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SYSTEMATIC REVIEW

Multi-omics Approaches for Biomarker Discovery of Uterine Fibroids: A Systematic Review

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ABSTRACT

Introduction: Uterine fibroids are the most common benign gynecological tumors, affecting up to 70–80% of women, yet still lack clinically validated biomarkers for disease stratification, monitoring, or therapeutic targeting. Advances in multi-omics technologies offer unprecedented opportunities for biomarker discovery; however, their application in fibroid research remains fragmented across platforms, biological samples, and study designs, limiting translational progress.

Methods: The PubMed, Embase, and Scopus databases were searched from 2000 to July 15, 2025, to identify relevant studies. Original, peer-reviewed articles using high-throughput omics technologies to discover fibroid-associated biomarkers were included. A narrative synthesis approach was employed to summarize the findings of included studies and map candidate biomarkers to their biological pathways. The methodological quality of the studies was assessed using the QUADOMICS tool. The protocol of this review was registered in PROSPERO platform (registration number CRD420251125813).

Results: Thirty-two studies met the inclusion criteria, including genomics ($n=10$, 31.3%), transcriptomics ($n=8$, 25.0%), epigenomics ($n=3$, 9.4%), proteomics ($n=4$, 12.5%), metabolomics ($n=3$, 9.4%), and multi-omics ($n=4$, 12.5%) approaches for biomarker discovery of uterine fibroids. Single-omics designs predominated (87.5%) over integrated multi-omics (12.5%). Six convergent pathways emerged across the different omics layers—extracellular matrix remodeling, hormone signaling, cell cycle regulation, apoptosis resistance, oxidative stress/metabolism, and genome stability—anchored by recurrent biomarker candidates.

Conclusion: This review provides the first systematic synthesis of high-throughput omics-based biomarker discovery in uterine fibroids across multiple biological samples. The convergence of multi-omics findings on a small set of

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interconnected pathways supports the view of fibroids as a systems-level disease and highlights tractable molecular targets that could inform non-invasive biomarker panels and guide the translation of new pathway-directed therapeutics beyond surgery. Critical gaps still include insufficient utilization of non-invasive samples, limited integration of multi-omics data, and inconsistent validation. Future research requires large-scale, integrated approaches that prioritize circulating biomarkers for clinical translation.

Keywords: Biomarkers; Multi-omics; Precision MEDICINE; Translational research; Uterine fibroids

Key Summary Points

Uterine fibroids are the most common benign tumors in women, yet they still lack clinically validated biomarkers, with diagnosis and management relying largely on imaging and surgical intervention rather than biomarker-guided molecular stratification, disease monitoring, or personalized therapeutic strategies.

Omics-based studies in uterine fibroids remain dominated by single-omics, tissue-based approaches, with limited integration of multi-omics data and minimal use of non-invasive biological samples.

Across genomics, epigenomics, transcriptomics, proteomics, metabolomics, and multi-omics studies, six convergent pathways—including extracellular matrix remodeling, hormone signaling, cell cycle regulation, oxidative stress, genome stability, and apoptosis resistance—emerge as core drivers of fibroid biology.

Recurrent biomarker candidates and pathways spanning multiple omics layers suggest possible targets for novel, targeted therapeutics beyond surgery and highlight the potential for composite, systems-level biomarker panels rather than single-marker strategies.

Future progress in uterine fibroid management will require large-scale, integrated multi-omics studies that prioritize non-invasive samples, standardized validation, and pathway-informed biomarker panels to enable biomarker-guided diagnosis, monitoring, and precise therapy strategies.

INTRODUCTION

Uterine fibroids (UFs), also known as leiomyomas or myomas, are the most common benign tumors of the female reproductive system, affecting up to 70–80% of women by age 50 [1–3]. Although the etiology of fibroid development remains incompletely understood, it is thought to involve a complex interplay of genetic susceptibility and environmental factors [4]. While many fibroids are asymptomatic, approximately 25–50% of affected women experience significant morbidity, including heavy menstrual bleeding, pelvic pain, infertility, and pregnancy complications, resulting in a substantial reduction in quality of life [3]. Despite the high prevalence and burden, early diagnosis of UFs remains challenging because of the lack of early symptoms and validated, non-invasive biomarkers for early detection, disease stratification, or treatment monitoring [5]. It is also important to note that the delayed diagnosis and treatment of UF is multifactorial and not solely biological. Although the absence of validated non-invasive biomarkers contributes to delayed detection, it represents only one component of a system-level clinical challenge. Delays in fibroid diagnosis are also influenced by healthcare access disparities, normalization or underrecognition of symptoms, and social stigma surrounding menstrual and reproductive health, which may discourage early clinical evaluation [3, 6, 7].

Diagnosis currently relies on imaging modalities such as transvaginal ultrasound and MRI, which effectively detect fibroids anatomically but cannot inform on disease progression, recurrence risk, or underlying molecular subtypes [5, 8]. Importantly, UFs are biologically and clinically heterogeneous, varying widely

in molecular subtype, tumor size, number, and anatomical location, as well as in symptom severity and reproductive impact [9–11]. This heterogeneity underscores the limitations of single-marker approaches and highlights the need for comprehensive molecular profiling strategies capable of capturing the complex biology underlying fibroid development and progression. The identification of reliable non-invasive biomarkers could facilitate not only early detection but also guide personalized, pathway-directed therapies, ultimately leading to reduced dependence on surgery, most notably hysterectomy, which is associated with both short- and long-term health and psychological risks [5, 8, 12]. In addition to potential medical side effects, current treatment strategies also largely conflict with many patients' reproductive goals. Surgical options such as hysterectomy permanently eliminate fertility, while hormonal therapies can suppress ovulation or require prolonged treatment courses that are not always compatible with pregnancy planning. Consequently, there is a critical clinical need for non-invasive tools and individualized management strategies that better align with reproductive preferences [13–15].

Recent advances in omics technologies, including genomics, epigenomics, transcriptomics, proteomics, and metabolomics, have transformed the ability to comprehensively characterize the molecular landscape of complex diseases [16, 17]. These high-throughput approaches can identify differentially expressed genes, proteins, and metabolites that may serve as biomarkers for early diagnosis, prognosis, or therapeutic targeting [17]. Preliminary studies have identified candidate biomarkers for fibroids, including *PLP1*, *FOS*, *LDH*, and *IGF*-1 [18–20]. In relation to each omic layer, transcriptomic profiling can reveal dysregulated signaling pathways that drive fibroid growth, while proteomic analysis can uncover extracellular matrix components that contribute to tumor stiffness and symptom severity [9, 21, 22]. Additionally, metabolic studies can highlight altered metabolic pathways that sustain fibroid proliferation. On the other hand, integrative multi-omics approaches offer additional advantages by linking genomic alterations, such as the most studied fibroids variants *MED12* mutations and *HMGA2* rearrangements, to downstream epigenetic regulation, transcriptional changes, protein expression, and metabolic phenotypes. By jointly interrogating multiple molecular layers, multi-omics strategies can elucidate how known UF pathways such as dysregulated extracellular matrix (ECM) remodeling and estrogen–progesterone signaling interact to drive fibroid growth, hormonal responsiveness, and clinical heterogeneity, all of which are relationships that single-omics studies cannot capture [9, 23, 24]. Despite these advances, the application of multi-omics approaches in fibroid research remains limited, and most studies rely on invasive tissue samples, with relatively few investigating circulating biomarkers from non-invasive biological specimens.

In this systematic review, we systematically synthesize current omics-based studies to identify candidate biomarkers associated with uterine fibroids across multiple molecular layers, including genomics, epigenomics, transcriptomics, proteomics, metabolomics, and integrated multi-omics approaches. Specifically, we characterize the distribution of omics platforms and biological sample types used in fibroid biomarker discovery and map reported candidate biomarkers to their key biological pathways. Through this integrative analysis, we highlight convergent molecular pathways underlying fibroid pathophysiology and identify critical gaps that must be addressed to advance biomarker-guided diagnosis, patient stratification, and future therapeutic development.

METHODS

Ethical Approval

This article is based on previously conducted studies and does not contain any new studies with human participants or animals performed by any of the authors.

Search Strategy

This systematic review was conducted following the Preferred Reporting Items for Systematic

Reviews and Meta-Analyses (PRISMA) 2020 Statement [25]. The review was also registered in the International Prospective Register of Systematic Reviews (PROSPERO) platform (registration number CRD420251125813), with the protocol available online (https://www.crd.york.ac.uk/PROSPERO/view/CRD420251125813).

A study search was conducted using three databases: PubMed, Embase, and Scopus. All English publications from the year 2000 until 15 July 2025 were searched without any restriction on the population's country of study. No additional restrictions were applied to the search in order to capture the broadest possible range of relevant studies and ensure comprehensive coverage of the literature. The search strategy employed used a combination of relevant controlled keywords and Boolean operators such as "uterine fibroids" or "leiomyoma" and "biomarker" or "diagnostic marker" and "omics" or "proteomics "or "metabolomics" or "transcriptomics" or "epigenomics" or "genomics". The full search strategy for each database is provided in Table S1 in the electronic supplementary material.

Eligibility Criteria and Selection of Studies

Original, peer-reviewed studies that used high-throughput omics technologies to discover biomarkers in uterine fibroids were included. The study's primary objective is to identify, validate, or characterize biomarkers associated with fibroids for diagnostic, prognostic, classification, or therapeutic purposes. Therefore, eligible studies met the following inclusion criteria: (a) the use of at least one omics-based technique, such as genomics, transcriptomics, proteomics, metabolomics, or epigenomics; (b) the study analyzes primary human biological samples, including tissue, blood, plasma, or urine; and (c) the study must clearly show that fibroids are the primary condition of interest for biomarker identification.

In addition, studies were excluded if they met any of the following conditions: (a) did not include any omics-based methodology or relied solely on low-throughput molecular techniques without prior omics discovery; (b) used exclusively non-human data, such as cell lines or animal models, without validation in human samples; (c) were not original research articles, including reviews, editorials, conference abstracts without full methodology; (d) studies that lacked a clearly defined comparison between fibroid cases and appropriate controls (e.g., normal myometrium or healthy individuals).

Data Collection and Extraction

All retrieved articles were uploaded into EndNote reference management software, and duplicates were removed prior to screening. Two reviewers independently assessed titles and abstracts for relevance. A second round of full-text review was then conducted on all studies that met the inclusion criteria. Data extraction was performed using a standardized Excel form. One reviewer independently extracted data from the full-text articles, and a second reviewer verified the accuracy of the extracted information. Disagreements or discrepancies in data interpretation were resolved through discussion.

For each included study, the following information was extracted: (a) the identity of candidate biomarkers proposed or validated in the results, (b) the omics technique used (e.g., genomics, transcriptomics, proteomics, metabolomics, or epigenomics), (c) the type of biological sample analyzed (e.g., tissue, blood, plasma, urine), (d) the comparison group (e.g., fibroid vs. normal myometrium), (e) the number of samples analyzed, and (f) the year of publication. Only biomarkers explicitly stated as statistically significant or validated biologically were included in the extraction results table. In cases of missing or unclear data, attempts were made to contact the corresponding authors for clarification or additional information.

