A Uniquely Human Mechanism Regulating the Inflammatory Response to Injury

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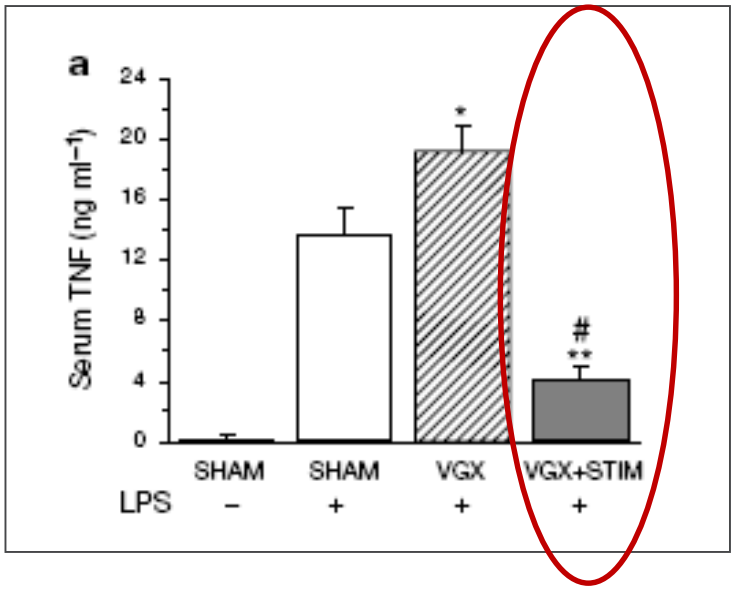
* No Disclosures

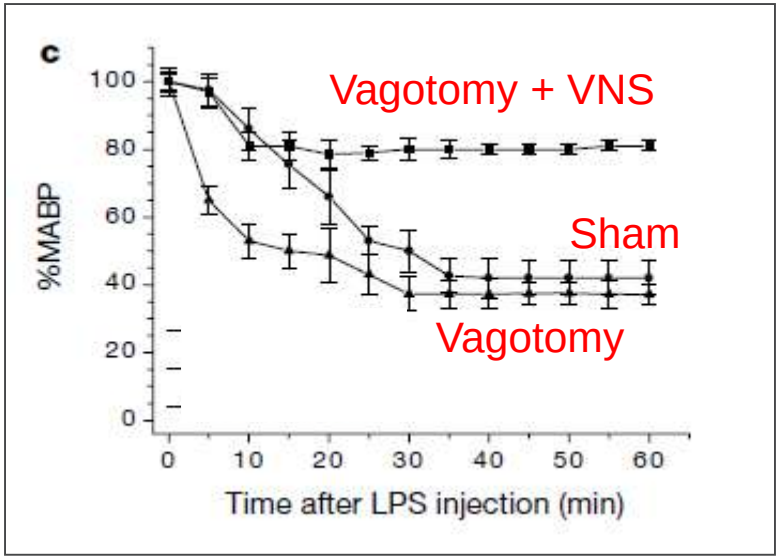
Vagus nerve stimulation attenuates the systemic inflammatory response to endotoxin

Lyudmila V. Borovikova*, Svetlana Ivanova*, Minghuang Zhang*, Huan Yang*, Galina I. Botchkina*, Linda R. Watkins†, Haichao Wang‡, Naji Abumrad§, John W. Eaton¶ & Kevin J. Tracey§

Serum TNF-α



Development of Shock (Mean Arterial BP)



Borovikova LV, et al. Nature 2000;405:458-462

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Vagal nerve stimulation protects against burn-induced intestinal injury through activation of enteric glia cells

Todd W. Costantini,[1] Vishal Bansal,[1] Michael Krzyzaniak,[1] James G. Putnam,[1] Carrie Y. Peterson,[1] William H. Loomis,[1] Paul Wolf,[2] Andrew Baird,[1] Brian P. Eliceiri,[1] and Raul Coimbra[1]

Targeting α-7 Nicotinic Acetylcholine Receptor in the Enteric Nervous System

A Cholinergic Agonist Prevents Gut Barrier Failure after Severe Burn Injury

Todd W. Costantini, Michael Krzyzaniak, Gerald A. Cheadle, James G. Putnam, Ann-Marie Hageny, Nicole Lopez, Brian P. Eliceiri, Vishal Bansal, and Raul Coimbra

A pharmacologic approach to vagal nerve stimulation prevents mesenteric lymph toxicity after hemorrhagic shock

Koji Morishita, MD, Todd W. Costantini, MD, Akinori Ueno, PhD, Vishal Bansal, MD, Brian Eliceiri, PhD, *and* **Raul Coimbra, MD, PhD,** *San Diego, California*

Costantini, et al. Am J Physiol-GI Liver Physiol. 2010;299:G1308-18
Costantini, et al. Am J Pathol. 2012;181:478-486
Morishita et al. J Trauma Acute Care Surg. 2015;78:52-59

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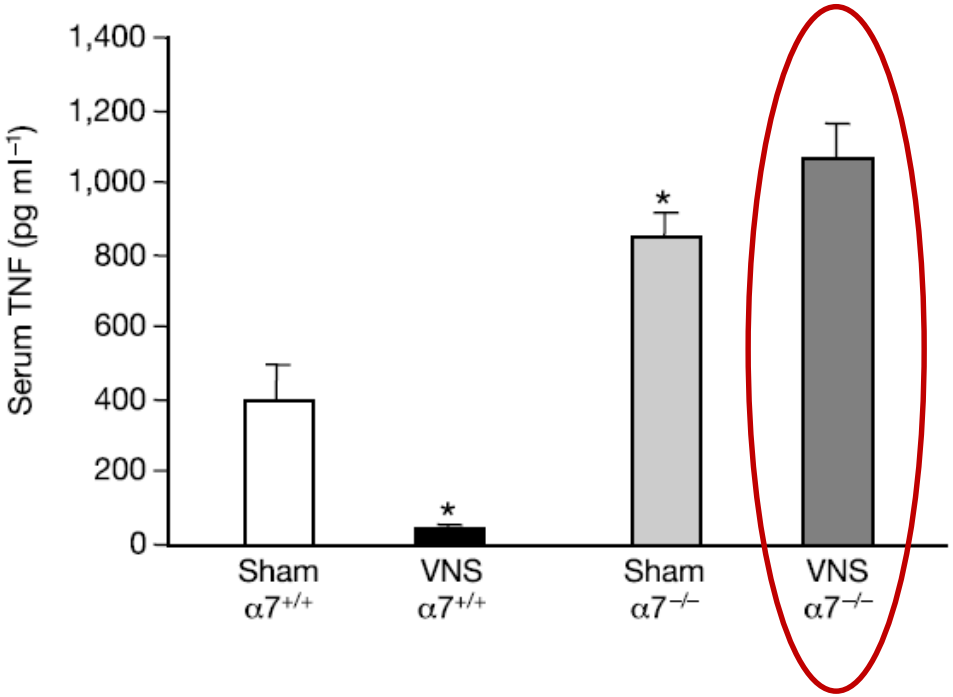
letters to nature

