

Supplementary data

Supplementary Table 1. Cytokine/chemokine/biomarker profiles in *CCR5*^{+/+} and *CCR5*^{-/-} mice[§]

| Parameter* | units | LLD [◇] | <i>CCR5</i> ^{+/+} | | | | <i>CCR5</i> ^{-/-} | | | | p [△] |
|-------------------|--------------|------------------|----------------------------|-------------|-------------|----------------|----------------------------|-------------|-------------|-------------|----------------|
| | | | 1 [•] | 3 | 5 | median | 2 | 4 | 6 | median | |
| Apo A1 | ug/mL | 0.21 | 0.00 ^V | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.011 | 0.00 | 0.3173 |
| CD40 | pg/mL | 2.4 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 |
| CD40 L | pg/mL | 18 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | . |
| CRP | ug/mL | 0.0042 | 0.0043 | 0.0035 | 0.0025 | 0.00 | 0.0024 | 0.0022 | 0.0053 | 0.00 | 0.5127 |
| EGF | pg/mL | 7.8 | 3.1 | 3.4 | 4.0 | 3.42 | 2.8 | 2.2 | 4.0 | 2.81 | 0.3758 |
| Endothelin-1 | pg/mL | 13 | 5.9 | 5.9 | 7.8 | 5.87 | 5.9 | 5.9 | 6.9 | 5.87 | 0.7963 |
| Eotaxin | pg/mL | 2.5 | 1.7 | 2.4 | 2.5 | 2.41 | 0.95 | 0.88 | 1.9 | 0.95 | 0.1266 |
| Factor VII | ng/mL | 0.19 | 0.76 | 1.1 | 1.1 | 1.06 | 0.54 | 0.54 | 0.96 | 0.54 | 0.1157 |
| FGF-9 | ng/mL | 0.20 | 0.00 | 0.064 | 0.00 | 0.00 | 0.11 | 0.00 | 0.00 | 0.00 | 0.7963 |
| FGF-basic | ng/mL | 0.12 | 0.70 | 0.75 | 0.82 | 0.75 | 0.47 | 0.40 | 0.62 | 0.47 | 0.0495 |
| Fibrinogen | ug/mL | 0.85 | 0.42 | 0.87 | 1.0 | 0.87 | 0.69 | 0.00 | 1.6 | 0.69 | 0.8273 |
| GCP-2 | ng/mL | 0.0049 | 0.0014 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.3173 |
| GM-CSF | pg/mL | 1.7 | 32 | 73 | 63 | 63.00 | 8.5 | 66 | 26 | 26.00 | 0.2752 |
| GST-alpha | ng/mL | 0.084 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | . |
| Haptoglobin | ug/mL | 0.013 | 0.20 | 0.22 | 0.16 | 0.20 | 0.20 | 0.22 | 0.17 | 0.20 | 0.8222 |
| IFN-gamma | pg/mL | 14 | 101 | 207 | 1030 | 207.00 | 14 | 423 | 202 | 202.00 | 0.5127 |
| IgA | ug/mL | 0.038 | 0.15 | 0.29 | 0.22 | 0.22 | 0.27 | 0.19 | 0.37 | 0.27 | 0.5127 |
| IL-10 | pg/mL | 22 | 205 | 184 | 386 | 205.00 | 111 | 186 | 162 | 162.00 | 0.1266 |
| IL-11 | pg/mL | 17 | 5.3 | 4.1 | 6.5 | 5.32 | 3.0 | 0.00 | 0.00 | 0.00 | 0.0463 |
| IL-12p70 | ng/mL | 0.11 | 0.011 | 0.020 | 0.011 | 0.01 | 0.020 | 0.011 | 0.011 | 0.01 | 1 |
| IL-17 | ng/mL | 0.030 | 0.038 | 0.25 | 0.17 | 0.17 | 0.0021 | 0.087 | 0.029 | 0.03 | 0.1266 |
| IL-18 | ng/mL | 0.13 | 0.13 | 0.24 | 0.19 | 0.19 | 0.13 | 0.068 | 0.19 | 0.13 | 0.2612 |
| IL-1alpha | pg/mL | 9.0 | 24 | 59 | 59 | 58.80 | 8.1 | 29 | 31 | 28.50 | 0.2683 |
| IL-1beta | ng/mL | 0.089 | 0.92 | 0.92 | 1.2 | 0.92 | 0.65 | 0.35 | 1.00 | 0.65 | 0.2683 |
| IL-2 | pg/mL | 13 | 162 | 73 | 116 | 116.00 | 12 | 30 | 26 | 25.90 | 0.0495 |
| IL-3 | pg/mL | 4.3 | 4.8 | 7.0 | 12 | 7.02 | 1.3 | 4.0 | 2.0 | 1.98 | 0.0495 |
| IL-4 | pg/mL | 15 | 12 | 20 | 57 | 19.70 | 0.00 | 42 | 31 | 31.00 | 0.8273 |
| IL-5 | ng/mL | 0.039 | 0.20 | 0.31 | 0.39 | 0.31 | 0.064 | 0.076 | 0.20 | 0.08 | 0.0765 |
| IL-6 | pg/mL | 2.8 | 5.1 | 6.8 | 17 | 6.81 | 0.00 | 4.2 | 2.0 | 1.97 | 0.0495 |
| IL-7 | ng/mL | 0.062 | 0.016 | 0.013 | 0.018 | 0.02 | 0.014 | 0.014 | 0.012 | 0.01 | 0.2683 |
| IP-10 | pg/mL | 8.1 | 24 | 30 | 66 | 30.20 | 4.2 | 23 | 24 | 23.00 | 0.0765 |
| KC/GROalpha | ng/mL | 0.035 | 0.00 | 0.00 | 0.0018 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.3173 |
| LIF | pg/mL | 8.7 | 485 | 368 | 640 | 485.00 | 131 | 414 | 285 | 285.00 | 0.1266 |
| Lymphotactin | pg/mL | 17 | 191 | 214 | 651 | 214.00 | 81 | 266 | 264 | 264.00 | 0.8273 |
| MCP-1 | pg/mL | 3.4 | 0.78 | 0.62 | 2.9 | 0.78 | 0.53 | 0.00 | 0.00 | 0.00 | 0.0463 |
| MCP-3 | pg/mL | 6.3 | 1.2 | 1.5 | 2.2 | 1.53 | 0.57 | 0.70 | 1.1 | 0.70 | 0.0495 |
| MCP-5 | pg/mL | 9.3 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | . |
| M-CSF | ng/mL | 0.0036 | 0.0071 | 0.0092 | 0.010 | 0.01 | 0.0028 | 0.0038 | 0.0069 | 0.00 | 0.0495 |
| MDC | pg/mL | 4.4 | 1310 | 2300 | 3090 | 2300.00 | 664 | 783 | 1820 | 783.00 | 0.1266 |
| MIP-1alpha | ng/mL | 0.045 | 0.91 | 0.84 | 3.3 | 0.91 | 0.23 | 1.5 | 1.1 | 1.14 | 0.8273 |
| MIP-1beta | pg/mL | 16 | 1640 | 1220 | 5290 | 1640.00 | 344 | 2370 | 1720 | 1720 | 0.8273 |

