Advances in proteomics in diffuse large B-cell lymphoma (Review)

ZIHAN GUO*, CHENCHEN WANG*, XINYI SHI, ZIXUAN WANG, JINGYI TAO, JIAYING MA and LINTAO BI

Department of Hematology and Oncology, China-Japan Union Hospital of Jilin University, Changchun, Jilin 130033, P.R. China

Received October 21, 2023; Accepted April 17, 2024

DOI: 10.3892/or.2024.8746

Abstract. Diffuse large B-cell lymphoma (DLBCL) is the most common pathological type of non-Hodgkin's lymphoma. Although the development of monoclonal antibodies, small-molecule-targeted drugs and novel chemotherapeutic agents, and the increased use of immunotherapy have markedly improved the outcomes of DLBCL, ~40% of patients cannot be cured following the use of standardized first-line treatment. In addition, the specific mechanisms of drug resistance and potential factors associated with a poor prognosis in these patients remain unclear. Proteomics research is used to determine potential associations between changes in DLBCL protein expression levels and different stages of disease occurrence and development. Proteomics may aid in the identification of novel molecular mechanisms and drug resistance mechanisms, through identifying multiple associated proteins and monitoring changes in expression levels. Thus, proteomics research may exhibit potential in the development of therapeutic targets and in improving prognostic evaluation in patients with DLBCL. The present study aimed to review the use of proteomic methods for the investigation of DLBCL, including the mechanisms underlying disease progression and drug resistance in DLBCL, and the function of the tumor microenvironment in lymphoma growth. The present review also demonstrated the potential of proteomic-guided therapeutic strategies for DLBCL and discussed the synergistic benefits of using proteomic methods in DLBCL research.

Contents

Correspondence to: Dr Lintao Bi, Department of Hematology and Oncology, China-Japan Union Hospital of Jilin University, 126 Xiantai Street, Changchun, Jilin 130000, P.R. China
E-mail: bilt@jlu.edu.cn

*Contributed equally

Key words: diffuse large B-cell lymphoma, proteomics, diagnosis, treatment, prognosis, tumor microenvironment, drug resistance, multi-omics

1. Introduction

Lymphoma is a malignant tumor of the blood system (1). Lymphomas are classified as non-Hodgkin's lymphoma (NHL) and Hodgkin's lymphoma, according to their histological and cytological characteristics (2). Diffuse large B-cell lymphoma (DLBCL) is the most common form of NHL, accounting for 30-40% of all lymphomas. DLBCL is distinguished from other tumor types through high levels of heterogeneity, invasiveness, and diverse clinical, pathological and biological features (3). Notably, DLBCL develops as a result of the uncontrolled growth of malignant B cells, in the absence of external stimulation from the tumor microenvironment (TME) (4).

Proteins play key roles in numerous cellular activities and functions (5). Proteomics research focuses on the existence and activity patterns of all proteins in cells (6). Proteomic techniques are used to determine potential associations between changes in protein expression levels in DLBCL and disease progression (7); thus, offering a tool for DLBCL detection and management. Proteomics may also aid in identifying novel therapeutic targets and determining patient prognosis (8). Proteomics techniques include protein-separation technology based on two-dimensional gel electrophoresis, and protein identification using bioinformatics analysis and mass spectrometry (9). The strengths and limitations of proteomics techniques, including gel electrophoresis, mass spectrometry and microarray analysis are listed in Table I (10).

2. Proteomic analysis of DLBCL disease progression and drug resistance

Proteomic techniques are used to identify and quantify changes in proteins (11), and determine potential associations between changes in protein expression and different stages of lymphoma development (12). Thus, proteomics provide valuable insights into alterations in the levels of proteins (13) and protein-related signaling molecules (14).

Ednersson *et al* (15) examined protein expression in formalin-fixed, paraffin-embedded tumor samples using quantitative proteomics in 202 patients with DLBCL. A total of 6,430 proteins were successfully identified. Of these proteins, a subset of 498 proteins were significantly differentially expressed between germinal center B-cell-like (GCB) and non-GCB cells. Notably, these proteins included guanylate-binding protein 1 (GBP1), CD64, CD85A and interferon-inducible protein with tetrapeptide repeat 2 and mixed lineage kinase domain-like protein (MLKL). In addition, immunohistochemical staining revealed the upregulation of GBP1 and MLKL protein expression in patients with DLBCL. Results of a previous clinical study demonstrated that human immunodeficiency virus (HIV)-related lymphoma is aggressive, with an increased incidence of drug resistance and a poor prognosis. Zhuang *et al* (16) used proteomics to screen 84 proteins that were differentially expressed between patients with AIDS and AIDS-NHL. Enrichment analysis of the differentially expressed proteins using the Gene Ontology and Kyoto Encyclopedia of Genes and Genomes databases indicated that the majority of proteins were closely associated with essential biological functions, including the humoral immune response and complement system activation. Protein-protein interaction analysis revealed extensive interactions among the proteins, including β_2-microglobulin, cathepsin D and various complement subunits. Collectively, these results highlighted the molecular changes occurring in patients with AIDS-NHL compared with patients with HIV infection alone; thus, demonstrating the differing molecular pathogenesis of AIDS-NHL.

Lymphomas are malignant tumors in which lymphocytes in the human body undergo different stages of development and differentiation (17). Lymphomas exhibit high levels of heterogeneity and a complex pathological classification, with different treatment responses among different pathological types. In addition, treatment responses may differ between patients with the same pathological type. Numerous factors, including cell proteomics and molecular features may impact the prognosis of patients (18). In clinical practice, patients with DLBCL often develop drug resistance (19), which is a barrier to treatment within clinical practice. Liu *et al* (20) examined samples from 14 patients with untreated DLBCL using mass spectrometry and two-dimensional (2D) gel electrophoresis, and quantitatively identified differentially expressed proteins between patients who were susceptible to CHOP treatment and those who were resistant. This approach allowed the comprehensive characterization of the proteomic landscape associated with chemotherapy response in DLBCL; thus, providing valuable insights into potential biomarkers and therapeutic targets for improving treatment outcomes. Results of the previous study demonstrated that the protein expression levels of histone H2A.2, S100A9, Ezrin and Pleckstrin were significantly increased. In addition, the protein expression levels of 61 kD protein, collagen alpha 1 (VI), glutathione S-transferase pi-1 and heat shock protein beta 1 were significantly lower in patients who were susceptible to CHOP treatment, compared with those that were resistant. Analyzing the protein network associated with resistance to CHOP chemotherapy may aid in identifying patients with DLBCL with CHOP resistance; thus, providing a novel theoretical basis for the identification of therapeutic targets.

