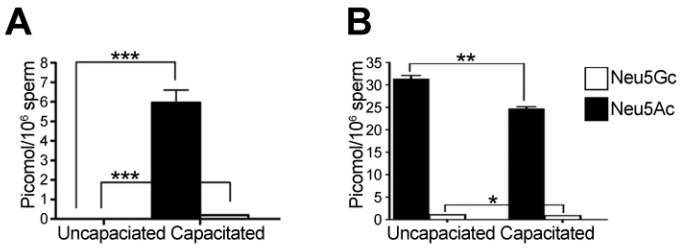
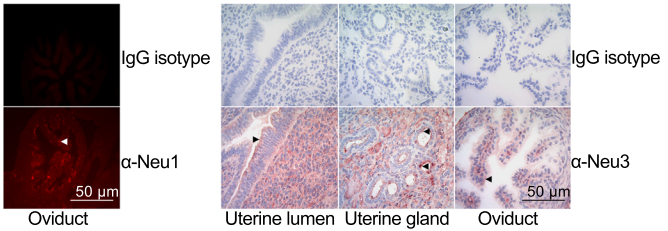
Supplemental Figure 1



Loss of Sia during mouse sperm capacitation:
A. sialoconjugates released by sperm into the capacitation buffer supernatant.
B. change in Sia on the sperm membrane pre and post capacitation.

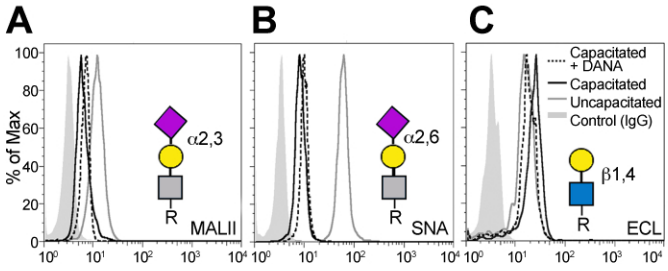
Supplemental Figure 2



NEU1/3 in female genital tract.

White arrow head arrow indicate positive staining for Neu1 with fluorescently labeled antibody in mouse oviduct. Black arrow heads indicate positive staining for Neu3 in mouse uterus and oviduct.

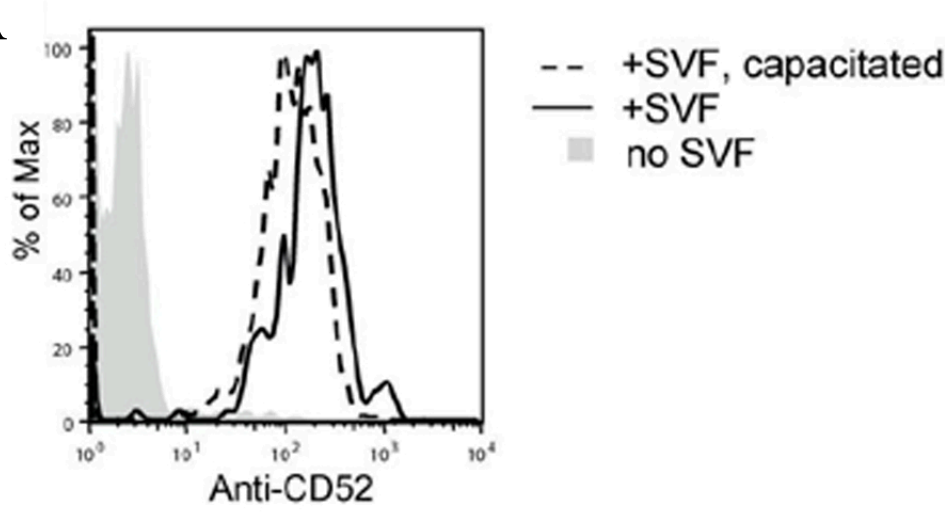
Supplemental Figure 3



Sia change on sperm during capacitaion.
A. Loss of α2-3 Siad as stained by lectin MAL II and weak inhibition by DANA.
B. Loss of α2-6 Sia as strained by lectin SNA and weak inhibition by DANA.
C. Gain of uncapped terminal galactose as stained by lectin ECL and weak inhibition by DANA.
Purple diamond = sialic acid; yellow circle = galactose; blue square = N-Acetylglucosamine; grey square = N-Acetylgalactoseamine/N-Acetylglucoseamne.