| | | | | | | | | | | | |
|---------------|--------------|------------|------------|------------|-----------|-------------|-------------|------------|------------|-------------|---------------|
| MIP-1gamma | ng/mL | 0.015 | 0.021 | 0.027 | 0.097 | 0.03 | 0.0072 | 0.013 | 0.014 | 0.01 | 0.0495 |
| MIP-2 | pg/mL | 1.4 | 5.2 | 16 | 27 | 15.70 | 1.3 | 3.2 | 6.1 | 3.16 | 0.1266 |
| MIP-3beta | ng/mL | 0.093 | 0.24 | 0.37 | 0.42 | 0.37 | 0.12 | 0.12 | 0.29 | 0.12 | 0.1212 |
| MMP-9 | ng/mL | 0.10 | 0.25 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.3173 |
| MPO | ng/mL | 0.19 | 0.56 | 0.38 | 1.7 | 0.56 | 0.12 | 0.066 | 0.19 | 0.12 | 0.0495 |
| Myoglobin | ng/mL | 0.24 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | . |
| OSM | ng/mL | 0.026 | 0.024 | 0.026 | 0.030 | 0.03 | 0.022 | 0.022 | 0.023 | 0.02 | 0.0463 |
| RANTES | pg/mL | 9.6 | 2.3 | 3.6 | 14 | 3.59 | 0.89 | 2.6 | 4.3 | 2.61 | 0.5127 |
| SAP | ug/mL | 0.027 | 0.041 | 0.059 | 0.043 | 0.04 | 0.040 | 0.063 | 0.051 | 0.05 | 0.8273 |
| SCF | pg/mL | 15 | 9.7 | 12 | 15 | 11.50 | 4.5 | 6.2 | 8.8 | 6.17 | 0.0495 |
| SGOT | ug/mL | 0.37 | 0.00 | 0.00 | 1.6 | 0.00 | 3.5 | 1.1 | 0.00 | 1.06 | 0.4867 |
| TIMP-1 | ng/mL | 0.036 | 0.051 | 0.051 | 0.064 | 0.05 | 0.037 | 0.047 | 0.045 | 0.05 | 0.0463 |
| Tissue Factor | ng/mL | 0.10 | 0.71 | 1.0 | 1.1 | 1.04 | 0.33 | 0.26 | 0.71 | 0.33 | 0.0765 |
| TNF-alpha | ng/mL | 0.027 | 0.050 | 0.034 | 0.073 | 0.05 | 0.00 | 0.043 | 0.024 | 0.02 | 0.1266 |
| TPO | ng/mL | 0.53 | 2.1 | 3.0 | 3.7 | 2.99 | 0.67 | 0.67 | 2.3 | 0.67 | 0.1212 |
| VCAM-1 | ng/mL | 0.19 | 0.00 | 0.068 | 0.080 | 0.07 | 0.00 | 0.034 | 0.00 | 0.00 | 0.2463 |
| VEGF | pg/mL | 7.6 | 92 | 52 | 96 | 91.80 | 19 | 61 | 41.9 | 41.90 | 0.1266 |
| vWF | ng/mL | 0.99 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | . |

