IMPERIAL

Adjuvancy effects of a RIG-I agonist (RIG-101) on H1N1 HA antigen intranasally vaccinated mice and human nasal epithelium/PBMCs co-culture

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Honoraria or consultation fees:	-
Participation in a company sponsored bureau:	-
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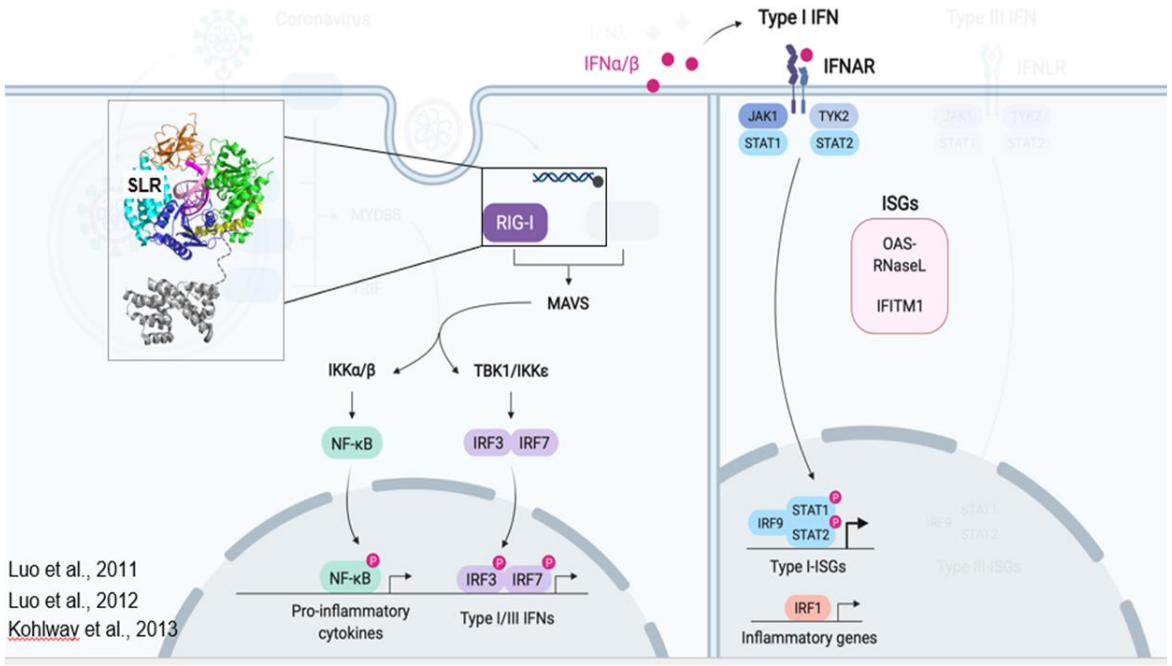
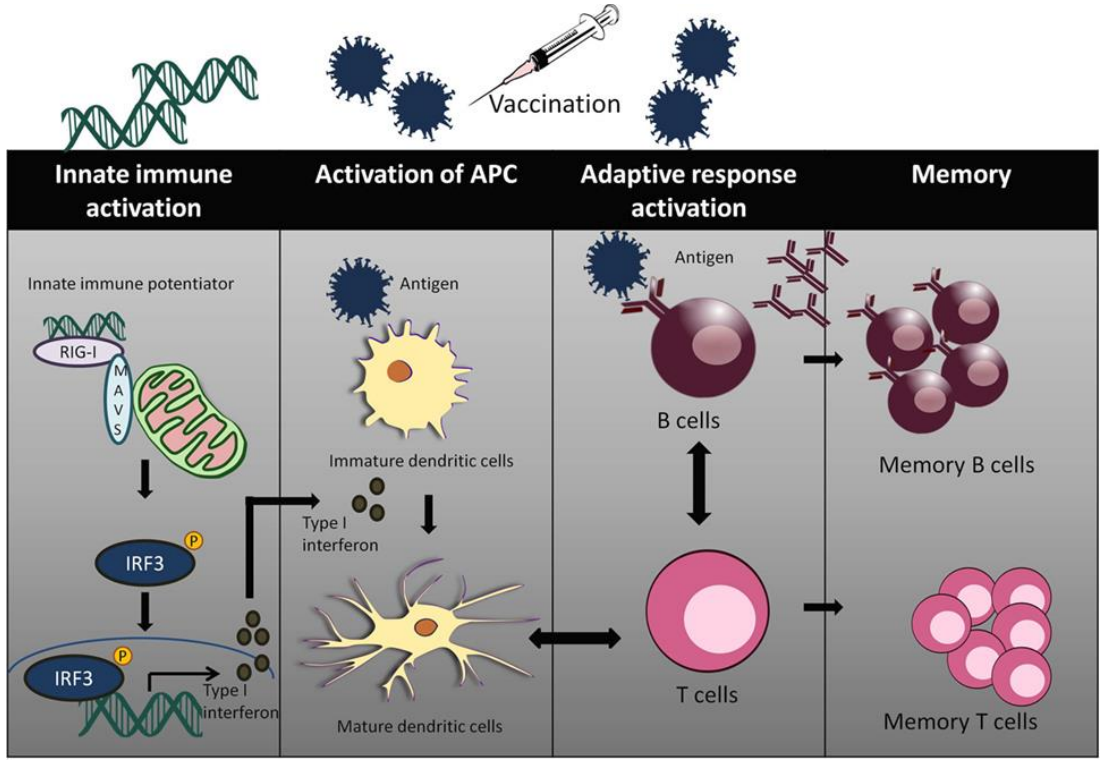
RIG-I – first line of defence against RNA viral pathogens, and also innate immune potentiator as adjuvant

