

Supplemental data Fig. 1 Calretinin detection in C57Bl/6 (WT) mice and competition of the antibody (214 102, Synaptic Systems) in the presence of purified calretinin protein (214-1P, Synaptic Systems, Germany). Detection of calretinin (red) in cerebellum (a) and spermatozoa (c,e) from WT mice by indirect immunofluorescence. Detection using the antibody incubated with calretinin in cerebellum (b) and spermatozoa (d,f) from WT mice by indirect immunofluorescence. Phase contrast detection of spermatozoa (i,j,m). Negative controls were performed on spermatozoa (k) and cerebellum (l) from WT mice. Overlay images of calretinin (c,d) or rabbit immunoglobulins (k) and the acrosome labelled with PNA-FITC (green). Nuclei were labelled with DAPI (blue). Scale: 10µm (c,d,e,f,g,h,i,j,k,m) and 50µm (a,b,l)

Supplemental data Fig. 2 Calretinin detection in calretinin knockout ($CR^{-/-}$) mice and competition of the antibody (214 102, Synaptic Systems) in the presence of purified calbindin D-28k protein (214-0P, Synaptic Systems, Germany). Detection of calretinin (red) in cerebellum (a) and spermatozoa (c) from $CR^{-/-}$ mice by indirect immunofluorescence. Detection using the antibody incubated with calbindin D-28k in cerebellum (b) and spermatozoa (d) from $CR^{-/-}$ mice by indirect immunofluorescence. Phase contrast detection of spermatozoa (e,f,i). Negative controls were performed on spermatozoa (g) and cerebellum (h) from $CR^{-/-}$ mice. Nuclei were labelled with DAPI (blue). Scale: 10µm (c,d,e,f,g,i) and 50µm (a,b,h)

Supplemental data Fig. 3 : Calretinin detection in C57Bl/6 (WT) and calretinin knockout mice ($CR^{-/-}$) with the calretinin antibody from Swant® (7696, Swant, Switzerland). Detection of calretinin (red) in spermatozoa (a,c) and cerebellum (b,d) from WT (a,b) and $CR^{-/-}$ (c,d) mice by indirect immunofluorescence. Negative controls were performed on spermatozoa (e,g)

and cerebellum (f,h) from WT (e,f) and $CR^{-/-}$ (g,h) mice. Overlay images of calretinin (a,c) or rabbit immunoglobulins (e,g) and the acrosome labelled with PNA-FITC (green). Nuclei were labelled with DAPI (blue). Scale: 10 μ m (a,c,e,g) and 50 μ m (b,d,f,h)

Supplemental data Fig. 4 Calretinin detection in C57Bl/6 (WT) and calretinin knockout mice ($CR^{-/-}$) with the calretinin antibody from Bioprime® (Bioprime CA520, BioLogo, Germany). Detection of calretinin (red) in spermatozoa (a,c) and cerebellum (b,d) from WT (a,b) and $CR^{-/-}$ (c,d) mice by indirect immunofluorescence. Negative controls were performed on spermatozoa (e,g) and cerebellum (f,h) from WT (e,f) and $CR^{-/-}$ (g,h) mice. Overlay images of calretinin (a,c) or rabbit immunoglobulins (e,g) and the acrosome labelled with PNA-FITC (green). Nuclei were labelled with DAPI (blue). Scale: 10 μ m (a,c,e,g) and 50 μ m (b,d,f,h)







