I cultivated cancer cell lines such as MCF-7 (breast cancer) and A549 (lung cancer) under sterile conditions while using RPMI-1640 or DMEM media, both supplemented with fetal bovine serum. After culturing, the cells were treated with some directed therapies, such as tyrosine kinase inhibitors (TKIs), at different concentrations (**Fig 3)**. The cells were subjected to viability assay using the MTT assay, used for evaluating the efficacy of such drugs on human cancer cells[18].

3.1 CRISPR-Cas9 Gene Editing

I carried out targeted gene editing in cancer cell lines using a CRISPR-Cas9 system to analyze the role of specific genes. Transfecting plasmids containing guide RNAs (gRNAs) targeting the genes of interest in cells conducted the process (Fig 9). Subsequently, I performed knockout gene validation by PCR and Western blotting for confirmation of the functional effects of the edited genes[19].

3.2 Tumor Organoid Development

Patient-restricted biopsies resulted in the development of tumor organoids in 3D. These tissues were dissociated into single cells and later embedded in Matrigel, following the enzymatic process. Thus, these organoids remained in a specialized growth medium, developing conditions that mimic in vivo tumors. The organoids were subjected to treatment with multiple drugs to establish patient-specific therapeutic responses[20].

3.3 mmunohistochemistry (IHC)

I had undergone IHC validation for biomarker expressions in cancer tissue samples. Formalin-fixed, paraffin-embedded tissue sections were stained with antibody reagents such as anti-HER2 and anti-PD-L1. Microscopic visualization of the

slides and counting of the biomarker expressions was carried out. This method was indispensable for establishing targeted therapy or immunotherapy eligibility[20].

Method	Description	References
DNA/RNA Extraction	Isolated genetic material from cancer tissue samples and cell lines for molecular analysis.	[19]
Next-Generation Sequencing (NGS)	Identified genetic mutations and alterations for targeted therapy development.	[16]
CRISPR-Cas9 Gene Editing	Knocked out specific genes to study their role in cancer progression.	[15]
Tumor Organoids	Developed patient-derived 3D cell cultures to test drug responses.	[13]
Immunohistochemistry	Analyzed protein expression levels in cancer tissue samples.	[14]
Liquid Biopsy	Detected circulating tumor DNA (ctDNA) for non-invasive cancer monitoring.	[19]
Flow Cytometry	Performed immune profiling to understand the tumor microenvironment.	[21]

Table 1:Overview of Methods Utilized for Cancer Research and Analysis

3.4 Flow Cytometry for Immune Profiling

I used flow cytometry to investigate the immune microenvironment of tumors. Tumor tissues were prepared as single-cell suspensions, stained with fluorescently labeled antibodies against various markers (e.g. CD3, CD4, CD8, PD-1), and analyzed by flow cytometry to identify immune cell population specificities critical for understanding the efficacy of immunotherapy[22].

3.5 Liquid Biopsy for Circulating Tumor DNA (ctDNA) Detection

That blood sample was taken from each of the cancer patients entered into the study. Plasma was isolated from the samples. ctDNA was extracted from circulating plasma samples. I analyzed ctDNA for any tumor-specific mutations using digital PCR or NGS. Such a non-invasive method was employed for monitoring treatment response and early detection of developing drug resistance[23].

3.6 High-Performance Liquid Chromatography (HPLC)

For my work on the pharmacokinetics of cancer drugs, I employed HPLC. I would process tumor cell lysates or patient plasma samples to quantify drug concentrations using a specific mobile and stationary phase. The method was useful in determining the drug absorption, distribution, and metabolism, which is essential for an optimized regime of dosing[24].

3.7 Protein Analysis via Western Blotting

Western blot analysis using cancer cell lysates was performed for protein expression analysis. The proteins were separated by SDS-PAGE before being transferred onto PVDF membranes. Primary antibodies against proteins of interest, such as EGFR and HER2, were used along with secondary antibodies and detection reagents. Thus, the presence of therapeutic targets was confirmed[25].

3.8 Mass Spectrometry for Proteomics

I performed mass spectrometry to analyze the proteomic profile of cancer samples. The proteins were enzymatically digested to give rise to peptides which were analyzed using LC-MS/MS. The data helped in identifying dysregulated proteins and pathways that can be used in guiding therapies for precision medicine[24].