3. Proteomic analysis of the DLBCL TME

Tumors form dynamic, complex and heterogeneous environments with various cells and surrounding components, known as the TME (21). The heterogeneity of DLBCL is associated with the types of cells in the TME (22), including matrix components, dendritic cells, macrophages, monocytes, fibroblasts and T cells (23). Notably, the extracellular matrix interacts with lymphoma cells (24), and high numbers of M2 macrophages, natural killer cells and plasma cells are associated with lower survival rates in patients with DLBCL (25). The TME plays a significant role in the initiation, development and treatment resistance of DLBCL, and these factors ultimately impact the prognosis of patients (19). In total, ~75% of patients with DLBCL possess aberrations in genes associated with immune escape (26), and the TME includes numerous inhibitory immune detection points (27). Liu *et al* (28) suggested that adaptor-related protein complex 2 subunit mu1 subunit may contribute to the resistance of DLBCL to chemotherapy and targeted medications through controlling the TME. Notably, results of previous studies highlighted that multiple components in the TME may impact the occurrence and development of DLBCL. Spatial proteomics analysis may provide location information for cells in the tissue (29), and this method may be used to explore the interaction between DLBCL cells and the TME (30).

Through the transcriptome analysis of 4,655 DLBCL microenvironments, Kotlov *et al* (31) identified four main types of lymphoma microenvironments. The composition of the DLBCL microenvironment was investigated using proteomics analysis and the establishment of a patient-driven tumor xenograft model. Results of these studies indicated that novel therapeutic options for the treatment of DLBCL should target tumor cells with specific genotypes, and consider the impact of different microenvironment types on lymphoma progression. Bouwstra *et al* (32) also used proteomics analysis, and results of the previous study demonstrated that the poor prognosis of patients with non-GCB type DLBCL following R-CHOP treatment may be associated with the upregulation of CD47. These results highlighted the occurrence of different DLBCL microenvironments derived from different cell sources that were regulated by intracellular genes and signal transduction. Different microenvironments may lead to the breakdown of homeostasis and microenvironment alterations in the tissue, ultimately resulting in lymphoma progression. Bram *et al* (15) used quantitative proteomics analysis to demonstrate that multiple proteins are involved in the development of DLBCL, including the upregulation of proteins in Activated B cell-like (ABC) DLBCL. Results of a cluster analysis demonstrated that the most common clusters contained proteins involved in the control of the immune system and TME, including MLKL. These clusters also included several damage-related molecular pattern proteins, including S-100A8, S100A9, fibrinogen-α and particulate lysin. Xu-Monette *et al* (33) investigated potential associations between MYC/BCL2 and microenvironment biomarkers in DLBCL isoforms. The results of the previous study revealed that the genotype, TME and high MYC/BCI2 double expression all played independent and interdependent roles in predicting the prognosis of DLBCL. Feng *et al* (34) used proteomics analysis to examine exosomes in the serum

Table I. Strengths and limitations of three key proteomics techniques used in the analysis of diffuse large B-cell lymphoma.

	Two-dimensional gel electrophoresis	Mass spectrometric protein detection technology	Microarrays
Classification	Two-dimensional immobilized metal affinity electrophoresis, serological proteome analysis, three-dimensional blue native/IEF/SDS-PAGE, two-dimensional zymography (91)	Liquid chromatography-mass spectrometry, matrix-assisted laser desorption/ionization, surface-enhanced laser desorption, targeted/directed mass spectrometry (10)	Protein microarray, antibody/antigen microarrays, tissue microarrays, protein domain microarray (10)
Functions	Proteins are fractionated based on the molecular weight dimension and the first-dimensional isoelectric points. Separation divides the protein network into numerous protein spots	The mass-to-charge ratio of one or more proteins is calculated in samples (10). This technique is used for locating and measuring proteins in biological materials	This technique includes a collection of biomolecules that are placed on a solid support, and these are utilized to determine interaction partners via affinity
Strengths and limitation	This technique exhibits a high level of precision in differentiating intact proteins (91); however, it exhibits limited precision for highly intricate protein samples with a wide concentration dynamic range (92)	This is a high-throughput technique that measures hundreds of proteins at once (93). In addition, levels of sensitivity are high (94). However, low abundance protein detection does not exhibit high levels of reliability (95)	This method exhibits a high sensitivity in identifying weak interactions, and a high adaptability for proteins with a low abundance. This technique may not accurately represent the binding occurrences in the cellular milieu (96)

of patients with DLBCL, and the results demonstrated that chemotherapy-resistant DLBCL cells exhibited increased CA1 expression levels in exosomes, compared with chemotherapy-sensitive cells. In addition, results of the previous study demonstrated an association between the increased protein expression of CA1 in the TME and the prognosis of patients with DLBCL. Collectively, these results highlighted the potential of CA1 as a biomarker for assessing treatment efficacy and the prognosis of patients with DLBCL. In addition, further investigations are required to determine the specific role of the TME in the development, diagnosis, classification, treatment and prognosis of DLBCL.

4. Using proteomics to explore potential therapeutic options for the treatment of DLBCL

The discovery and application of anti-CD20 monoclonal antibodies in the early 20th century led to a new era of DLBCL treatment (35). At present, global treatment guidelines recommend first-line therapy with R-CHOP, comprising rituximab with cyclophosphamide, doxorubicin, vincristine and prednisone (36), leading to a cure in ~60% of patients (37). However, a small number of patients continue to exhibit refractory disease or relapse following complete remission (38), and traditional salvage immunochemotherapy combined with autologous hematopoietic stem cell transplantation only achieves a cure in ~10% of these patients (39). Thus, the remaining 90% of patients exhibit poor treatment outcomes (Fig. 1). Thus, improving the prognosis of these patients is complex (40), and further proteomic analyses are required to determine the signaling pathways associated with the onset and progression of DLBCL (41). Novel developments in proteomics technology have led to the discovery of multiple drug resistance mechanisms in lymphoma (42); thus, strategies and methods that eliminate the drug resistance of lymphoma cells and improve the therapeutic effects are also required (43).

Proteomics includes the identification of differentially expressed proteins in DLBCL tissues (44), obtaining 2D electrophoresis profiles (45), and the use of mass spectrometry to identify associated proteins (46) (Fig. 2). Bioinformatics analysis is also used to identify differentially expressed proteins for further validation at the tissue level, which may provide a theoretical basis for subsequent experiments (47). In addition, further investigations are required to determine the specific mechanisms of DLBCL resistance and verify the feasibility of differentially expressed proteins as drug-resistance-related targets (48). Chen *et al* (49) carried out mRNA/protein analysis of clinicopathological samples, and the results demonstrated that inhibitors of bromodomain and extraterminal (BET) protein inhibited the progression of DLBCL. BET inhibition led to upregulation of GTPase regulatory protein (IQGAP3), which inhibited RAS protein activity in DLBCL cells, indicating that patients with DLBCL with low IQGAP3 expression levels exhibited a poor prognosis. In addition, BET inhibitors

