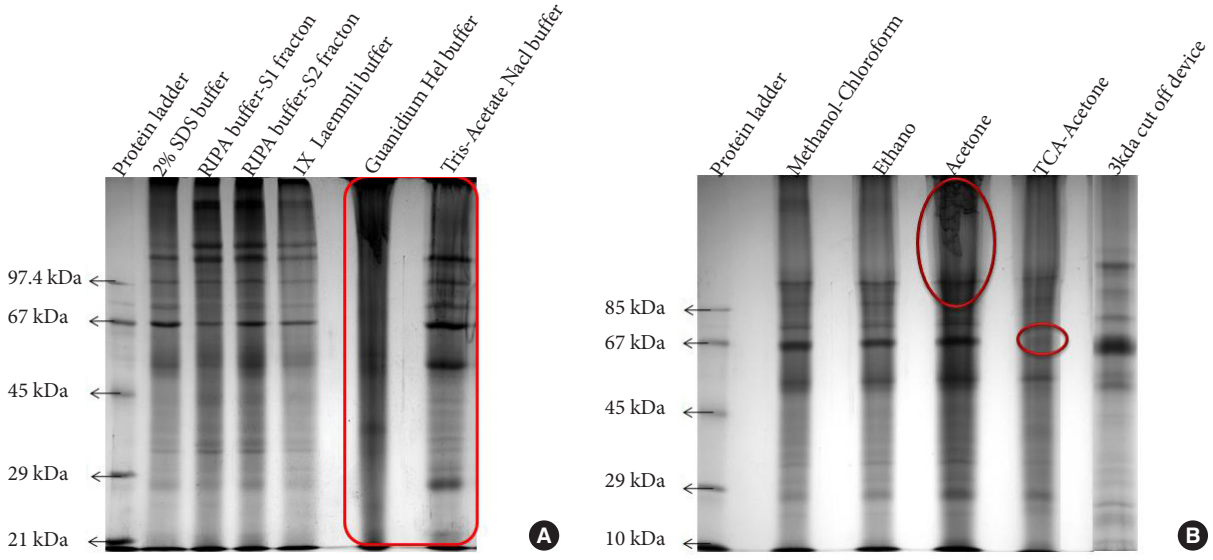
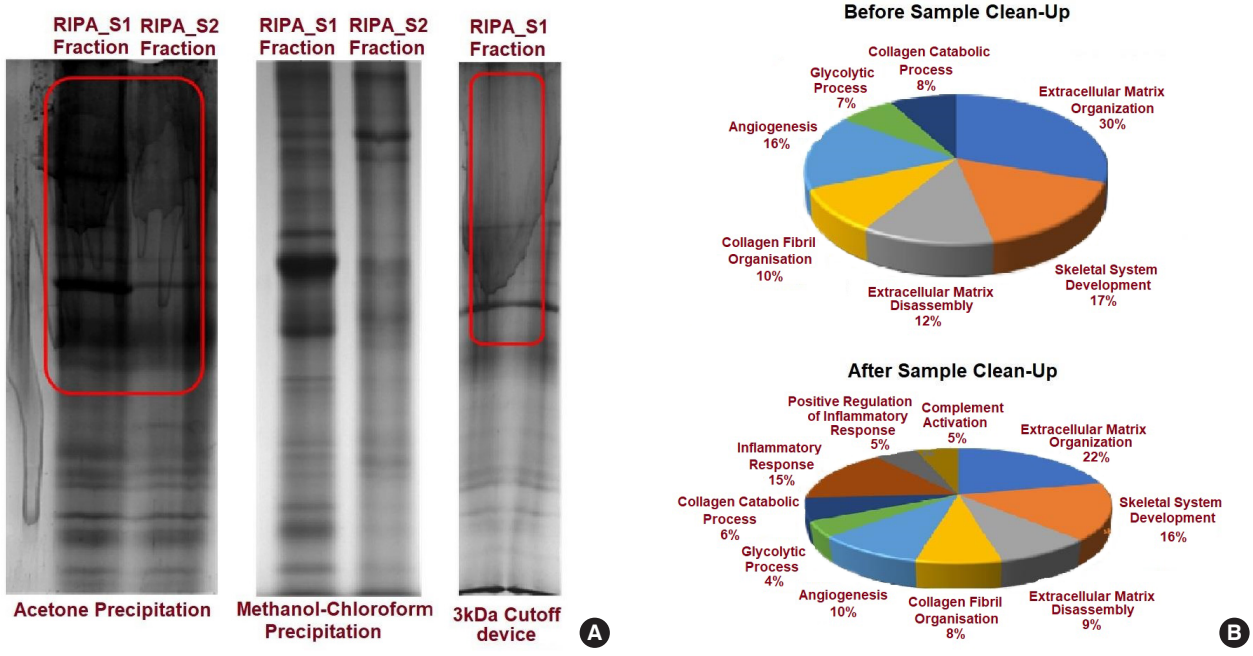


Supplementary Fig. 1. Comparative proteome profiling of nucleus pulposus observed during protein extraction and protein clean-up followed by prefractionation on 10% sodium dodecyl sulfate (SDS)-polyacrylamide gel electrophoresis. (A) Total protein extraction using various buffers: 2% SDS; radio immunoprecipitation assay buffer (RIPA) buffer (S1 fraction) + 2% SDS (S2 fraction); 1X laemmli buffer; guanidium-HCl buffer; tris-acetate buffer, red-colored circle indicates spillage pattern due to interfering glycans. (B) Sample clean-up using various organic solvents after extraction of proteins to remove the interfering glycans before proteome analysis by ESI-LC-MS/MS. The red circle indicates the interfering glycans which were not removed after precipitation with acetone. With tricholoroacetic acid (TCA)-acetone, the loss of proteins is shown with a circle in the respective lane. ESI-LC-MS/MS, electrospray ionization-liquid chromatography tandem mass spectrometry.



Supplementary Fig. 2. Comparative profile of the intervertebral disc nucleus pulposus tissue on a 10% sodium dodecyl sulfate-polyacrylamide gel electrophoresis after sample clean-up using: acetone; methanol-chloroform; 3-kDa membrane cutoff device. (A) Presence of interfering glycan's during prefractionation (circled in red), in acetone precipitation and 3 kDa, cutoff device compared to methanol-chloroform. (B) Biological processes of proteins identified before and after sample clean-up with methanol-chloroform organic solvent. RIPA, radio immunoprecipitation assay buffer.

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