Additional file 2. Supplementary figures 1 to 3

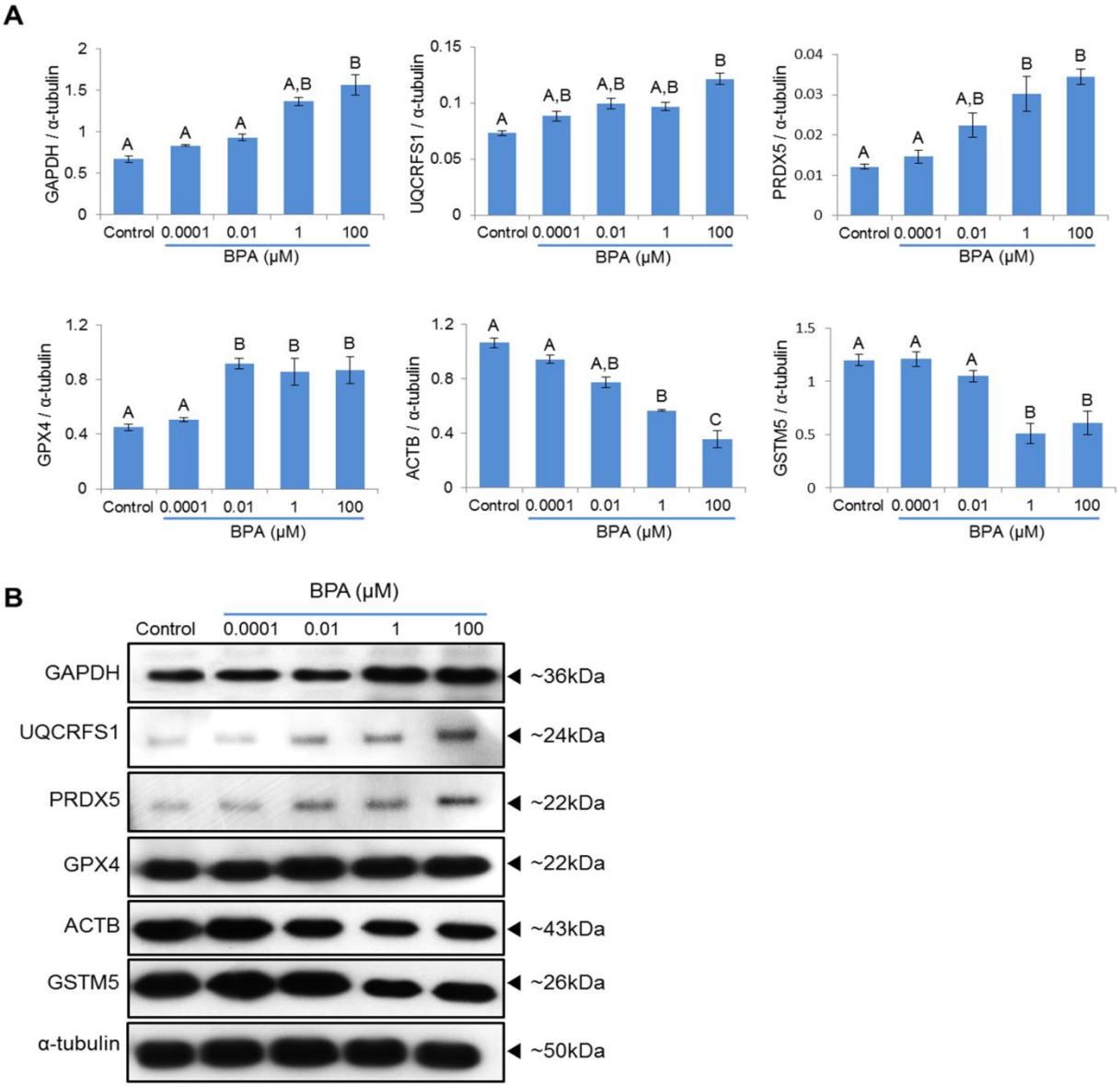

Fig. S1 Validation of the 2-DE results by western blot analysis. (**A**) The bars represent the densities of particular proteins bands, such as GAPDH, UQCRFS1, PRDX5, GPX4, ACTB, and GSTM5. Data are presented as mean ± SEM (n=3). Values with different superscript characters (^{A,B,C}) indicate significant differences between the control and treatment samples as determined by one-way ANOVA ($P < 0.05$). (**B**) Representative western blot image of GAPDH, UQCRFS1, PRDX5, GPX4, ACTB, and GSTM5 probed with the respective antibodies.