Study Risk of Bias and Quality Assessment

The methodological quality and risk of bias of the included studies were assessed using the QUADOMICS methodology criteria, specifically designed to evaluate studies that apply omics-based technologies [26]. Quality

assessment was conducted independently by two reviewers. The QUADOMICS tool consists of a 16-item checklist, and each study was evaluated against these criteria using a Yes/No/Unclear scoring system, where *Yes* received a single point and *No* or *Unclear* received zero points. A total quality score was then calculated for each study, with scores of 13–16 classified as *high quality*, scores of 9 or below as *low quality*, and scores in between considered *moderate quality*. In addition to assigning an overall score per study, the proportion of studies fulfilling each individual criterion was calculated, providing a comprehensive overview of the methodological strengths and weaknesses across all included studies.

Data Synthesis and Visualization

Given the diversity of study designs and outcomes, a narrative synthesis approach was employed. This synthesis highlighted the most promising biomarker candidates within each omics layer. Studies were grouped into synthesis categories according to the omics layer investigated: genomics, epigenomics, transcriptomics, proteomics, metabolomics, and multi-omics. Extracted study data were entered into a standardized Excel spreadsheet and harmonized to ensure consistency in terminology and format. The results of biomarker discovery were organized in summary tables for each omics category, detailing key study characteristics, sequencing methods, validation status, and identified biomarker candidates. Additionally, visual representations were generated, including bar charts that illustrate the distribution of biological sample types, the number of studies conducted per omics layer, and Sankey plot for pathway convergence across all included studies.

Heterogeneity among studies was explored qualitatively, focusing on differences in omics platforms, biological sample sources, and the geographical origin of study populations. This approach allowed the identification of methodological trends and gaps that may inform future research priorities.

Omics Category Definitions and Classification Rules

Omics classifications were defined according to the primary molecular layer directly interrogated in biological samples rather than downstream computational interpretation. In this review, genomics studies were defined as those identifying DNA-level variation such as genome-wide association studies (GWAS), whole-exome sequencing (WES), single nucleotide polymorphism (SNP) genotyping, and candidate gene polymorphism analyses. In silico functional analyses derived from genetic variants, including expression quantitative trait loci (eQTL) mapping and transcriptome-wide association studies (TWAS), were also classified under genomics because they infer transcriptional consequences from genotype. Transcriptomics studies included those that directly quantified RNA expression in biological samples using techniques such as RNA sequencing, microarrays, or targeted expression profiling. Epigenomics comprised studies assessing DNA methylation or chromatin regulatory modifications in fibroid-related samples. Proteomics and metabolomics included studies directly measuring proteins and metabolites, respectively. Multi-omics studies were defined as investigations experimentally measuring two or more molecular layers within the same biological samples. Computational integration of independent datasets without simultaneous experimental measurement was not classified as multi-omics. To avoid inflation of study numbers, multi-omics studies were counted exclusively within the multi-omics category and were not duplicated across individual omics layers.

Additional Sensitivity Search for Genomics Studies

To assess whether the use of the term 'biomarker' in the primary search strategy may have excluded genomics studies—such as GWAS, targeted genetic association studies, and candidate gene studies reporting statistical associations without explicitly using biomarker-related terminology—we conducted an additional sensitivity targeted genomics search. This search was performed in the same primary databases

in PubMed, Embase, and Scopus (2000–15 July 2025) using the original search strategy with the sole removal of the term 'biomarker' and its related synonyms (Table S1). This analysis was undertaken to determine whether the biomarker-focused search restriction would influence the genomics biological interpretation and pathway convergence reported in this review.

RESULTS

Study Selection and Characteristics

The detailed flowchart of the study selection process is shown in Fig. 1. A total of 2730 records were identified from three databases: PubMed ($n=905$), Embase ($n=953$), and Scopus ($n=872$). After removal of 1667 duplicates, 1063 unique records were retained for title and abstract screening. Of these, 919 were excluded. The remaining 144 articles were retrieved for full-text assessment. Following full-text review, 112 articles were excluded. Ultimately, 32 studies met the inclusion criteria and were included in the synthesis. More details of the screening process are in the PRISMA flow diagram (Fig. 1).

The included studies spanned five omics categories: genomics ($n=10$; 31.2%), transcriptomics ($n=8$; 25.0%), epigenomics ($n=3$; 9.4%), proteomics ($n=4$; 12.5%), metabolomics ($n=3$; 9.4%), and multi-omics ($n=4$; 12.5%). Single-omics approaches predominated (87.5%), with

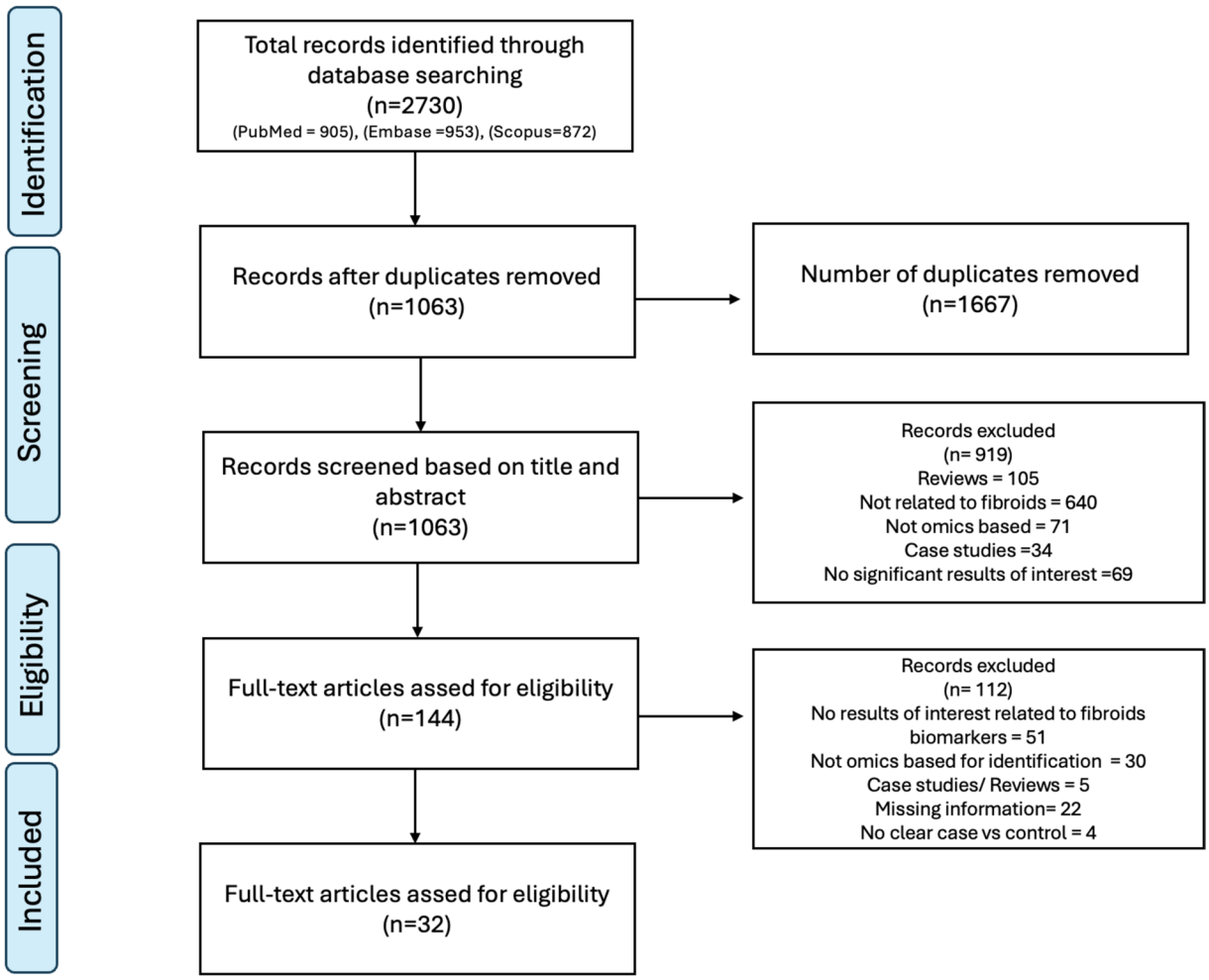


Fig. 1 PRISMA 2020 flow diagram summarizing the study selection process for the systematic review

only 12.5% employing integrated multi-omics analyses. Biological sample sources varied, with tissue being the most common ($n = 18$), followed by blood ($n = 11$), a combination of tissue and blood ($n = 2$), and saliva ($n = 1$). Notably, no included studies utilized urine as a biological sample for fibroid biomarker discovery using omics (Fig. 2). The full details of study characteristics for all 32 included papers are found in Table S2 in the electronic supplementary material.

According to the QUADOMICS tool's scoring, of the included studies, 7 were ranked high quality, 18 moderate quality, and 7 low quality. Several methodological criteria were strongly adhered to across the studies; 93.8% of the studies clearly described their selection criteria, and 84.4% used a range of well-representative patient sample types. In addition, 81.2% of