Nicotinic acetylcholine receptor α7 subunit is an essential regulator of inflammation

Hong Wang*, Man Yu*, Mahendar Ochani*, Carol Ann Amella*, Mahira Tanovic*, Seenu Susarla*, Jian Hua Li*, Haichao Wang*, Huan Yang*, Luis Ulloa*, Yousef Al-Abed†, Christopher J. Czura* & Kevin J. Tracey*

Alpha-7 nAchR Gene Knock Out Eliminates Vagal Responsiveness



Wang et al. Nature. 2003;421:384-388

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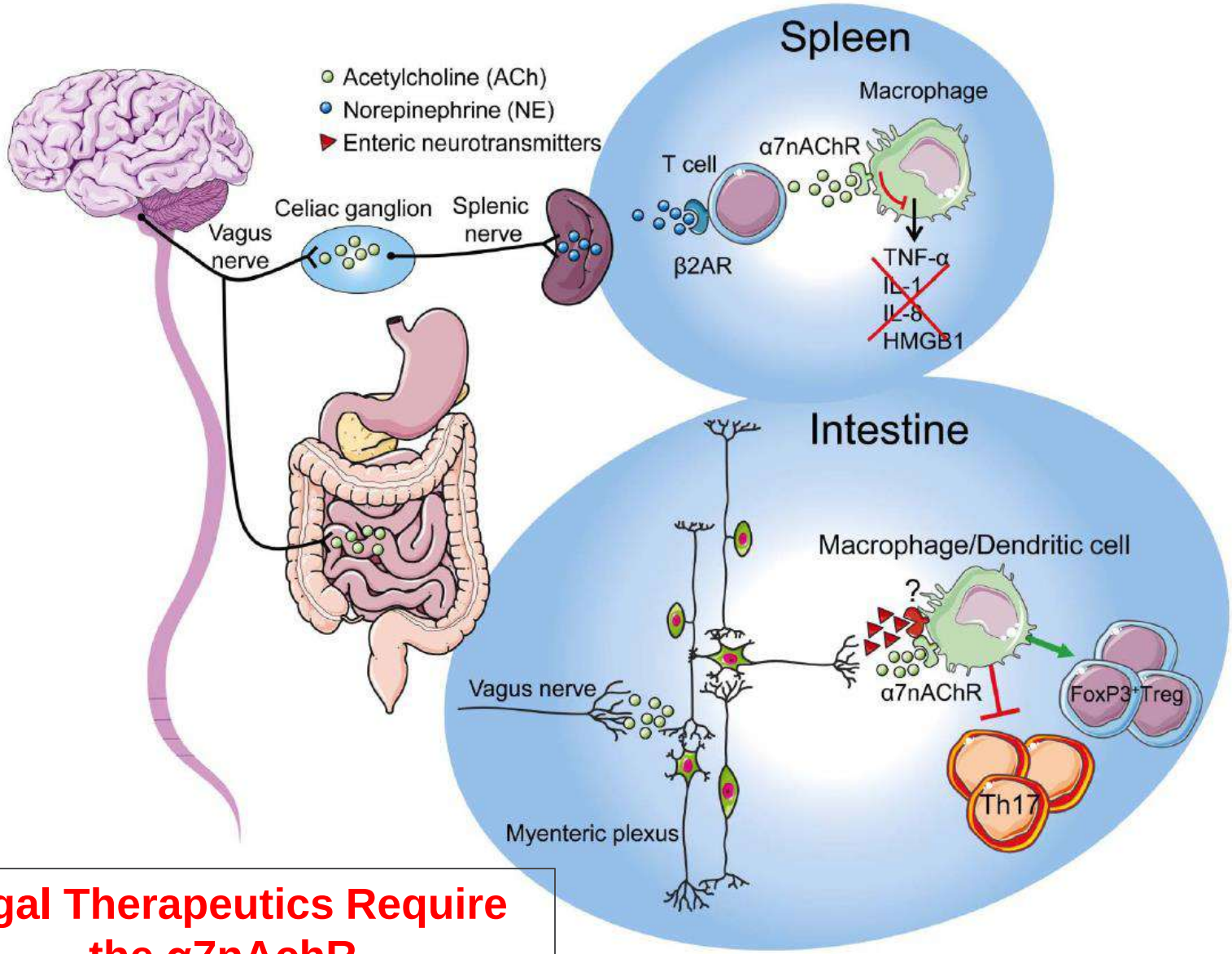

Vagal Therapeutics Require the α7nAchR

Matteoli and Boeckxstaens. Gut. 2013;62:1214-1222

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Delivery of Ligands to Mimic the Natural Biology of α7nAchR

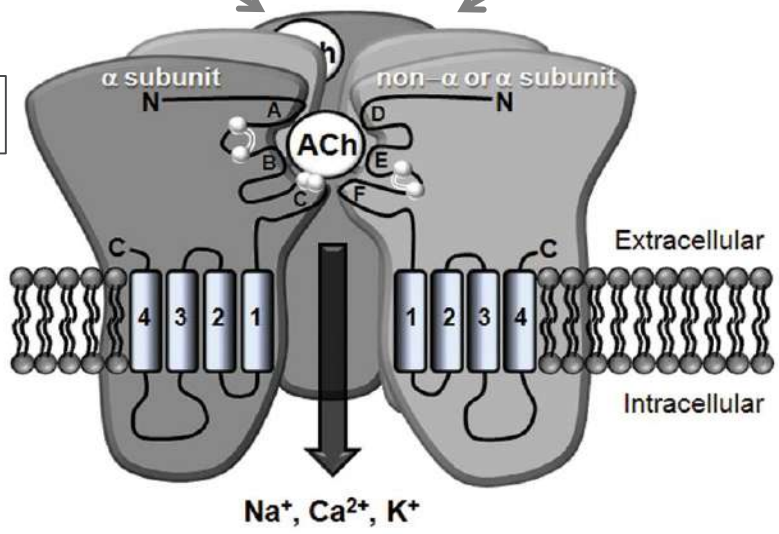
NATURE
(from vagus nerve)

Pharmaceutics
(nicotine, etc…)

Ligand

Agonist

α7nACh Receptor



ANTI-INFLAMMATION

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Vagal Agonists Have Had Limited Effectiveness in Human Trials Thus Far…

Vagus nerve stimulation inhibits cytokine production and attenuates disease severity in rheumatoid arthritis

Frieda A. Koopman[a], Sangeeta S. Chavan[b], Sanda Miljko[c], Simeon Grazio[d], Sekib Sokolovic[e], P. Richard Schuurman[f], Ashesh D. Mehta[g], Yaakov A. Levine[h], Michael Faltys[h], Ralph Zitnik[h], Kevin J. Tracey[b], and Paul P. Tak[a,1,2,3,4]