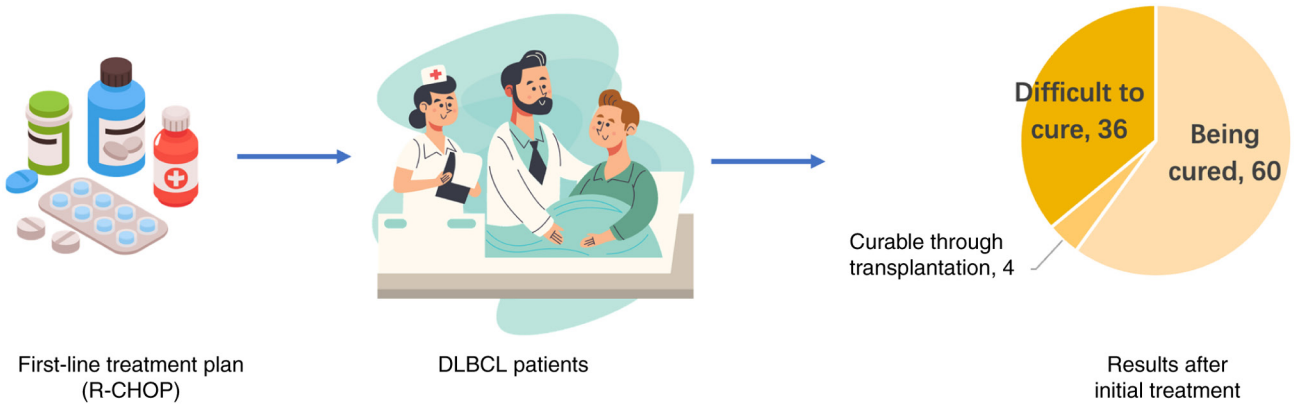

Figure 1. First-line treatment and prognosis of patients with DLBCL. First-line R-CHOP treatment leads to a cure in 60% of patients; however, the remaining 40% of patients develop relapsing or resistant disease, despite initial complete remission. Traditional salvage immunochemotherapy combined with autologous hematopoietic stem cell transplantation leads to a cure in ~10% of patients with refractory and relapsed disease; however, the remaining 90% of patients experience unfavorable treatment outcomes, highlighting the need for alternative therapeutic strategies to improve prognosis. DLBCL, diffuse large B-cell lymphoma.

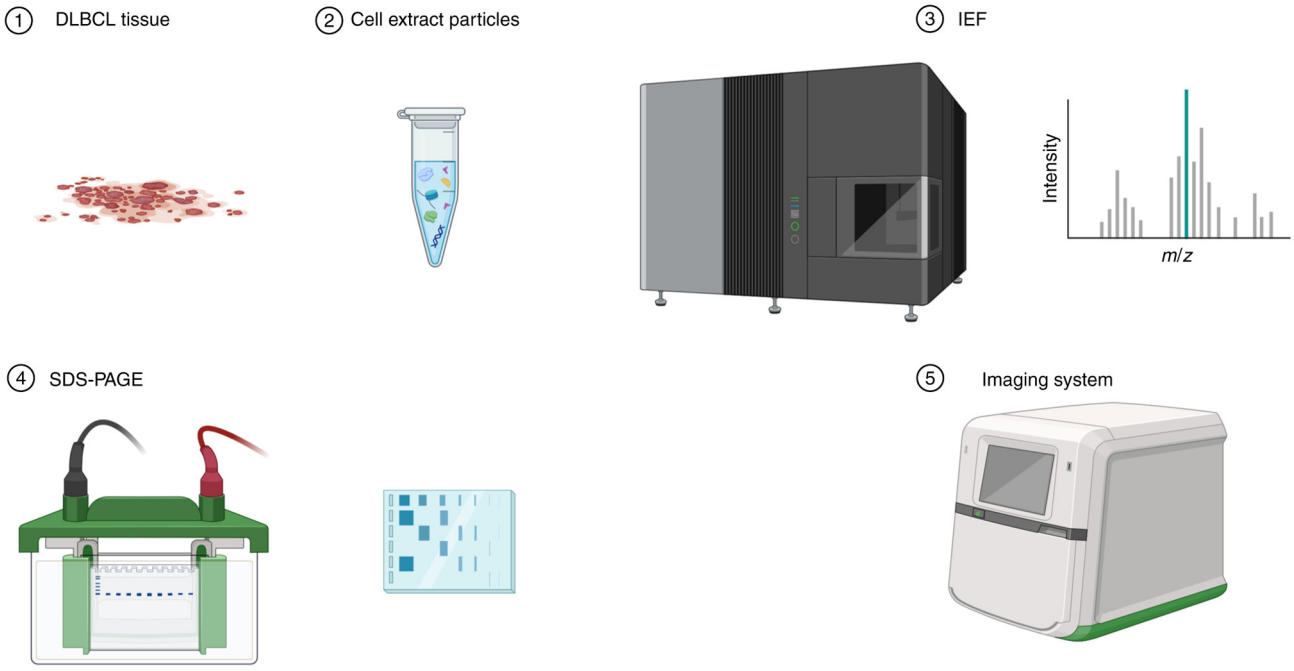

Figure 2. Experimental proteomics workflow using two-dimensional gel electrophoresis. Two-dimensional gel electrophoresis involves IEF and SDS-PAGE. Proteins are separated according to their isoelectric points using IEF, and subsequently separated according to their molecular sizes using SDS-PAGE. Using two-dimensional gel electrophoresis, high-resolution protein separation is achieved, and valuable information about protein abundance, post-translational modifications and protein isoforms is obtained. IEF and SDS-PAGE are widely used in proteomics studies to investigate complex protein interactions and identify potential biomarkers or protein targets in various biological samples. The process produces a two-dimensional protein map. IEF, isoelectric focusing.

effectively controlled the progression of DLBCL. Collectively, these results provided a theoretical basis for targeting the BET protein (50) as a potential treatment strategy for DLBCL.

Advances in proteomics-associated technologies have demonstrated that the emergence of DLBCL chemoresistance is closely associated with signaling pathways (51), including the PI3K/Akt pathway. Akt promotes cell survival and proliferation (52), as well as dysregulation of key effectors controlling cell metabolism (53). A proteomics analysis conducted by Xu *et al* (52) revealed that removal of the PI3K/Akt signaling pathway antagonist, PTEN, led to inactivation of the PI3K/Akt pathway in GCB DLBCL. Thus, PI3K/Akt activation may play a key role in the development of GCB DLBCL, and these findings demonstrate the potential value of PTEN as a therapeutic target. PTEN acts as a lipoprotein phosphatase, dephosphorylating the 3' position of phosphatidylinositol triphosphate, thereby reducing Akt activation (Fig. 3). Measurement of phosphorylated Akt levels indicated that PTEN expression was negatively correlated with PI3K/Akt activation in both a GCB DLBCL model and primary DLBCL samples (52). Bisserier and Wajapeyee (54) demonstrated that DLBCL cells resistant to Enhancer of Zeste Homolog 2 inhibitors exhibited activation of insulin-like growth factor I receptor, PI3K, and mitogen-activated protein kinase pathways. Feng *et al* (34)