Viral dsRNA directly activate RIG-I to activate antiviral ISGs (interferon stimulating genes)



Innate immune potentiator as adjuvant

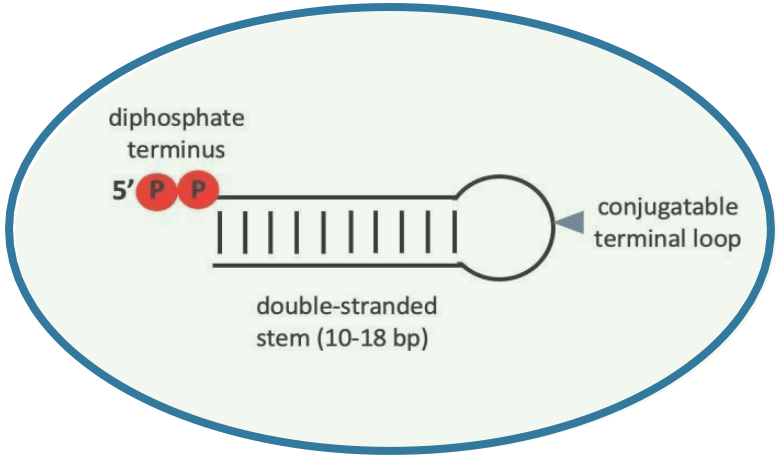


Yong and Luo, 2018, Front Immunol

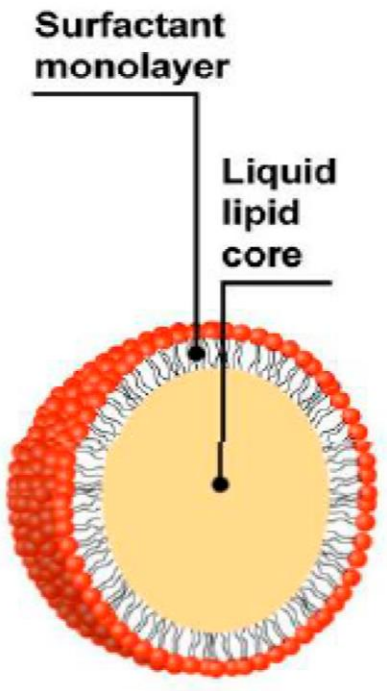
RIG-101 a synthetic RIG-I agonist delivered in novel NEED™ formulation

RIG-101
(Synthetic stem loop RNA)

