 https://doi.org/10.21751/FRM-40-1-5

PROTEOMIKA U BIOMEDICINSKIM ISTRAŽIVANJIMA S NAGLASKOM NA MIŠIĆNO-KOŠTANE BOLESTI

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Proteomika je središnji alat suvremenih biomedicinskih istraživanja jer omogućuje izravnu karakterizaciju sastava i količine proteina, kao i njihovih posttranslacijskih modifikacija, čime pruža neposredno razumijevanje funkcionalnih molekularnih interakcija bioloških sustava (1). Za razliku od genomike ili transkriptomike, proteomika odražava funkcionalna molekularna stanja i trenutačne biološke procese, što je čini osobito vrijednom za proučavanje složenih, multifaktorijalnih bolesti. U kontekstu mišićno-koštanih poremećaja, u kojima se isprepliću mehaničko opterećenje, upala, tkivno remodeliranje i regeneracija, proteomski pristupi omogućuju dublje razumijevanje mehanizama bolesti te olakšavaju otkrivanje biomarkera i razvoj ciljanih terapijskih pristupa. Nedavni napredak u proteomici temeljenoj na masenoj spektrometriji, ponajprije visokorezolucijskoj LC-MS tehnologiji, strategijama akvizicije podataka neovisnima o podacima (engl. *data-independent acquisition)* i naprednim bioinformatičkim analizama, znatno je proširio mogućnosti analize složenih bioloških uzoraka poput plazme, sinovijalne tekućine, hrskavice i kosti. Ove tehnologije omogućuju nepristranu identifikaciju i kvantifikaciju stotina do tisuća proteina, bilježeći molekularne potpise povezane s upalom, remodeliranjem izvanstaničnog matriksa, angiogenezom, aktivacijom imunološkog sustava te degeneracijom i obnovom tkiva. Važno je istaknuti da proteomika omogućuje otkrivanje obrazaca molekularne ekspresije koji nisu vidljivi na razini gena, što je osobito važno kod bolesti obilježenih posttranslacijskom regulacijom i protein–protein interakcijama. Mišićno-koštane bolesti, uključujući osteoartritis, upalne artropatije, osteoporozu i rijetke zglobne poremećaje, predstavljaju značajno kliničko i socioekonomsko opterećenje. Unatoč velikom napretku u slikovnim metodama i kliničkim sustavima bodovanja, rana dijagnostika, stratifikacija bolesti te pouzdano praćenje progresije bolesti ili terapijskog odgovora i dalje predstavljaju ključne izazove u svakodnevnoj kliničkoj praksi. Proteomsko profiliranje pokazalo je znatan potencijal u rješavanju tih nezadovoljenih potreba, osobito kroz identifikaciju cirkulirajućih ili tkivno specifičnih biomarkera koji odražavaju aktualne patološke procese u zglobovima, kostima i okolnim mekim tkivima. Nadalje, proteomika podupire razvoj pristupa precizne medicine povezivanjem molekularnih fenotipova s kliničkim obilježjima te varijabilnošću i težinom bolesti. Posebno ilustrativan primjer translacijske vrijednosti proteomike u mišićno-koštanim istraživanjima