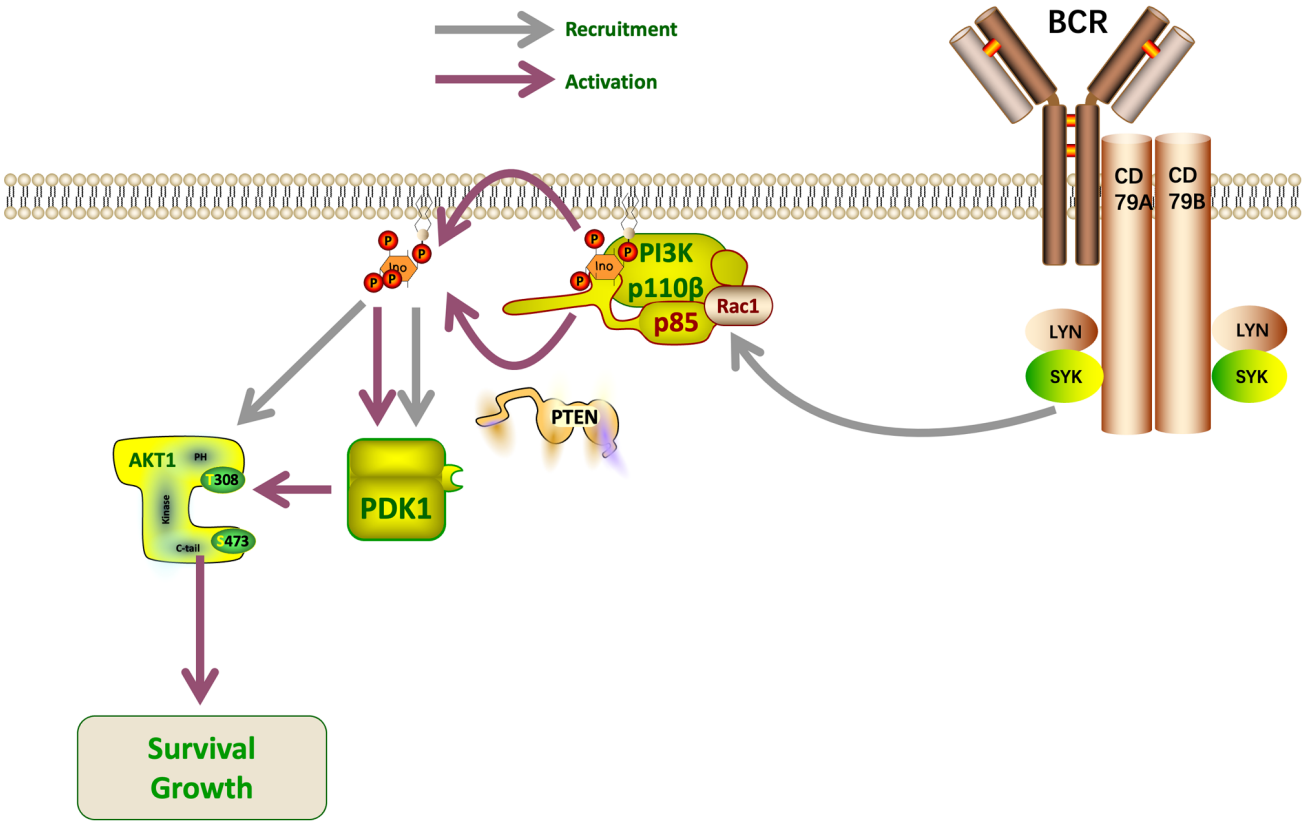

Figure 3. Schematic diagram demonstrating the mechanisms underlying the negative regulator PTEN. The PI3K/Akt pathway is an essential signaling mechanism controlling the onset and progression of DLBCL. PI3K activation transforms phosphatidylinositol bisphosphate into PIP3, which in turn recruits PDK1 and Akt to the cell membrane; thus, leading to PDK1-dependent activation of Akt. Akt promotes cell survival and proliferation. PTEN acts as a lipoprotein phosphatase, dephosphorylating the 3' position of PIP3, thereby reducing Akt activation. PIP3, phosphatidylinositol triphosphate; PDK1, pyruvate dehydrogenase kinase 1; DLBCL, diffuse large B-cell lymphoma.

used proteomics technology to demonstrate the increased expression levels of exocrine carbonic anhydrase (CA)1, and the role of this protein as a biomarker for the prognosis of DLBCL. Notably, CA1 expression levels were also associated with an increased resistance to chemotherapy via the signal transducer and activator of transcription 3 signaling pathways and nuclear factor-κB.

Collectively, these results indicated that proteomic techniques exhibit potential in the differential and enrichment analyses of DLBCL-associated proteins for the subsequent discovery of novel therapeutic targets. Specific signaling pathway inhibitors also exhibit potential in highlighting the molecular mechanisms underlying drug resistance; thus, leading to the development of novel therapeutic options.

5. Application of proteomics combined with other omics technologies in DLBCL

Proteomics technologies have improved the current understanding of the molecular changes associated with DLBCL (55,56). Storage of large amounts of proteomics data (57) is challenging; however, these often contain biologically significant results (7). Data storage may be aided through a combination of multiple omics techniques (58), and numerous biological techniques are used to examine lymphomas (59), including genomics (60), proteomics (61), epigenetics (62) and radiomics (63).

Fornecker *et al* (64) conducted a large-scale differential multi-group analysis of samples obtained from patients with DLBCL, with the main goal of identifying novel targets to overcome chemotherapeutic resistance and potential biomarkers for early recurrence risk. Through targeted RNA sequencing and non-labeled quantitative proteomics, results of the previous study revealed significant differences in the expression levels of 22 proteins and corresponding RNA between patients with typical DLBCL and patients with recurrent DLBCL. Notably, multiple key targets have successfully been identified using proteomics and transcriptomics techniques. Hexokinase 3 expression was significantly increased in patients with chemotherapeutic resistance, indicating that this protein may play a key role in the chemotherapeutic resistance of DLBCL. In addition, IDO1 is highly expressed in patients with chemotherapeutic resistance, and may exhibit potential as a novel immune checkpoint target. CXCL13 is overexpressed in patients with chemotherapeutic resistance and may play a crucial role in the microenvironment of DLBCL. The S100 protein is involved in regulating the proliferation, migration and invasion of cancer cells, and dysregulation of this protein is present in the majority of human cancers, such as breast, prostate, melanoma and colorectal (65). Results of a previous study demonstrated that the S100 protein may exhibit potential as a therapeutic target in R/R DLBCL. CD79B expression was significantly reduced at both protein and transcriptional levels; thus, a combination

of transcriptome and proteome techniques (66) may aid in processing large datasets (67).

Moreover, results of previous studies revealed a regulatory role of interleukin-1 receptor-associated kinase (IRAK4) in lymphoma cell proliferation and inflammation through proteome and phosphorylation modifications (68). A series of targeted degradation agents of IRAK4 were used to study the effects of impaired IRAK4 function on the phosphorylation levels of downstream signaling proteins, and the results demonstrated that IRAK4 only partially participated in the regulation of ABC DLBCL cell proliferation and inflammatory signals (69). The survival of ABC DLBCL cells was not solely dependent on the function of IRAK4; thus, highlighting a requirement for the development of other drug targets in ABC DLBCL (70).