- RIG-101 is optimized to be a highly selective RIG-I agonist

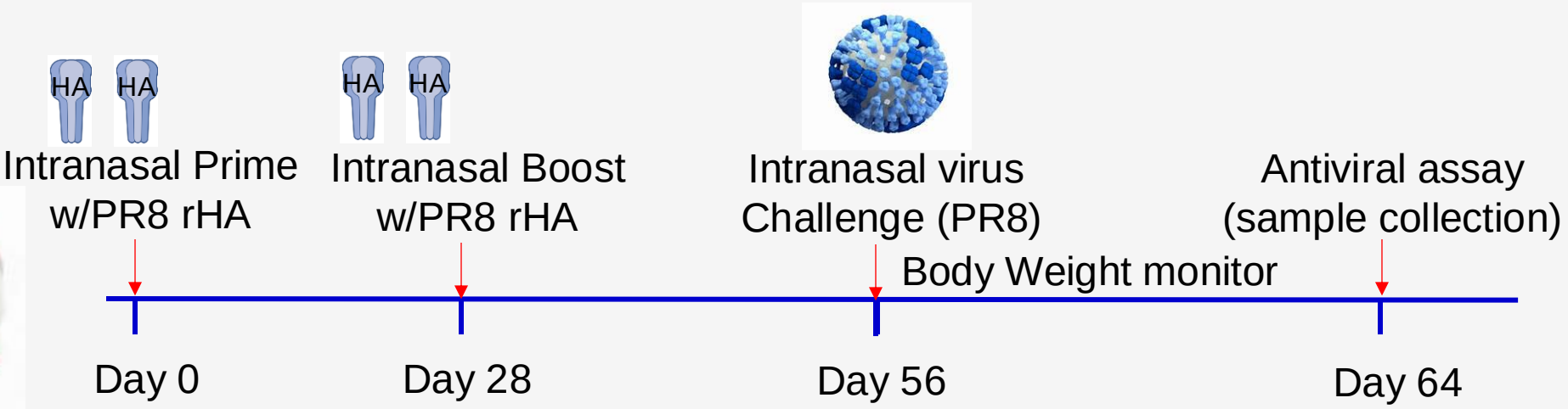


NEED™
(Nano-Emulsion Enhanced Delivery)



- Novel non-LNP delivery system
- Proprietary transformation of surfactants and fatty acids into a nano-emulsion complex (non-LNP) that encapsulates a nucleic acid payload with control of particle size and charge (RIGImmune Inc. patent pending)

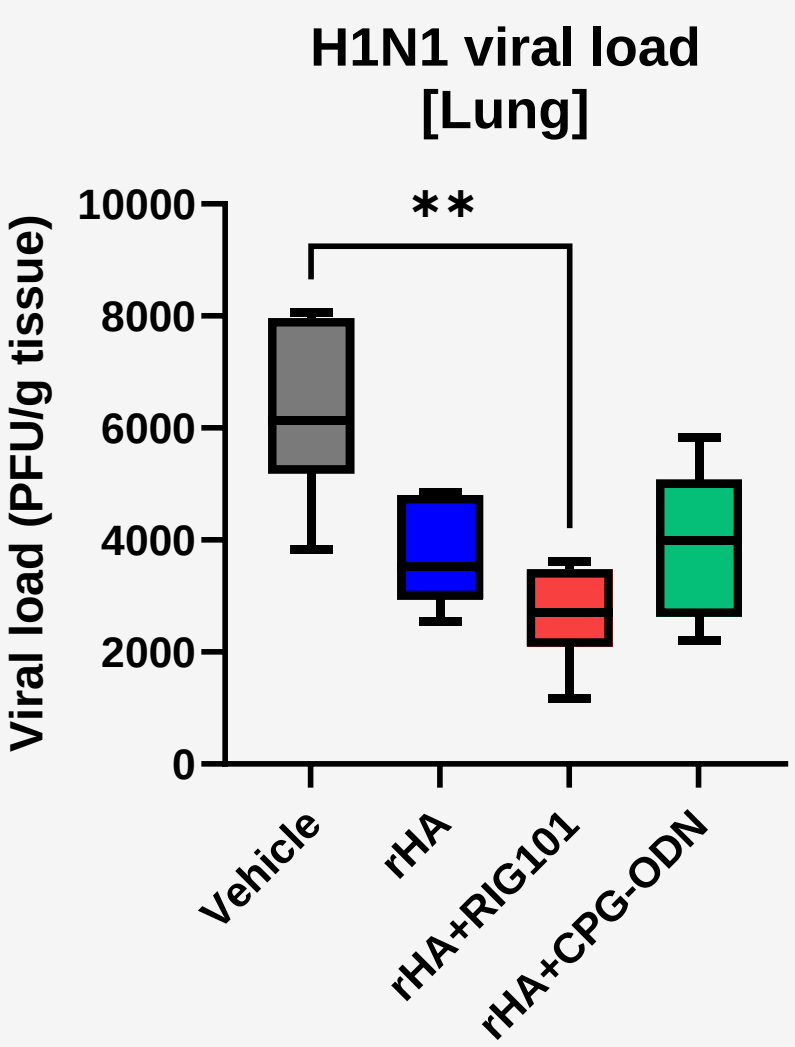
Study protocol: H1N1 HA intranasal (IN) vaccination

