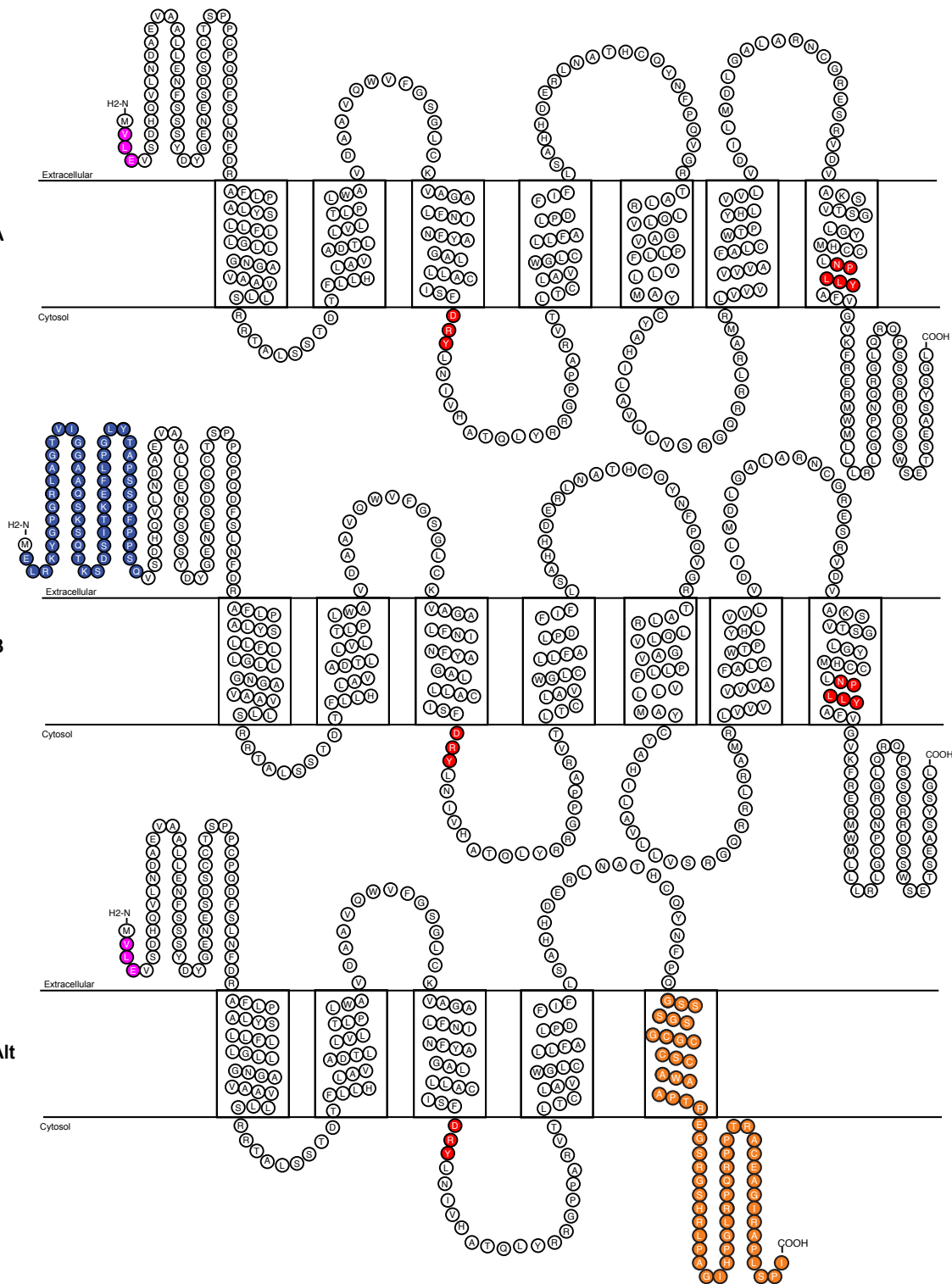


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Journal title: Molecular Pharmacology

