

## human N-Formyl Peptide FPRL1 Receptor

Product No.: ES-610-M400UA

Lot No.: 1739636

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### Material Provided

Membranes: 1 x 400 units / 400  $\mu$ L frozen aliquot

### Product Information

Cellular Background: CHO-K1

GenBank Accession Number: M84562

Unit Size: 0.5  $\mu$ g protein / unit

Storage Buffer: 50 mM Tris-HCL (pH 7.4), 0.5mM EDTA, 10mM MgCl<sub>2</sub>, 10% sucrose.

Storage Conditions: Store at -80°C. Freeze-thaw is not recommended as it can affect product performance and homogeneity. In order to minimize negative impact of freeze-thawing, flash freeze in liquid nitrogen for 30 seconds prior to transferring to -80°C.

Stability: This product is stable for at least 3 years from reception if used and stored under recommended conditions.

### Quality Control

$B_{max}$  and  $K_d$  are determined using radioactive saturation binding assays (Figure 1). Protein concentration is determined using the BCA method <sup>(1)</sup>. Ratio-to-Reference (RTR) is determined by dividing the maximal signal of the current lot ( $B_{max}$  in fmoles) by the maximal signal of a pre-defined reference tested in parallel. RTR is an indicator of lot-to-lot consistency. \*We certify that these results meet our quality release criteria.

Ratio-to-Reference (RTR): N/A

Expression Level ( $B_{max}$ ): 15 pmol/mg membrane protein.

$K_d$  for [<sup>125</sup>I]-WKYMVm: 0.13 nM

Protein Concentration: 0.5  $\mu$ g/ $\mu$ L

(1) Smith, P.K., et al. (1985). *Anal. Biochem.* 150, 76-85.

Recommended Assay Conditions

Assay Buffer:	50 mM Tris-HCl pH 7.4, 0.3% BSA
Wash Buffer:	50 mM Tris-HCl pH 7.4
Binding Protocol:	Binding assays are performed in 200 $\mu$ L total volume according to the following conditions:
1 - Membrane dilution:	0.05 mL of membranes + 7.45 mL assay buffer (1:150 dilution)
2 - Incubation:	25 $\mu$ L of incubation buffer or WKYMV(D-Met)-NH <sub>2</sub> (Phoenix 072-12) 50 $\mu$ M final for non specific binding (Saturation binding assay)
	<i>For competition binding assay: 25 <math>\mu</math>L of reference compounds at decreasing concentrations (see figure 2)</i>
	25 $\mu$ L of radioligand at the appropriate concentration (see graph below)
	150 $\mu$ L of diluted membranes
3 - Incubation time:	60 minutes at 27°C
4 - Filtration:	aspirate and wash 9 x 500 $\mu$ L with ice cold wash buffer over GF/C filter (presoaked in 50 mM Tris-HCl pH 7.4, 0.3% BSA).

Lot Specific Data

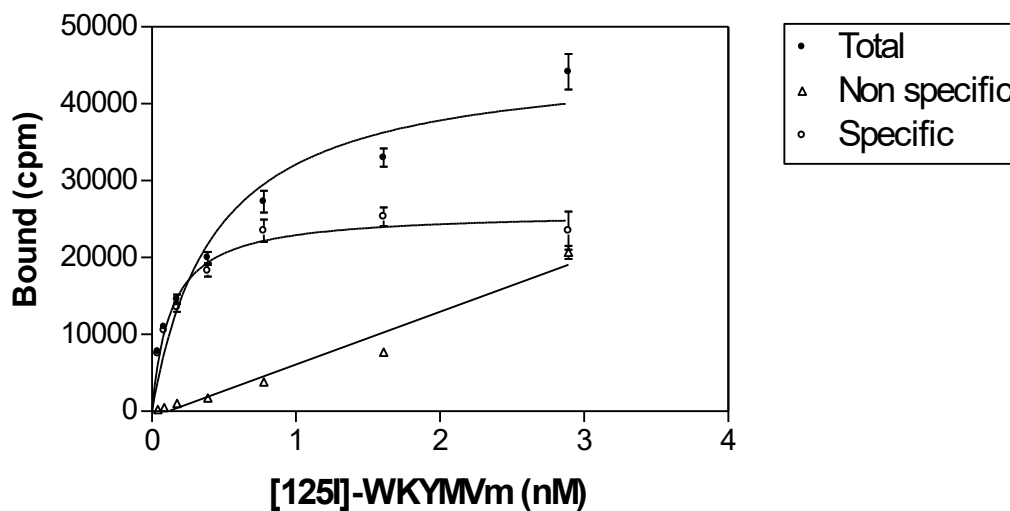


Figure 1: Saturation binding assay curve (filtration)  
 96-well saturation binding assay curve (0.5  $\mu$ g membranes/well, TopCount®) using [125I]-WKYMMm (Revvity NEX386 Lot No.: JV62970)

Typical Product Data

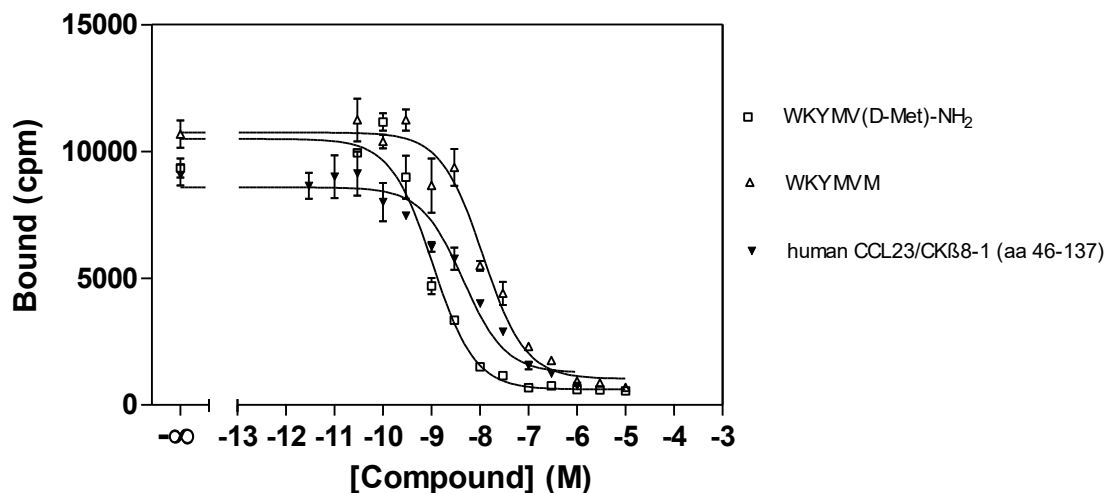


Figure 2: Competition binding assay curve (filtration)  
 96-well competition binding assay curve (0.5 µg membranes/well, TopCount®). Recommended radioligand concentration = 0.2 nM.

\*Even though two sites can be observed occasionally with some ligands, the data presented is derived from single site fitting.

Reference Compounds	Ki (nM)
WKYMV(D-Met)-NH <sub>2</sub>	0.51
WKYMVM	5.6
human CCL23/CKβ8-1 (aa 46-137)	2.2

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