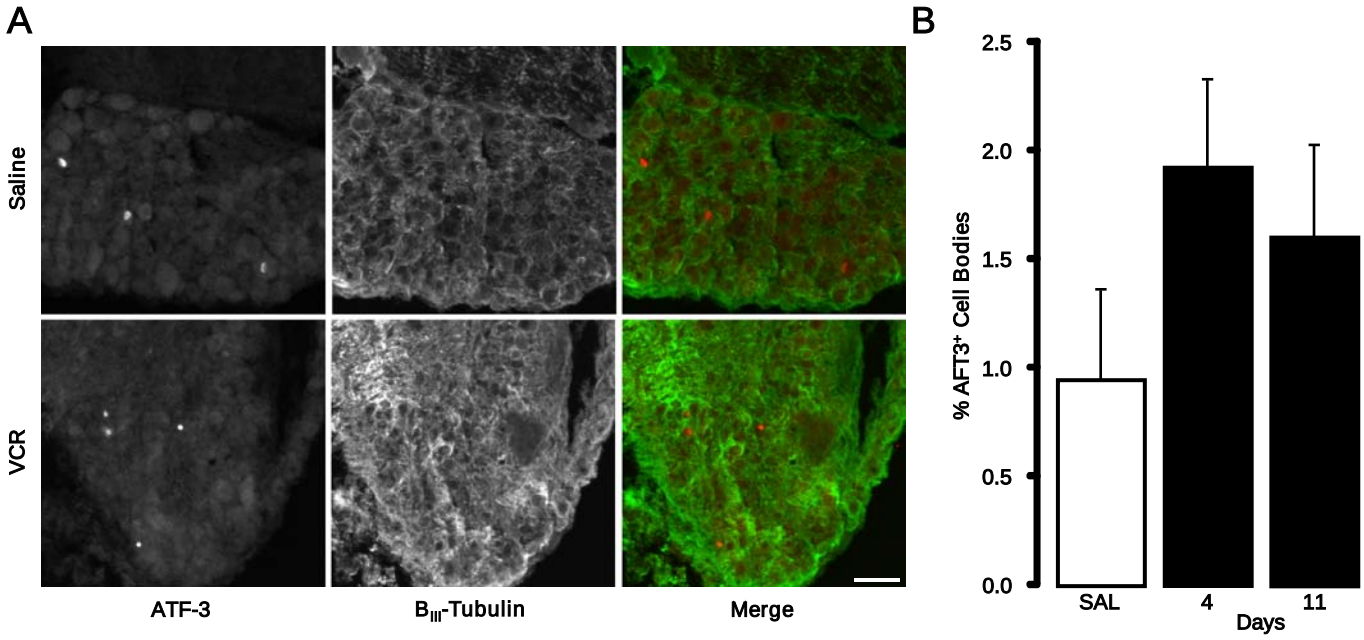
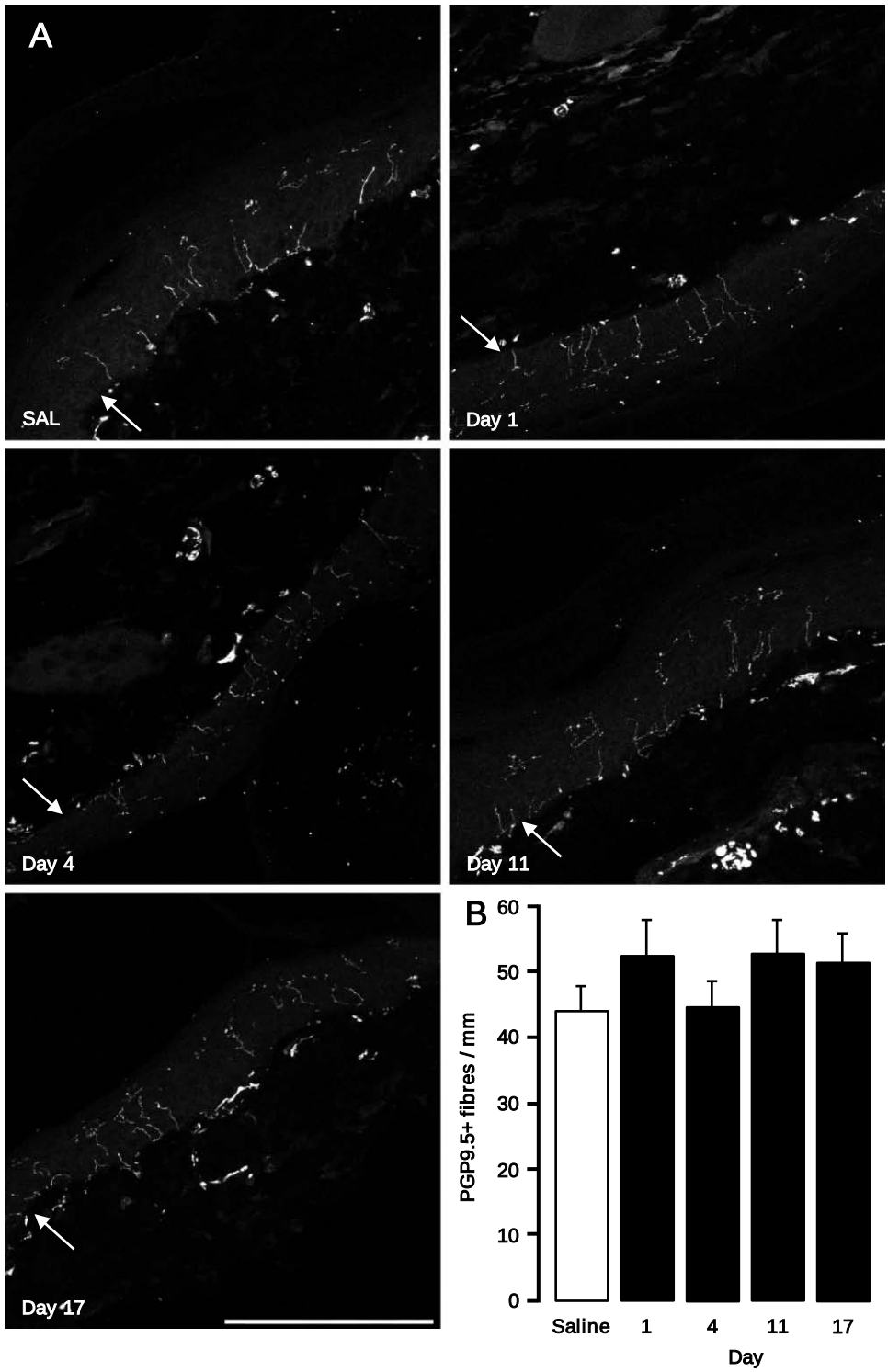
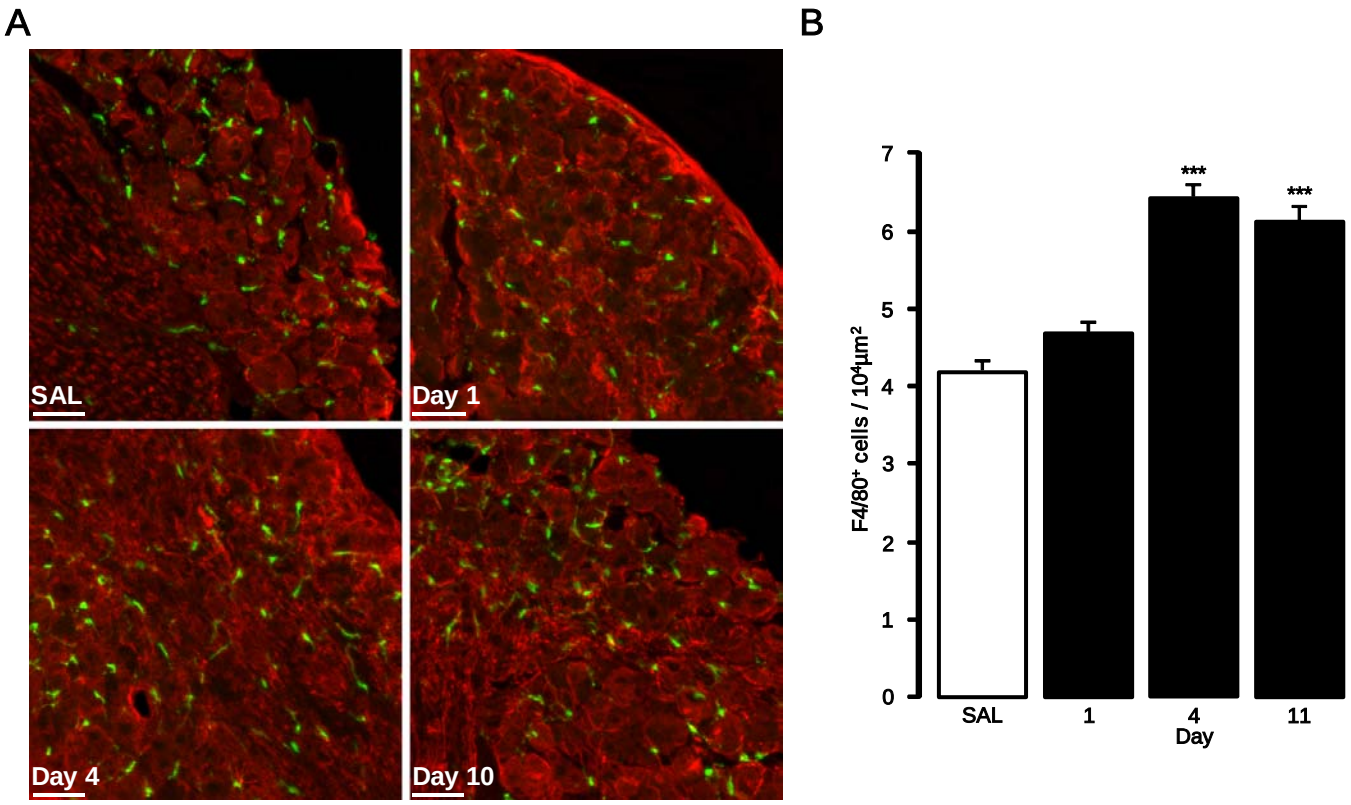


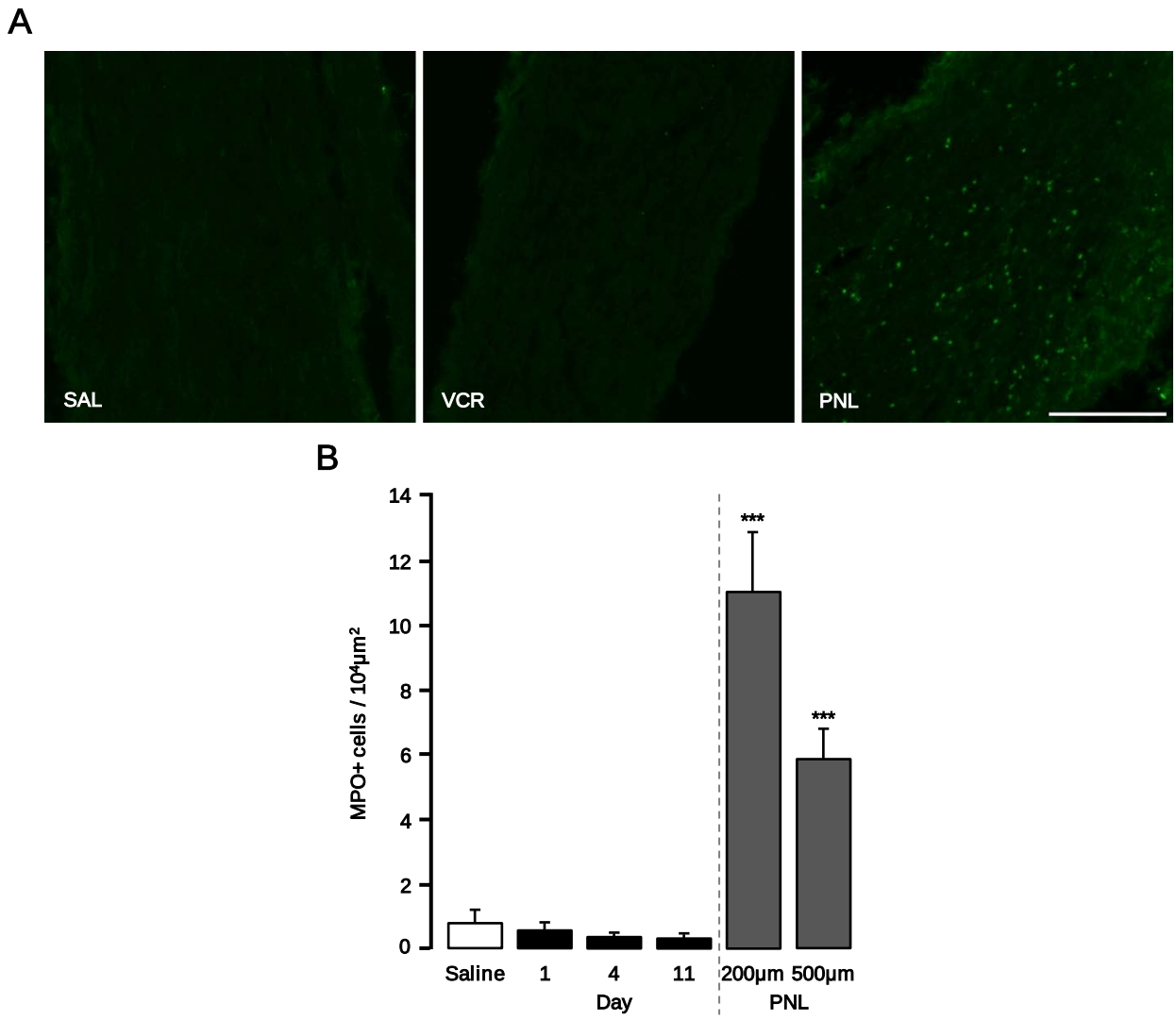
Supplementary Figure 1. No evidence of neuronal stress in dorsal root ganglia during vincristine treatment. (A) Representative photomicrographs of lumbar DRG sections obtained after either saline or VCR treatment (2x5day cycles; 0.5mg/kg/day i.p.) representing ATF-3 (red) and B_{III}tubulin (green) expression, scale bar equal 100µm. **(B)** DRG were collected after saline treatment, first VCR cycle (day4) and second VCR cycle (day11). The percentage of ATF3 positive cell bodies was counted in $10^4 \mu m^2$ boxes and data are shown as mean ±SEM, n=4 mice/group.