A combination of protein genomics, and proteome, transcriptome and genome data (71) has demonstrated potential in the discovery of novel biomarkers (72) and drug targets (73). A previous study used protein genomics to analyze the N-glycoprotein spectrum of 13 subtypes of lymphoma, spanning 32 cell lines (74). Using unsupervised clustering analysis, results of the previous study revealed that the N-glycoprotein spectrum categorized these cell lines according to lineage and cell origin. These conformed to the subtypes identified by the World Health Organization, and demonstrated that the N-glycoprotein spectrum of clinicopathological lymphoma samples may correspond with traditional pathological classification, providing a key theoretical basis for the discovery of novel drug targets (74). A computational biology tool; namely, Drug Combo Explorer, was developed to identify lymphoma signaling pathways. This tool integrated numerous existing DLBCL pharmacogenomics and proteomics data to provide effective and synergistic drug combinations for the treatment of lymphoma (75).

The integration of multi-omics technologies exhibits potential in the treatment of DLBCL (76). Proteomics may also be used in conjunction with other omics techniques, such as transcriptomics, metabolomics and genomics, to further the current understanding of the molecular landscape and mechanisms underlying DLBCL (77). This integrative approach exhibits potential in the discovery of novel biomarkers, therapeutic targets and personalized treatment strategies for patients with DLBCL.

6. Conclusions

In conclusion, proteomics techniques are widely established in the study of DLBCL (78), and proteomics have been used in investigating the pathogenesis, drug resistance and mechanisms of lymphoma, the evaluation of prognosis, and guiding treatment plans (79). Further developments in proteomics-associated technologies are required for the identification of novel drugs and drug targets for the treatment of DLBCL (80). For example, Maurer *et al* (81) found that DLBCL patients with elevated serum free light chain (sFLC) had a relatively poor prognosis using FREELITE analysis. Then, Witzig *et al* (82) conducted a 6-year monitoring of FLC concentrations in patients with DLBCL and found that patients with DLBCL belonged to the FLC monoclonal and polyclonal groups; and the results revealed that elevated FLC was an adverse factor in the poor prognosis of DLBCL patients, and the aforementioned study provides new ideas for the treatment of DLBCL (7). Spatial proteomics, also known as spatiomics technology, is advancing (83). This technique is used to examine biological components, such as RNA and proteins, and adds 'location' dimensional information to further the current understanding of the microenvironment (84). Spatial proteomics has been used in breast cancer research and treatment; Cords *et al* (85) used highly multiplexed imaging mass cytometry on breast cancer samples matched to single-cell RNA sequencing datasets to confirm their cancer-associated fibroblast phenotypes defined at the protein level, and used spatial proteomics to analyze their spatial distributions in tumors, which provided a new strategy for this treatment. Notably, spatial proteomics is being used in lymphoma research (86), and may exhibit potential in the treatment of DLBCL. Spatial proteomics involves analysis of the subcellular localization of proteins in a systematic and high-throughput manner (87), where proteins simultaneously exist in different subcellular locations (88) and travel between them (89). For example, spatial proteomics may be used to demonstrate the spatial profile of proteins in the liver of patients with obesity, and these results are compared with healthy individuals to determine the localization of hepatocytes. Thus, spatial proteomics may aid in the treatment of patients with liver disease (90). In an era of rapid advances in medical technology, the use of spatial proteomics for the analysis and precise treatment of DLBCL can help to develop personalized treatment plans for patients and improve the cure and survival rates of DLBCL patients. However, due to the limitations of research methods and research data, there is still a lot of space for the wide application of spatial proteomics.

Acknowledgements

Not applicable.

Funding

The present study was supported by the Natural Science Foundation of Jilin (grant no. YDZJ202201ZYTS117).

Availability of data and materials

Not applicable.

Authors' contributions

ZG and CW authored or reviewed drafts of the manuscript, and approved the final draft. XS, ZW, JT and JM provided figures and helped with proofreading of draft. LB prepared tables and approved the final draft. All authors read and approved the final manuscript. Data authentication is not applicable.

Ethics approval and consent to participate

Not applicable.

Patient consent for publication

Not applicable.

Competing interests

The authors declare that they have no competing interests.