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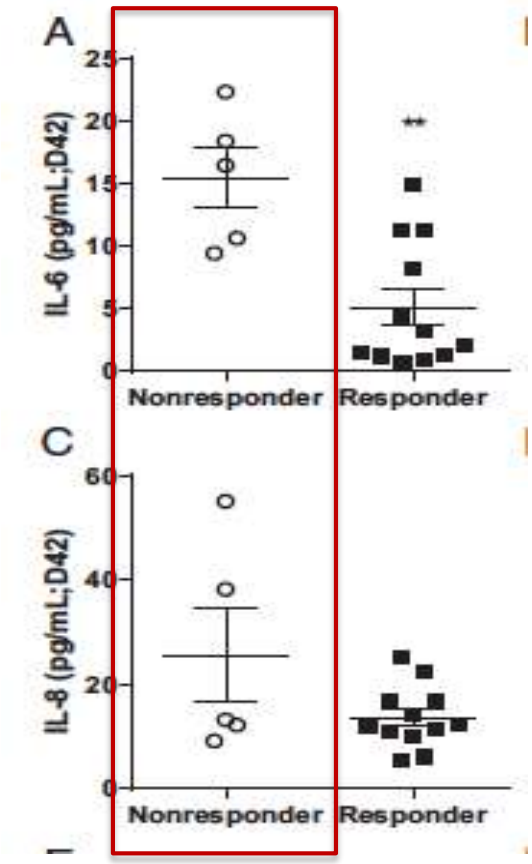

Koopman, et al. PNAS. 2016;113;8284-9

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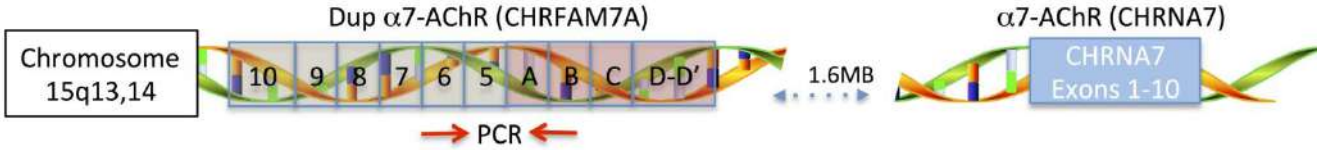
Does the Uniquely Human Gene CHRFAM7A Regulate a Human Response to Injury?

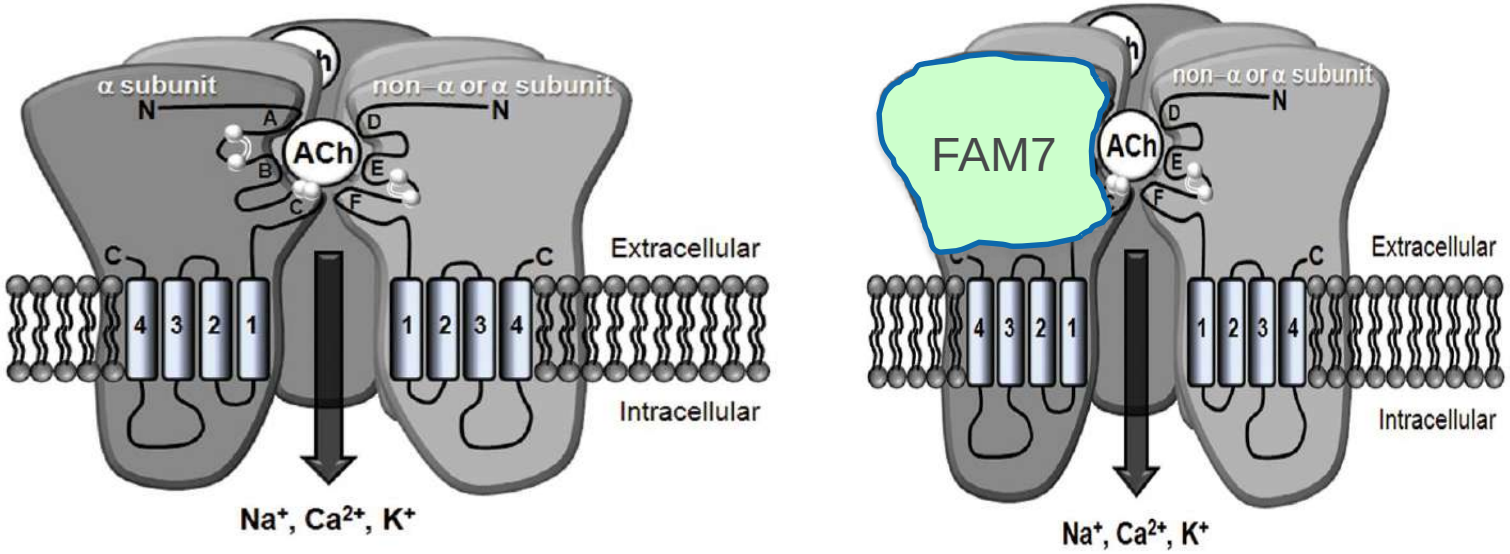


JLB

Review

CHRFAM7A, a human-specific and partially duplicated α7-nicotinic acetylcholine receptor gene with the potential to specify a human-specific inflammatory response to injury

Todd W. Costantini, Xitong Dang, Raul Coimbra, Brian P. Eliceiri, and Andrew Baird[1]



Costantini et al. J Leukoc Biol. 2015;97:247-257

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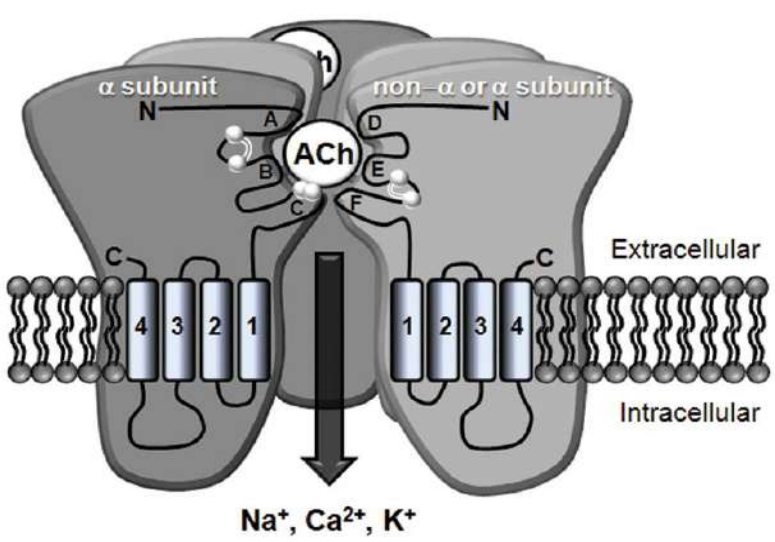
Agonist