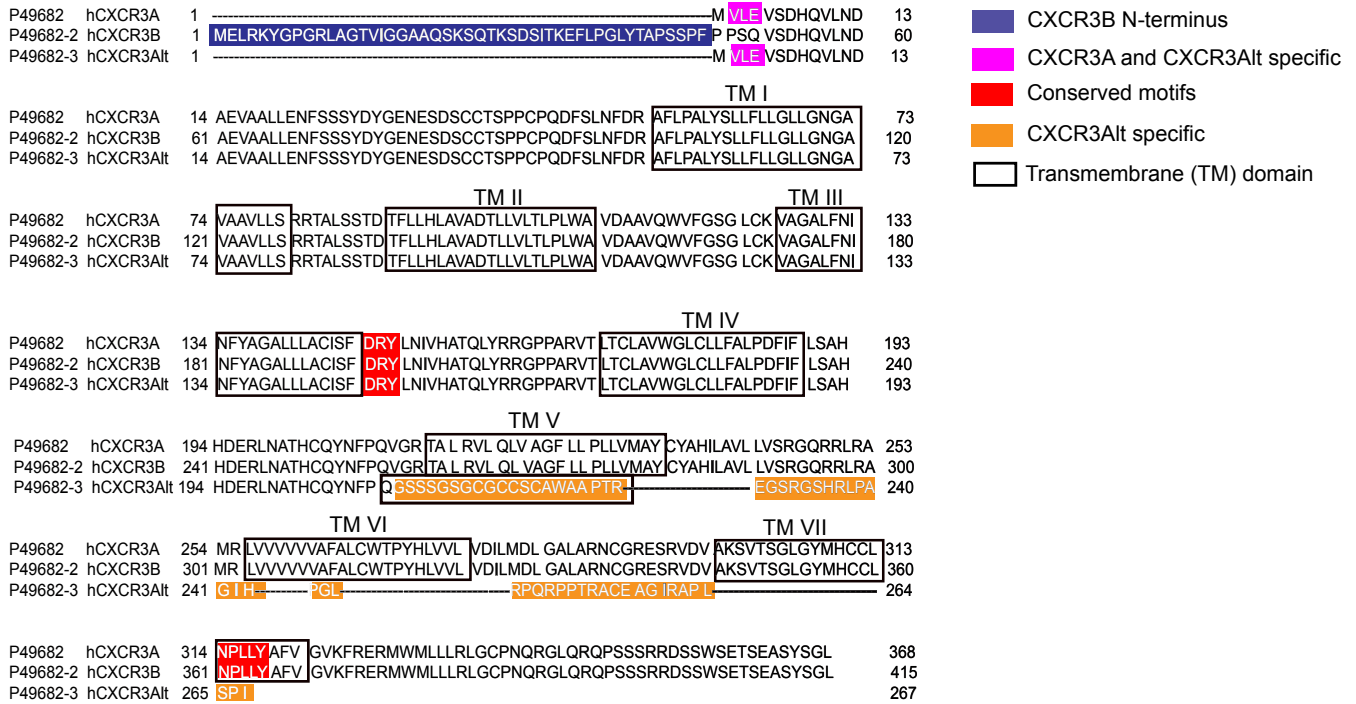


Supplemental Figure 1. Schematic representation of CXCR3 alternative splice variants. Residues in blue are unique to CXCR3B, in orange to CXCR3Alt, residues in red are conserved motifs and residues in magenta are common to CXCR3A and CXCR3Alt. The predicted positions of the TM domains common for all three variants is according to www.uniprot.org data base. The position of TM V of CXCR3Alt was predicted by an algorithm based analysis of TM domains (http://www.ch.embnet.org/software/TMPRED_form.html).

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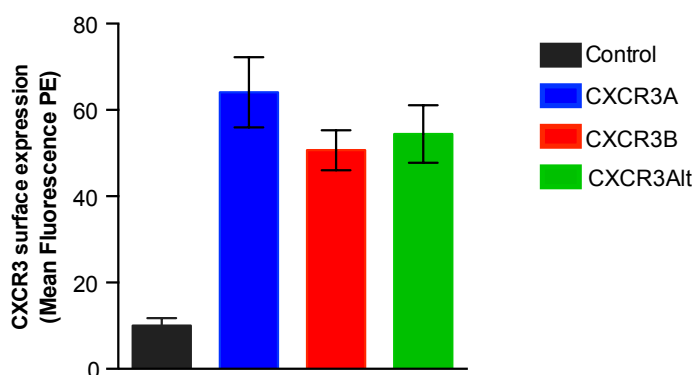
Supplemental Figure 2. Amino acid sequence alignment of CXCR3 alternative splice variants.

Identically to Supplemental Figure 1, the predicted positions of the TM domains common for all three variants is based on www.uniprot.org data base. The position of TM V of CXCR3Alt was predicted by an algorithm based analysis of TM domains (http://www.ch.embnet.org/software/TMPRED_form.html).

