

Supplementary figures:

S1. Motor function of Control ($n=7$) and Primed ($n=7$) mice assessed on the rotarod demonstrating no differences between groups in (A) rotations per minute, (B) latency to fall and (C) distance travelled ($P>0.05$ for all time points 2 way RM ANOVA Primed vs Control).

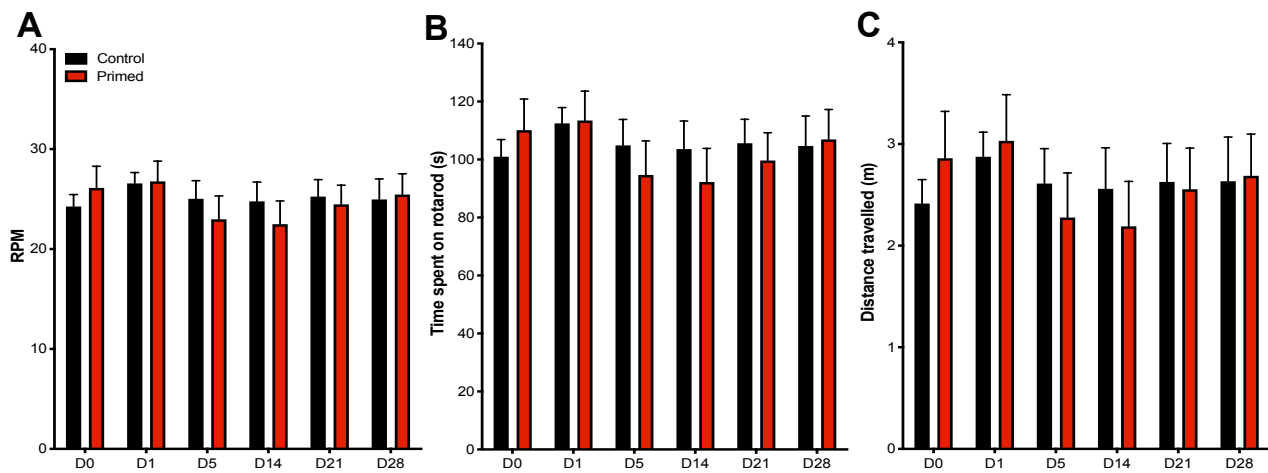
S2. A hyperalgesic priming model of widespread pain was induced in all mice with intramuscular injections of carrageenan on D0 and D4 (green arrows) and neutrophil sequestering antibody ($50\mu\text{g}$ i.v. 1A8 Clone) administered either 24 hours before D0 (A-C) or on D28 (D-E), indicated by the vertical dotted line ($n=6$ per group). **(A)** Ipsilateral mechanical hypersensitivity to von Frey stimuli in mice pretreated with Vehicle or Clone 24 hours prior to induction of the priming model. **(B)** Contralateral mechanical hypersensitivity to von Frey stimuli in mice pretreated with Vehicle or Clone 24 hours prior to induction of the priming model. ($***P<0.001$ 2-way RM ANOVA Vehicle vs Clone following administration of antibody). **(C)** Hindleg inflammation measured through gastrocnemius muscle size ($P>0.05$ all timepoints 2-way RM ANOVA). **(D)** Ipsilateral mechanical hypersensitivity to von Frey stimuli in mice administered Vehicle or Clone on D28 following induction of the priming model. **(E)** Contralateral mechanical hypersensitivity to von Frey stimuli in mice administered Vehicle or Clone on D28 following induction of the priming model. ($***P<0.001$ 2-way RM ANOVA Vehicle vs Clone following administration of antibody).

S3. (i-ii) tSNE analysis of Ly6G/Ly6C double positive cells derived from (i) blood and (ii) DRG (of both Control and Primed mice) illustrating heterogeneous expression of CXCR2, CD62L, CD11b, Ly6G and Ly6C. (iii) tSNE analysis of Ly6C/Ly6G double positive neutrophils obtained from blood ($n=8$ mice) and DRG ($n=6$ mice) of Primed animals. Differential expression of CXCR2, CD62L, CD11b, Ly6G and Ly6C based on DRG specific clusters also illustrated.