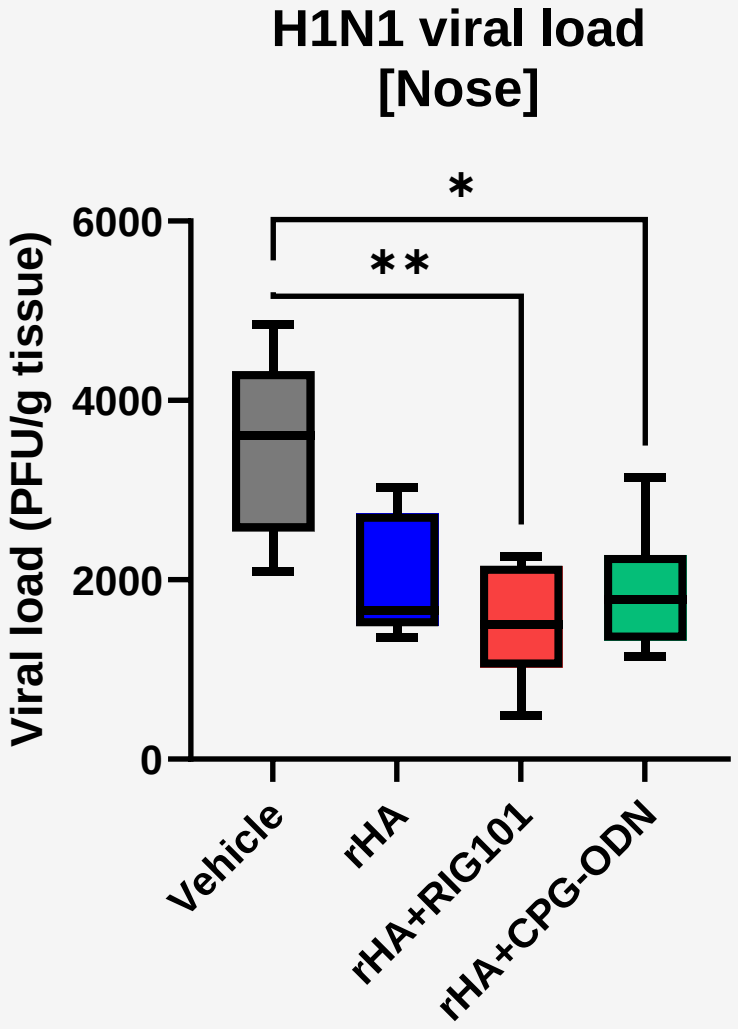


Group	Prime Dosing (Day 0)	Boost Dosing (Day 21)
A	Vehicle IN	Vehicle IN
B	Recombinant Hemagglutinin (rHA) IN only	rHA IN only
C	rHA & RIG-101 NEED™ (both IN)	rHA & RIG-101 (both IN)
D	rHA & CPG-ODN (both IN)	rHA & CPG-ODN (both IN)