§ Biomarker concentrations were measured in the culture supernatants of purified mouse T cells after 48 hrs of stimulation with anti-CD3/28 Abs. Three mice per CCR5 genotype group were analyzed, and data from each mouse is shown. Data were obtained by Rules Based Medicine, a multi-analyte profile bioassay (http://www.rulesbasedmedicine.com/services_rodent_antigen.asp). The sensitivity of this bioassay is comparable to that from ELISA. Notably, we and others have used this approach previously to establish murine phenotypes at the protein level (1-4).

*Apo A1 (Apolipoprotein A1); CD40L (CD40 Ligand); CRP (C Reactive Protein); EGF (Epidermal Growth Factor); FGF-9 (Fibroblast Growth Factor-9); FGF-basic (Fibroblast Growth Factor-basic); GCP-2 (Granulocyte Chemotactic Protein-2); GM-CSF (Granulocyte Macrophage-Colony Stimulating Factor); GST-alpha (Glutathione S-Transferase alpha); IFN-gamma (Interferon-gamma); IgA (Immunoglobulin A); IL- (Interleukin-); IP-10 (Inducible Protein-10); KC/GROalpha (Melanoma Growth Stimulatory Activity Protein); LIF (Leukemia Inhibitory Factor); MCP- (Monocyte Chemoattractant Protein-); M-CSF (Macrophage-Colony Stimulating Factor); MDC (Macrophage-Derived Chemokine); MIP- (Macrophage Inflammatory Protein-); MMP-9 (Matrix Metalloproteinase-9); MPO (Myeloperoxidase); OSM (Oncostatin M); RANTES (Regulation Upon Activation, Normal T-Cell Expressed and Secreted); SAP (Serum Amyloid P); SCF (Stem Cell Factor); SGOT (Serum Glutamic-Oxaloacetic Transaminase); TIMP-1 (Tissue Inhibitor of Metalloproteinase Type-1); TNF-alpha (Tumor Necrosis Factor-alpha); TPO (Thrombopoietin); VCAM-1 (Vascular Cell Adhesion Molecule-1); VEGF (Vascular Endothelial Cell Growth Factor); vWF (von Willebrand Factor). **CCR5 ligands are highlighted in yellow.**

◇ The least detectable dose (LDD) was determined as the mean \pm 3 standard deviations of 20 blank readings. Results below the LDD will be more variable than results above the LDD.

• Numbers in this row (1 to 6) correspond to the mouse identification number

^Statistical significance (p value) for comparison of the median values of $CCR5^{+/+}$ and $CCR5^{-/-}$ mice using Mann-Whitney test. Significant differences ($p < 0.05$) are shown in the main text (Table 1).

∇Undetectable levels are shown as zero.

References

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