References

1. Thandra KC, Barsouk A, Saginala K, Padala SA, Barsouk A and Rawla P: Epidemiology of non-hodgkin's lymphoma. Med Sci (Basel) 9: 5, 2021.
2. de Leval L and Jaffe ES: Lymphoma classification. Cancer 26: 176-185, 2020.
3. Harrington F, Greenslade M, Talaulikar D and Corboy G: Genomic characterisation of diffuse large B-cell lymphoma. Pathology 53: 367-376, 2021.
4. Opinto G, Vegliante MC, Negri A, Skrypets T, Loseto G, Pileri SA, Guarini A and Ciavarella S: The tumor microenvironment of DLBCL in the computational era. Front Oncol 10: 351, 2020.
5. McCarthy L, Bentley-DeSousa A, Denoncourt A, Tseng YC, Gabriel M and Downey M: Proteins required for vacuolar function are targets of lysine polyphosphorylation in yeast. FEBS Lett 594: 21-30, 2020.
6. Kanduc D: The role of proteomics in defining autoimmunity. Expert Rev Proteomics 18: 177-184, 2021.
7. Liang XJ, Song XY, Wu JL, Liu D, Lin BY, Zhou HS and Wang L: Advances in multi-omics study of prognostic biomarkers of diffuse large B-cell lymphoma. Int J Biol Sci 18: 1313-1327, 2022.
8. Stegemann M, Denker S and Schmitt CA: DLBCL 1L-what to expect beyond R-CHOP? Cancers (Basel) 14: 1453, 2022.
9. McArdle AJ and Menikou S: What is proteomics? Arch Dis Child Educ Pract Ed 106: 178-181, 2021.
10. Punetha A and Kotiya D: Advancements in oncoproteomics technologies: Treading toward translation into clinical practice. Proteomes 11: 2, 2023.
11. Huang Z, Ma L, Huang C, Li Q and Nice EC: Proteomic profiling of human plasma for cancer biomarker discovery. Proteomics 17, 2017.
12. Kothalawala WJ, Barták BK, Nagy ZB, Zsigrai S, Szigeti KA, Valcz G, Takács I, Kalmár A and Molnár B: A detailed overview about the single-cell analyses of solid tumors focusing on colorectal cancer. Pathol Oncol Res 28: 1610342, 2022.
13. Gao HX, Li SJ, Niu J, Ma ZP, Nuerlan A, Xue J, Wang MB, Cui WL, Abulajiang G, Sang W, *et al*: TCL1 as a hub protein associated with the PI3K/AKT signaling pathway in diffuse large B-cell lymphoma based on proteomics methods. Pathol Res Pract 216: 152799, 2020.
14. Bingham GC, Lee F, Naba A and Barker TH: Spatial-omics: Novel approaches to probe cell heterogeneity and extracellular matrix biology. Matrix Biol 91-92: 152-166, 2020.
15. Ednersson SB, Stern M, Fagman H, Nilsson-Ehle H, Hasselblom S, Thorsell A and Andersson PO: Proteomic analysis in diffuse large B-cell lymphoma identifies dysregulated tumor microenvironment proteins in non-GCB/ABC subtype patients. Leuk Lymphoma 62: 2360-2373, 2021.
16. Zhuang K, Zhang Y, Mo P, Deng L, Jiang Y, Yu L, Mei F, Huang S, Chen X, Yan Y, *et al*: Plasma proteomic analysis reveals altered protein abundances in HIV-infected patients with or without non-Hodgkin lymphoma. J Med Virol 94: 3876-3889, 2022.
17. Ysebaert L, Quillet-Mary A, Tosolini M, Pont F, Laurent C and Fournié JJ: Lymphoma heterogeneity unraveled by single-cell transcriptomics. Front Immunol 12: 597651, 2021.
18. Jiang M, Bennani NN and Feldman AL: Lymphoma classification update: T-cell lymphomas, Hodgkin lymphomas, and histiocytic/dendritic cell neoplasms. Expert Rev Hematol 10: 239-249, 2017.
19. Zhang J, Gu Y and Chen B: Drug-resistance mechanism and new targeted drugs and treatments of relapse and refractory DLBCL. Cancer Manag Res 15: 245-225, 2023.
20. Liu Y, Zeng L, Zhang S, Zeng S, Huang J, Tang Y and Zhong M: Identification of differentially expressed proteins in chemotherapy-sensitive and chemotherapy-resistant diffuse large B cell lymphoma by proteomic methods. Med Oncol 30: 528, 2013.
21. Xie M, Huang X, Ye X and Qian W: Prognostic and clinicopathological significance of PD-1/PD-L1 expression in the tumor microenvironment and neoplastic cells for lymphoma. Int Immunopharmacol 77: 105999, 2019.
22. Steen CB, Luca BA, Esfahani MS, Azizi A, Sworder BJ, Nabet BY, Kurtz DM, Liu CL, Khameneh F, Advani RH, *et al*: The landscape of tumor cell states and ecosystems in diffuse large B cell lymphoma. Cancer Cell 39: 1422-37.e10, 2021.
23. Cioroianu AI, Stinga PI, Sticlaru L, Cioplea MD, Nichita L, Popp C and Staniceanu F: Tumor microenvironment in diffuse large B-cell lymphoma: role and prognosis. Anal Cell Pathol (Amst) 2019: 8586354, 2019.
24. Ceccato J, Piazza M, Pizzi M, Manni S, Piazza F, Caputo I, Cinetto F, Pisoni L, Trojan D, Scarpa R, *et al*: A bone-based 3D scaffold as an in-vitro model of microenvironment-DLBCL lymphoma cell interaction. Front Oncol 12: 947823, 2022.
25. de Groot FA, de Groen RAL, van den Berg A, Jansen PM, Lam KH, Mutsaers PGNJ, van Noesel CJM, Chamuleau MED, Stevens WBC, Plaça JR, *et al*: Biological and clinical implications of gene-expression profiling in diffuse large B-cell lymphoma: A proposal for a targeted BLYM-777 consortium panel as part of a multilayered analytical approach. Cancers (Basel) 14: 1857, 2022.
26. Takahara T, Nakamura S, Tsuzuki T and Satou A: The immunology of DLBCL. Cancers (Basel) 15: 835, 2023.
27. Ofori K, Bhagat G and Rai AJ: Exosomes and extracellular vesicles as liquid biopsy biomarkers in diffuse large B-cell lymphoma: Current state of the art and unmet clinical needs. Brit J Clin Pharmaco 87: 284-294, 2021.
28. Liu X, Zhao X, Yang J, Wang H, Piao Y and Wang L: High expression of AP2M1 correlates with worse prognosis by regulating immune microenvironment and drug resistance to R-CHOP in diffuse large B cell lymphoma. Eur J Haematol 110: 198-208, 2023.
29. Ejtehadifar M, Zahedi S, Gameiro P, Cabeçadas J, da Silva MG, Beck HC, Carvalho AS and Matthiesen R: Meta-analysis of MS-based proteomics studies indicates interferon regulatory factor 4 and nucleobindin1 as potential prognostic and drug resistance biomarkers in diffuse large B cell lymphoma. Cells 12: 196, 2023.
30. Ma J, Pang X, Li J, Zhang W and Cui W: The immune checkpoint expression in the tumor immune microenvironment of DLBCL: Clinicopathologic features and prognosis. Front Oncol 12: 1069378, 2022.
31. Kotlov N, Bagaev A, Revuelta MV, Phillip JM, Cacciapuoti MT, Antysheva Z, Svekolkin V, Tikhonova E, Miheecheva N, Kuzkina N, *et al*: Clinical and biological subtypes of B-cell lymphoma revealed by microenvironmental signatures. Cancer Discov 11: 1468-1489, 2021.
32. Bouwstra R, He Y, de Boer J, Kooistra H, Cendrowicz E, Fehrmann RSN, Ammatuna E, Zu Eulenburg C, Nijland M, Huls G, *et al*: CD47 Expression defines efficacy of rituximab with CHOP in non-germinal center B-cell (non-GCB) diffuse large B-cell lymphoma patients (DLBCL), but not in GCB DLBCL. Cancer Immunol Res 7: 1663-1671, 2019.
33. Xu-Monette ZY, Wei L, Fang X, Au Q, Nunns H, Nagy M, Tzankov A, Zhu F, Visco C, Bhagat G, *et al*: Genetic subtyping and phenotypic characterization of the immune microenvironment and MYC/BCL2 double expression reveal heterogeneity in diffuse large B-cell lymphoma. Clin Cancer Res 28: 972-983, 2022.
34. Feng Y, Zhong M, Tang Y, Liu X, Liu Y, Wang L and Zhou H: The role and underlying mechanism of exosomal CA1 in chemotherapy resistance in diffuse large B cell lymphoma. Mol Ther Nucleic Acids 21: 452-463, 2020.
35. Klein C, Jamois C and Nielsen T: Anti-CD20 treatment for B-cell malignancies: Current status and future directions. Expert Opin Biol Ther 21: 161-181, 2021.
36. Poletto S, Novo M, Paruzzo L, Frascione PMM and Vitolo U: Treatment strategies for patients with diffuse large B-cell lymphoma. Cancer Treat Rev 110: 102443, 2022.
37. Susanibar-Adaniya S and Barta SK: 2021 Update on diffuse large B cell lymphoma: A review of current data and potential applications on risk stratification and management. Am J Hematol 96: 617-629, 2021.
38. Roider T, Seufert J, Uvarovskii A, Frauhammer F, Bordas M, Abedpour N, Stolarczyk M, Mallm JP, Herbst SA, Bruch PM, *et al*: Dissecting intratumour heterogeneity of nodal B-cell lymphomas at the transcriptional, genetic and drug-response levels. Nat Cell Biol 22: 896-906, 2020.
39. Ferreri AJM, Doorduijn JK, Re A, Cabras MG, Smith J, Ilariucci F, Luppi M, Calimeri T, Cattaneo C, Khwaja J, *et al*: MATRix-RICE therapy and autologous haematopoietic stem-cell transplantation in diffuse large B-cell lymphoma with secondary CNS involvement (MARIETTA): An international, single-arm, phase 2 trial. Lancet Haematol 8: e110-e121, 2021.