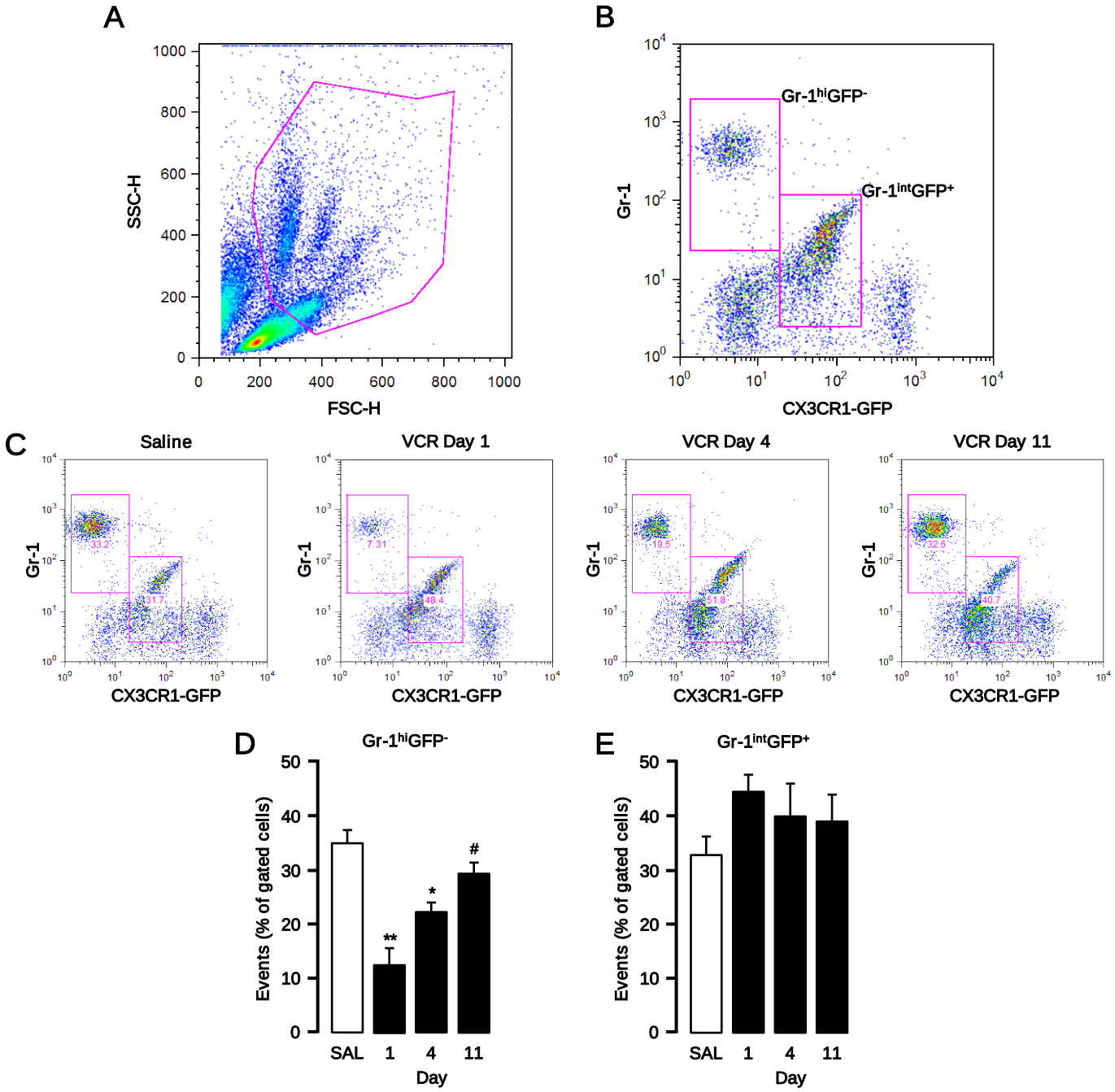
Supplementary Figure 2. No change in intraepidermal nerve fibre density in the plantar skin following vincristine treatment. Saline or VCR (0.5mg/kg/day i.p.) were administered for 2x5day-cycles and skin samples collected on day 1, at the end of first cycle (day4), second cycle (day11) and 6 days after cessation of treatment (day17). **(A)** Representative photomicrographs of PGP9.5^{+} fibres in glabrous skin sections. Scale bar equals 100µm and arrow shows epidermal border. **(B)** The number of PGP9.5^{+} fibres was counted throughout the whole skin section and data are shown as mean ±SEM, n=5 mice/group.