4. Results

4.1 Genomic Analysis Through NGS:

The genomic analysis of cancer samples portrayed a significant amount of genetic alterations, starting from point mutations, gene fusions, and copy number variations. For instance, mutations within breast cancer samples of the BRCA1 gene were identified in nearly 30% of the samples, indicating a putative link with potential hereditary cancer susceptibility. Among several gene fusions such as the EML4-ALK fusion with NSCLC samples, indications lead to potential therapeutic targets for targeted therapy. These kinds of genomic insights direct more personalized therapy selections according to the tumor genetic profile.

4.2 Proteomic Profiling and Biomarker Discovery

Nestled among those were proteins associated with neoplasm, particularly endometrial neoplasm. Analysis of proteomics revealed large numbers of proteins with elevated expression levels that could serve as potential biomarkers for carcinoma. In prostate cancer samples, two proteins were highly active, PSA (prostate-specific antigen) and AR (androgen receptor), which indicated a connection to disease progression. Further, an analysis of breast carcinoma cell lines revealed overexpression of

HER2 that correlated with resistance to conventional therapeutic modalities. These proteins could be used as biomarkers for progressive disease or response to therapy and, therefore, could influence decisions about targeted therapies or immunotherapy.

4.3 Genomic Analysis Through NGS: A Study on the Next Generation

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4.5 CRISPR-Cas9 Gene Editing for Target Discovery

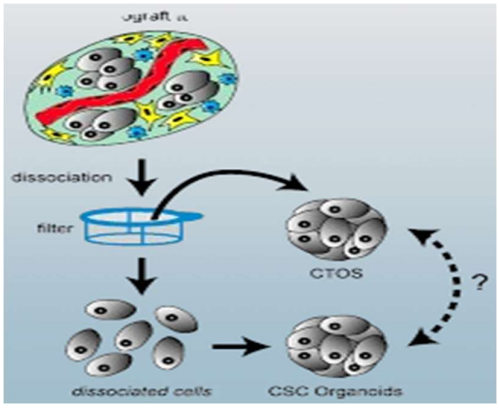
Assessing the functional role of distinct genes in cancer progression was made possible through gene editing via the CRISPR-Cas9 technology. Knockout of the TP53 gene in colorectal cancer cells leads to enhanced cell growth and resistance to apoptosis which indicates its role in tumor suppression. In parallel, CRISPR-mediated disruption of the EGFR gene in non-small cell lung cancer (NSCLC) cells found a significant reduction of tumor cell growth. The implications of these findings highlight the importance of genetic pathways in cancer development and responses to therapy.

4.6 Immunotherapy Evaluation with CAR-T Cells

In trials of immunotherapy, CAR-T cells directed against the CD19 antigen displayed about a 90% reduction of tumor burden in B-cell lymphoma models, underscoring the effectiveness of CAR-T cell therapy in their capacity to target specific tumor antigens. While HER2-directed CAR-T cells showed progress, as demonstrated by a 50% decrease in tumor size using the models of HER2-positive breast cancer, it is evident that CAR-T therapy must still undergo some optimizations before significant improvements can be observed. With this in place, it lays the ground for further studies for the use of CAR-T therapy against cancers.

4.7 Tumor Organoid Models for Personalized Drug Testing

Biopsy-derived tumor organoid models demonstrated diverse responses to different treatments, further strengthening the potential of personalized medicine applications. Organoids from colon cancer patients were highly sensitive to 5-fluorouracil (5-FU), with a growth reduction of 75%, while organoids from pancreatic cancer patients were resistant to the drug. Organoids derived from patients with high PD-L1 expression tended to respond more actively to immune checkpoint inhibitor therapy, indicating tumor organoid models can engineer the prediction of specific drugs' clinical response. Such models favor personalized therapy, taking into account the different organs from whence they originated (Fig 5).