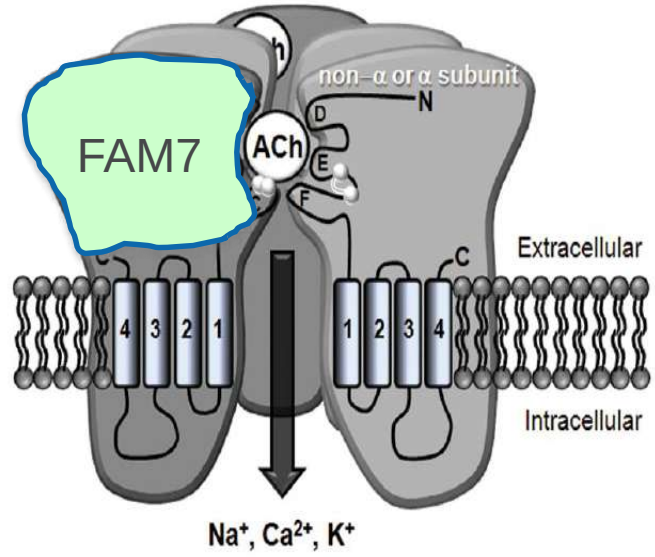
EFFECTIVE

- Mouse
- Humans with low CHRFAM7A

NOT EFFECTIVE

- Humans with high CHRFAM7A
- α7nAchR knock out mice





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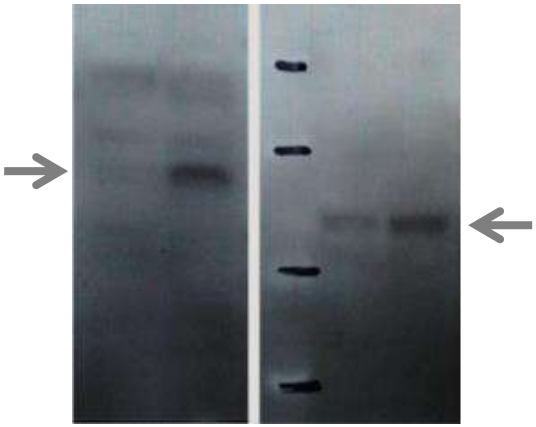
CHRFAM7A is a Uniquely Human Open Reading Frame that Produces Protein

A. anti-CHRFAM7A peptide
Immunoblotting

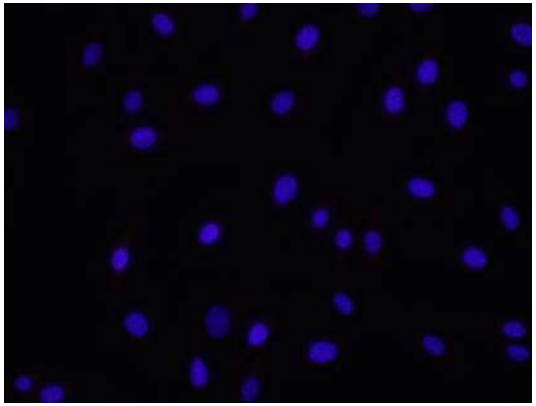
3T3 THP1



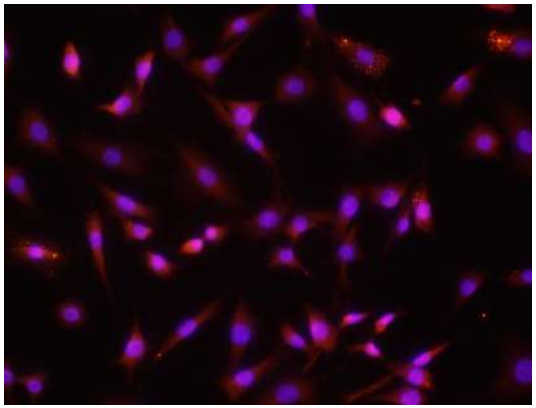
Forced Expression (Mouse) Over-Expressed (Human)