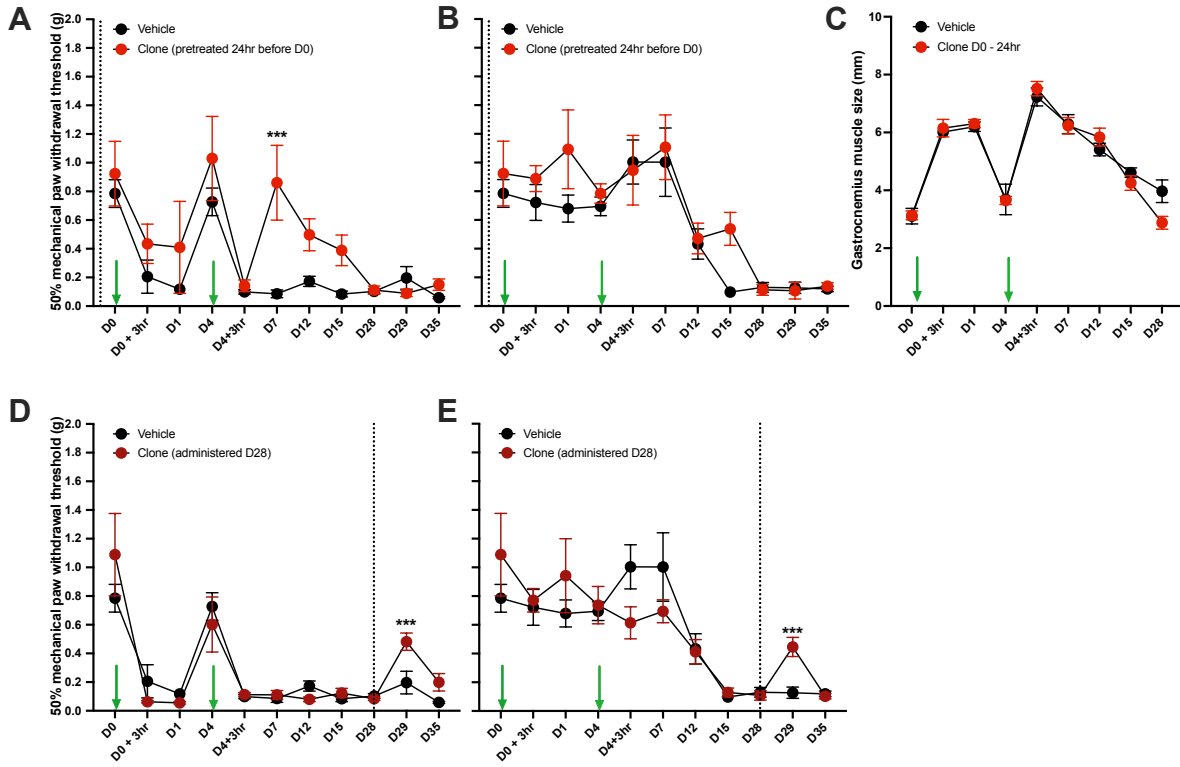
S4. Representative image of L4 DRG 24hr following i.v. administration of Patient neutrophils, illustrating endogenous neutrophils (mouse MRP14; green) and exogenous neutrophils (CellTracer dye 555; blue) infiltrating peripheral mouse ganglia. Some neutrophils are indicated by white arrows.

S5. Passive transfer of 8mg IgG derived from a fibromyalgia patient vs. pain free control into naïve recipient mice ($n=4$ mice per group). No differences in sensory thresholds were observed over a 7-day period to (i) punctate mechanical stimulation or (ii) noxious heat stimulation ($P>0.05$ for all time points 2 way RM ANOVA).