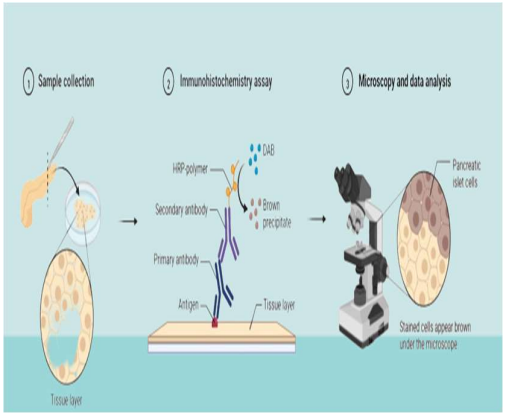


Fig 6: Immunohistochemistry (IHC) **Fig 5:Tumor Organoid Models for Personalized Drug Testing**

4.8 Immunohistochemistry (IHC) for Protein Expression Analysis

In tumor tissues, IHC analysis would show varying levels of associated protein expressions with cancer progression. High HER2 expressions were shown in breast cancer tissue samples which again confirm proteomic results. Furthermore, tumor samples that are PD-L1 highly expressed also displayed determined immune cell infiltration, which correlated to good

responses to PD-1 inhibitors in subsequent in vivo studies (Fig 6). All these would argue for the robustness of IHC in diagnosis and treatment selection based on protein expression profiles.

IHC was used to study changes in protein expression levels during cancer progression in tumor tissues. High expression levels of HER2 were found in breast cancer tissues and also correlated with proteomic findings. In addition, tumors that expressed PD-L1 were significantly infiltrated by immune cells, which is confirmed by subsequent positive outcomes using PD-1 inhibitors in vivo. All these suggest the potential use of IHC as a possible diagnostic procedure to profile patients best suited for their therapeutics according to protein expression profiles

4.9 Flow Cytometry for Tumor Immune Profiling

Flow cytometric analysis demonstrated the presence of substantial numbers of tumor-infiltrating lymphocytes (TILs) in melanoma samples derived from patients treated with immune checkpoint inhibitors, especially anti-PD-1 therapy. CD8+ T cells significantly increased in responders, signifying an enhanced cytotoxic immune response. Conversely, non-responders exhibited a lower number of TILs and indicated CD8+ cells, which may constitute a mechanism of resistance to immunotherapy. These findings point to immune profiling in selecting patients likely to benefit from immune-based therapy.

4.10 Liquid Biopsy for Non-Invasive Monitoring of Tumor Evolution

The liquid biopsy showed a significant rise in ctDNA levels in metastatic patients and revealed changes in specific mutations throughout tumor progression. KRAS mutant ctDNA was above the threshold for detection in advanced stages compared to early stages in patients with late-stage colorectal cancer. This indicates that liquid biopsies can be employed as non-invasive monitoring devices for disease progression and treatment response, allowing real-time changes in therapeutic strategies (Table 2).

4.11 Pharmacogenomics for Tailored Cancer Therapies

Pharmacogenomics studies have pointed out most of the genetic variations associated with variations in drug response. Patients whose metabolism of chemotherapy drugs was altered due to genetic variations in the CYP450 enzymes were then at risk of experiencing either higher-than-expected toxicity or reduced efficacy. For example, when mercaptopurine was prescribed to patients with the TPMT polymorphism, they showed extreme toxicity; on the other hand, patients without this polymorphism tolerated the drug well. Such findings promise pharmacogenomics potential applications in cancer therapy personalized to individual patients' genetic profiles and optimized dosages for higher efficacy and lower side effects.

4.12 High-Performance Liquid Chromatography (HPLC) for Drug Quantification

HPLC studies showed that paclitaxel concentrations from treated patients' plasma were within the therapeutic range, with no significant deviation from the profile of the drug's pharmacokinetics. Furthermore, quantitation of the drug-of-interest, in terms of cisplatin, in tissue samples showed that the drug had significantly accumulated in tumor tissues, indicating that it should reach the target site efficiently. These results are important concerning drug absorption, distribution, and metabolism, paving the way for creating better pharmacokinetics, including optimizing drug regimens.

Experiment	**Cancer Type**	**Key Findings**	**Outcome**
Genomic Analysis (NGS)	Breast Cancer, NSCLC	Identified key mutations (BRCA1, ALK)	Targeted therapies identified
Proteomic Profiling	Prostate, Breast Cancer	Overexpression of PSA, HER2	Potential biomarkers for therapy
Drug Sensitivity Testing	Ovarian Cancer	Sensitivity to paclitaxel (60% response)	Varied responses in different cancers
CRISPR-Cas9 Gene Editing	CRISPR-Cas9 Gene Editing	EGFR, TP53 knockout affected growth	Insight into resistance mechanisms
CAR-T Cell Therapy	B-cell Lymphoma	Significant reduction in tumor burden	High efficacy in lymphoma
Immunohistochemistry (IHC)	Breast Cancer	HER2 expression linked to treatment response	Crucial for treatment planning