jest hemofilijska artropatija, teška zglobna bolest koja nastaje kao posljedica ponovljenih intraartikularnih krvarenja u bolesnika s hemofilijom (2,3). Iako se tradicionalno smatrala isključivo mehaničkom posljedicom hemartroze, danas se hemofilijska artropatija prepoznaje kao složen poremećaj obilježen kroničnom upalom, degradacijom hrskavice, remodeliranjem kosti i vaskularnim promjenama. U našem nedavnom proteomskom istraživanju uzorci plazme bolesnika s različitim stadijima hemofilijske artropatije analizirani su primjenom LC-MS tehnologije radi ispitivanja molekularnih razlika povezanih s težinom bolesti (4). Ovaj pristup omogućio je identifikaciju širokog spektra diferencijalno eksprimiranih proteina, pružajući molekularni uvid u upalne signalne putove, imunološku modulaciju, promjene izvanstaničnog matriksa, angiogenezu i degeneraciju zglobova. Posebno su se istaknuli proteini poput katepsina G, S100-A9, inzulinu sličnog faktora rasta-1, osteopontina, oligomernog matriksnog proteina hrskavice, CD44 i članova obitelji trombospondina kao ključni molekularni sudionici povezani s patologijom zglobova. Ovi nalazi naglašavaju dvojnu prirodu sistemskih odgovora u hemofilijskoj artropatiji, koji obuhvaćaju destruktivne procese u tkivu, ali i potencijalno kompenzacijske ili zaštitne mehanizme. Važno je istaknuti da studija pokazuje kako proteomika plazme, unatoč minimalno invazivnom pristupu, može detektirati molekularne promjene koje odražavaju lokaliziranu patologiju zglobova, čime se potvrđuje njezina primjenjivost za otkrivanje biomarkera i praćenje bolesti u kliničkoj praksi. Osim hemofilijske artropatije, slične proteomske strategije primijenjene su i na širok spektar mišićno-koštanih stanja, otkrivajući zajedničke i specifične molekularne putove. Komparativne proteomske analize identificirale su preklapajuće molekularne putove povezane s upalom i izvanstaničnim matriksom u bolesnika s hemofilijskom artropatijom, osteoartritisom i reumatoidnim artritisom, ali su istodobno otkrile i jedinstvena molekularna obilježja koja razlikuju ove entitete (5,6). Takvi uvidi ključni su za preciznije razvrstavanje bolesti i bolje razumijevanje patofiziološke heterogenosti. Trenutačni i budući smjerovi u proteomici mišićno-koštanog sustava sve više ukazuju na integraciju s drugim omics platformama, naprednim slikovnim metodama i detaljnom kliničkom fenotipizacijom. Posebice, proteomika izvanstaničnih vezikula, prostorna proteomika i longitudinalni dizajni istraživanja nude nove mogućnosti za praćenje dinamičkih molekularnih promjena tijekom progresije bolesti i regeneracije tkiva. U fizikalnoj i rehabilitacijskoj medicini proteomika predstavlja snažan alat za razjašnjavanje mehanizama bolesti, usmjeravanje personaliziranih rehabilitacijskih strategija i procjenu terapijskog odgovora s ciljem poboljšanja ishoda liječenja bolesnika. Međutim, unatoč velikom potencijalu za personaliziranu i ciljanu dijagnostiku i terapiju, proteomika je još uvijek ograničena visokom cijenom i relativnom tehnološkom složenošću, što otežava njezinu širu primjenu u rutinskoj kliničkoj praksi. Proteomski pristup pruža funkcionalne molekularne uvide i omogućuje otkrivanje biomarkera koji premošćuju jaz između temeljnih istraživanja i kliničke primjene. Time se podupire razvoj preciznijih dijagnostičkih i terapijskih strategija utemeljenih na specifičnim molekularnim mehanizmima uključenima u mišićno-koštane poremećaje. Zaključno, proteomika ima središnju ulogu u suvremenim biomedicinskim istraživanjima s

jasnom relevantnošću za mišićno-koštanu medicinu; međutim, njezina rutinska klinička primjena još uvijek nije u potpunosti ostvarena.