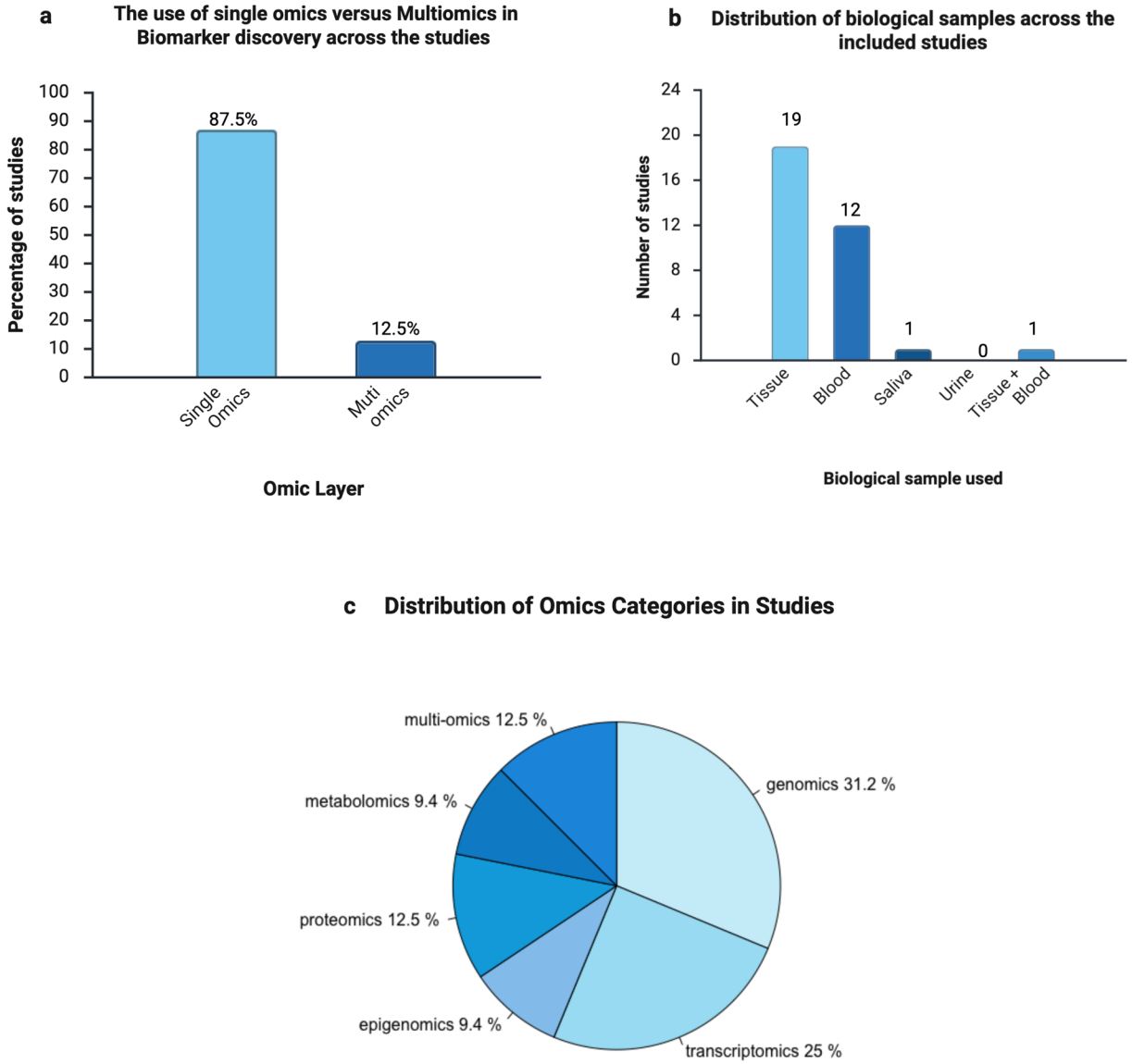
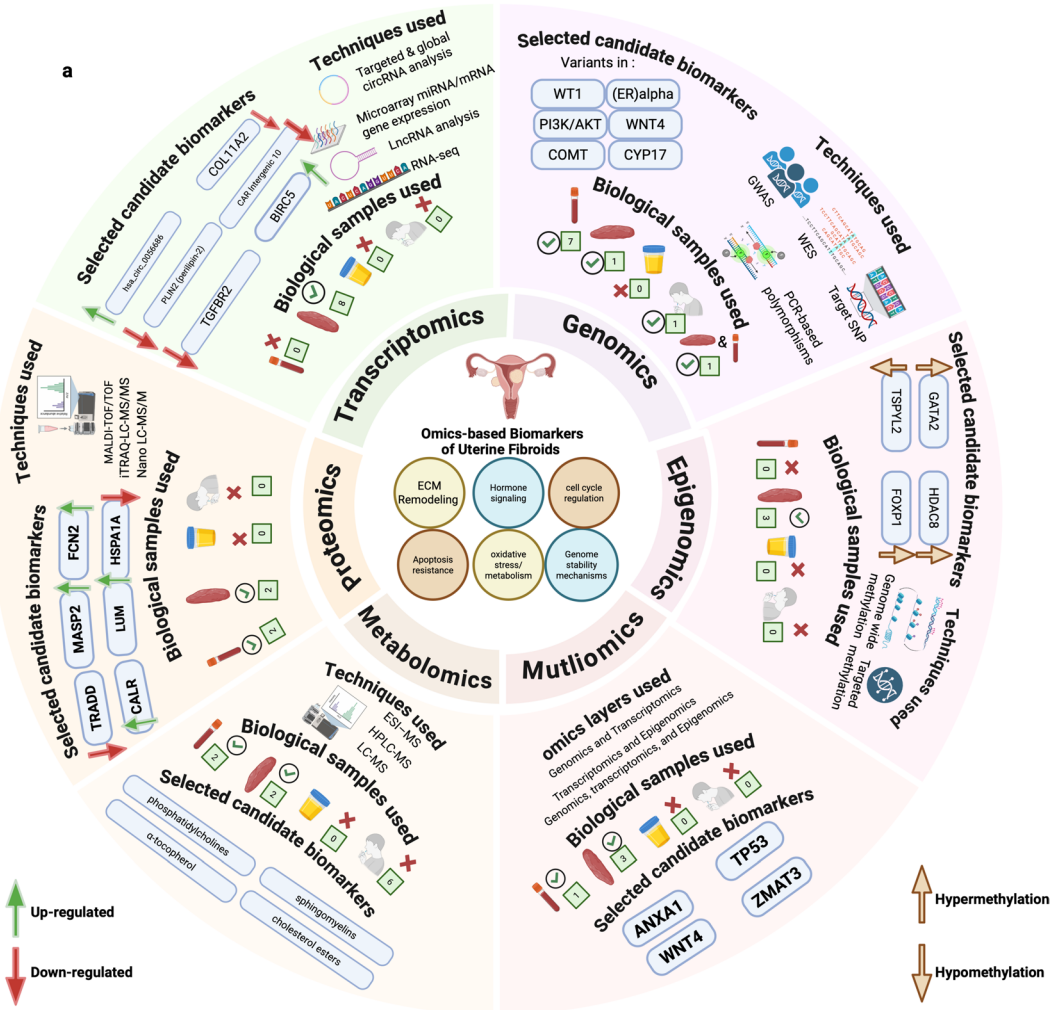
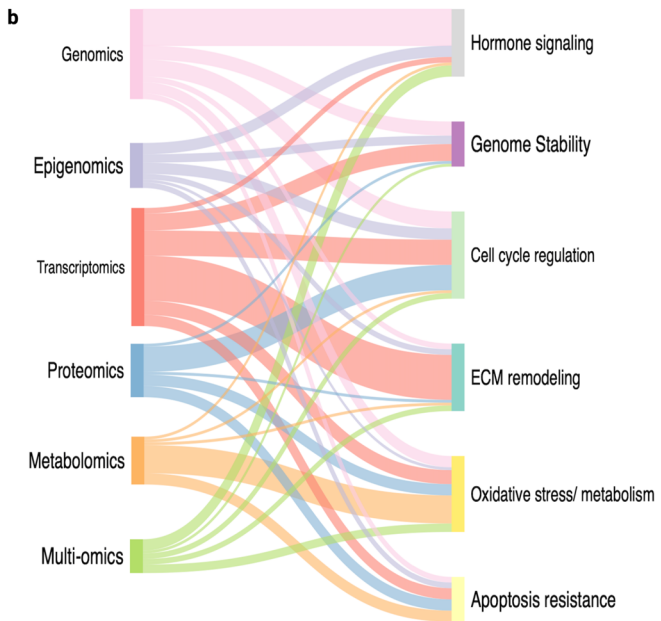


Fig. 2 Overview of study characteristics: distribution of omics and biological samples across included studies. **a** Proportion of studies applying single omics versus multi-omics approaches. **b** Distribution of biological sample types used across studies. **c** Distribution of omics layers investigated across the included studies


a
Transcriptomics
Genomics
Epigenomics
Mutliomics
Metabolomics
Proteomics
Omics-based Biomarkers of Uterine Fibroids
ECM Remodeling
Hormone signaling
cell cycle regulation
Apoptosis resistance
oxidative stress/ metabolism
Genome stability mechanisms
Techniques used
Targeted & global circRNA analysis
Microarray miRNA/mRNA gene expression
LncRNA analysis
RNA-seq
Selected candidate biomarkers
COL11A2
CAR intergenic 10
BIRC5
TGFBR2
PLIN2 (perilipin 2)
Biological samples used
Selected candidate biomarkers
Variants in :
WT1
(ER)alpha
PI3K/AKT
WNT4
COMT
CYP17
Techniques used
GWAS
WES
PCR-based polymorphisms
Target SNP
Biological samples used
Selected candidate biomarkers
TSPYL2
GATA2
FOXP1
HDAC8
Biological samples used
Techniques used
Genome wide methylation
Targeted methylation
omics layers used
Genomics and Transcriptomics
Transcriptomics and Epigenomics
Genomics, transcriptomics, and Epigenomics
Biological samples used
Selected candidate biomarkers
TP53
ZMAT3
ANXA1
WNT4
Techniques used
ESI-MS
HPLC-MS
LC-MS
Biological samples used
Selected candidate biomarkers
phosphatidylcholines
α-tocopherol
sphingomyelins
cholesterol esters
Techniques used
MALDI-TOF/TOF
iTRAQ-LC-MS/MS
Nano LC-MS/M
Biological samples used
Selected candidate biomarkers
FCN2
HSPA1A
MASP2
LUM
TRADD
CALR
Up-regulated
Down-regulated
Hypermethylation
Hypomethylation


b
Genomics
Epigenomics
Transcriptomics
Proteomics
Metabolomics
Multi-omics
Hormone signaling
Genome Stability
Cell cycle regulation
ECM remodeling
Oxidative stress/ metabolism
Apoptosis resistance

◂**Fig. 3** Integration of multi-omics studies on biomarker discovery of uterine fibroids. (**a**) Each sector represents one omics layer, highlighting the primary techniques used, the types of biological samples, and the selected candidate biomarkers. The central hub illustrates the convergence of omics findings onto key dysregulated biological pathways. (**b**) Sankey plot illustrating convergence of biological pathways across omics layers in uterine fibroids. Each ribbon links an omics layer (left) to a key dysregulated biological pathway (right). Pathways include hormone signaling, genome stability, cell cycle regulation, oxidative stress/metabolism, ECM remodeling, and apoptosis resistance. Ribbon widths indicate the relative number of biomarkers mapped to each pathway. *SNP* single nucleotide polymorphism, *WES* whole-exome sequencing, *GWAS* genome-wide association study, *PCR* polymerase chain reaction, *circRNA* circular RNA, *miRNA* microRNA, *lncRNA* long noncoding RNA, *RNA-seq* RNA sequencing, *MALDI TOF/TOF* matrix-assisted laser desorption/ionization time-of-flight, *iTRAQ-LC–MS/MS* isobaric tags for relative and absolute quantitation-liquid chromatography–mass spectrometry, *Nano LC–MS/MS* nano liquid chromatography–mass spectrometry, *ESI–MS* electrospray ionization mass spectrometry, *HPLC–MS* high-performance liquid chromatography–mass spectrometry, *LC–MS* liquid chromatography–mass spectrometry, *ECM* extracellular matrix

studies reported handling and pre-analytical procedures in detail. However, several weaknesses were also evident across the studies. The lowest-performing domains were blinding of the index test to the reference standard (only 9.4%), detailed reporting of diagnostic and treatment procedures (40.6%), and the failure of most studies (62.5%) to report failed/invalid samples, masking potential selection bias. These shortcomings suggest potential risks of bias, particularly in the study's execution and data analysis. A full breakdown of the detailed quality scoring for each included study is provided in Table S3 in the electronic supplementary material.