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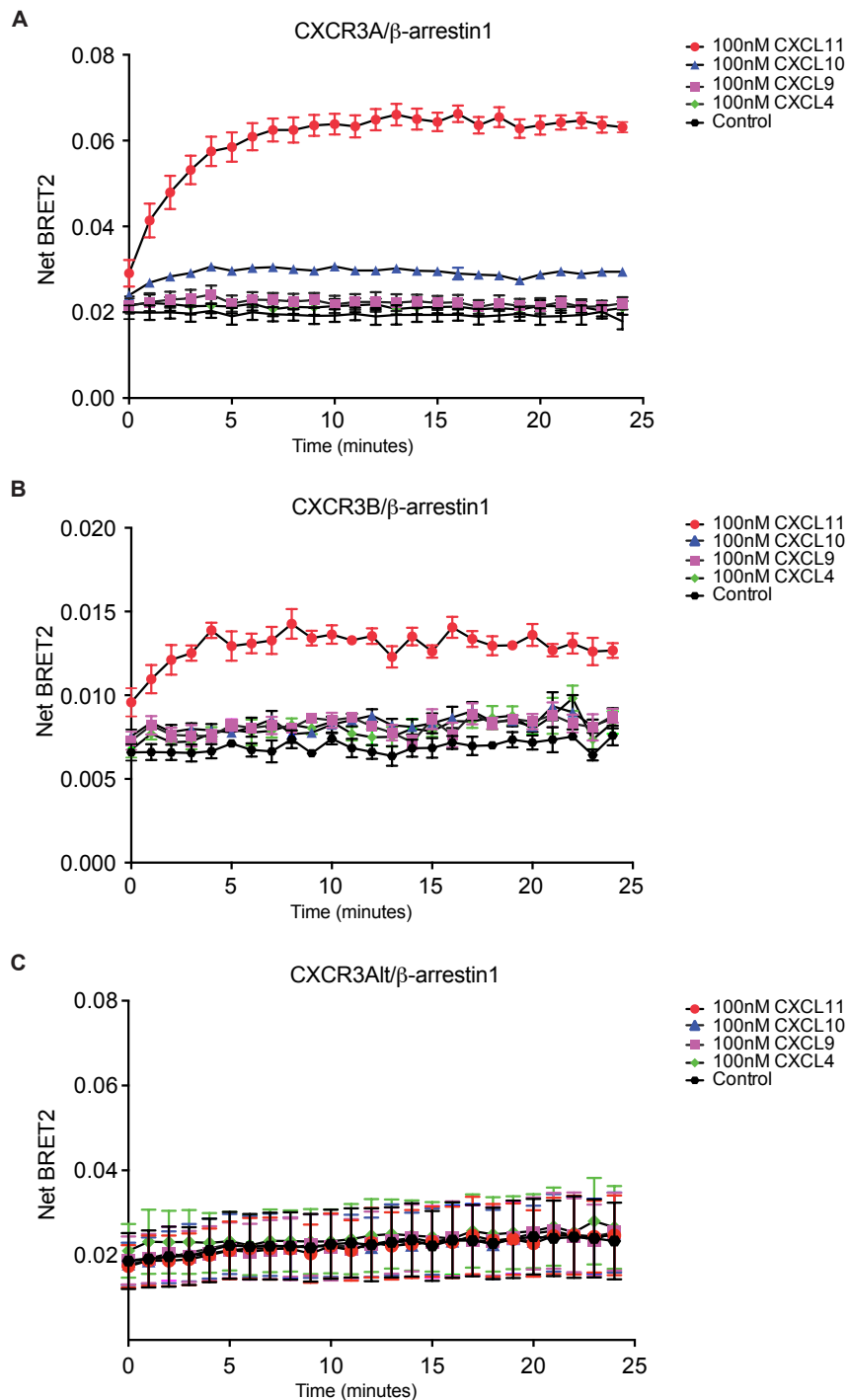


Supplemental Figure 3. Cell surface expression of CXCR3 splice variants. HEK 293T cells were transfected with 1.0 μ g of CXCR3A, 1.5 μ g of CXCR3B, 1.0 μ g of CXCR3Alt and 2 μ g of empty vector pcDNA3.1+ serving as a control. 24 hours after transfection cells were stained with anti-CXCR3 PE (clone 1C6) antibody and cell surface expression was measured by flow cytometry. Data are mean of nine to ten independent experiments \pm S.E.M.