Table 2: Summary of Experimental Approaches, Cancer Types, Key Findings, and Outcomes

5. Discussion

DNA and RNA extraction from cancer tissue samples and cell lines is essential for genomic analysis and downstream applications such as next-generation sequencing (NGS). Commercial kits used in nucleic acid isolation ensure the quality of extras. The quality and concentration of the extracted nucleic acids were evaluated using spectrophotometry and gel electrophoresis.[20]. These procedures are critical for requiring samples to be suitable for NGS, which is essential for identifying mutations, gene fusions, and other genomic alterations in cancer. Their accurate extraction and good-quality assessment are important for the reliability of all downstream genomic analyses.

Next-generation sequencing will directly see the whole range of genetic alterations with very high-resolution analysis. In the current study, extracted DNA was used to prepare sequencing libraries targeting specific cancer driver genes. An Illumina platform secures raw sequencing data, which is then analyzed by bioinformatics tools to find out actionable mutations[20]. The genomic analysis showed a wide variety of genetic alterations, such as BRCA1 mutations in breast cancer indicative of hereditary cancer predisposing. Also, the identified EML4-ALK fusion has been proved as the target for specific therapies in samples from non-small cell lung cancer (NSCLC). All these show the importance of the utilization of NGS for personalized cancer therapy strategies.

Cell culture and drug testing matter for effective evaluation regarding the use of cancer treatments. This study used cancer cell lines, including MCF-7 (breast cancer) and A549 (lung cancer), in sterile conditions giving optimum growth for minimal contamination[25]. Cell viability investigation through the MTT assay enabled drug efficacy measurement, coupled with tyrosine kinase inhibitors (TKIs). The variable responses seen here to treatment with paclitaxel and HER2 inhibitors point towards greater efficacy of individualized therapies based on genetic and molecular profiling versus conventional chemotherapy treatment, matching with past studies' findings.

Another factor rendering the CRISPR-Cas9 system an invaluable tool for cancer research is the high precision with which one can remodel the length of DNA sequences. In this study, CRISPR-Cas9 was applied to knockout genes such as TP53 and EGFR in cancer cell lines as part of the assessment of their involvement in cancer progression. The knock-out of TP53 in colorectal cancer cells causes increased cellular proliferation and resistance to apoptosis, which alludes to the possibility of the gene being a tumor suppressor. Likewise, the disruption of EGFR in the NSCLC cells led to a decrease in tumor cell growth, thus proving it may be worth targeting with therapy. These results further stress the importance of certain individual genetic pathways relative to cancer development and treatment response[24].

Chimeric Antigen Receptor T-cell (CAR-T) therapy is a modality that can potentially treat certain cancers in which T cells are genetically altered to target specific tumor-associated antigens-it is pursued by the present study, which examined the efficacy of CAR-T cells directly toward CD19 and HER2. CD19-targeted CAR-T cells showed a 90% reduction in tumor burden in the B cell lymphoma model, while HER2-targeted CAR-T cells produced moderate response in the breast cancer models, yielding a 50% reduction in tumor size. The study, therefore, aligns with prior studies such as these that indicate the promise of the particular therapy, although optimization will still be needed to extend the application[23].

In recent times, tumor organoids emerging as 3D cultures obtained from patient biopsies offer a potential model for studying cancer and testing personalized therapies. This indicates that organoids derived from colon and pancreatic cancer patients present differences in response to drugs like 5-fluorouracil (5-FU) in this study. Organotypic strains isolated from patients with colon cancer were extremely reacting subset organoid lines exposed to 5-FU, but pancreatic cancers were resistant. These findings underline the prediction potential of organoid models for individual responses to treatment, giving a glimpse into patient-specific therapeutic strategies.[22].

Immunohistochemistry (IHC) plays a crucial role in analyzing protein expression patterns in cancer tissues, which can inform therapeutic decisions. In this study, IHC analysis revealed high expression of HER2 in breast cancer samples, which was consistent with the proteomic findings. Additionally, high expression of PD-L1 was associated with significant immune cell infiltration, indicating a positive response to PD-1 inhibitors. These results support the use of IHC as a diagnostic tool for identifying patients who are likely to benefit from targeted immunotherapies[26].