B. anti-CHRFAM7A peptide
Immunostaining

Parental BALB-3T3

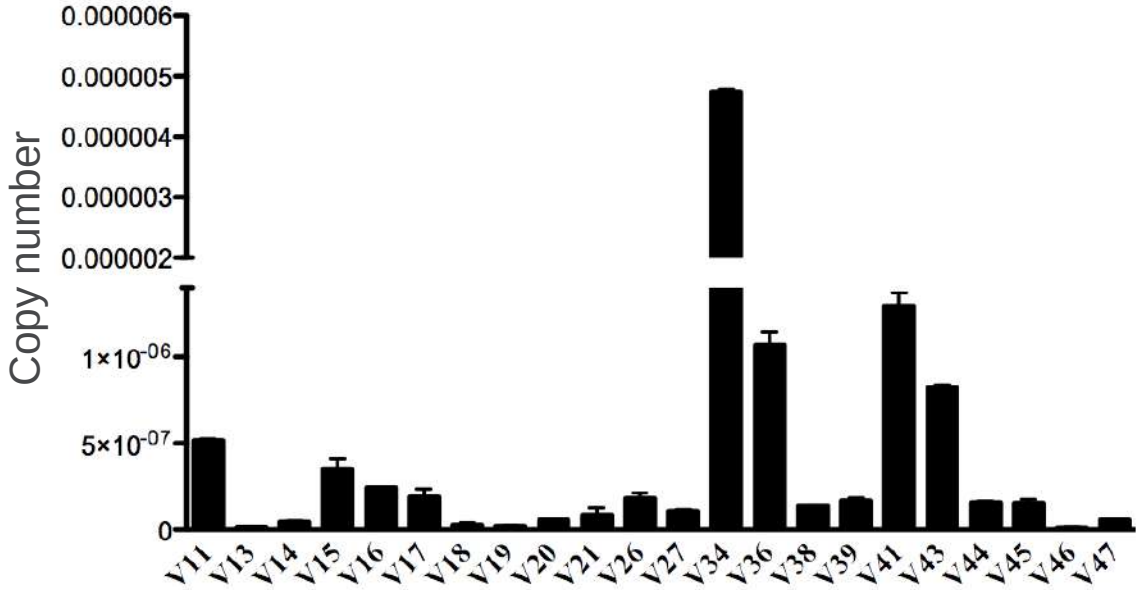


CHRFAM7A BALB-3T3

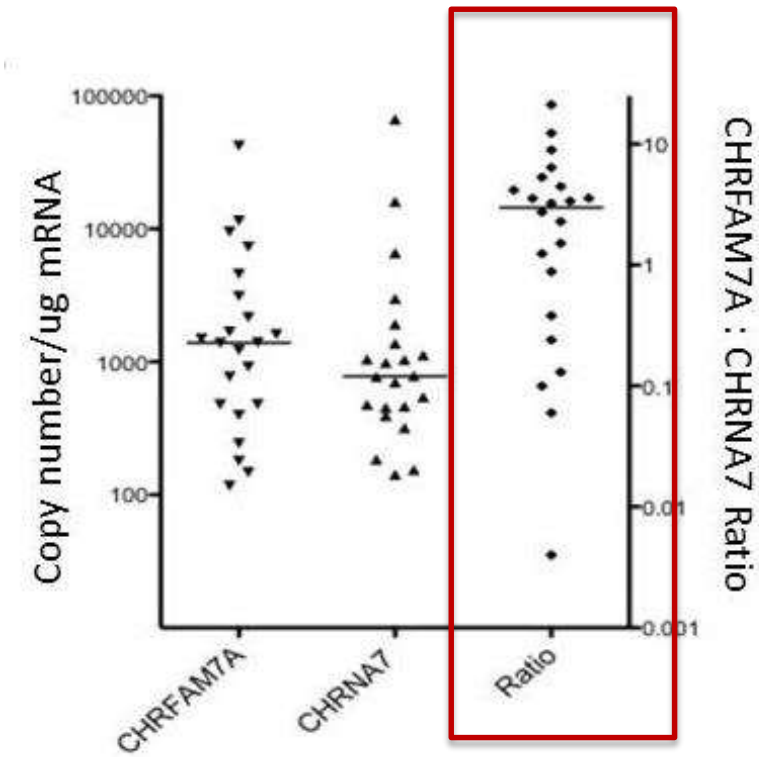


CHRFAM7A Expression is Variable in Donor Leukocytes

200-fold Differences in CHRFAM7A Expression Between Normal Human Leukocyte Donors



CHRFAM7A to CHRNA7 Expression Ratio in Human Leukocytes



Up to ***10,000-fold*** Differences in CHRFAM7A/CHRNA7 Ratio Between Normal Human Leukocyte Donors

Costantini et al. Mol Med. 2015;21:323-36

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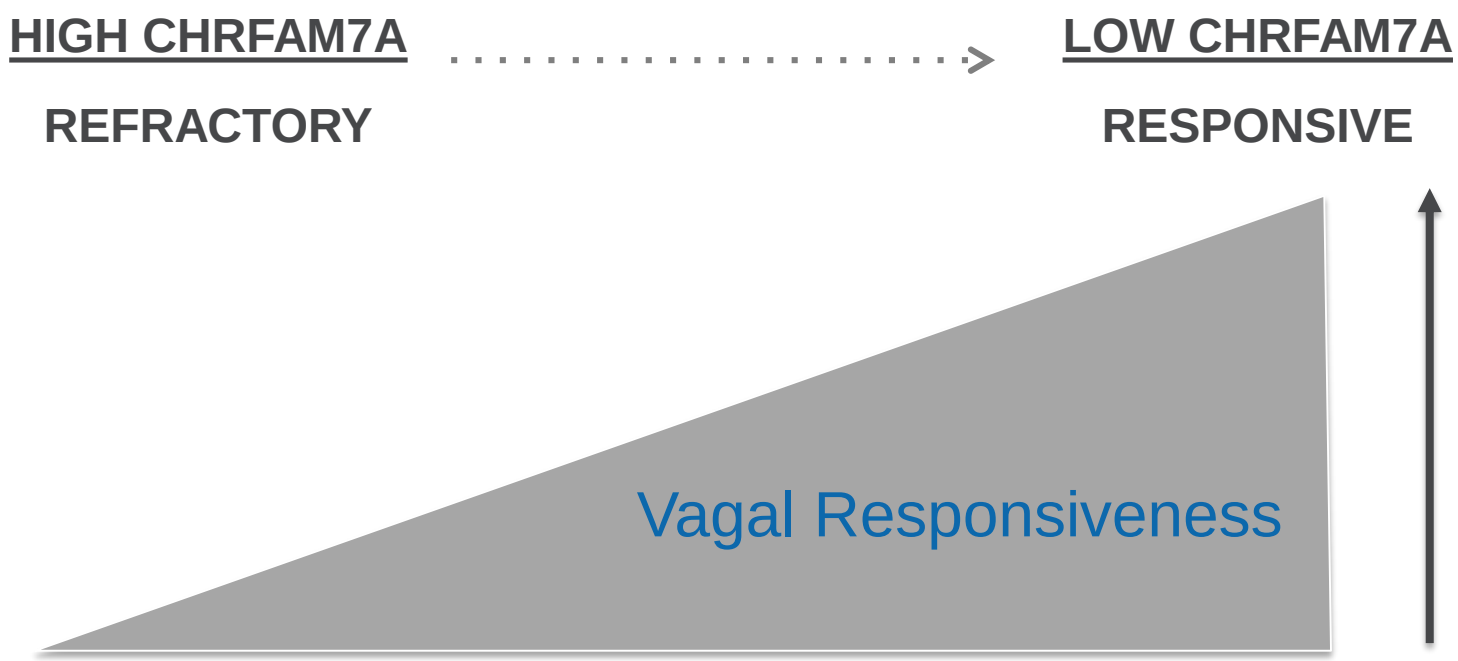
Hypothesis:

CHRFAM7A contributes to human variability in the inflammatory response and modulates the response to vagal therapeutics

CHRFAM7A: Modulator of Human Vagal Responsiveness?

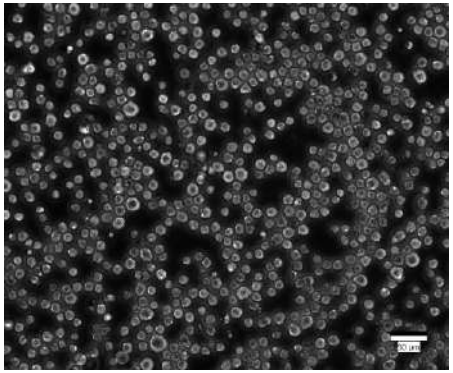


Does CHRFAM7A have Biologic Activity?

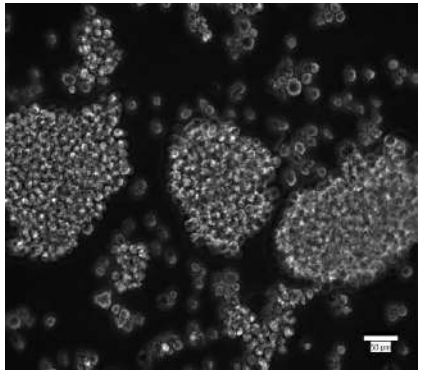
A Human-Specific α7-Nicotinic Acetylcholine Receptor Gene in Human Leukocytes: Identification, Regulation and the Consequences of *CHRFAM7A* Expression

Todd W Costantini,[1] Xitong Dang,[1,2*] Maryana V Yurchyshyna,[1] Raul Coimbra,[1] Brian P Eliceiri,[1] and Andrew Baird[1]*

Vector THP1



THP1-CHRFAM7A



Pathways altered by CHRFAM7A gene expression

Enriched Pathways	P-value
TGF-beta signaling pathway	2.76E-002
Insulin signaling pathway	2.23E-002
Leukocyte transendothelial migration	1.78E-002
RIG-I-like receptor signaling pathway	1.78E-002
Osteoclast differentiation	1.74E-002
Cytokine-cytokine receptor interaction	1.64E-002

