



figure e-1: Plexin D1-IgG binding to pain-conducting small DRG neurons and IgG subclass analysis by TBA.
(A) Immunostaining of IB4 (red), a marker of pain-conducting unmyelinated C-fiber type DRG neurons, mostly colocalized with IgG (green) from all eight SFN patients with Plexin D1-IgG. The immunostaining (green) of small DRG neurons by IgG from iSFN Cases 1–6 and sSFN Cases 1 and 2 was prevented by pre-absorption with rhPlexin D1 (2 µg/ml). HC IgG showed no significant immunoreactivity to mouse DRG. Nuclei are counterstained with 4',6-diamidino-2-phenylindole (DAPI) (blue). **(B)** IgG subclass analysis for Plexin D1-IgG by TBA showed a predominance of the IgG2 subclass in iSFN Cases 1, 2, 4, 5, and 6, and sSFN Cases 1 and 2. iSFN Cases 4 and 6 also had weak IgG1 reactivity. iSFN Case 3 had only strong IgG1 reactivity against Plexin D1. HC IgG1–4 showed no significant immunoreactivity to mouse DRG. Scale bars = 50 µm. DRG = dorsal root ganglia; HC = healthy control; IB4 = isolectin B4; IgG = immunoglobulin G; iSFN = idiopathic small fiber neuropathy; OD = optical density; rhPD1 = recombinant human Plexin D1; SFN = small fiber neuropathy; sSFN = secondary SFN; TBA = tissue-based indirect immunofluorescence assay.