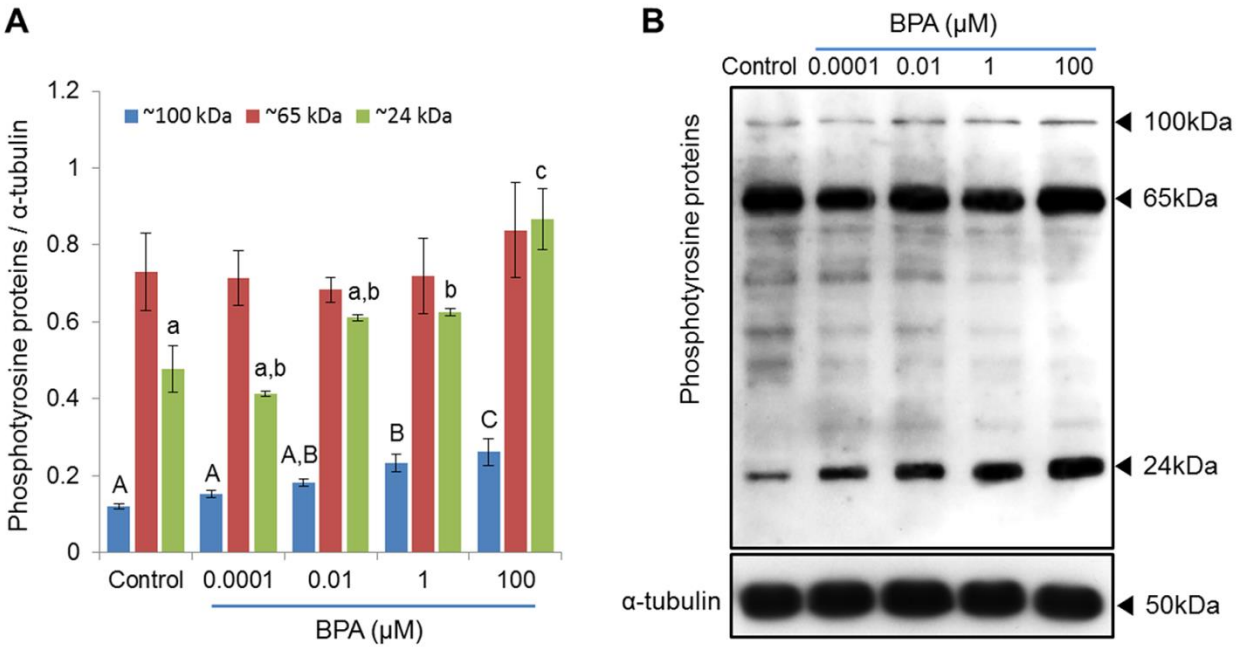

Fig. S2 Effect of BPA on tyrosine phosphorylation status of spermatozoa. (**A**) Density of phosphotyrosine proteins in control and BPA-treated samples. Data are the mean of four replicates ± SEM. Values with different upper and lower case letters (^{A,B,C,a,b,c}) are significantly different between control and treatment groups by one-way ANOVA. (**B**) Tyrosine-phosphorylated proteins were probed with PY 20. Lane 1: control; lane 2: 0.0001 µM BPA; lane 3: 0.01 µM BPA; lane 4: 1 µM BPA; and lane 5: 100 µM BPA.