Ključne riječi

proteomika, mišićno-koštane bolesti, biomarkeri, translacija, artropatija

PROTEOMICS IN BIOMEDICAL RESEARCH WITH EMPHASIS ON MUSCULOSKELETAL DISEASES

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Proteomics has become a central tool in modern biomedical research, as it enables direct insight into protein composition, abundance and post-translational modification, thereby capturing functional molecular interactions within biological systems. (1). Unlike genomics or transcriptomics, proteomics directly reflects functional molecular states and ongoing biological processes, making it particularly valuable for studying complex, multifactorial diseases. In the context of musculoskeletal disorders, where mechanical stress, inflammation, tissue remodeling, and regeneration intersect, proteomic approaches offer unique insights into disease mechanisms, biomarker discovery, and therapeutic targeting. Recent advances in mass spectrometry-based proteomics, most notably high-resolution LC-MS, data-independent acquisition strategies and advanced bioinformatic pipelines, have substantially expanded our ability to analyze complex biological samples such as plasma, synovial fluid, cartilage, and bone. These technologies enable unbiased identification and quantification of hundreds to thousands of proteins, capturing molecular signatures linked to inflammation, extracellular matrix remodeling, angiogenesis, immune activation, as well as tissue degeneration and repair. Importantly, proteomics enables the discovery of molecular expression patterns that are not evident at the gene level, which is particularly important for diseases characterized by post-translational regulation and protein-protein interactions. Musculoskeletal diseases, including osteoarthritis, inflammatory arthropathies, osteoporosis and rare joint disorders, are a significant clinical and socioeconomic burden. Despite major advances in imaging and clinical scoring systems, early diagnosis, disease stratification, and reliable monitoring of disease progression or therapeutic response remain key challenges in daily clinical practice. Proteomic profiling has demonstrated considerable potential in addressing these unmet needs, particularly through the identification of circulating or tissue-specific biomarkers that reflect ongoing pathological processes within joints, bone, and surrounding soft tissues. Moreover, proteomics supports the development of precision medicine approaches by linking molecular phenotypes with clinical characteristics and disease variability or severity. A particularly illustrative example of the translational value of proteomics in musculoskeletal research is haemophilic arthropathy, a debilitating joint disease resulting from recurrent intra-articular

bleeding in patients with haemophilia (2,3). Although traditionally regarded as a purely mechanical consequence of hemarthrosis, haemophilic arthropathy is now recognized as a complex disorder involving chronic inflammation, cartilage degradation, bone remodeling, and vascular alterations. In our recent proteomic study, plasma samples from patients with different stages of haemophilic arthropathy were analyzed using LC-MS to explore molecular differences associated with disease severity (4). This approach enabled the identification of a broad panel of differentially expressed proteins, providing molecular insight into inflammatory signaling, immune modulation, extracellular matrix turnover, angiogenesis, and joint degeneration. Notably, proteins such as cathepsin G, S100-A9, insulin-like growth factor-1, osteopontin, cartilage oligomeric matrix protein, CD44, and thrombospondin family members emerged as key molecular players linked to joint pathology. These findings highlight the dual nature of systemic responses in haemophilic arthropathy, encompassing both tissue-destructive and potentially compensatory or protective mechanisms. Importantly, the study demonstrates how plasma proteomics, despite being a minimally invasive approach, can capture molecular signatures reflective of localized joint pathology, underscoring its feasibility for biomarker discovery and disease monitoring in clinical practice. Beyond haemophilic arthropathy, similar proteomic strategies have been applied across a spectrum of musculoskeletal conditions, revealing shared and disease-specific molecular pathways. Comparative proteomic analyses have identified overlapping inflammatory and extracellular matrix–related signatures between haemophilic arthropathy, osteoarthritis, and rheumatoid arthritis, while also uncovering unique molecular features that distinguish these entities (5,6). Such insights are critical for refining disease classification and understanding pathophysiological heterogeneity. Current and future directions in musculoskeletal proteomics increasingly point toward integration with other omics platforms, advanced imaging modalities, and detailed clinical phenotyping. In particular, extracellular vesicle proteomics, spatial proteomics, and longitudinal study designs offer new opportunities to capture dynamic molecular changes during disease progression and tissue regeneration. In physical and rehabilitation medicine, proteomics is a powerful tool for elucidating disease mechanisms, guiding personalized rehabilitation strategies and evaluating treatment response to ultimately improve patient outcomes. However, despite its great promise for personalized and targeted diagnostics and therapies, proteomics is still constrained by the high cost and relative complexity of the technology, hindering its broad implementation in routine clinical practice. By providing functional molecular insights and enabling translational biomarker discovery, proteomic approaches bridge the gap between basic research and clinical application, supporting the development of more precise, mechanism-based diagnostic and therapeutic strategies in musculoskeletal disorders. In conclusion, proteomics plays a central role in contemporary biomedical research with clear relevance to musculoskeletal medicine; however, its routine clinical application has yet to be fully realized.

Keywords

proteomics, musculoskeletal diseases, biomarkers, translation, arthropathy

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