S1

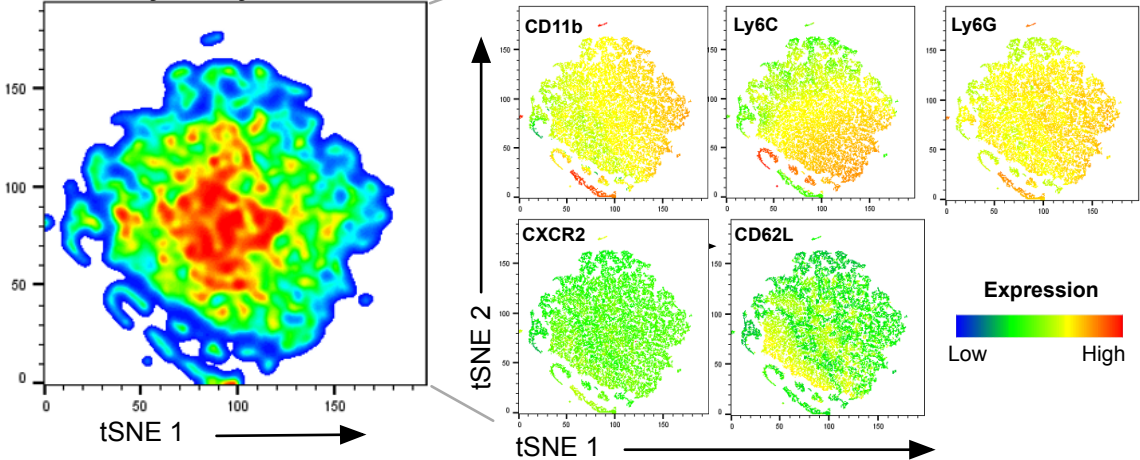


S2



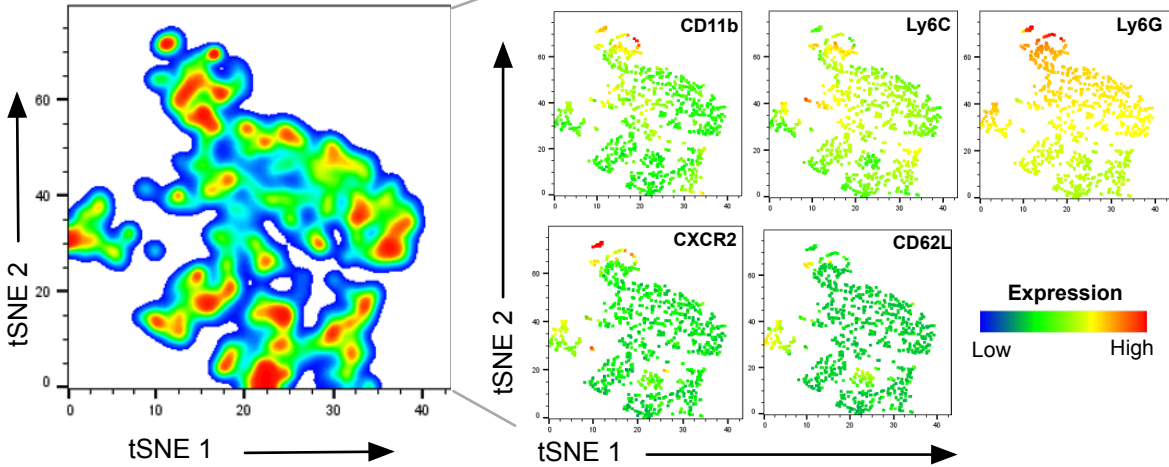
S3i

Blood Ly6G+/Ly6C+ cells



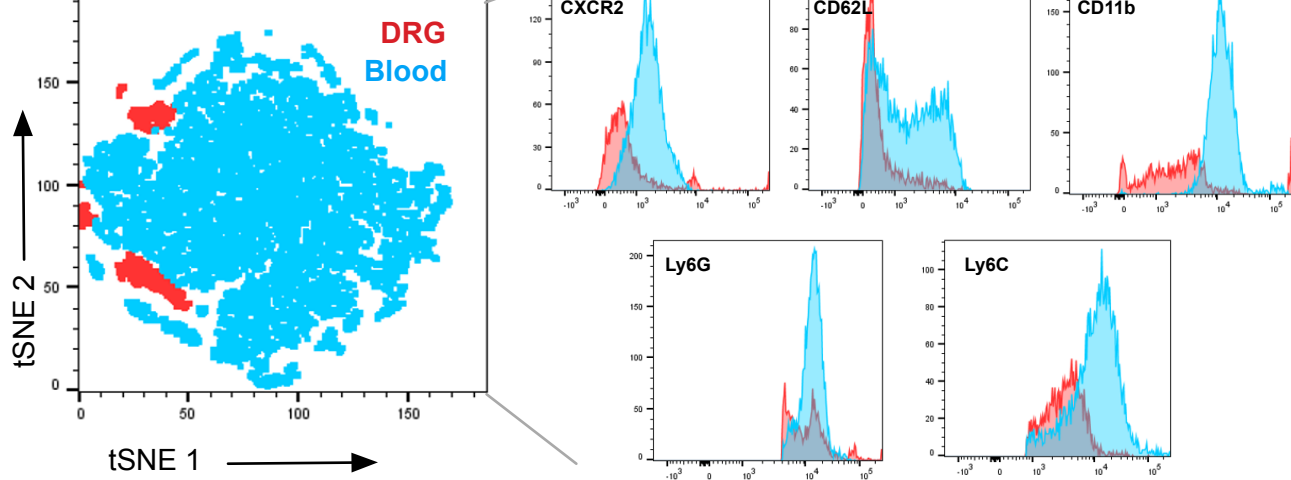
S3ii

DRG Ly6G+/Ly6C+ cells

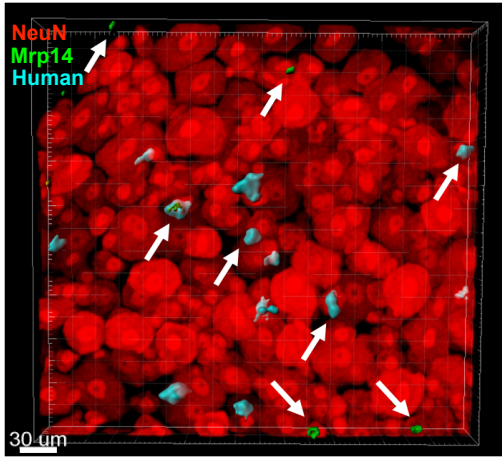


S3iii

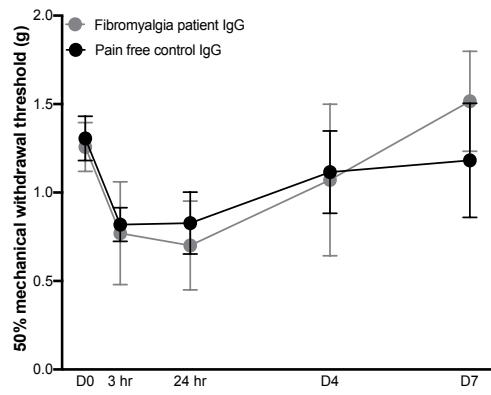
Differentially expressed clusters of Ly6G+/Ly6C+ cells in blood and DRG



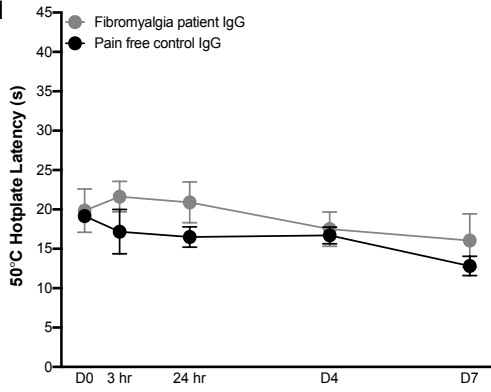
S4



S5 i



ii



Supplementary Tables

Supplementary Table 1 tSNE clusters of neutrophils in DRG derived from Control and Primed mice

Cluster	Control	Primed	% Change
1	3.74%	1.52%	↓59%
2	5.75%	13.50%	↑135%
3	11.50%	4.55%	↓60%
4	9.48%	15.80%	↑66%
5	4.02%	11.20%	↑178%
6	21.60%	14.60%	↓32%
7	8.33%	10.80%	↑30%
8	7.76%	5.16%	↓33%
9	11.80%	5.01%	↓57%
10	6.90%	7.44%	↑8%
11	8.62%	10%	↑16%

tSNE analysis was performed with Ly6G/Ly6C double positive neutrophils sorted from DRG derived from Control and Primed mice, producing 11 clusters of neutrophil subsets based on expression of CXCR2, CD62L, CD11b, Ly6G and Ly6C markers. Column 2 and Column 3 show percentages of each cluster in the total population of neutrophils in Control or Primed samples, respectively. The