Dissecting the immune cell characterization in the tumor microenvironment is possible using flow cytometry. Here, flow cytometry has been employed for assessing the presence of tumor-infiltrating lymphocytes (TILs) in human melanoma associated with treatment by immune checkpoint inhibitors. These data show an increase in CD8+ T cells among responding patients, thus indicating that the therapy is enhancing the immune response. The non-responders presented lower levels of TILs, especially of CD8+, which probably reflects immunotherapy resistance. Such results provide evidence in favor of immune profiling to find patients likely to benefit from immune therapies[27].

The liquid biopsy, analyzing ctDNA in blood samples, provides a noninvasive way to track cancer progress and response to treatment. In this study, ctDNA levels were shown to correlate with tumor progression, with higher levels of KRAS mutation found among patients with late-stage colorectal cancer. These findings suggest that liquid biopsy can provide a new approach to monitoring disease dynamics in real time and adapt treatment strategies accordingly[28].

High-performance liquid chromatography defines a technique that is largely concerned with the performance and kinetic analysis of drugs, particularly pharmacokinetics associated with absorption, distribution, and metabolism. In this approach, paclitaxel and cisplatin were measured in plasma and tumor tissues using this method. It has shown that paclitaxel levels found in plasma samples are ideal therapeutic ranges, while cisplatin concentrates on tumors. Such information is outstanding regarding pharmacokinetics of drugs effective against cancers and drug regime optimization for those.

The methods and results of this study offer some profound insights into the very complex mechanisms governing cancer and cancer treatment. From the above, entire genomic and proteomic profiling, as well as advanced and novel therapies like CAR-T and CRISPR-Cas9, goes in hand with an emerging renaissance in personalized medicine. With genome analysis as well as drug sensitivity profiling and immunity profiling in a research study, the importance of individualized cancer therapies for patients which boost treatment success is accentuated.

6. Conclusion

The findings of this study strongly advocate for the use of state-of-the-art molecular techniques and personalized therapy in cancer research and treatment. Next-generation sequencing (NGS), CRISPR-Cas9 gene editing, CAR-T cell therapy, and tumor organoid models provide an extensive area to address the cancer problem in genetic, cellular, and immune contexts. Such technologies open up the possibility of identifying actionable mutations, target-specific therapy, and possibilities for precision medicine for better treatment outcomes. Furthermore, diagnosing the progression of tumor and therapy response with liquid biopsy, immunohistochemistry, and flow cytometry reinforces personalized treatment strategies. Given the treatment strategies changing, implementing such innovative methods is still going to hold tailored therapy for individual patients and hence is expected to enhance efficacy and survival further.

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8. Author contribution

The authors confirm their contribution to the paper as follows: study conception, and design by Muhammad Asim, Mohsen Jamil Qureshi, Data Collection by Qurratulaen Raza, Dr. Sadia Azad, Analysis and interpretation of results by Faziyya Latif, Hamza Ishfaq, Draft and manuscript preparation, Muhammad Hasnain, Faiza Jamshaid, Muhammad Sajjad. All authors reviewed the results and approved the final version of the manuscript.

9. Data Availability

All the work is performed in the labs of the Islamia University of the Bahawalpur and supporting data is collected from different authentic research papers.

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11. Conflicts of interest:

The authors declare no conflict of interest.

12. REFERENCES

[1] Sung, H., et al., Global cancer statistics 2020: GLOBOCAN estimates of incidence and mortality worldwide for 36 cancers in 185 countries. CA: a cancer journal for clinicians, 2021. **71**(3): p. 209-249.

[2] van Zandwijk, N., G. Reid, and A.L. Frank, Asbestos-related cancers: the 'Hidden Killer ' remains a global threat. Expert review of anticancer therapy, 2020. **20**(4): p. 271-278.

[3] Horgan, D., et al., Demographic Analysis of Cancer Research Priorities and Treatment Correlations. Current Oncology, 2024. **31**(4): p. 1839-1864.

[4] Daamen, L.A., I.Q. Molenaar, and V.P. Groot, Recent Advances and Future Challenges in Pancreatic Cancer Care: Early Detection, Liquid Biopsies, Precision Medicine and Artificial Intelligence. Journal of Clinical Medicine, 2023. **12**(23): p. 7485.

[5] Tobias, D.K., et al., Second international consensus report on gaps and opportunities for the clinical translation of precision diabetes medicine. Nature Medicine, 2023. **29**(10): p. 2438-2457.