Costantini et al. Mol Med. 2015;21:323-36

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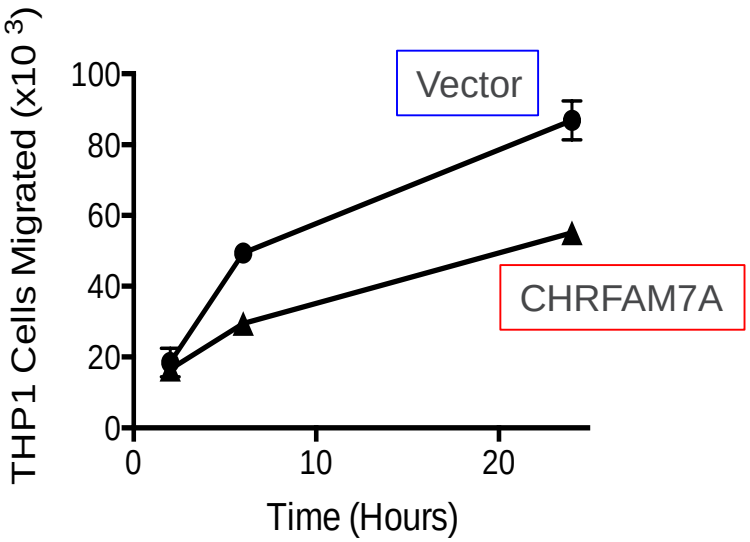
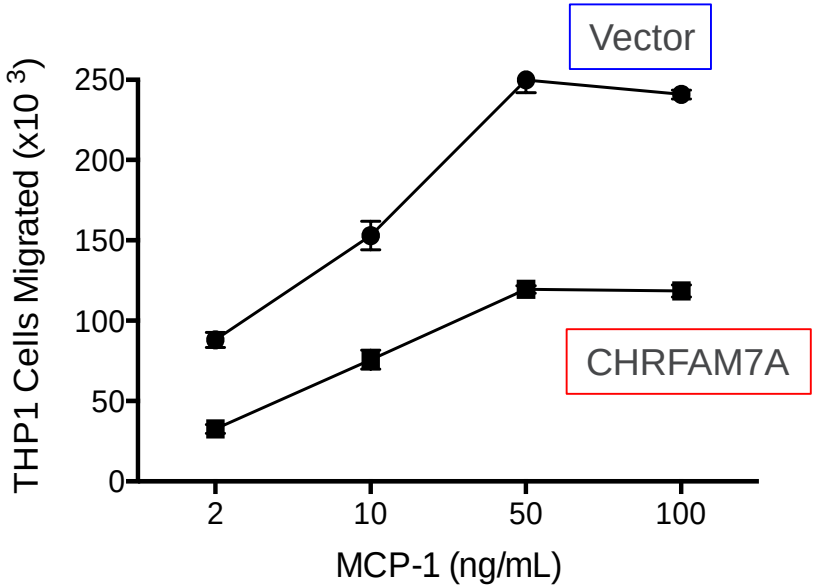
CHRFAM7A Over-expression Decreases Leukocyte Migration

Migration Assay





Costantini et al, unpublished data

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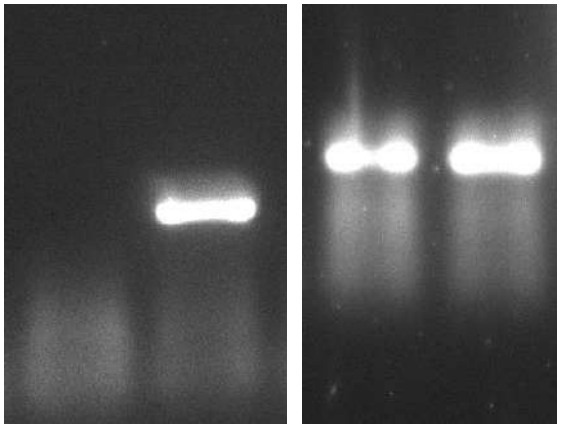
CHRFAM7A introduced into PC12 cells

PCR

CHRFAM7A CHRNA7



Vector CHRFAM7A Vector CHRFAM7A

Immunoblot

42kDa

Vector CHRFAM7A

Costantini et al, unpublished data

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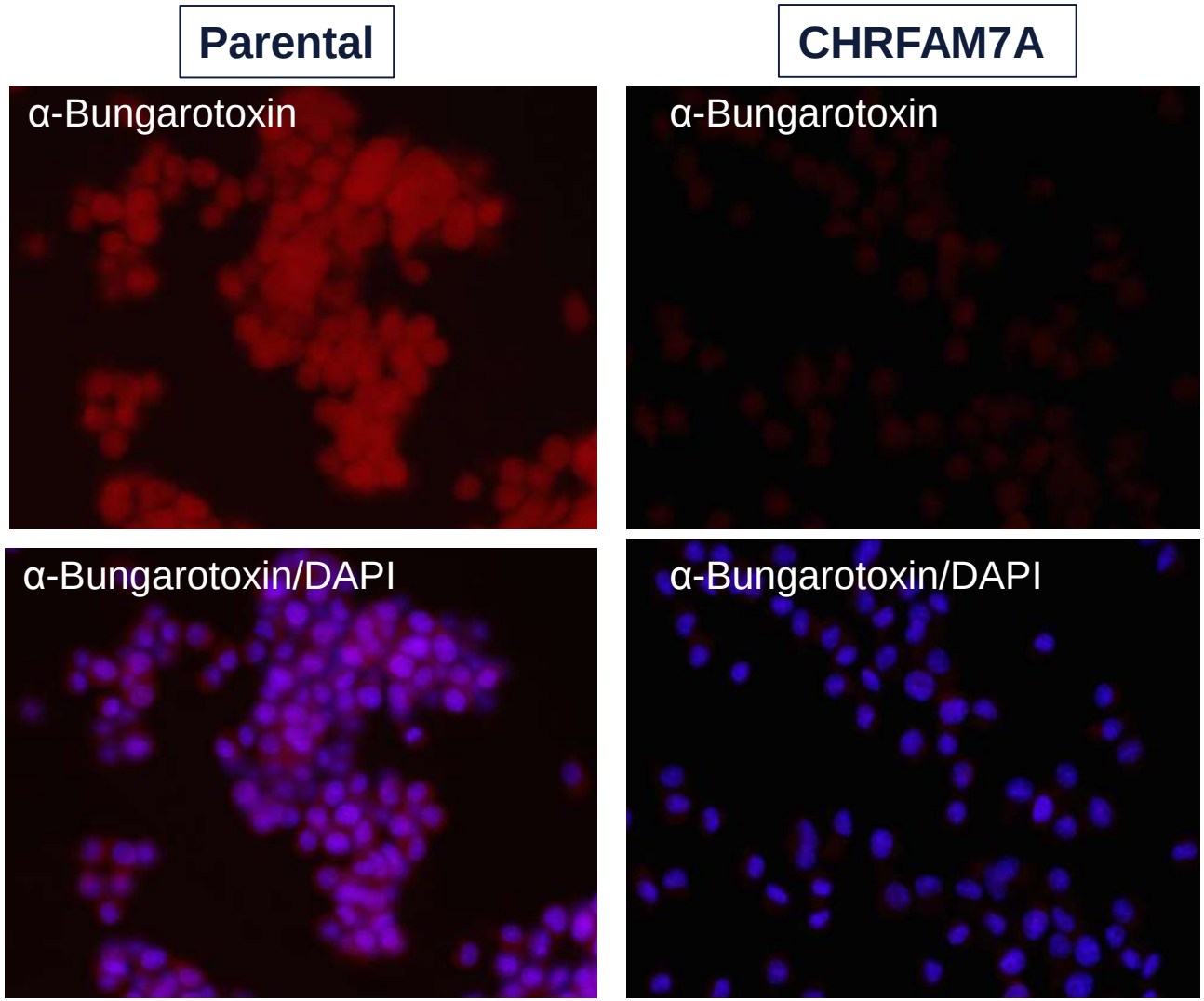
CHRFAM7A decreases ligand binding to the a7nAchR