Genomics: Diverse Genetic Technologies Reveal Fibroid Susceptibility Loci

Across the ten included genomics studies (Fig. 3), diverse genetic technologies were applied. Two studies (20%) used GWAS [27, 28], one study (10%) analyzed WES [29], 3 studies (30%) used targeted SNP genotyping [30–32], and 4 studies (40%) used PCR-based polymorphism detection [33–36]. Regarding biological samples, seven studies (70%) used blood samples, one (10%) used saliva, one (10%) used tissue only, and one (10%) used both tissue and blood. Across these studies, direct biological validation of candidate biomarker variants was largely absent, except for one study (10%) [29], which incorporated molecular validation, while the remainder relied solely on statistical significance (see Table S4 in the electronic supplementary material for details).

Collectively, the primary genomic studies have revealed an interplay between inherited susceptibility loci and somatic alterations, converging on biological pathways related to hormone signaling, cell proliferation, genome stability, and ECM remodeling, which serve as consistent genetic candidate markers of fibroid risk. Notably, candidate variants such as *WT1* (transcriptional regulation), *ESR1* (estrogen receptor alpha), *CDC42/WNT4* (cell signaling pathway), *OBFC1* (telomere maintenance), *LIN28B* (apoptosis inhibition/PI3K/AKT pathways), *YEATS4* (p53 pathway inhibition/cell proliferation), and *ZNHIT1* (p53-mediated DNA damage response) were all linked to fibroids risk across the different studies [28–31, 34]. In addition, polymorphisms in colocalization with the vitamin D receptor (*VDR*) genes, such as *DHCR7* and *ASIP*, were also proposed as genetic markers of fibroid risk [32]. A multi-ancestry GWAS further identified novel associations at *VIP* and *FOXO3*, implicating vasoactive and stress-response transcriptional programs: *VIP* variants may link fibroid risk to vascular tone, blood-pressure regulation, and fibrosis, while *FOXO3* is connected to estrogen-related receptor signaling and *TP53*-mediated cell cycle control, reinforcing cross talk between hormonal, proliferative, and tumor-suppressor networks [27]

Interestingly, one large multi-ancestry GWAS meta-analysis included 74,294 fibroid cases and 465,810 controls (27.7% non-European), enabling ancestry-stratified analyses in European, East Asian/Central/South Asian, and African ancestry populations [27]. Using an LDAK Sum-Her approach, the authors estimated SNP-based

heritability at 5% in the combined multi-ancestry sample and 7% in Europeans, with a notably higher estimate of 15.9% in African-ancestry individuals, highlighting a substantial inherited component of fibroid risk in this population. Ancestry-stratified analyses replicated *WT1* as a fibroid risk gene in African-ancestry individuals and identified a novel association at *COL22A1*, a collagen gene upregulated in fibroid tissue, linking African-ancestry genetic risk to ECM remodeling processes observed in other omics layers [27].

Moreover, an additional targeted sensitivity search was performed to address the possibility that requiring 'biomarker/diagnostic marker' as a search term may have underrepresented genomics studies such as GWAS and targeted genetic association analyses. This search yielded 3223 (PubMed), 1941 (Embase), and 1818 (Scopus) records. After deduplication and title/abstract screening prioritizing genomics-focused designs (GWAS, SNP-based genetic association, candidate gene studies), 32 full-text articles were assessed against the original eligibility criteria, and four additional genomics studies met inclusion criteria and were incorporated further into the synthesis (Table S4). The additional sensitivity targeted genomics search identified further genetic association studies reporting susceptibility loci without explicit biomarker terminology. These studies also consistently highlighted loci related to hormone signaling (*GREB1*, *WNT4/CDC42*), proliferative/inflammatory pathways (*WNT4*, *WT1*, *MED12*, *TNRC6B*, *BET1L*, *COX-2*), and genome stability/telomere maintenance (*TERT*, *TP53*) [37–40], reinforcing the same biological themes observed in the primary analysis. Large-scale association analyses similarly linked fibroid risk to hormone-responsive and tumor-suppressor pathways. Importantly, the inclusion of these additional genomics studies did not alter the pathway convergence or the primary interpretation of genomic contributions to fibroid biology described in this review.

Altogether, the genomic biomarker variants described in this review mechanistically contribute to fibroid pathogenesis through disrupted transcriptional regulation (e.g., *WT1*), estrogen hypersensitivity (e.g., *ESR1*), aberrant Wnt/β-catenin signaling (e.g., *CDC42/WNT4*), telomere maintenance defects (e.g., *OBFC1*, *TERT*), and p53 pathway inhibition (e.g., *YEATS4*, *ZNHIT1*), creating a proliferative, survival-advantaged cellular state. Vitamin D pathway polymorphisms (e.g., *DHCR7*, *ASIP*) may impair antiproliferative effects, while ECM collagen variants (*COL22A1*) can link genetic risk to the fibrotic phenotype, establishing inherited susceptibility as a foundational driver of fibroid molecular heterogeneity.

Epigenomics: DNA Methylation and Chromatin Remodeling as Biomarker Signatures

Regarding epigenomics, three studies investigated DNA methylation changes in fibroids, with a primary focus on genome-wide methylation profiling and targeted methylation analysis of candidate biomarker genes. Across the three studies (100%), the methylation changes implicated genes involved in transcriptional regulation, cell signaling, and hormonal response [41–43]. Notably, unlike genomic studies, these epigenomic investigations consistently incorporated various types of biological validation for candidate biomarkers (see Table S5 in the electronic supplementary material for details). All the epigenomics studies (100%) used patient-matched paired tissue samples [41–43]

Overall, epigenetic studies consistently implicated aberrant DNA methylation and chromatin regulation as candidate markers in fibroid development. Across genome-wide and candidate-gene analyses, both hypermethylation and hypomethylation patterns emerged, with recurrent alterations affecting regulatory genes, such as *GATA2* and *NTRK2* [42], as well as X-linked loci, including *FAM9A* and *CPXCR1* [43]. These changes converged on pathways related to cell adhesion, transcriptional regulation, and cell signaling, processes tightly linked to proliferation and differentiation [41, 43]. Importantly, differential methylation signatures distinguished fibroids from matched myometrial tissues, identifying significant biomarker candidates, such as *TSPYL2* (an assembly protein required for cell cycle maintenance under stress) and *OCRL* (inositol 5-phosphatase), in 68.2% and 50% of fibroid specimens, respectively [41]. Despite

methodological heterogeneity, the findings collectively highlight epigenetic remodeling as a hallmark of fibroid biology. Mechanistically, epigenomic alterations silence tumor suppressors (e.g., *TSPYL2*) and activate growth signaling (e.g., *GATA2*, *NTRK2*) through hyper/hypomethylation, disrupting cell cycle maintenance and adhesion. Chromatin remodeling creates heritable transcriptional memory that sustains the pro-proliferative, ECM-producing fibroid phenotype independently of coding mutations.

Transcriptomics: Noncoding RNAs and Differential Gene Expression in Fibroid Pathogenesis

This systematic review included eight studies that used transcriptomics. Of these, two studies (25%) analyzed circular RNAs (circRNAs) [44, 45], one using a global [44] and one a targeted [45] approach. One study (12.5%) utilized a DNA microarray of gene expression screening [46], two studies (25%) analyzed microarray miRNA/mRNA gene expression [47, 48], two studies (25%) analyzed long noncoding RNAs (lncRNAs) [49, 50], and one study (12.5%) used RNA-sequencing (RNA-seq) [51]. Regarding the biological samples, in all eight papers (100%), the core expression analysis and biomarker discovery were tissue-based (Fig. 3). In seven studies, differential expression findings were biologically validated using techniques such as quantitative RT-PCR, western blotting, immunohistochemistry, in situ hybridization, and functional in vitro assays in primary fibroid cells or leiomyoma cell lines, whereas one study relied solely on statistical significance without experimental validation. Notably, none of the included transcriptomic papers incorporated in vivo animal models; all functional follow-up was limited to ex vivo human tissue or in vitro cellular systems (see Table S6 in the electronic supplementary material for details).

Overall, transcriptomic studies consistently implicated dysregulation of ECM organization, cell cycle regulation, and metabolic pathways in fibroid pathogenesis, with noncoding RNAs (ncRNAs), particularly circRNAs and lncRNAs, emerging as promising biomarker classes [44, 45, 49, 50]. Circular RNA profiling identified a panel (hsa_circ_0083920, hsa_circ_0056686, hsa_circ_0062558, hsa_circ_0020376, hsa_circ_0043597) with high diagnostic sensitivity and specificity accuracy [44], while hsa_circ_0060927 was ectopically expressed in one-third (33.33%) of fibroids but absent in controls [45]. In addition, integrated miRNA/mRNA microarray analysis identified hub differentially expressed genes (DEGs) (e.g., *ASPM*, *SNAI2*, *COL3A1*) and hub differentially expressed molecules (DEMs) (hsa-miR-181b-5p, hsa-miR-381-3p) linked to cell cycle progression, ECM remodeling, and epithelial–mesenchymal transition (EMT), validated via qRT-PCR and functional assays (proliferation, migration, apoptosis) (see Table S6 in the electronic supplementary material for details) [47]. Concurrent lncRNA profiling revealed widespread dysregulation of lncRNAs in leiomyoma versus normal myometrium. In RNA-seq studies, many dysregulated miRNAs in leiomyoma are reported to target genes involved in angiogenesis, inflammation, ECM accumulation, and tissue fibrosis, and to link progesterone signaling to altered lipid metabolism, all of which are characteristic processes of leiomyoma [51, 52]. Another lncRNA study further shows that lncRNA-mediated sponging of miR-29c and miR-200c increases COL1A1, COL3A1, and FN1 protein levels, directly promoting collagen-rich ECM deposition and fibrotic remodeling in fibroid smooth muscle spheroids [53]. Protein-coding transcriptomics further highlighted changes associated with metabolism and the ECM [42].

Collectively, transcriptomic dysregulation mechanistically drives ECM deposition through collagen upregulation (e.g., COL1A1, COL3A1) and lncRNA/miRNA sponging, cell cycle acceleration via CCNB2/BIRC5/FOXM1 overexpression, and EMT promotion (e.g., SNAI2, miR-181b-5p). Noncoding RNAs (circRNAs, lncRNAs) form regulatory hubs that maintain fibrotic stiffness, proliferation, and survival signaling, positioning them as master regulators linking genetic predisposition to the extracellular and proliferative hallmarks of fibroid tumors.