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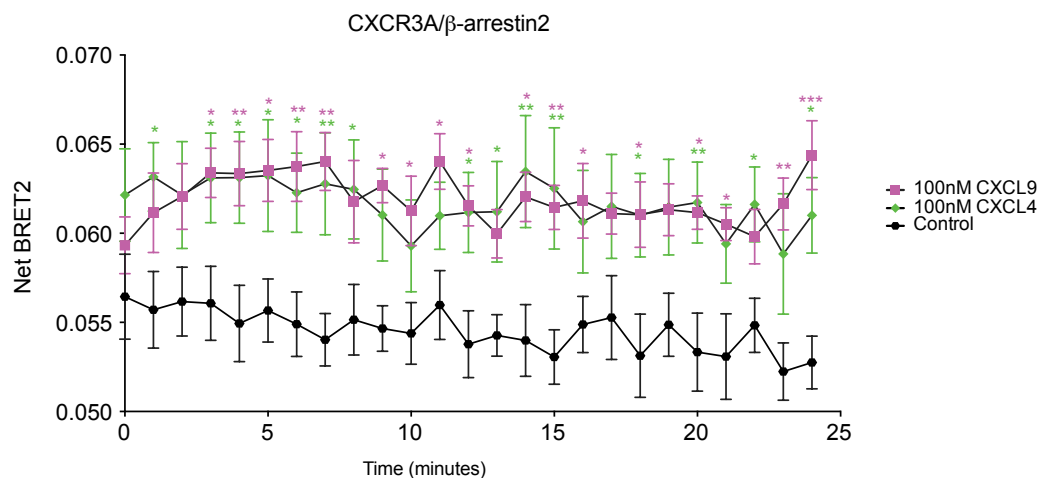


Supplemental Figure 4. Selective β -arrestin1 recruitment to CXCR3 variants in response to 100nM of chemokine. HEK293T cells transiently co-expressing the indicated CXCR3 variant GFP10 fusion with β -arrestin1-Rluc3 at BRETmax were incubated with the indicated ligands at room temperature. BRET was measured immediately following ligand addition. Data are reported as mean values of three to four independent experiments performed in duplicate \pm S.E.M.

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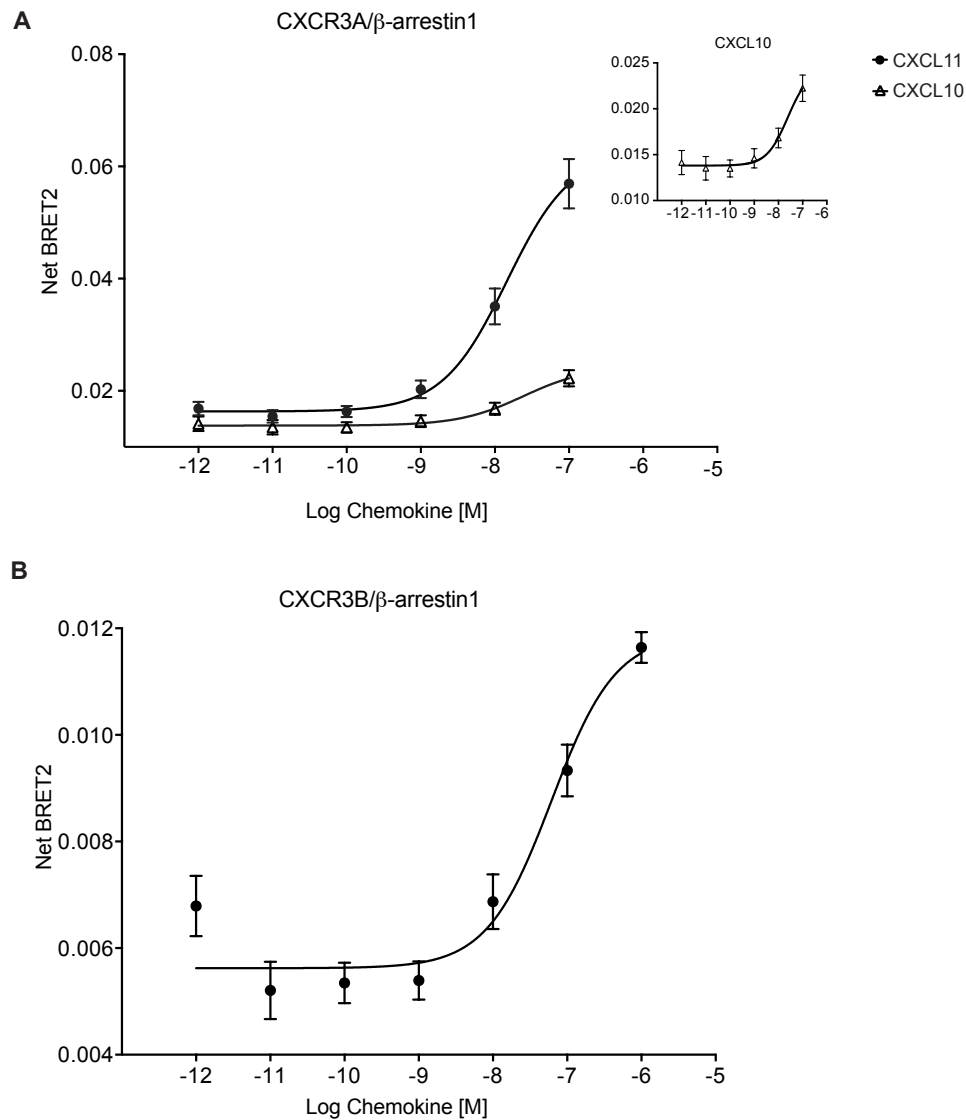
Supplemental Figure 5. β-arrestin2 recruitment to CXCR3A in response to 100nM CXCL9 and CXCL4.