40x; exposure 1/3 sec

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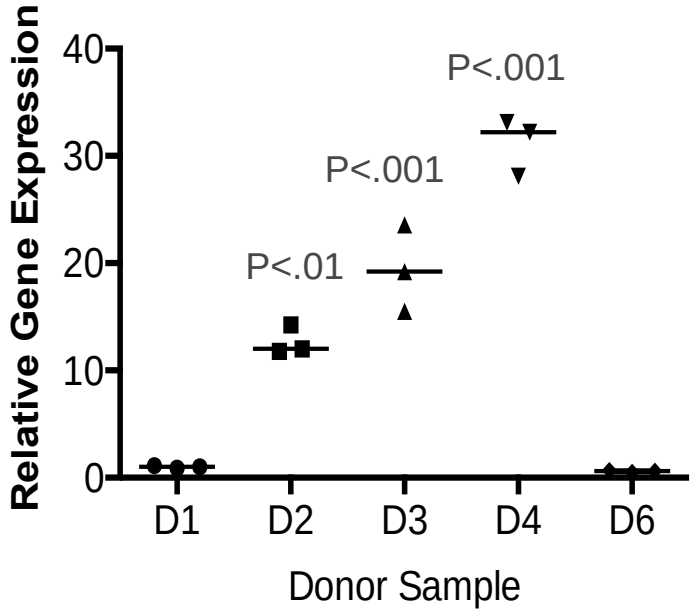
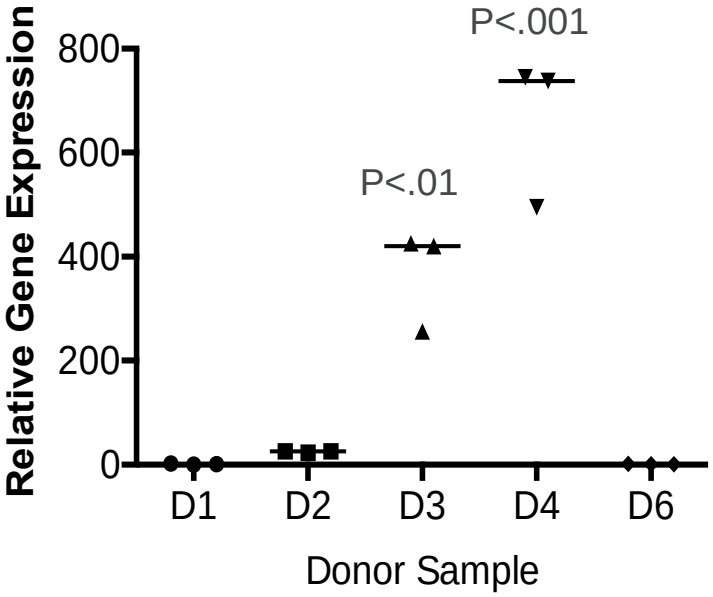
CHRFAM7A Gene Expression is Variable in Human Macrophages

CHRFAM7A



CHRFAM7A:CHRNA7 Ratio



Costantini et al, unpublished data

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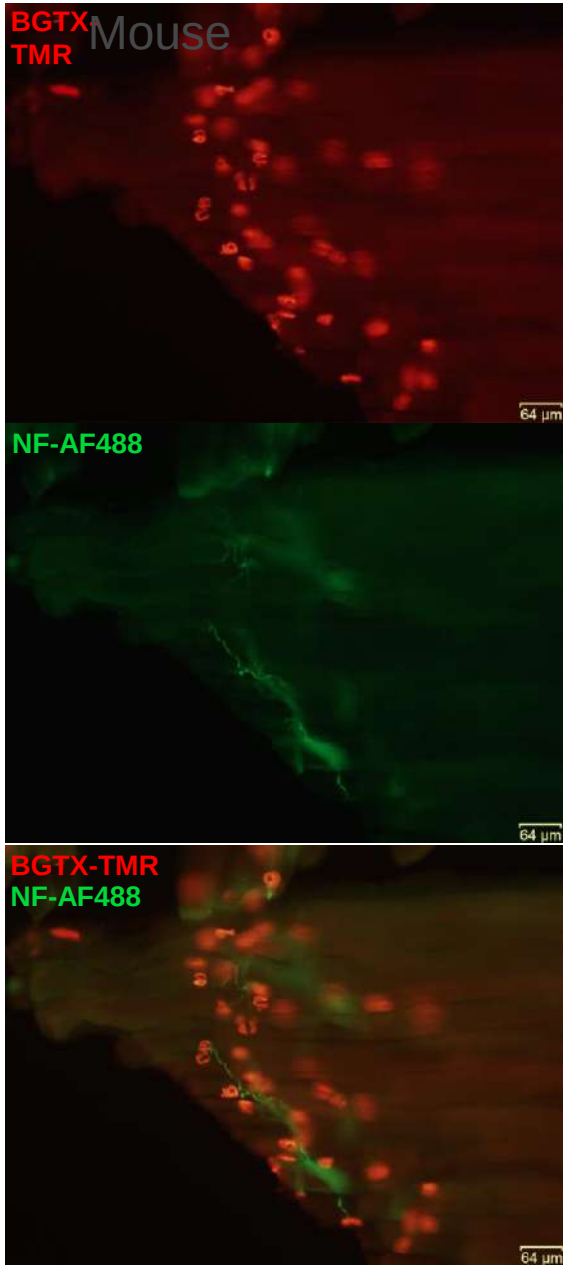
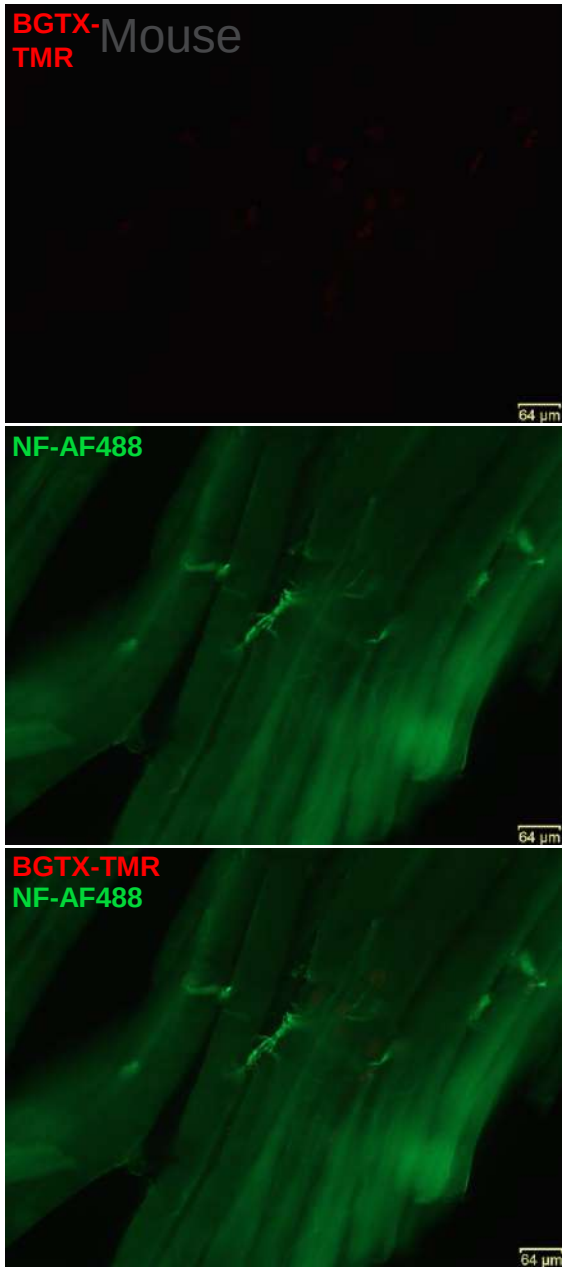
CHRFAM7A Transgenic Mice