Proteomics and Metabolomics: Protein and Metabolic Signatures of Fibroid Biology

This systematic review included seven studies on the discovery of fibroid biomarkers using proteomics and metabolomics: four studies (57.1%) used proteomics, and three studies (42.9%) used metabolomics techniques.

With regards to proteomics, all the included studies (100%) used mass spectrometry-based platforms as the core technology for protein identification and quantification, with variations in labeling strategies and fractionation methods such as MALDI-TOF/TOF [54], iTRAQ-LC–MS/MS [55], Nano LC–MS/MS [56], and 2D-DIGE/MALDI-TOF MS [57]. Three of the studies used unbiased approaches, while one focused on targeted chaperone proteins (Fig. 3). In addition, two studies used paired tissue samples [45, 46], while two used serum or plasma [56, 57] for proteomic identification. Collectively, the studies' findings depict a proteomic shift toward a pro-growth, pro-angiogenic, and apoptosis-resistant fibroid phenotype [54–56]. This was evident by the altered expression of chaperones (CALR, HSPA5, PDIA3, VCP) [54], apoptosis regulators (TRADD) [55], secreted proteins (MASP2, lumican, ficolin-2, kallistatin, EMSY) [56], as well as transportation/coagulation proteins such as apolipoprotein A-I, fibrinogen chains, serotransferrin, and vitamin D-binding protein [57]. These proteins were implicated in cell proliferation, evasion of apoptosis, ECM remodeling, and angiogenesis. All proteomics candidates were experimentally validated using various methods, including western blot, immunohistochemistry, RT-qPCR, and bio-layer interferometry (see Table S7 in the electronic supplementary material for details). The recurrent detection of ECM-related and apoptosis-regulatory proteins across different proteomics platforms and sample types strengthens their potential as candidate biomarker and therapeutic targets.

Similarly, across all three metabolomics studies (100%), unbiased mass spectrometry-based platforms were used to characterize the metabolic alterations associated with uterine fibroids. One study utilized electrospray ionization mass spectrometry (ESI–MS) for tissue samples and high-performance liquid chromatography–mass spectrometry (HPLC–MS) for plasma [58]. The other study used tissue samples, which were analyzed by liquid chromatography–mass spectrometry (LC–MS) [59], and the last study analyzed plasma by gas chromatography–mass spectrometry (GC–MS) [60]. Only one study incorporated biological validation, while the others relied solely on statistical significance (see Table S7 in the electronic supplementary material for details). Collectively, the metabolomics findings reveal consistent metabolic signatures associated with lipid remodeling, energy metabolism, and oxidative stress pathways during fibroid development. Metabolite candidates, such as phosphatidylcholines, sphingomyelins, and cholesterol esters, were proposed as biomarker candidates for fibroids [58]. Additional alterations included reduced plasma levels of branched-chain amino acids (L-isoleucine, L-valine), pyroglutamic acid, fatty acids (arachidonic, palmitic, stearic acids, α-tocopherol), and carbohydrates (*myo*-inositol, D-threitol), alongside increased α-linolenic acid and L-glutamine. These metabolite shifts suggest systemic metabolic reprogramming that may underpin the growth, recurrence, and progression of fibroids [59, 60].

Overall, proteomic shifts toward chaperone-mediated growth (e.g., CALR, HSPA5), apoptosis evasion (e.g., TRADD downregulation), and angiogenic ECM remodeling (e.g., lumican, MASP2) create a pro-tumorigenic niche, while metabolomic signatures reveal lipid reprogramming (phosphatidylcholines, sphingomyelins) and oxidative stress (BCAA depletion, α-tocopherol reduction) that fuel proliferation and survival. These protein/metabolite alterations reflect systems-level adaptation to hormonal/growth stimuli characteristic of fibroid pathophysiology.

Multi-omics Integration: Convergent Biomarker Pathways Across Omics Layers

This review included four studies that used multi-omics approaches to discover fibroid biomarkers. All four (100%) studies included transcriptomics as one of their layers. Two studies (50%) investigated genomics and transcriptomics [61, 62], one study

(25%) utilized transcriptomics and epigenomics [63], and the last study employed three omics layers: genomics, transcriptomics, and epigenomics [64]. Moreover, three studies (75%) analyzed tissue samples, while one (25%) used blood samples. Similarly, only one study (25%) employed biological validation, while the rest (75%) relied solely on statistical significance, which limits confidence that the findings reflect true biological mechanisms and potentially reduces their reproducibility (see Table S8 in the electronic supplementary material for details).

Collectively, the multi-omics findings converge to biomarker candidates related to oxidative stress, telomere regulation, ECM remodeling, and hormone signaling as central pathways in fibroids. Transcriptome–Mendelian randomization identified oxidative stress-related biomarkers (ANXA1, CD36, MICB, PRDX6) [61] as mediators of fibroid development. Large-scale genomic analyses revealed susceptibility loci linked to telomere maintenance (*TERT*, *TERC*, *OBFC1*, *TP53*, *ATM*) and hormone-regulated pathways (*WNT4*, *WT1*, *MED12*, *ESR1*, *FOXO1*) [64]. Integrative analyses also pinpointed *PTGS2* and *TACSTD2* as key genetic biomarkers connecting prostaglandin metabolism and epithelial adhesion to fibroid biology [63]. Subtype-specific markers further refined disease heterogeneity, where *ZMAT3*, a tumor-suppressor-associated gene that works downstream of p53, was upregulated across all fibroids [62]. Together, multi-omics integration mechanistically links genetic risk variants (e.g., *TERT*, *ESR1*) to downstream oxidative stress (e.g., ANXA1, PRDX6), telomere dysregulation (e.g., OBFC1), and hormone-responsive ECM deposition (e.g., PTGS2), revealing causal pathways from germline susceptibility through epigenomic/transcriptomic mediation to proteomic/metabolic phenotypes that sustain fibroid growth and persistence.

Biological Pathways Convergence: Systems-Level Insights into Fibroid Pathophysiology

To synthesize findings across the included studies, we mapped candidate biomarkers from genomics, epigenomics, transcriptomics, proteomics, metabolomics, and multi-omics investigations to their key biological pathways. Results showed a convergence in pathways related to hormone signaling, genome stability, cell cycle regulation, oxidative stress/metabolism, ECM remodeling, and apoptosis resistance. A Sankey plot (Fig. 3b) illustrates how each omics layer contributes to multiple pathways, emphasizing the systems-level dysregulation characteristic of uterine fibroids.

Genomic variants and epigenetic changes were most frequently linked to hormone signaling and genome stability. Transcriptomic and proteomic data converged strongly on cell cycle regulation, apoptosis resistance, and ECM remodeling, reflecting proliferative and survival phenotypes. Proteomic and metabolomic studies highlighted oxidative stress and metabolic reprogramming.

DISCUSSION

To our knowledge, this review is the first systematic synthesis focused specifically on omics-based biomarker discovery in uterine fibroids. By consolidating findings across genomics, epigenomics, transcriptomics, proteomics, metabolomics, and multi-omics studies, a comprehensive overview of molecular candidates and pathways implicated in fibroid pathogenesis was provided. Unlike previous narrative reviews that have separately highlighted genetic or molecular alterations, our study integrates evidence from multiple omics layers, thereby offering an updated landscape of candidate biomarkers and mechanisms underlying fibroids development. Importantly, this integrative perspective reframes fibroid biomarker discovery as a translational tool to support non-invasive diagnosis, molecular subtyping, and the rational development of targeted medical therapies beyond surgical management.

Six primary pathways consistently emerged across multiple omics platforms, demonstrating a cross-layer validation in fibroid biomarker research. ECM remodeling has emerged as a common pathway, present across nearly all omics layers. Transcriptomic analyses

revealed upregulation of multiple collagen genes (*COL3A1*, *COL11A2*, *COL4A5*, *COL4A6*), while proteomic investigations detected altered expression of lumican and other ECM-related proteins. From a novel therapeutics view, modulators of transforming growth factor (TGF)-β signaling, which regulate ECM deposition, have been investigated in preclinical studies and show promise in reducing fibroid size and proliferation [65]. Functional studies using in vitro and animal models have validated the role of specific ECM proteins, such as collagen and lumican, in promoting fibroid cell proliferation and ECM accumulation, supporting their relevance as both mechanistic biomarkers and potential intervention points [66–69]. The cross-layer consistency suggests that ECM dysregulation has a translational potential and represents a fundamental pathophysiological mechanism driving fibroid growth and tissue architecture changes, extending beyond simple structural alterations to encompass complex regulatory networks governing cellular adhesion, migration, and differentiation.

Hormone signaling pathways have also demonstrated cross-omics validation, appearing in genomic studies through *ESR1* (estrogen receptor alpha) and *LIN28B* variants, in epigenomic investigations via hormone-responsive gene methylation patterns, and in multi-omics analyses that identify *ESR1*, *FOXO1*, and related hormone-regulated pathways. This convergence highlights the well-established role of hormonal regulation in fibroid biology, while also revealing previously unrecognized layers of hormonal control through genetic predisposition, epigenetic modification, and metabolic reprogramming. Clinically, these insights align with the efficacy of hormone-targeting therapies such as gonadotropin-releasing hormone (GnRH) antagonists and selective progesterone receptor modulators, which shrink fibroid volume and reduce bleeding by suppressing estrogen–progesterone signaling [70, 71]. Functional studies in fibroid cells and animal models show that blocking progesterone and estrogen receptors reduces proliferation, induces apoptosis, and downregulates ECM-related and inflammatory genes [70–73]. Supporting the translational value of targeting hormonal pathways are the phase III clinical trials of relugolix combination therapy. This oral GnRH antagonist regimen has demonstrated significant symptom control and fibroid volume reduction while substantially lowering overall healthcare costs compared with standard care, offering a medical alternative to invasive procedures [74].

The consistent identification of cell cycle regulation and proliferation pathways across omics layers, from genomic variants in *WT1* and *YEATS4* to transcriptomic upregulation of *CCNB2*, *BIRC5*, and *FOXM1*, to proteomic alterations in chaperone proteins, provides mechanistic insight into the proliferative advantage characteristic of fibroids. From a translational perspective, this convergence supports pharmacologic targeting of proliferative signaling, as experimental inhibition of dysregulated chaperones reduces leiomyoma cell growth and promotes apoptosis [54]. Although CDK4/6 inhibitors such as palbociclib have so far been evaluated mainly in leiomyosarcoma and other sarcomas, where they induce G1 arrest and reduce proliferation via RB-dependent blockade of the CDK4/6–RB pathway, these agents provide a proof-of-concept for pharmacologic targeting of cyclin–CDK-driven cell cycle dysregulation that could be repurposed or adapted for high-proliferation fibroid subtypes in future trials [75–77].

