CHARACTERIZATION OF LIH383, AN ANALGESIC PEPTIDE TARGETING THE NEWLY IDENTIFIED OPIOID RECEPTOR ACKR3

Alexis Lavallée[1], Ester Cassano[2], Marc-André Dansereau[1], Ulrike Froehlich[1], Alexandre Murza[1], Martyna Szpakowska[2], Andy Chevigné[2], Philippe Sarret[1], Pierre-Luc Boudreault[1]

1. Institut de Pharmacologie de Sherbrooke, Université de Sherbrooke, Sherbrooke, Canada. 2. Department of Infection and Immunity, Luxembourg Institute of Health, Luxembourg

INTRODUCTION

ACKR3 (Atypical Chemokine Receptor 3) is considered atypical because, unlike other chemokine receptors, it does not recruit G proteins. Recently, ACKR3 was identified as having an affinity for endogenous opioid peptides. It is speculated that ACKR3 may act as a scavenger receptor for these endogenous opioids, thereby reducing their analgesic effects. Following this discovery, LIH383, a peptide agonist with low nanomolar potency, was developed[1].We believe that modulating ACKR3 will lead to opioid-dependent analgesia by increasing the availability of endogenous opioids

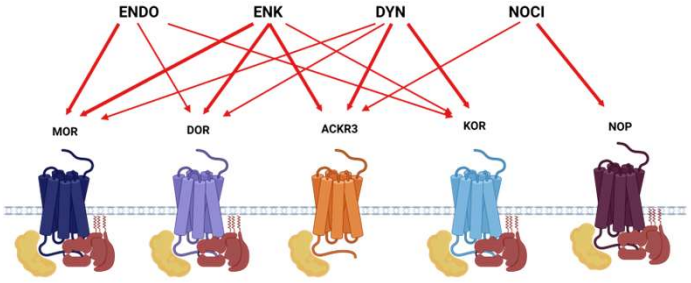


Figure 1. Binding of endogenous opioids to opioid receptors.

OBJECTIVES

- Characterize the effect of LIH383 and other ACKR3 modulators in the formalin pain model
- Determine the key molecular determinant for LIH383 binding and activity

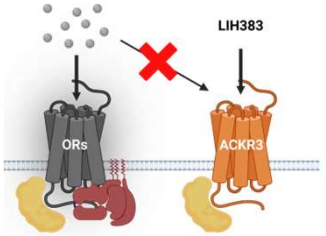


Figure 2. Proposed mechanism of action for ACKR3-mediated analgesia

FORMALIN PAIN MODEL

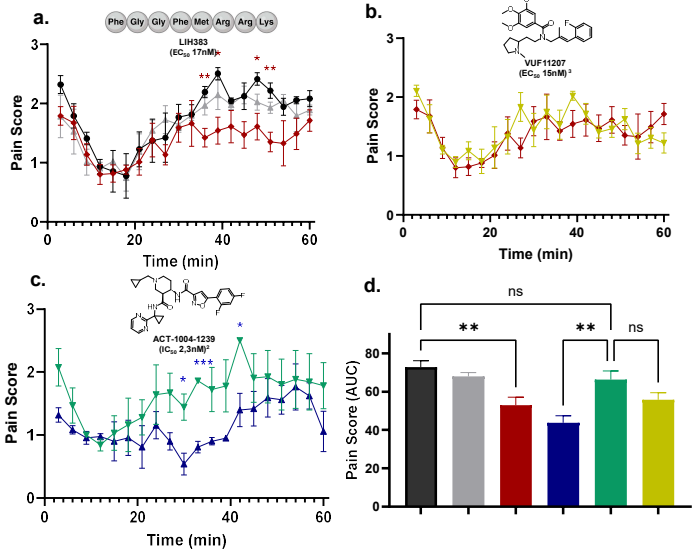


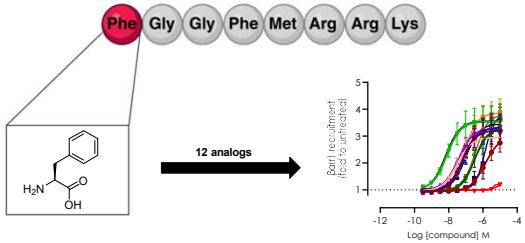
Figure 3. Antinociceptive action of ACKR3 modulators. **a-c.** Time-dependent antinociceptive action of LIH383 (agonist), VUF11207 (agonist), and ACT-1004-1239 (antagonist) in the formalin pain model. **d.** Area under the curve of ACKR3 modulators in the inflammatory phase of the formalin pain model.

SAR STUDY

Table 1. Effect of d-Amino Acid Substitution and N-Methylation on Activity and Binding

Position	pEC_{50} (nM ± CI) (%Max)	pIC_{50} (nM ± CI)
LIH383	7,86 ± 0,36	8,05 ± 0,58
d-Amino Acids Scan		
1	6,22 ± 0,12 (118)	5,91 ± 0,76
4	7,56 ± 0,20 (110)	7,43 ± 0,53
5	6,11 ± 0,18 (113)	5,61 ± 0,89
6	6,78 ± 0,13 (116)	5.95 ± 0,81
7	7,44 ± 0,18 (109)	6,86 ± 0,62
8	6,86 ± 0,28 (110)	6,80 ± 0,53
N-Methylation Scan		
1	7.79 ± 0,46 (89)	7,44 ± 0,57
2	7,16 ± 0,30 (102)	6,49 ± 0,69
3	6,10 ± 0,34 (103)	5,77 ± 0,75
4	5,89 ± 0,24 (109)	5,48 ± 0,75
5	6,26 ± 0,21 (106)	6,05 ± 0,90
6	5.79 ± 0,25 (109)	5,89 ± 0,91
7	7,30 ± 0,35 (112)	7,09 ± 1,1
8	8,02 ± 0,50 (102)	7.92 ± 0,42

P1 SUBSTITUTION



Phenylalanine derivatives				
pEC_{50} (nM ± CI)	8,20 ± 0,59	8,10 ± 0,52	6,50 ± 0,35	5,89 ± 0,33
pIC_{50} (nM ± CI)	8,75 ± 0,14	8,66 ± 0,26	6,20 ± 0,38	5,65 ± 0,77

Figure 3. Effect of Phenylalanine Analogs on Activity and Binding

CONCLUSION

- It was shown for the first time that modulation of ACKR3 results in analgesia.
- The antagonist demonstrated greater efficacy in inhibiting ACKR3's scavenger activity.
- The N-terminal of LIH383 exhibited high sensitivity to modification.
- The positive charge at the N-terminal of LIH383 is essential for its binding to ACKR3

REFERENCES

1. Meyrath, M., Szpakowska. Nat Commun. (2020)
2. S., Richard-Bildstein. J. Med. Chem. (2020)
3. M., Wijtmans. Eur. J Med. Chem. (2012)