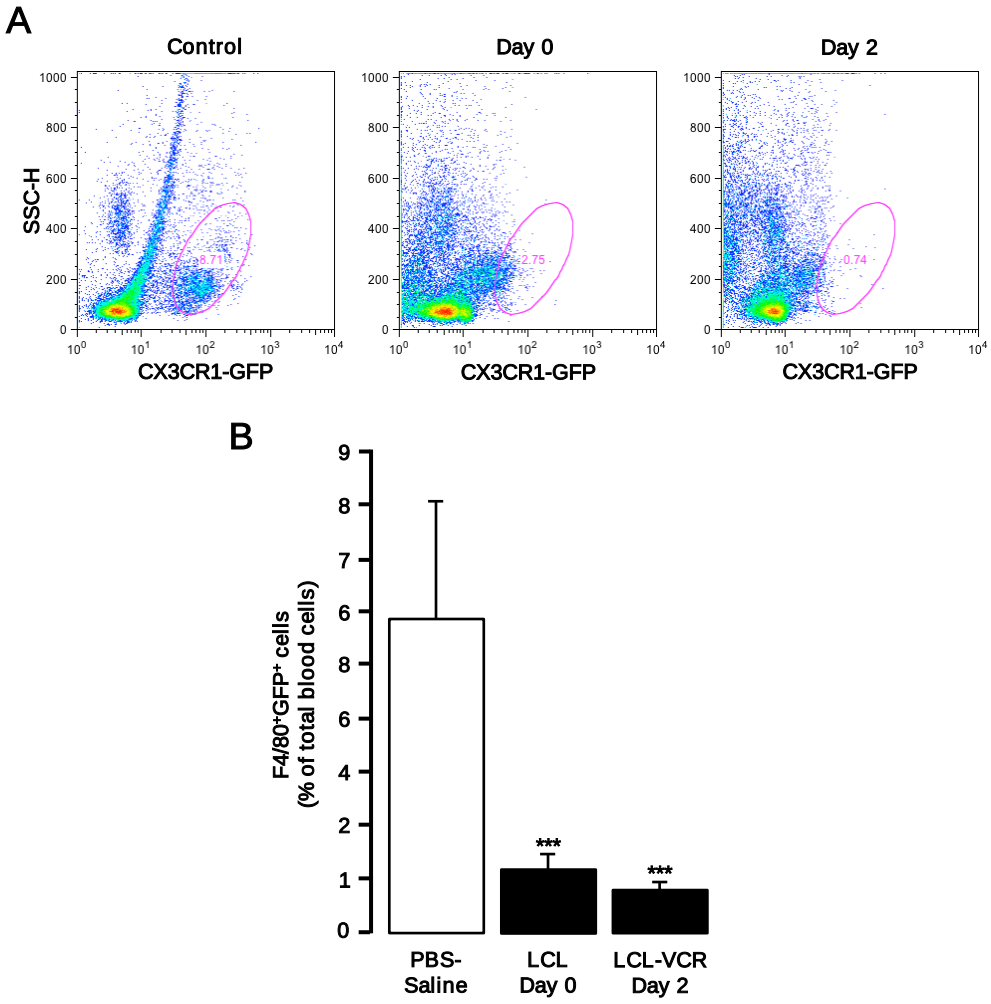
Supplementary Figure 3. Significant monocyte-macrophage infiltration of the dorsal root ganglia during vincristine treatment. Saline or VCR (0.5mg/kg/day i.p.) were administered for 2x5day-cycles and DRG collected on day 1 and at the end of first cycle (day4) and second cycle (day11) **(A)** Representative photomicrographs of F4/80$^+$ (green) cells in lumbar DRG sections, scale bars equal 100µm (B_{III}tubulin-red). **(B)** The number of cells expressing F4/80 were counted in 10^4µm^2 boxes in DRG sections and data are shown as mean ±SEM, n=4 mice/group. ***P<0.001 compared to saline, one-way ANOVA, post-hoc Tukey test.