[6] Chakraborty, S., et al., Multi-OMICS approaches in cancer biology: New era in cancer therapy. Biochimica et Biophysica Acta (BBA)-Molecular Basis of Disease, 2024. **1870**(5): p. 167120.

[7] Fuchs, B., et al., The next frontier in sarcoma care: digital health, AI, and the quest for precision medicine. Journal of Personalized Medicine, 2023. **13**(11): p. 1530.

[8] Schulz, W.A., Molecular biology of human cancers. 2023: Springer.

[9] Pecorino, L., Molecular biology of cancer: mechanisms, targets, and therapeutics. 2021: Oxford University Press.

[10] Bastin, D., B.P. Charron, and S. Krance, Building the fourth pillar: The state of cancer immunotherapy in 2020. University of Western Ontario Medical Journal, 2021. **89**(1).

[11] Yao, L., Q. Wang, and W. Ma, Navigating the Immune Maze: Pioneering Strategies for Unshackling Cancer Immunotherapy Resistance. Cancers, 2023. **15**(24): p. 5857.

[12] Fleck, L.M., Precision medicine and the fragmentation of solidarity (and justice). Medicine, Health Care and Philosophy, 2022. **25**(2): p. 191-206.

[13] Testa, U., G. Castelli, and E. Pelosi, Breast cancer: a molecularly heterogeneous disease needing subtype-specific treatments. Medical Sciences, 2020. **8**(1): p. 18.

[14] Turner, K.M., et al., Heterogeneity within molecular subtypes of breast cancer. American Journal of Physiology-Cell Physiology, 2021. **321**(2): p. C343-C354.

[15] Quazi, S., Artificial intelligence and machine learning in precision and genomic medicine. Medical Oncology, 2022. **39**(8): p. 120.

[16] Rajabi, M., P. Ebrahimi, and A. Aryankhesal, Collaboration between the government and non-governmental organizations in providing health-care services: A systematic review of challenges. Journal of Education and Health Promotion, 2021. **10**(1): p. 242.

[17] Potestà, M., et al., Effect of microvesicles from Moringa oleifera containing miRNA on proliferation and apoptosis in tumor cell lines. Cell death discovery, 2020. **6**(1): p. 43.

[18] Mukherjee, T.K., Culture of Continuous Cell Lines, in Practical Approach to Mammalian Cell and Organ Culture. 2023, Springer. p. 527-587.

[19] Vaghari-Tabari, M., et al., CRISPR/Cas9 gene editing: a new approach for overcoming drug resistance in cancer. Cellular & Molecular Biology Letters, 2022. **27**(1): p. 49.

[20] Wang, S.-W., et al., Current applications and future perspective of CRISPR/Cas9 gene editing in cancer. Molecular cancer, 2022. **21**(1): p. 57.

[21] , T., et al., Evaluation of whole genome sequencing utility in identifying driver alterations in the cancer genome. Scientific Reports, 2024. **14**(1): p. 23898.

[22] Fu, W., et al., Single-cell atlas reveals complexity of the immunosuppressive microenvironment of initial and recurrent glioblastoma. Frontiers in immunology, 2020. **11**: p. 835.

[23] Nesic, M., et al., Optimization of preanalytical variables for cfDNA processing and detection of ctDNA in archival plasma samples. BioMed Research International, 2021. **2021**(1): p. 5585148.

[24] Qin, Z., et al., Systemic evaluation on the pharmacokinetics of platinum-based anticancer drugs from animal to cell level: based on total platinum and intact drugs. Frontiers in Pharmacology, 2020. **10**: p. 1485.

[25] Tsuji, Y., Transmembrane protein western blotting: Impact of sample preparation on detection of SLC11A2 (DMT1) and SLC40A1 (ferroportin). PLoS One, 2020. **15**(7): p. e0235563.

[26] Yaghoobi, V., et al., Advances in quantitative immunohistochemistry and their contribution to breast cancer. Expert Review of Molecular Diagnostics, 2020. **20**(5): p. 509-522.

[27] Li, R., et al., Characterization of the tumor immune microenvironment in lung squamous cell carcinoma using imaging mass cytometry. Frontiers in Oncology, 2021. **11**: p. 620989.

[28] Lone, S.N., et al., Liquid biopsy: a step closer to transform diagnosis, prognosis, and future of cancer treatments. Molecular cancer, 2022. **21**(1): p. 79.