last column refers to percentage change in the cluster proportion in Primed samples compared to Control samples. Cluster 3 and Cluster 5 represent the most downregulated and upregulated clusters, respectively.

Supplementary Table2. tSNE clusters of neutrophils in whole blood derived from Control and Primed mice.

Cluster	Control	Primed	% Change
1	3.35%	5.54%	↑65%
2	2.45%	3.18%	↑30%
3	19.40%	8.74%	↓55%
4	5.98%	8.62%	↑44%
5	8.64%	17.40%	↑101%
6	2.53%	1.78%	↓30%
7	5.34%	8.12%	↑52%
8	7.54%	9.30%	↑23%
9	17%	12.60%	↓26%
10	7.59%	10.40%	↑37%
11	19.70%	12.80%	↓35%

tSNE analysis was performed with Ly6G/Ly6C double positive neutrophils sorted from blood derived from Control and Primed mice, producing 11 clusters of neutrophil subsets based on expression of CXCR2, CD62L, CD11b, Ly6G and Ly6C markers. Column 2 and Column 3 show percentages of each cluster in the total population of neutrophils in Control or Primed samples, respectively. The last column refers to percentage change in the cluster proportion in Primed samples compared to Control samples. Cluster 3 and Cluster 5 represent the most downregulated and upregulated clusters, respectively.