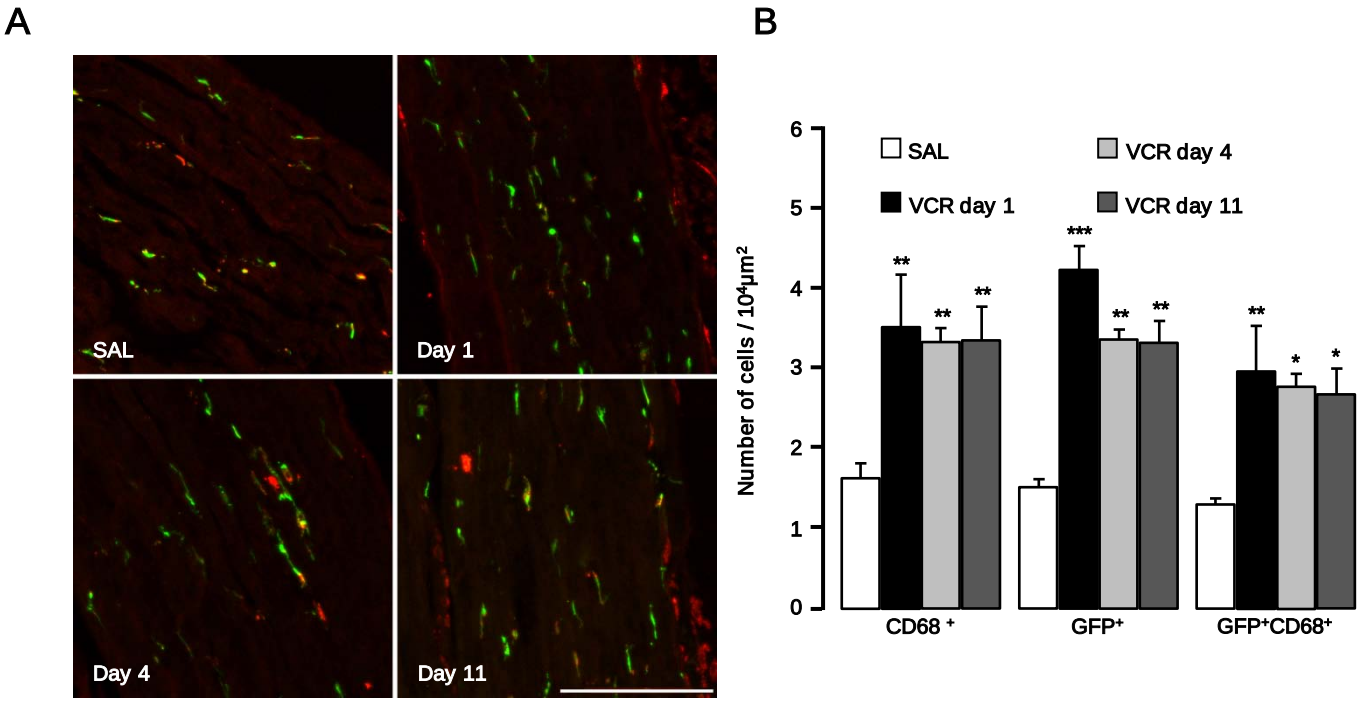
Supplementary Figure 4. No infiltration of neutrophils into the sciatic nerve after VCR treatment. Saline or VCR (0.5mg/kg/day i.p.) were administered for 2x5day-cycles and sciatic nerve segments collected on day 1 and at the end of first cycle (day4) and second cycle (day11). For comparison purposes, perfuse-fixed sciatic nerve segments were also obtained from mice 11 days after sciatic nerve partial (surgical) injury [PNL] (25). **(A)** Representative photomicrographs of myeloperoxidase $(MPO)^+$ cells in sciatic nerve sections, scale bar equal to 200µm. **(B)** The number of MPO^+ cells were counted in $10^4\mu m^2$ boxes, data are shown as mean ±SEM, n=4 mice/group. ***P<0.001 compared to saline control, one-way ANOVA, post-hoc Tukey test.



