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ORIGINAL ARTICLE

Update on the proteomics of male infertility: A systemic review

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KEYWORDS

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Abstract ***Objective:*** To assess the role of differentially expressed proteins as a resource for potential biomarker identification of infertility, as male infertility is of rising concern in reproductive medicine and evidence pertaining to its aetiology at a molecular level particularly proteomic as spermatozoa lack transcription and translation. Proteomics is considered as a major field in molecular biology to validate the target proteins in a pathophysiological state. Differential expression analysis of sperm proteins in infertile men and bioinformatics analysis offer information about their involvement in biological pathways.

Materials and methods: Literature search was performed on PubMed, Medline, and Science Direct databases using the keywords 'sperm proteomics' and 'male infertility'. We also reviewed the relevant cross references of retrieved articles and included them in the review process. Articles written in any language other than English were excluded.

Results: Of 575 articles identified, preliminary screening for relevant studies eliminated 293 articles. At the next level of selection, from 282 studies only 80 articles related to male infertility condition met the selection criteria and were included in this review.

Conclusion: In this molecular era, sperm proteomics has created a platform for enhanced understanding of male reproductive physiology as a potential tool for identification of novel protein biomarkers related to sperm function in infertile

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men. Therefore, it is believed that proteomic biomarkers can overcome the gaps in information from conventional semen analysis that are of limited clinical utility.

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Introduction

Currently, infertility is one of the most addressed issues related to male reproductive dysfunction. Amongst the 9% of the world's infertility cases, ~20% is contributed by the male population alone [1]. There are multiple factors that govern and regulate male factor infertility, although most of these cases remain idiopathic. Andrology laboratories rely mainly on semen analysis to evaluate male infertility in patients with poor semen quality. Conventional tests such as basic semen analysis to determine sperm concentration, motility, vitality, and morphology are used for diagnosing male infertility based on reference values established by the WHO [2]. However, advanced laboratory tests such as quantification of reactive oxygen species (ROS)[1] and antioxidants in semen by chemiluminescence assay [3], oxidation–reduction potential in semen by Male Infertility Oxidative System (MiOXSYS®; Aytu BioScience Inc., Englewood, CO USA) [4], and sperm DNA fragmentation assessment by terminal deoxynucleotidyl transferase-mediated dUTP nick-end labelling (TUNEL) assay [5], are clinically used to identify the specific cause of infertility for further utilisation in assisted reproductive technology. However, the aetiological changes at the subcellular level of the spermatozoa remain unknown.

New generation techniques, such as proteomics, are poised to help researchers identify the molecular aspects of spermatozoa that are affected in infertility conditions. The majority of protein biomolecules are involved in cell signalling pathways. Generally, spermatozoa are transcriptionally and translationally silent, so they depend on their proteins to carry out their biological functions [6]. Proteomics is an emerging tool that could potentially help identify protein alterations in spermatozoa and seminal plasma of male infertile patients [7]. Differential expression of sperm proteins in fertility compromised patients is an indicator of defective spermatogenesis, motility, capacitation, hyperactivation, acrosome reaction, and fertilisation processes at a molecular level.

Aberrant expression of proteins in spermatozoa of men causes changes in physiological functions due to post-translational modifications, such as phosphorylation, glycosylation, proteolytic cleavage, and mutations [8,9]. Therefore, it is important to understand the changes in the proteins and cellular pathways affected in infertile patients for better diagnosis in a clinical perspective.

Materials and methods

An extensive search of studies published until October 2017 was performed using PubMed, MedLine, and Science Direct databases. The search was limited to full articles published in the English language and studies on human semen were only included. 'Sperm proteomics' and 'male infertility' were the two primary keywords used to retrieve articles from different databases. Combination of the following keywords relevant to infertility and proteomics was used to extract the articles: 'spermatozoa', 'sperm proteomics', 'varicocele proteomics', 'proteome', 'oxidative stress and proteomics', '2D-PAGE, mass spectrometry'. Search terms such as 'azoospermia', 'asthenozoospermia', 'mitochondrial dysfunction', 'testicular cancer and proteomics' were also used. Cross referencing was also referred to and used in the review process.

Results

Comprehensive literature collection via electronic search resulted in a total of 575 review and original research articles. Preliminary screening resulted in 282 articles that included different proteomic studies from human sperm (Fig. 1). In the subsequent screening, 206 studies were rejected as many ($n = 84$) were not related to high-throughput proteomics. Finally, 80 full-text articles (review, original research, and book chapters) met the inclusion criteria and were found to be eligible for the review.

[1] *Abbreviations:* ACPP, prostatic acid phosphatase/prostatic-specific acid phosphatase; ANXA7, annexin A7; AZGP1, zinc α2-glycoprotein 1; CLU, clusterin; CRISP(1)(3),cysteine-rich secretory protein (1) (3); CRISPLD(1)(2), CRISP LCCL domain-containing (1) (2); CST3, cystatin 3; 2D, two dimensional; DEP, differentially expressed protein; FN1, fibronectin 1; GO, gene ontology; HIST1H2BA, histone cluster 1 H2B family member A; HSP, heat shock protein; HSPA(2)(5), HSP family A (HSP70) member (2) (5); KLK3, kallikrein 3; LC–(MS), liquid chromatography–(mass spectrometry); LGALS3BP, galectin-3 binding protein; MALDI-TOF, matrix-assisted laser desorption ionisation time-of-flight; MIF, macrophage migration inhibitory factor; NSAF, normalised spectral abundance factor; (N)OA, (non-) obstructive azoospermia; OAT, oligoasthenozoospermia; ODF(1), outer dense fibre of sperm tails (1) (2); OS, oxidative stress; PAEP, progestogen-associated endometrial protein/glycodelin S; PGK2, phosphoglycerate kinase 2; PIP, prolactin induced protein; ROS, reactive oxygen species; SDS, sodium dodecyl sulphate; SEMG(1)(2), semenogelin (1) (2); SOD1, superoxide dismutase 1; SpC, spectral count; TEX101, testis-expressed protein 101; TUNEL, terminal deoxynucleotidyl transferase-mediated dUTP nick-end labelling; VDAC(2)(3), voltage-dependent anion channel (2) (3); ZPBP2, zona pellucida binding protein 2.

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