These results are also shown in Fig4. of the manuscript on a different scale. HEK293T cells transiently co-expressing the indicated CXCR3 variant GFP10 fusion with β-arrestin2-Rluc3 at BRETmax were incubated with 100nM of the indicated ligands at room temperature. BRET was measured immediately following ligand addition in one minute intervals. Statistical significance of the differences between stimulated and control condition at a specific time: *, $p < 0.05$, **, $p < 0.01$, ***, $p < 0.001$, (Two-way ANOVA, Bonferroni's multiple comparison test). Data are reported as mean values of four independent experiments performed in duplicate \pm S.E.M.

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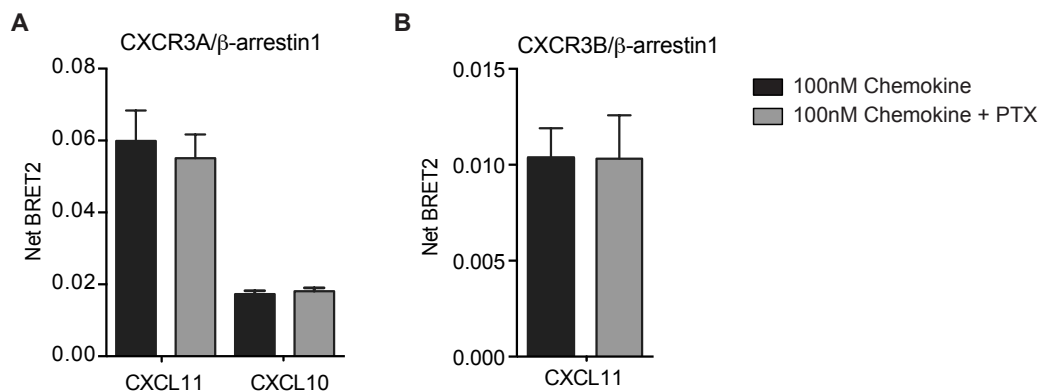


Supplemental Figure 6. CXCR3A and CXCR3B recruit β -arrestin1 in the presence of increasing concentrations of chemokine. HEK293T cells transiently co-expressing CXCR3A-GFP10 or CXCR3B-GFP10 with β -arrestin2-Rluc3 at BRETmax were incubated with increasing concentrations of the indicated ligands for 5 minutes at 37°C. BRET was measured five minutes following ligand addition. Data are reported as the mean values of three independent experiments performed in triplicate \pm S.E.M (see Table 1 for curve fitting parameters).

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Supplemental Figure 7. PTX treatment does not inhibit chemokine induced β -arrestin1 recruitment to CXCR3A and CXCR3B. HEK293T cells transfected with CXCR3-GFP10 fusion with β -arrestin1-Rluc3 were incubated with 100ng/ml PTX for 16 hours at 37°C. β -arrestin1 recruitment to A, CXCR3A and B, CXCR3B induced with 100nM of the indicated ligands in the absence or presence of PTX. Results represent data obtained from three to four independent experiments performed in triplicate \pm S.E.M.