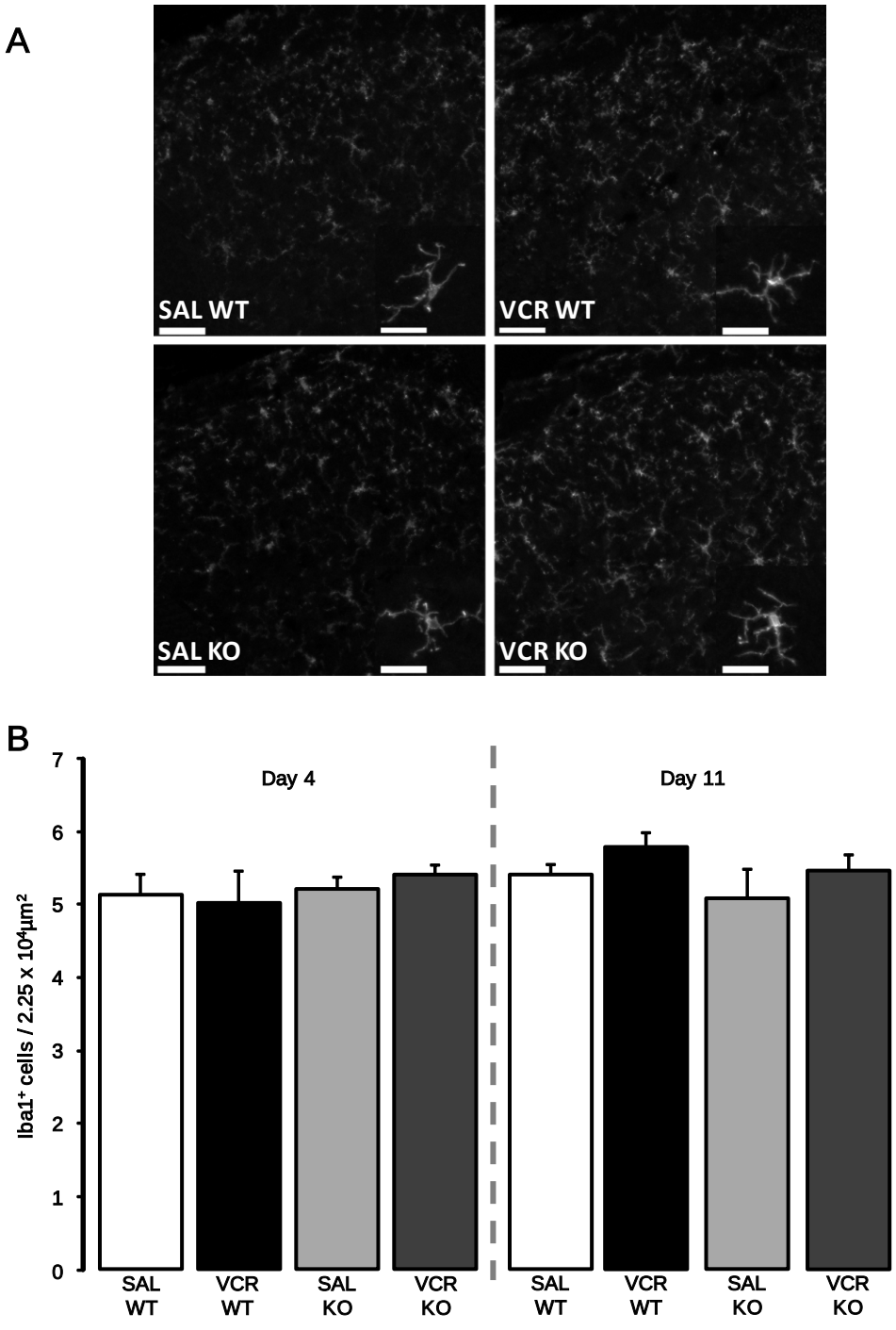
Supplementary Figure 5. VCR treatment is associated with no change in circulating monocyte and decrease in granulocyte population. The monocyte blood population Gr-1^{+}/CX3CR1-GFP^{+} did not change over VCR (0.5mg/kg/day i.p.) 2x5day-cycles. Whole blood samples were taken on days 1, 4 and 11, as well as from saline controls. Blood samples were labelled with Gr-1. **(A)** Cells were gated as neutrophil and monocyte population according to forward and side scatter profiles (note the classical spread of mouse granulocyte populations). **(B)** Gated cells were subsequently clustered into 2 distinct groups namely: Gr-1^{hi}CX3CR1-GFP^{-}, Gr-1^{int}CX3CR1-GFP^{+}. **(C)** Representative plots shown for each time point. **(D-E)** Bar graphs reporting summary data (mean ±SEM) as quantified in the specific gates, indicates initial depletion of neutrophils/Gr-1^{hi}CX3CR1-GFP^{-} (D) but no change in monocytes/Gr-1^{int}CX3CR1-GFP^{+} (E). ∗ P<0.05 compared to saline # P<0.05 compared to day 1, one-way ANOVA with post-hoc Tukey test, n=3 mice/group.