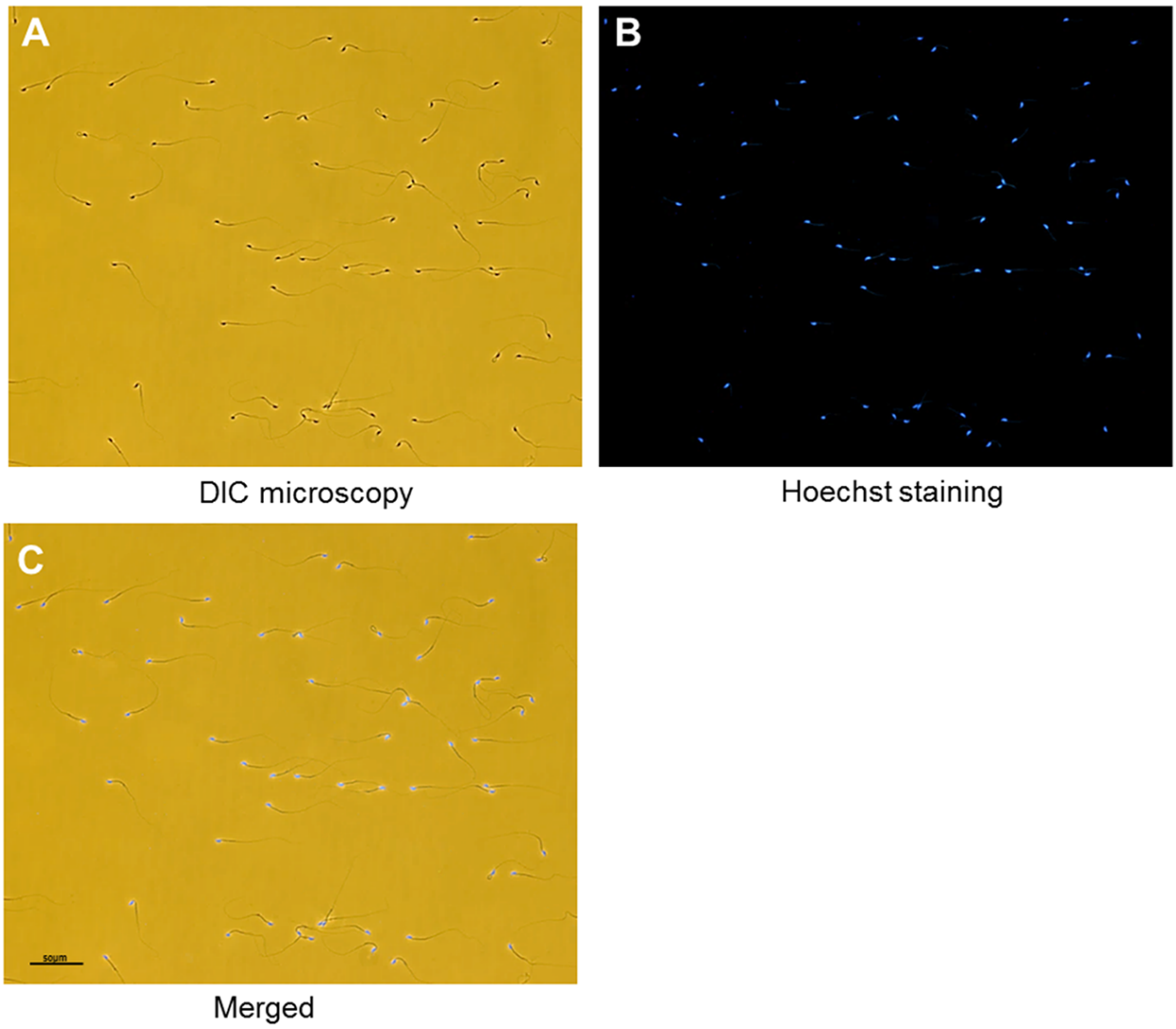

Fig. S3 Validation of separation technique of spermatozoa for proteomic analysis. Spermatozoa examined for the non-appearance of immature sperm cells and somatic cells using microscopy. (**A**) Differential interference contrast microscopic (DIC) image of spermatozoa used for proteomic analyses. (**B**) Image of hoechst staining spermatozoa. (**C**) Merged image.