WT C57/BL6



CHRFAM7A



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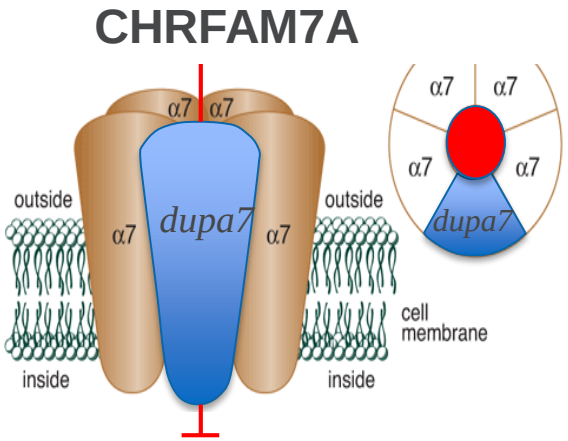
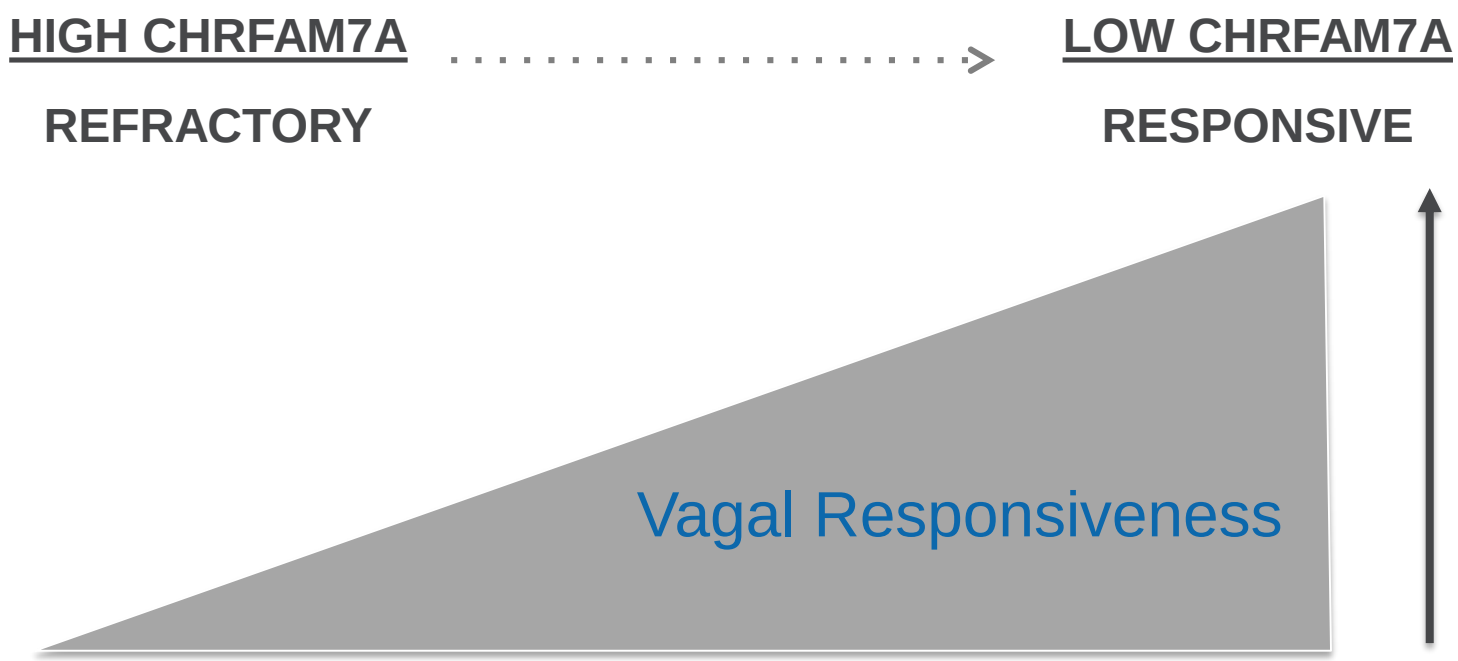
CHRFAM7A: Modulator of Human Vagal Responsiveness

- CHRFAM7A decreases binding to the α7nAchR
- CHRFAM7A expression levels vary widely between individuals
- CHRFAM7A is biologically active in macrophages
- CHRFAM7A alters macrophage gene expression in pathways related to inflammation and the immune response
- CHRFAM7A expression modulates the human anti-inflammatory reflex



CHRFAM7A: Modulator of Human Vagal Responsiveness?



UC San Diego Division of Trauma, Surgical Critical Care, Burns and Acute Care Surgery

Research Team:

- Andrew Baird, PhD
- Brian Eliceiri, PhD
- Raul Coimbra, MD, PhD
- Theresa Chan, MD
- Elliot Williams, MD
- Simone Langness, MD
- Emelie Amburn
- Olga Cohen
- Ann-Marie Hageny

Research Support:

- NIH 1R01GM121530: The Human-Specific Gene CHRFAM7A in Leukocytes

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- American Surgical Association Foundation Research Fellowship
- American College of Surgeons C. James Carrico Faculty Research Fellowship
- American Association for the Surgery of Trauma Faculty Scholarship Award

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