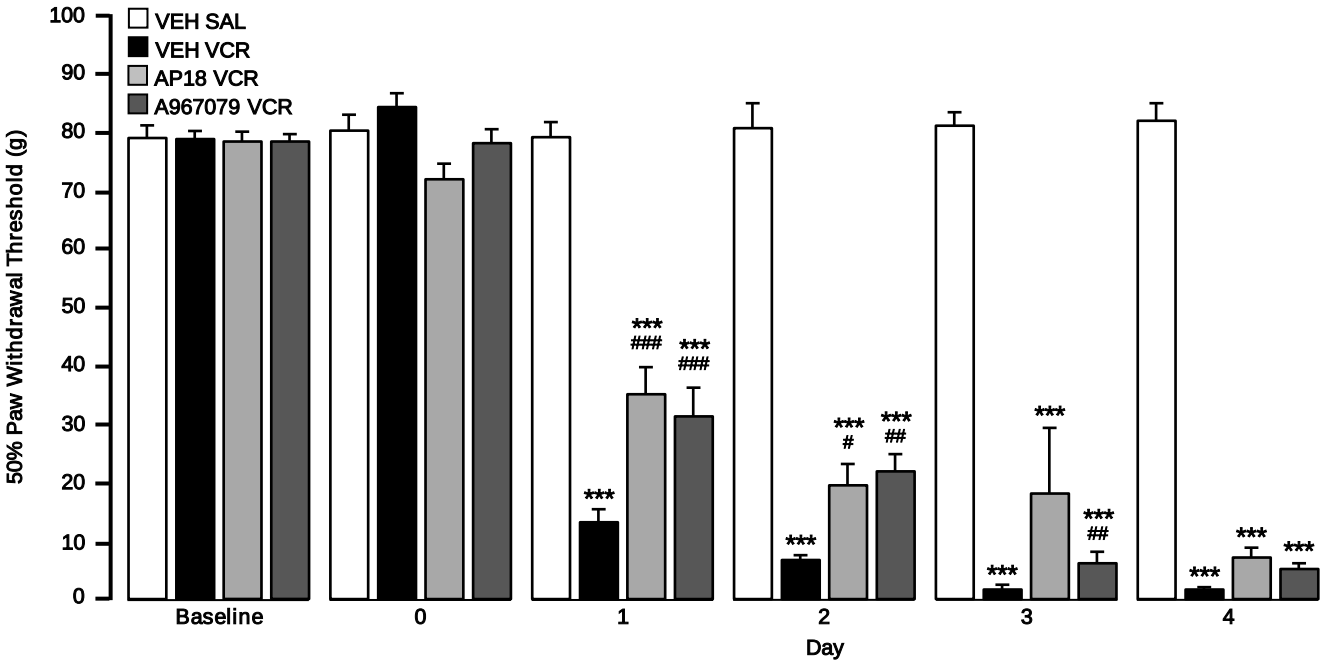
Supplementary Figure 6. Depletion of blood monocytes following liposome chlodronate treatment. LCL was administered twice (2x100µl/10g injections of 5mg/ml in PBS i.p.) and blood samples collected to be analysed by flow cytometry after depletion only (Day0) or depletion plus two days of VCR (0.5mg/kg/day i.p.) treatment (Day2). **(A)** Dot plot analysis of side scatter and CX3CR1-GFP$^+$ blood cells. **(B)** Cumulative data for relative percentage of total blood cells that are F4/80$^+$CX3CR1-GFP$^+$. Data shown are mean ±SEM, **P<0.01, One way ANOVA followed by Tukey post-hoc test; n=3 mice/group.



Supplementary Figure 7. Vincristine treatment induces infiltration of CD68$^+$ monocyte-macrophages in the sciatic nerve. CD68$^+$ monocyte-macrophages populate the sciatic nerve immediately after VCR (0.5mg/kg/day i.p.) treatment (day1) and in the first cycle (days0-4) and second cycle (days7-11). **(A)** Representative photomicrographs demonstrating CX3CR1-GFP$^+$ cells (green) and CD68$^+$ cells (red) in sciatic nerve sections where the scale bars equal to 200µm. **(B)** Positive cells were counted in 10^4µm^2 boxes and data are shown as mean ±SEM, n=5 mice/group, * P<0.05, ** P<0.001compared to saline, one-way ANOVA, post-hoc Tukey test**.**



Supplementary Figure 8. No change in microglial cell numbers in the dorsal horn of *Cx3cr1*-deficient or wildtype mice during or at the end of VCR cycles. Saline or VCR (0.5mg/kg/day i.p.) were administered for 2x5day-cycles and perfuse-fixed lumbar spinal cord obtained at the end of first cycle (day4) and second cycle (day11). **(A)** Representative photomicrographs of dorsal horn sections showing microglia ($Iba1^+$ cells) in this area, scale bars equal 100µm and 20µm in the insets, respectively. **(B)** The number of $Iba1^+$ cells was counted in $2.25x10^4\mu m^2$ boxes and data are shown as mean ±SEM, n=4 mice/group.



Supplementary Figure 9. TRPA1 antagonists attenuate VCR-induced mechanical allodynia. Both AP-18 and A9670079 partially prevented the development of VCR-induced allodynia. AP-18 (100mg/kg twice a day p.o.) and A967079 (100mg/kg twice a day p.o.) were administered 1 hour prior to VCR (0.5mg/kg/day i.p.) and paw withdrawal thresholds (PWT) measured 1 hour after VCR. Data are expressed as 50% PWT and shown as mean ±SEM, n=10 mice/group. ***P<0.001 compared to vehicle (VEH SAL) thresholds, $^{\#}$P<0.05, $^{\#\#}$P<0.001, $^{\#\#\#}$P<0.001 compared VCR (VEH VCR) treated thresholds, two-way RM ANOVA, post-hoc Holm-Sidak test.