Similarly, apoptosis and cell death pathway dysregulation, evidenced by *YEATS4* and *ZNHIT1* genomic alterations, *CASP1* transcriptomic changes, and TRADD proteomic downregulation, suggests that fibroid persistence results from coordinated resistance to programmed cell death across multiple regulatory levels. Translationally, several agents have been shown to partially reverse this apoptosis resistance: selective progesterone receptor modulators such as ulipristal acetate and asoprisnil increase cleaved caspase-3, reduce BCL-2 expression, and significantly raise apoptosis indices in fibroid cells and myoma tissue, in parallel with reductions in fibroid volume and vascularity [78, 79]. In addition, small molecules such as fisetin, EGCG, and anthocyanin-rich strawberry extracts induce caspase-dependent apoptosis and cell cycle arrest in leiomyoma cells, supporting pro-apoptotic pathways and markers such as CASP1 and TRADD as

mechanistic biomarkers and promising targets for future anti-fibroid therapies [80–82].

The emergence of oxidative stress and metabolic pathways across metabolomic studies and multi-omics analyses (including genes *ANXA1*, *CD36*, *MICB*, and *PRDX6*) indicates that fibroids may exert systemic metabolic effects extending beyond local uterine tissue. These insights support antioxidant and metabolic-targeted strategies. Clinical and preclinical data show that vitamin D and polyphenol-rich compounds such as green tea extract and fisetin can reduce fibroid burden and improve oxidative stress markers in leiomyoma cells [81, 83, 84]. In line with this, an in vivo leiomyoma model in Wistar rats demonstrated that vitamin D supplementation attenuated oxidative stress, reinforcing the potential of targeting oxidative-metabolic imbalance as a disease-modifying strategy in fibroids [85].

Finally, this review also showed a consistent alteration in genome stability and DNA repair mechanisms (*OBFC1*, *TERT*, *TERC*, *TP53*, *ATM*), suggesting that fibroid pathogenesis involves fundamental disruptions to cellular homeostasis that may have broader physiological consequences. Although no DNA-repair-targeted drugs are yet approved specifically for fibroids, preclinical work in uterine fibroid smooth muscle cells indicates that mTOR inhibition (e.g., rapamycin) and senolytic or telomerase-modulating strategies can influence telomere maintenance, growth arrest, and senescence, highlighting genome-stability pathways as an emerging therapeutic axis that may be exploitable using agents already in development for other proliferative and DNA-damage-driven disorders [86]

From another translational perspective, the recurrence of specific biomarker candidates across omics layers underscores the opportunity to move toward composite biomarker panels rather than single-molecule approaches. When integrated, these markers could enhance diagnostic accuracy, stratify fibroid subtypes, and provide a foundation for tailoring interventions to individual patients. Such an approach aligns directly with the goals of precision medicine, offering the potential not only to improve detection but also to guide prognosis and therapy in a condition that has long been dominated by surgical management. The clinical importance of biomarker development is further underscored by the limitations of current treatment approaches. Many available therapies are now symptom-directed rather than disease-modifying, and both surgical and hormonal options may be incompatible with reproductive goals for a substantial proportion of patients [14, 15]. Non-invasive biomarkers could therefore support earlier intervention, monitoring of progression, and fertility-preserving management strategies rather than reliance on late-stage surgical treatment.

Although this review presents valuable insights, the findings reveal a significant gap between current research practice and clinical needs, particularly regarding the sample types used. Our findings reveal a significant disconnect between the predominant use of tissue samples (58% of studies) and the clinical need for non-invasive diagnostic approaches. This gap reflects fundamental challenges in biomarker translation that must be systematically addressed. The biological validity question centers on whether tissue-derived biomarkers accurately reflect systemic disease signatures detectable in circulation. For fibroids, emerging evidence suggests the disease may have systemic metabolic and vascular manifestations [87, 88], indicating that circulating biomarkers may indeed capture meaningful disease signatures. The technical translation challenge involves developing assays that can detect molecular changes and biomarkers in the more dilute, heterogeneous environment of blood or urine. This requires not only analytical sensitivity improvements but also an understanding of which molecular classes (proteins, metabolites, circulating nucleic acids) best preserve disease-specific signatures during transition from tissue to circulation.

There also remains a lack of true multi-omics integration, with most studies analyzing individual omics layers in isolation. This reduces the potential to uncover synergistic mechanisms or cross-validated biomarkers. Also, validation strategies were inconsistent across studies, with many relying solely on statistical associations rather than functional or experimental confirmation, thereby limiting the robustness of proposed biomarkers. This is specifically important when interpreting the candidate variants of genomics. While many genomics findings originate from statistical association studies, several loci demonstrate progression toward biological relevance through functional annotation

and cross-omics concordance. In particular, eQTL overlap, expression concordance with transcriptomic datasets, and replication across independent cohorts support the likelihood that a subset of susceptibility loci represents biologically meaningful candidates rather than isolated statistical signals. Nevertheless, only a limited number of variants have undergone direct experimental validation, and most remain candidate biomarkers requiring further functional and clinical confirmation. An additional consideration is the potential overlap of study populations across large association analyses. Some investigations build upon shared cohorts or previously published datasets, meaning reported loci may not represent entirely independent discoveries. Consequently, recurrence of specific variants across studies should be interpreted as supportive rather than strictly independent evidence, highlighting the need for prospective validation in well-characterized populations.

A major strength of this review lies in its comprehensive, omics-wide approach, which enabled the integration of findings across genomics, epigenomics, transcriptomics, proteomics, and metabolomics and facilitated the identification of recurrent molecular pathways implicated in uterine fibroid biology. An additional strength is the strict adherence to PRISMA and QUADOMICS criteria, ensuring methodological rigor, transparency, and consistency in study identification, selection, and quality assessment. Despite these strengths, several limitations must be acknowledged. First, the small number of included studies may limit the generalizability of results. Another key limitation is methodological heterogeneity across included studies, including diverse omics platforms (GWAS vs. WES vs. SNP arrays), biological sample types (tissue, blood, saliva), study designs (discovery vs. validation), and analytical approaches (unbiased vs. targeted). This heterogeneity prevented quantitative meta-analysis, restricting the ability to derive pooled estimates of biomarker performance. Additionally, many candidate biomarkers were identified through association-based analyses without independent validation cohorts, reducing immediate clinical applicability. In addition, population ancestry differences across studies may influence genetic findings and biomarker generalizability. Therefore, while convergent biological pathways were consistently observed, individual biomarkers should be interpreted cautiously pending standardized validation studies. Finally, the additional targeted search specifically evaluated the impact of biomarker-related terminology on genomics studies, where association analyses frequently omit such terminology. Although other omics fields more commonly describe findings using molecular measurement terms, we cannot exclude the possibility that some studies across other omics layers were not captured because of differences in terminology.

Looking forward, the integration of multi-omics holds the greatest promise for advancing biomarker discovery in fibroids. Future work should include larger cohort studies, the inclusion of circulating matrices (plasma/urine) with appropriate normalization, and externally validated models that report sensitivity, specificity, and calibration. Combining genetic risk variants, epigenetic signatures, protein expression patterns, and metabolic alterations could enable early disease detection, patient stratification, and the development of tailored interventions within a precision medicine framework. Nevertheless, many critical questions remain unanswered, including (1) which multi-omics biomarkers can be validated to reliably detect uterine fibroids, predict clinical outcomes, or be translated to a novel therapeutic; (2) whether non-invasive circulating biomarkers accurately reflect tissue-level disease biology; and (3) whether non-invasive circulating biomarkers accurately reflect the molecular and functional heterogeneity of fibroids observed in tissue. Large, well-designed prospective studies are needed to validate candidate biomarkers and establish causality through longitudinal designs. Integrative multi-omics pipelines should be adopted to better capture the complexity of fibroid biology, while prioritizing non-invasive sampling approaches to enhance translational potential. Importantly, future research should also incorporate diverse populations to account for ethnic variability in fibroid prevalence and biology.

CONCLUSION

This systematic review synthesized current evidence from high-throughput omics studies investigating candidate biomarkers in uterine fibroids and provided an integrated overview of the emerging molecular landscape of the disease. Across genomics, epigenomics, transcriptomics, proteomics, metabolomics, and multi-omics investigations, convergent biological pathways—including ECM remodeling, hormone signaling, cell cycle regulation, oxidative stress, genome stability, and apoptosis resistance—were consistently implicated. The recurrence of these pathways across multiple molecular layers supports a systems-level view of uterine fibroids. Although most identified biomarkers remain at an early discovery stage and methodological heterogeneity persists, the collective findings highlight the potential of omics-based approaches to advance understanding of fibroid pathophysiology and to inform future biomarker development. Future research should focus on large, well-characterized cohorts, incorporate circulating biological matrices, and adopt integrated multi-omics pipelines coupled with functional validation to enable the development of clinically actionable biomarker panels. Such efforts are essential to bridge the gap between discovery-level findings and biomarker-guided precision medicine approaches that enable personalized, nonsurgical management of uterine fibroids.

Author Contributions. Fatimah Hussein and Mira Mousa were involved in all aspects of the study: conception, study design, data acquisition, information extraction, quality assessment, analysis, drafting the manuscript, and final approval. Ola Elamin was involved in the study design and data acquisition. Ayman Al-Hendy and Mira Mousa were involved in the conception, manuscript review, and final approval. All authors approved the submitted version.

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Data Availability. The datasets generated during and/or analyzed during the current study are available from the corresponding author on reasonable request.

Declarations

Conflict of Interest. All authors (Fatimah Hussein, Ola Elamin, Ayman Al-Hendy, and Mira Mousa) declare that they have no competing interests.

Ethical Approval. This article is based on previously conducted studies and does not contain any new studies with human participants or animals performed by any of the authors.

REFERENCES

1. Yang Q, Ciebiera M, Bariani MV, et al. Comprehensive review of uterine fibroids: developmental origin, pathogenesis, and treatment. Endocr Rev. 2022. https://doi.org/10.1210/endrev/bnab039.

2. Stewart EA, Laughlin-Tommaso SK, Catherino WH, Lalitkumar S, Gupta D, Vollenhoven B. Uterine

fibroids. Nat Rev Dis Primers. 2016. https://doi.org/10.1038/nrdp.2016.43.

3. Marsh EE, Al-Hendy A, Kappus D, Galitsky A, Stewart EA, Kerolous M. Burden, prevalence, and treatment of uterine fibroids: a survey of U.S. women. J Womens Health (Larchmt). 2018;27(11):1359–67. https://doi.org/10.1089/jwh.2018.7076.

4. Al-Hendy A, Myers ER, Stewart E. Uterine fibroids: burden and unmet medical need. Semin Reprod Med. 2017;35(6):473–80. https://doi.org/10.1055/s-0037-1607264.

5. Mukherjee P. Genetic and biomarker approaches to uterine fibroids: toward precision medicine. Front Glob Womens Health. 2025. https://doi.org/10.3389/fgwh.2025.1581823.