40. Yan J, Yuan W, Zhang J, Li L, Zhang L, Zhang X and Zhang M: Identification and validation of a prognostic prediction model in diffuse large B-cell lymphoma. Front Endocrinol (Lausanne) 13: 846357, 2022.
41. Stanwood SR, Chong LC, Steidl C and Jefferies WA: Distinct gene expression patterns of calcium channels and related signaling pathways discovered in lymphomas. Front Pharmacol 13: 795176, 2022.
42. Frontzek F, Karsten I, Schmitz N and Lenz G: Current options and future perspectives in the treatment of patients with relapsed/refractory diffuse large B-cell lymphoma. Ther Adv Hematol 13: 20406207221103321, 2022.
43. Li S, Young KH and Medeiros LJ: Diffuse large B-cell lymphoma. Pathology 50: 74-87, 2018.
44. Gao HX, Nuerlan A, Abulajiang G, Cui WL, Xue J, Sang W, Li SJ, Niu J, Ma ZP, Zhang W and Li XX: Quantitative proteomics analysis of differentially expressed proteins in activated B-cell-like diffuse large B-cell lymphoma using quantitative proteomics. Pathol Res Pract 215: 152528, 2019.
45. Robotti E, Calà E and Marengo E: Two-dimensional gel electrophoresis image analysis. Methods Mol Biol 2361: 3-13, 2021.
46. Rotello RJ and Veenstra TD: Mass spectrometry techniques: Principles and practices for quantitative proteomics. Curr Protein Pept Sci 22: 121-133, 2021.
47. Yang J, Li Y, Zhang Y, Fang X, Chen N, Zhou X and Wang X: Sirt6 promotes tumorigenesis and drug resistance of diffuse large B-cell lymphoma by mediating PI3K/Akt signaling. J Exp Clin Cancer Res 39: 142, 2020.
48. Zhang X, Duan YT, Wang Y, Zhao XD, Sun YM, Lin DZ, Chen Y, Wang YX, Zhou ZW, Liu YX, *et al*: SAF-248, a novel PI3Kδ-selective inhibitor, potently suppresses the growth of diffuse large B-cell lymphoma. Acta Pharmacol Sin 43: 209-219, 2022.
49. Chen CC, Hsu CC, Chen SL, Lin PH, Chen JP, Pan YR, Huang CE, Chen YJ, Chen YY, Wu YY and Yang MH: RAS mediates BET inhibitor-endued repression of lymphoma migration and prognosticates a novel proteomics-based subgroup of DLBCL through its negative regulator IQGAP3. Cancers (Basel) 13: 5024, 2021.
50. Wang N, Wu R, Tang D and Kang R: The BET family in immunity and disease. Signal Transduct Target Ther 6: 23, 2021.
51. Sun F, Fang X and Wang X: Signal pathways and therapeutic prospects of diffuse large B cell lymphoma. Anticancer Agents Med Chem 19: 2047-2059, 2019.
52. Xu W, Berning P and Lenz G: Targeting B-cell receptor and PI3K signaling in diffuse large B-cell lymphoma. Blood 138: 1110-1119, 2021.
53. Dunleavy K, Erdmann T and Lenz G: Targeting the B-cell receptor pathway in diffuse large B-cell lymphoma. Cancer Treat Rev 65: 41-46, 2018.
54. Bisserier M and Wajapeyee N: Mechanisms of resistance to EZH2 inhibitors in diffuse large B-cell lymphomas. Blood 131: 2125-2137, 2018.
55. Coronado BNL, da Cunha FBS, de Toledo Nobrega O and Martins AMA: The impact of mass spectrometry application to screen new proteomics biomarkers in ophthalmology. Int Ophthalmol 41: 2619-2633, 2021.
56. Dallavalasa S, Beeraka NM, Basavaraju CG, Tulimilli SV, Sadhu SP, Rajesh K, Aliev G and Madhunapantula SV: The role of tumor associated macrophages (TAMs) in cancer progression, chemoresistance, angiogenesis and metastasis-current status. Curr Med Chem 28: 8203-8236, 2021.
57. Kelly RT: Single-cell proteomics: Progress and prospects. Mol Cell Proteomics 19: 1739-1748, 2020.
58. Hasin Y, Seldin M and Lusis A: Multi-omics approaches to disease. Genome Biol 18: 83, 2017.
59. Wang L, Li LR and Young KH: New agents and regimens for diffuse large B cell lymphoma. J Hematol Oncol 13: 175, 2020.
60. Xiong J, Cui BW, Wang N, Dai YT, Zhang H, Wang CF, Zhong HJ, Cheng S, Ou-Yang BS, Hu Y, *et al*: Genomic and transcriptomic characterization of natural killer T cell lymphoma. Cancer Cell 37: 403-419.e6, 2020.
61. van der Meeren LE, Kluiver J, Rutgers B, Alsagoor Y, Kluin PM, van den Berg A and Visser L: A super-SILAC based proteomics analysis of diffuse large B-cell lymphoma-NOS patient samples to identify new proteins that discriminate GCB and non-GCB lymphomas. PLoS One 14: e0223260, 2019.
62. Zhang P and Zhang M: Epigenetic alterations and advancement of treatment in peripheral T-cell lymphoma. Clin Epigenetics 12: 169, 2020.
63. Jiang H, Li A, Ji Z, Tian M and Zhang H: Role of radiomics-based baseline PET/CT imaging in lymphoma: Diagnosis, prognosis, and response assessment. Mol Imaging Biol 24: 537-549, 2022.
64. Fornecker LM, Muller L, Bertrand F, Paul N, Pichot A, Herbrecht R, Chenard MP, Mauvieux L, Vallat L, Bahram S, *et al*: Multi-omics dataset to decipher the complexity of drug resistance in diffuse large B-cell lymphoma. Sci Rep 9: 895, 2019.
65. Bresnick AR, Weber DJ and Zimmer DB: S100 proteins in cancer. Nat Rev Cancer 15: 96-109, 2015.
66. Ye X, Wang L, Nie M, Wang Y, Dong S, Ren W, Li G, Li ZM, Wu K and Pan-Hammarström Q: A single-cell atlas of diffuse large B cell lymphoma. Cell Rep 39: 110713, 2022.
67. Wang N, Li X, Wang R and Ding Z: Spatial transcriptomics and proteomics technologies for deconvoluting the tumor microenvironment. Biotechnol J 16: e2100041, 2021.
68. Cumming IA, Degorce SL, Aagaard A, Braybrooke EL, Davies NL, Diène CR, Eatherton AJ, Felstead HR, Groombridge SD, Lenz EM, *et al*: Identification and optimisation of a pyrimidopyridone series of IRAK4 inhibitors. Bioorg Med Chem 63: 116729, 2022.
69. Yoon SB, Hong H, Lim HJ, Choi JH, Choi YP, Seo SW, Lee HW, Chae CH, Park WK, Kim HY, *et al*: A novel IRAK4/PIM1 inhibitor ameliorates rheumatoid arthritis and lymphoid malignancy by blocking the TLR/MYD88-mediated NF-κB pathway. Acta Pharm Sin B 13: 1093-1109, 2023.
70. Zhang J, Fu L, Shen B, Liu Y, Wang W, Cai X, Kong L, Yan Y, Meng R, Zhang Z, *et al*: Assessing IRAK4 functions in ABC DLBCL by IRAK4 kinase inhibition and protein degradation. Cell Chem Biol 27: 1500-1509.e13, 2020.
71. Boșoteanu M, Cristian M, Așchie M, Deacu M, Mitroi AF, Brînzan CS and Bălțătescu GI: Proteomics and genomics of a monomorphic epitheliotropic intestinal T-cell lymphoma: An extremely rare case report and short review of literature. Medicine (Baltimore) 101: e31951, 2022.
72. Coradduzza D, Ghironi A, Azara E, Culeddu N, Cruciani S, Zinellu A, Maioli M, De Miglio MR, Medici S, Fozza C and Carru C: Role of polyamines as biomarkers in lymphoma patients: A pilot study. Diagnostics (Basel) 12: 2151, 2022.
73. Cheson BD, Nowakowski G and Salles G: Diffuse large B-cell lymphoma: New targets and novel therapies. Blood Cancer J 11: 68, 2021.
74. Rolland DCM, Basrur V, Jeon YK, McNeil-Schwalm C, Fermin D, Conlon KP, Zhou Y, Ng SY, Tsou CC, Brown NA, *et al*: Functional proteogenomics reveals biomarkers and therapeutic targets in lymphomas. Proc Natl Acad Sci USA 114: 6581-6586, 2017.
75. Huang L, Brunell D, Stephan C, Mancuso J, Yu X, He B, Zinner R, Kim J, Davies P and Wong STC: Driver network as a biomarker: systematic integration and network modeling of multi-omics data to derive driver signaling pathways for drug combination prediction. Bioinformatics 35: 3709-3717, 2019.
76. Chakraborty S, Hosen MI, Ahmed M and Shekhar HU: Onco-multi-OMICS approach: A new frontier in cancer research. Biomed Res Int 2018: 9836256, 2018.
77. Gohil SH, Iorgulescu JB, Braun DA, Keskin DB and Livak KJ: Applying high-dimensional single-cell technologies to the analysis of cancer immunotherapy. Nat Rev Clin Oncol 18: 244-256, 2021.
78. Yang D, Wang J, Hu M, Li F, Yang F, Zhao Y, Xu Y, Zhang X, Tang L and Zhang X: Combined multiomics analysis reveals the mechanism of CENPF overexpression-mediated immune dysfunction in diffuse large B-cell lymphoma in vitro. Front Genet 13: 1072689, 2022.
79. Landeira-Viñuela A, Diez P, Juanes-Velasco P, Lécrevisse Q, Orfao A, De Las Rivas J and Fuentes M: Deepening into intracellular signaling landscape through integrative spatial proteomics and transcriptomics in a lymphoma model. Biomolecules 11: 1776, 2021.
80. Jamil MO and Mehta A: Diffuse large B-cell lymphoma: Prognostic markers and their impact on therapy. Expert Rev Hematol 9: 471-477, 2016.
81. Maurer MJ, Micallef INM, Cerhan JR, Katzmann JA, Link BK, Colgan JP, Habermann TM, Inwards DJ, Markovic SN, Ansell SM, *et al*: Elevated serum free light chains are associated with event-free and overall survival in two independent cohorts of patients with diffuse large B-cell lymphoma. J Clin Oncol 29: 1620-1626, 2011.

