High Throughput LC-MS/MS Studies on Sperm Proteome of Malnad Gidda (Bos Indicus) Cattle

Authors : Kerekoppa Puttaiah Bhatta Ramesha, Uday Kannegundla, Praseeda Mol, Lathika Gopalakrishnan, Jagish Kour Reen, Gourav Dey, Manish Kumar, Sakthivel Jeyakumar, Arumugam Kumaresan, Kiran Kumar M., Thottethodi Subrahmanya Keshava Prasad

Abstract : Spermatozoa are the highly specialized transcriptionally and translationally inactive haploid male gamete. The understanding of proteome of sperm is indispensable to explore the mechanism of sperm motility and fertility. Though there is a large number of human sperm proteomic studies, in-depth proteomic information on Bos indicus spermatozoa is not well established yet. Therefore, we illustrated the profile of sperm proteome in indigenous cattle, Malnad gidda (Bos Indicus), using high-resolution mass spectrometry. In the current study, two semen ejaculates from 3 breeding bulls were collected employing the artificial vaginal method. Using 45% percoll purification, spermatozoa cells were isolated. Protein was extracted using lysis buffer containing 2% Sodium Dodecyl Sulphate (SDS) and protein concentration was estimated. Fifty micrograms of protein from each individual were pooled for further downstream processing. Pooled sample was fractionated using SDS-Poly Acrylamide Gel Electrophoresis, which is followed by in-gel digestion. The peptides were subjected to C18 Stage Tip clean-up and analyzed in Orbitrap Fusion Tribrid mass spectrometer interfaced with Proxeon Easy-nano LC II system (Thermo Scientific, Bremen, Germany). We identified a total of 6773 peptides with 28426 peptide spectral matches, which belonged to 1081 proteins. Gene ontology analysis has been carried out to determine the biological processes, molecular functions and cellular components associated with sperm protein. The biological process chiefly represented our data is an oxidation-reduction process (5%), spermatogenesis (2.5%) and spermatid development (1.4%). The highlighted molecular functions are ATP, and GTP binding (14%) and the prominent cellular components most observed in our data were nuclear membrane (1.5%), acrosomal vesicle (1.4%), and motile cilium (1.3%). Seventeen percent of sperm proteins identified in this study were involved in metabolic pathways. To the best of our knowledge, this data represents the first total sperm proteome from indigenous cattle, Malnad Gidda. We believe that our preliminary findings could provide a strong base for the future understanding of bovine sperm proteomics.

Keywords : Bos indicus, Malnad Gidda, mass spectrometry, spermatozoa

Conference Title : ICAGB 2018 : International Conference on Animal Genetics and Breeding

Conference Location : Amsterdam, Netherlands

Conference Dates : January 22-23, 2018