6. Orellana M, Riggan KA, DSouza K, et al. Perceptions of ethnoracial factors in the management and treatment of uterine fibroids. J Racial Ethn Health Disparities. 2022;9(4):1184–91. https://doi.org/10.1007/s40615-021-01059-8.

7. Orellana M, DSouza KN, Yap JQ, et al. "In our community, we normalize pain": discussions around menstruation and uterine fibroids with Black women and Latinas. BMC Womens Health. 2024. https://doi.org/10.1186/s12905-024-03008-z..

8. Levy G, Hill MJ, Plowden TC, Catherino WH, Armstrong AY. Biomarkers in uterine leiomyoma. Fertil Steril. 2013. https://doi.org/10.1016/j.fertnstert.2012.10.048.

9. Goad J, Rajkovic A. Uterine fibroids at single-cell resolution: unveiling cellular heterogeneity to improve understanding of pathogenesis and guide future therapies. Am J Obstet Gynecol. 2025. https://doi.org/10.1016/j.ajog.2024.08.037.

10. Tran AP, Al Naber J, Tambor ES, Myers ER. Addressing heterogenous outcomes in uterine fibroid research: a call to action. Am J Obstet Gynecol. 2020;223(1):75.e1-75.e5. https://doi.org/10.1016/j.ajog.2020.03.018.

11. Bulun SE, Yin P, Wei JJ, et al. Uterine fibroids. Physiol Rev. 2025. https://doi.org/10.1152/physrev.00010.2024.

12. Madueke-Laveaux OS, Elsharoud A, Al-Hendy A. What we know about the long-term risks of hysterectomy for benign indication—a systematic review. J Clin Med. 2021. https://doi.org/10.3390/jcm10225335.

13. Sánchez Martín MJ, Huerga López C, Cristóbal García I, Cristóbal Quevedo I. Efficacy of GnRH antagonists in the treatment of uterine fibroids: a meta-analysis. Arch Gynecol Obstet. 2025;311(3):685–96. https://doi.org/10.1007/s00404-025-07932-9.

14. Don EE, Mijatovic V, Huirne JAF. Infertility in patients with uterine fibroids: a debate about the hypothetical mechanisms. Hum Reprod. 2023. https://doi.org/10.1093/humrep/dead194.

15. Babalola O, Gebben D, Tarver ME, Sangha R, Roberts J, Price V. Patient preferences regarding surgical treatment methods for symptomatic uterine fibroids. Ther Innov Regul Sci. 2023;57(5):976–86. https://doi.org/10.1007/s43441-023-00525-1.

16. Chen C, Wang J, Pan D, et al. Applications of multi-omics analysis in human diseases. MedComm. 2023. https://doi.org/10.1002/mco2.315.

17. Hu ZZ, Huang H, Wu CH, et al. Omics-based molecular target and biomarker identification. Methods Mol Biol. 2011. https://doi.org/10.1007/978-1-61779-027-0_26.

18. Cai L, Liao Z, Li S, et al. PLP1 may serve as a potential diagnostic biomarker of uterine fibroids. Front Genet. 2022. https://doi.org/10.3389/fgene.2022.1045395.

19. Jacob M, Richter R, Sehouli J, David M. Evaluation of biomarkers in myoma patients: a prospective study investigating the role of LDH, CA 125, and IGF-1 after uterus-preserving surgical therapy. Gynecol Obstet Invest. 2021;86(1–2):100–7. https://doi.org/10.1159/000513045.

20. Cai L, Li J, Long R, et al. An autophagy-related diagnostic biomarker for uterine fibroids: FOS. Front Med. 2023. https://doi.org/10.3389/fmed.2023.1153537.

21. Jamaluddin MFB, Ko YA, Kumar M, et al. Proteomic profiling of human uterine fibroids reveals upregulation of the extracellular matrix protein periostin. Endocrinology. 2018;159(2):1106–18. https://doi.org/10.1210/en.2017-03018.

22. Boldu-Fernández S, Lliberos C, Simon C, Mas A. Mapping human uterine disorders through single-cell transcriptomics. Cells. 2025. https://doi.org/10.3390/cells14030156.

23. Song J, Wang C, Zhao T, et al. Multi-omics approaches for biomarker discovery and precision diagnosis of prediabetes. Front Endocrinol. 2025. https://doi.org/10.3389/fendo.2025.1520436.

24. Amendola I, Spann M, Segars J, Singh B. The mediator complex subunit 12 (MED-12) gene and uterine fibroids: a systematic review. Reprod Sci. 2024. https://doi.org/10.1007/s43032-023-01297-7.

25. Page MJ, Moher D, Bossuyt PM, et al. PRISMA explanation and elaboration: updated guidance and exemplars for reporting systematic reviews. The BMJ. 2020. https://doi.org/10.1136/bmj.n160.

26. Lumbreras B, Porta M, Márquez S, Pollán M, Parker LA, Hernández-Aguado I. QUADOMICS: an adaptation of the quality assessment of diagnostic accuracy assessment (QUADAS) for the evaluation of the methodological quality of studies on the diagnostic accuracy of '-omics'-based technologies. Clin Biochem. 2008;41(16–17):1316–25. https://doi.org/10.1016/j.clinbiochem.2008.06.018.

27. Kim J, Williams A, Noh H, et al. Genome-wide meta-analysis identifies novel risk loci for uterine fibroids within and across multiple ancestry groups. Nat Commun. 2025. https://doi.org/10.1038/s41467-025-57483-5.

28. Edwards TL, Giri A, Hellwege JN, et al. A trans-ethnic genome-wide association study of uterine fibroids. Front Genet. 2019. https://doi.org/10.3389/fgene.2019.00511.

29. Välimäki N, Jokinen V, Cajuso T, et al. Inherited mutations affecting the SRCAP complex are central in moderate-penetrance predisposition to uterine leiomyomas. Am J Hum Genet. 2023;110(3):460–74. https://doi.org/10.1016/j.ajhg.2023.01.009.

30. Liu B, Wang T, Jiang J, et al. Association of BET1L and TNRC6B with uterine leiomyoma risk and its relevant clinical features in Han Chinese population. Sci Rep. 2018. https://doi.org/10.1038/s41598-018-25792-z.

31. Ponomarenko I, Reshetnikov E, Polonikov A, et al. Candidate genes for age at menarche are associated with uterine leiomyoma. Front Genet. 2021. https://doi.org/10.3389/fgene.2020.512940.

32. Wise LA, Ruiz-Narváez EA, Haddad SA, Rosenberg L, Palmer JR. Polymorphisms in vitamin D-related genes and risk of uterine leiomyomata. Fertil Steril. 2014. https://doi.org/10.1016/j.fertnstert.2014.04.037.

33. Dzhemlikhanova LK, Efimova OA, Osinovskaya NS, et al. Catechol-O-methyltransferase Val158Met polymorphism is associated with increased risk of multiple uterine leiomyomas either positive or negative for MED12 exon 2 mutations. J Clin Pathol. 2017;70(3):233–6. https://doi.org/10.1136/jclinpath-2016-203976.

34. Govindan S, Shaik NA, Vedicherla B, Kodati V, Rao KP, Hasan Q. Estrogen receptor-α gene (T/C) Pvu II polymorphism in endometriosis and uterine fibroids. Dis Markers. 2009;26(4):149–54. https://doi.org/10.3233/DMA-2009-0625.

35. Amant F, Dorfling CM, De Brabanter J, et al. A possible role of the cytochrome P450c17α gene (CYP17) polymorphism in the pathobiology of uterine leiomyomas from black South African women: a pilot study. Acta Obstet Gynecol Scand. 2004;83(3):234–9. https://doi.org/10.1111/j.1600-0412.2004.00422.x.

36. Shaik Ahmad N, Govindan S, Kodati V, Prabhakar Rao K, Hasan Q. Polymorphic (CAG)n repeats in the androgen receptor gene: a risk marker for endometriosis and uterine leiomyomas. Hematol Oncol Stem Cell Ther. 2009. https://doi.org/10.1016/S1658-3876(09)50040-4.

37. Cha PC, Takahashi A, Hosono N, et al. A genome-wide association study identifies three loci associated with susceptibility to uterine fibroids. Nat Genet. 2011;43(5):447–51. https://doi.org/10.1038/ng.805.

38. Rafnar T, Gunnarsson B, Stefansson OA, et al. Variants associating with uterine leiomyoma highlight genetic background shared by various cancers and hormone-related traits. Nat Commun. 2018. https://doi.org/10.1038/s41467-018-05428-6.

39. Elsaid AM, Elsayed YM, Abu Almaaty AH, Eid MI, Hasan AK. Cycloxygenase-2 (–765G>C) gene polymorphism and risk of uterine fibroids in Egyptian women. Gene Rep. 2026;1:42. https://doi.org/10.1016/j.genrep.2025.102417.

40. Hellwege JN, Jeff JM, Wise LA, et al. A multistage genome-wide association study of uterine fibroids in African Americans. Hum Genet. 2017;136(10):1363–73. https://doi.org/10.1007/s00439-017-1836-1.

41. Sato S, Maekawa R, Yamagata Y, Asada H, Tamura I, Lee L, et al. Potential Mechanisms of Aberrant DNA Hypomethylation on the X Chromosome in Uterine Leiomyomas. J Reprod Dev. 2014;60(1):31–6. https://doi.org/10.1262/jrd.2013-095.

42. Sato S, Maekawa R, Yamagata Y, et al. Identification of uterine leiomyoma-specific marker genes based on DNA methylation and their clinical application. Sci Rep. 2016;6:6. https://doi.org/10.1038/srep30652.

43. Maekawa R, Yagi S, Ohgane J, et al. Disease-dependent differently methylated regions (D-DMRs) of DNA are enriched on the X chromosome in uterine leiomyoma. J Reprod Dev. 2011. https://doi.org/10.1262/jrd.11-035a.

44. Wang W, Zhou L, Wang J, Zhang X, Liu G. Circular RNA expression profiling identifies novel biomarkers in uterine leiomyoma. Cell Signal. 2020;76:76. https://doi.org/10.1016/j.cellsig.2020.109784.

45. Fazeli E, Piltan S, Sadeghi H, et al. Ectopic expression of CYP24A1 circular RNA hsa_circ_0060927 in uterine leiomyomas. J Clin Lab Anal. 2020. https://doi.org/10.1002/jcla.23114.

46. Litovkin KV, Ivanova OV, Bauer A, Hoheisel JD, Bubnov VV, Zaporozhan VN. Microarray study of gene expression in uterine leiomyoma. Exp Oncol. 2008;30(2):106–11.

47. Peng X, Mo Y, Liu J, Liu H, Wang S. Identification and validation of miRNA-TF-mRNA regulatory networks in uterine fibroids. Front Bioeng Biotechnol. 2022. https://doi.org/10.3389/fbioe.2022.856745.

48. Liu Y, Liu X, Liu Y, Zhao J. Screening of potential biomarkers in uterine leiomyomas disease via gene expression profiling analysis. Mol Med Rep. 2018;17(5):6985–96. https://doi.org/10.3892/mmr.2018.8756.