82. Witzig TE, Maurer MJ, Stenson MJ, Allmer C, Macon W, Link B, Katzmann JA and Gupta M: Elevated serum monoclonal and polyclonal free light chains and interferon inducible protein-10 predicts inferior prognosis in untreated diffuse large B-cell lymphoma. Am J Hematol 89: 417-422, 2014.
83. Grünwald BT, Devisme A, Andrieux G, Vyas F, Aliar K, McCloskey CW, Macklin A, Jang GH, Denroche R, Romero JM, *et al*: Spatially confined sub-tumor microenvironments in pancreatic cancer. Cell 184: 5577-5592.e18, 2021.
84. Akhtar M, Haider A, Rashid S and Al-Nabet ADMH: Paget's 'seed and soil' theory of cancer metastasis: An idea whose time has come. Adv Anat Pathol 26: 69-74, 2019.
85. Cords L, Tietscher S, Anzeneder T, Langwieder C, Rees M, de Souza N and Bodenmiller B: Cancer-associated fibroblast classification in single-cell and spatial proteomics data. Nat Commun 14: 4294, 2023.
86. Franciosa G, Kverneland AH, Jensen AWP, Donia M and Olsen JV: Proteomics to study cancer immunity and improve treatment. Semin Immunopathol 45: 241-251, 2023.
87. Gatto L, Breckels LM and Lilley KS: Assessing sub-cellular resolution in spatial proteomics experiments. Curr Opin Chem Biol 48: 123-149, 2019.
88. Liu X, Salokas K, Tamene F, Jiu Y, Weldatsadik RG, Öhman T and Varjosalo M: An AP-MS- and BioID-compatible MAC-tag enables comprehensive mapping of protein interactions and subcellular localizations. Nat Commun 9: 1188, 2018.
89. Pankow S, Martínez-Bartolomé S, Bamberger C and Yates JR: Understanding molecular mechanisms of disease through spatial proteomics. Curr Opin Chem Biol 48: 19-25, 2019.
90. Guilliams M, Bonnardel J, Haest B, Vanderborght B, Wagner C, Remmerie A, Bujko A, Martens L, Thoné T, Browaeys R, *et al*: Spatial proteogenomics reveals distinct and evolutionarily conserved hepatic macrophage niches. Cell 185: 379-396.e38, 2022.
91. Lee PY, Saraygord-Afshari N and Low TY: The evolution of two-dimensional gel electrophoresis-from proteomics to emerging alternative applications. J Chromatogr A 1615: 460763, 2020.
92. Strohkamp S, Gemoll T and Habermann JK: Possibilities and limitations of 2DE-based analyses for identifying low-abundant tumor markers in human serum and plasma. Proteomics 16: 2519-2532, 2016.
93. Lin TT, Zhang T, Kitata RB, Liu T, Smith RD, Qian WJ and Shi T: Mass spectrometry-based targeted proteomics for analysis of protein mutations. Mass Spectrom Rev 42: 796-821, 2023.
94. Noor Z, Ahn SB, Baker MS, Ranganathan S and Mohamedali A: Mass spectrometry-based protein identification in proteomics-a review. Brief Bioinform 22: 1620-1638, 2021.
95. Ren AH, Diamandis EP and Kulasingam V: Uncovering the depths of the human proteome: Antibody-based technologies for ultrasensitive multiplexed protein detection and quantification. Mol Cell Proteomics 20: 100155, 2021.
96. Syu GD, Dunn J and Zhu H: Developments and applications of functional protein microarrays. Mol Cell Proteomics 19: 916-927, 2020.