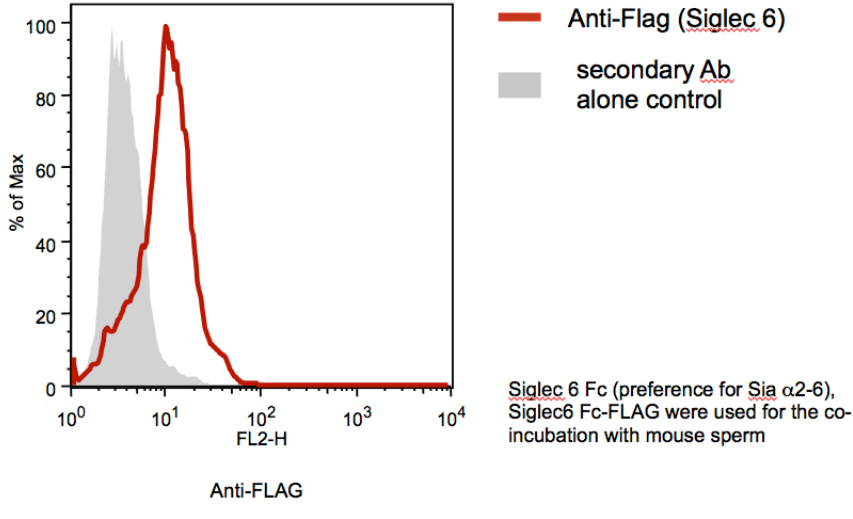
Supplemental Figure 4

A



Mouse epididymal sperm incubated with seminal vesicle fluid and then incubated under capacitating conditions or in BWW. There was little change in CD52 levels as measured with an anti-CD52 peptide Ab.

B



Determination of Siglec6 binding of mouse sperm. Staining with a chimeric, flag-tagged Siglec6-Fc probe reveals marked binding of mouse epididymal sperm.

Supplemental Methods

***In vitro* Seminal fluid (SVF) incubation with sperm.**

Seminal vesicles were isolated from blood vessels and accessory glands by careful dissection. The gland's fluid contents were manually expressed into microcentrifuge tubes. 20 μl of seminal fluid was then mixed with 100 μl of physiological BWW, and placed in a humidity chamber at 37°C for 10 min, centrifuged at 1000g for 5 min, before using the supernatant for sperm incubation. Sperm were added to the diluted seminal vesicle fluid. 10^6 sperm in 50 μl diluted seminal vesicle fluid were incubated at 37°C and 5%CO_2 for 15 min following (1) .

Analysis of Flow cytometry for CD52 and biding of Siglec-6

For detection of CD52, spermatozoa were fixed with freshly thawed 3% PFA for 20 min at RT, washed with PBS, and then blocked with 1%BSA PBS, anti- mouse CD52 (Santa Cruz) antibody with 1:400 at 4°C for 1 h, secondary donkey anti-mouse antibody conjugated with Alexa Fluor 647 was used at 1:500 at 4°C for 1 h. Fc of Siglec-6 tagged with FLAG(N-DYKDDDDK-C) was kindly provided by Dr Varki (UCSD), incubated with mouse sperm as 1ug/1,000,000 for 1h at 37°C, 5%CO2, followed by secondary incubation with anti-FLAG PE (Prozyme) antibody at 1:500 for 30min at 4°C, prior to detection by flow cytometry (FACSCalibur).

1. Buttke, D. E., Nelson, J. L., Schlegel, P. N., Hunnicutt, G. R. and Travis, A. J. (2006) Visualization of GM1 with cholera toxin B in live epididymal versus ejaculated bull, mouse, and human spermatozoa. *Biol Reprod* **74**, 889-895