49. Yang E, Xue L, Li Z, Yi T. Lnc-AL445665.1–4 may be involved in the development of multiple uterine leiomyoma through interacting with miR-146b-5p. BMC Cancer. 2019. https://doi.org/10.1186/S12885-019-5775-1.

50. Guo H, Zhang X, Dong R, et al. Integrated analysis of long noncoding RNAs and mRNAs reveals their potential roles in the pathogenesis of uterine leiomyomas. Oncotarget. 2014;5(18):8625–36. https://doi.org/10.18632/oncotarget.2349.

51. Okeigwe I, Bulun S, Liu S, et al. PLIN2 functions as a novel link between progesterone signaling and metabolism in uterine leiomyoma cells. J Clin Endocrinol Metab. 2019;104(12):6256–64. https://doi.org/10.1210/jc.2019-00762.

52. Chuang TD, Khorram O. Expression profiling of lncRNAs, miRNAs, and mRNAs and their differential expression in leiomyoma using next-generation RNA sequencing. Reprod Sci. 2018;25(2):246–55. https://doi.org/10.1177/1933719117711265.

53. Chuang TD, Rehan A, Khorram O. Functional role of the long noncoding RNA X-inactive specific transcript in leiomyoma pathogenesis. Fertil Steril. 2021;115(1):238–47. https://doi.org/10.1016/j.fertnstert.2020.07.024.

54. Ura B, Scrimin F, Arrigoni G, Aloisio M, Monasta L, Ricci G. Dysregulated chaperones associated with cell proliferation and negative apoptosis regulation in the uterine leiomyoma. Oncol Lett. 2018;15(5):8005–10. https://doi.org/10.3892/ol.2018.8325.

55. Liu Y, Lu D, Sheng J, Luo L, Zhang W. Identification of TRADD as a potential biomarker in human uterine leiomyoma through iTRAQ based proteomic profiling. Mol Cell Probes. 2017;36:15–20. https://doi.org/10.1016/j.mcp.2017.07.001.

56. Sahar T, Nigam A, Anjum S, Gupta N, Wajid S. Secretome profiling and computational biology of human leiomyoma samples unravel molecular signatures with potential for diagnostic and therapeutic interventions. Reprod Sci. 2021. https://doi.org/10.1007/s43032-021-00580-9.

57. Lin CP, Chen YW, Liu WH, et al. Proteomic identification of plasma biomarkers in uterine leiomyoma. Mol Biosyst. 2012;8(4):1136–45. https://doi.org/10.1039/c2mb05453a.

58. Tonoyan NM, Chagovets VV, Starodubtseva NL, et al. Alterations in lipid profile upon uterine fibroids and its recurrence. Sci Rep. 2021. https://doi.org/10.1038/s41598-021-89859-0.

59. Heinonen HR, Mehine M, Mäkinen N, et al. Global metabolomic profiling of uterine leiomyomas. Br J Cancer. 2017;117(12):1855–64. https://doi.org/10.1038/bjc.2017.361.

60. Barison GAS, D'amora P, Izidoro MA, et al. Metabolomic profiling of peripheral plasma by GC-MS and correlation with size of uterine leiomyomas. J Endocr Soc. 2022. https://doi.org/10.1210/jendso/bvac061.

61. Li Y, Chen H, Zhang H, Lin Z, Song L, Zhao C. Identification of oxidative stress-related biomarkers in uterine leiomyoma: a transcriptome-combined Mendelian randomization analysis. Front Endocrinol (Lausanne). 2024. https://doi.org/10.3389/fendo.2024.1373011.

62. Mehine M, Kaasinen E, Heinonen HR, et al. Integrated data analysis reveals uterine leiomyoma subtypes with distinct driver pathways and biomarkers. Proc Natl Acad Sci U S A. 2016;113(5):1315–20. https://doi.org/10.1073/pnas.1518752113.

63. Mallik S, Maulik U. MiRNA-TF-gene network analysis through ranking of biomolecules for multi-informative uterine leiomyoma dataset. J Biomed Inform. 2015;57:308–19. https://doi.org/10.1016/j.jbi.2015.08.014.

64. Välikmäki N, Kuisma H, Pasanen A, Heikinheimo O, Sjöberg J. Genetic predisposition to uterine leiomyoma is determined by loci for genitourinary development and genome stability. Elife. 2018. https://doi.org/10.7554/eLife.37110.

65. Obeagu EI, Obeagu GU. Unraveling the role of transforming growth factor-beta (TGF-β) in the progression of fibroids. Elite J Med. 2024;2(2):67–77.

66. Sun T, Xiao C, Yang Z, Deng J, Yang X. Transcriptome profiling analysis of uterus during chicken laying periods. BMC Genomics. 2023. https://doi.org/10.1186/s12864-023-09521-z.

67. Islam MS, Afrin S, Singh B, et al. Extracellular matrix and Hippo signaling as therapeutic targets of antifibrotic compounds for uterine fibroids. Clin Transl Med. 2021. https://doi.org/10.1002/ctm2.475.

68. Yan Y, Liu J, Lawrence A, et al. Prolonged cadmium exposure alters benign uterine fibroid cell behavior, extracellular matrix components, and TGFB signaling. FASEB J. 2021. https://doi.org/10.1096/fj.202100354R.

69. Saad EE, Michel R, Borahay MA. Immunosuppressive tumor microenvironment and uterine fibroids: role in collagen synthesis. Cytokine Growth Factor Rev. 2024. https://doi.org/10.1016/j.cytogfr.2023.10.002.

70. Ali M, Ciebiera M, Vafaei S, et al. Progesterone signaling and uterine fibroid pathogenesis; molecular mechanisms and potential therapeutics. Cells. 2023. https://doi.org/10.3390/cells12081117.

71. Borahay MA, Asoglu MR, Mas A, Adam S, Kilic GS, Al-Hendy A. Estrogen receptors and signaling in fibroids: role in pathobiology and therapeutic implications. Reprod Sci. 2017;24(9):1235–44.

72. Rabe T, Saenger N, Ebert AD, et al. Selective progesterone receptor modulators for the medical treatment of uterine fibroids with a focus on ulipristal acetate. BioMed Res Int. 2018. https://doi.org/10.1155/2018/1374821.

73. Nair HB, Santhamma B, Dileep KV, et al. EC313-a tissue selective SPRM reduces the growth and proliferation of uterine fibroids in a human uterine fibroid tissue xenograft model. Sci Rep. 2019. https://doi.org/10.1038/s41598-019-53467-w.

74. Al-Hendy A, Lukes AS, Poindexter AN, et al. Treatment of uterine fibroid symptoms with relugolix combination therapy. N Engl J Med. 2021;384(7):630–42. https://doi.org/10.1056/nejmoa2008283.

75. Böhm MJ, Marienfeld R, Jäger D, et al. Analysis of the CDK4/6 cell cycle pathway in leiomyosarcomas as a potential target for inhibition by palbociclib. Sarcoma. 2019;2019:3914232. https://doi.org/10.1155/2019/3914232.

76. Francis AM, Alexander A, Liu Y, et al. CDK4/6 inhibitors sensitize Rb-positive sarcoma cells to Wee1 kinase inhibition through reversible cell-cycle arrest. Mol Cancer Ther. 2017;16(9):1751–64. https://doi.org/10.1158/1535-7163.MCT-17-0040.

77. Martin-Broto J, Martinez-Garcia J, Moura DS, et al. Phase II trial of CDK4/6 inhibitor palbociclib in advanced sarcoma based on mRNA expression of CDK4/ CDKN2A. Signal Transduct Target Ther. 2023. https://doi.org/10.1038/s41392-023-01661-8.

78. Biglia N, Carinelli S, Maiorana A, D'Alonzo M, Lo MG, Marci R. Ulipristal acetate: a novel pharmacological approach for the treatment of uterine fibroids. Drug Design Dev Ther. 2014. https://doi.org/10.2147/DDDT.S54565.

79. Horak P, Mara M, Dundr P, et al. Effect of a selective progesterone receptor modulator on induction of apoptosis in uterine fibroids in vivo. Int J Endocrinol. 2012;2012:436174. https://doi.org/10.1155/2012/436174.

80. Zhang D, Al-Hendy M, Richard-Davis G, Montgomery-Rice V, Rajaratnam V, Al-Hendy A. Antiproliferative and proapoptotic effects of epigallocatechin gallate on human leiomyoma cells. Fertil Steril. 2010;94(5):1887–93. https://doi.org/10.1016/j.fertnstert.2009.08.065.

81. Lee JW, Choi HJ, Kim EJ, Hwang WY, Jung MH, Kim KS. Fisetin induces apoptosis in uterine leiomyomas through multiple pathways. Sci Rep. 2020. https://doi.org/10.1038/s41598-020-64871-y.

82. Islam S, Giampieri F, Janjusevic M, et al. An anthocyanin rich strawberry extract induces apoptosis and ROS while decreases glycolysis and fibrosis in human uterine leiomyoma cells. Oncotarget. 2017;8(14):23575–87. https://doi.org/10.18632/oncotarget.15333

83. AlAshqar A, Lulseged B, Mason-Otey A, et al. Oxidative stress and antioxidants in uterine fibroids: pathophysiology and clinical implications. Antioxidants Basel. 2023. https://doi.org/10.3390/antiox12040807.

84. Ploumaki I, El Sayed S, Asamoah-Mensah A, Macri VI, Segars JH, Islam MS. The potential of vitamin D and epigallocatechin gallate (EGCG) for the treatment of uterine fibroids: evidence from in vitro to clinical studies. Mol Nutr Food Res. 2025. https://doi.org/10.1002/mnfr.70231.

85. Moyosola Olumide O, Olatunji-Bello I, Morounke Saibu G, et al. Vitamin D modulator of oxidative stress, pro-inflammatory cytokines and hormonal disruptions in monosodium glutamate (MSG)-induced uterine leiomyoma in wistar rat models. Afr J Biomed Res. 2025;28(3s):857–65. https://doi.org/10.53555/AJBR.v28i3S.7761.

86. Suo G, Sadarangani A, Tang W, Cowan BD, Wang JYJ. Telomerase expression abrogates rapamycin-induced irreversible growth arrest of uterine fibroid smooth

muscle cells. Reprod Sci. 2014;21(9):1161–70. https://doi.org/10.1177/1933719114532839.

87. Kirschen GW, AlAshqar A, Miyashita-Ishiwata M, Reschke L, El Sabeh M, Borahay MA. Vascular biology of uterine fibroids: connecting fibroids and vascular disorders. Reproduction. 2021. https://doi.org/10.1530/REP-21-0087.

88. Han S, Choi EK, Ahn HJ, et al. Increased risk of atrial fibrillation in uterine fibroid patients: a nationwide population-based study. Sci Rep. 2025. https://doi.org/10.1038/s41598-025-12954-z.