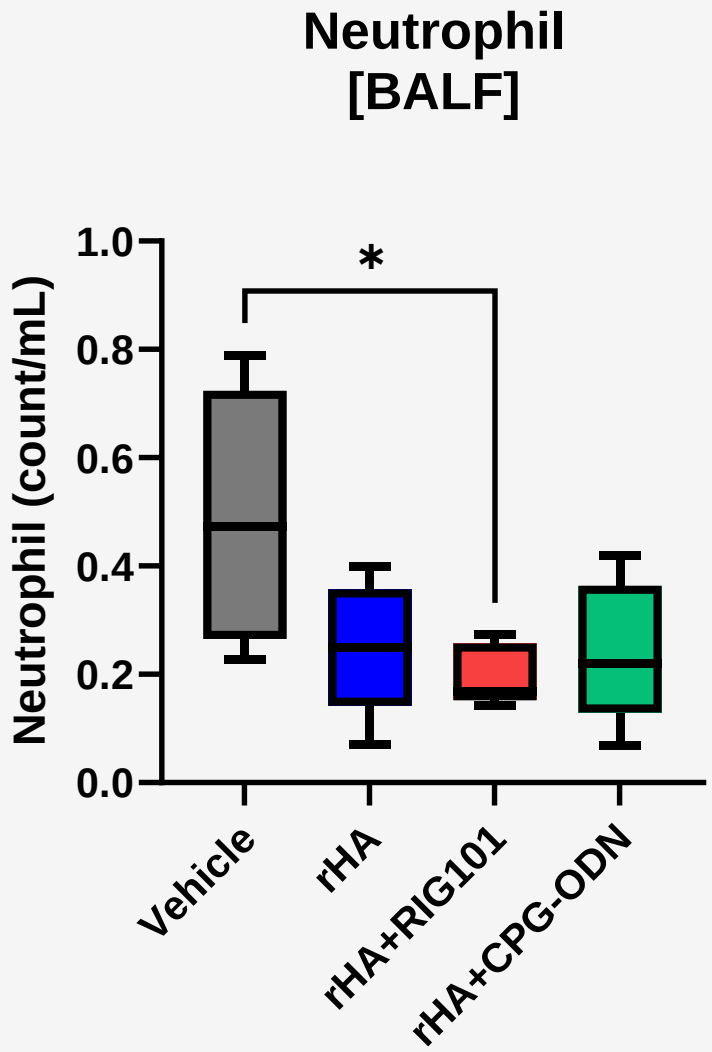
Saline/RIG-101/CPG-ODN 10µL intranasally + recombinant rHA 10µL intranasally

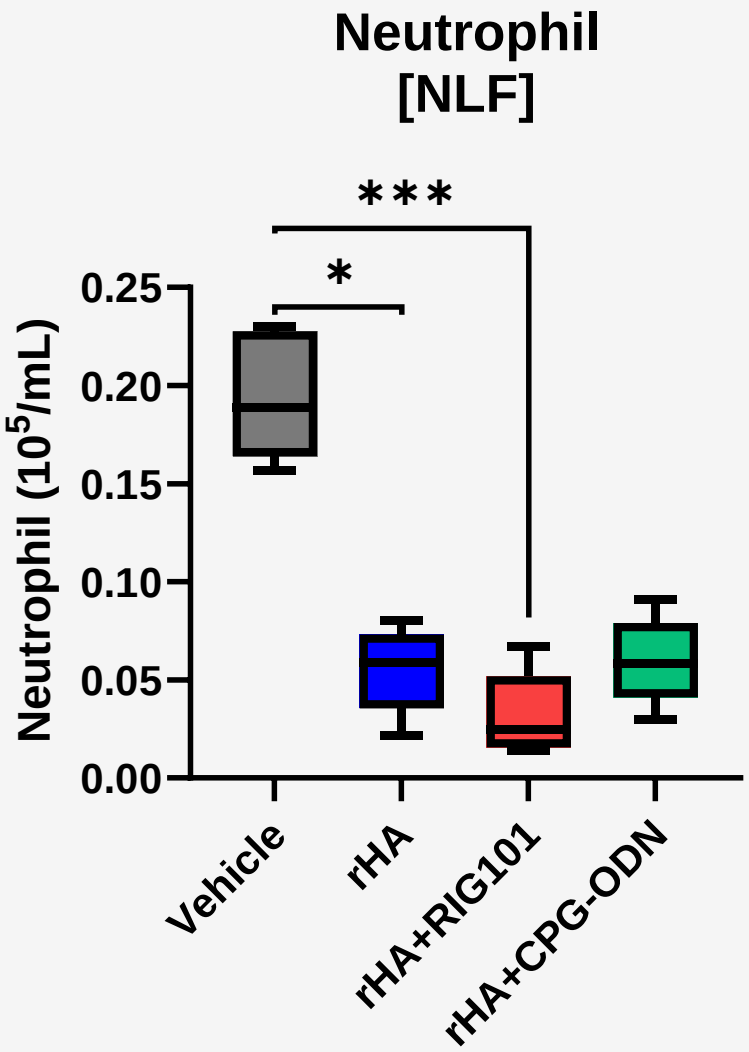
Intranasal treatment with RIG-101 NEED™, as an adjuvant, enhances H1N1 HA vaccination's *effects in vivo* in mice [Viral load]



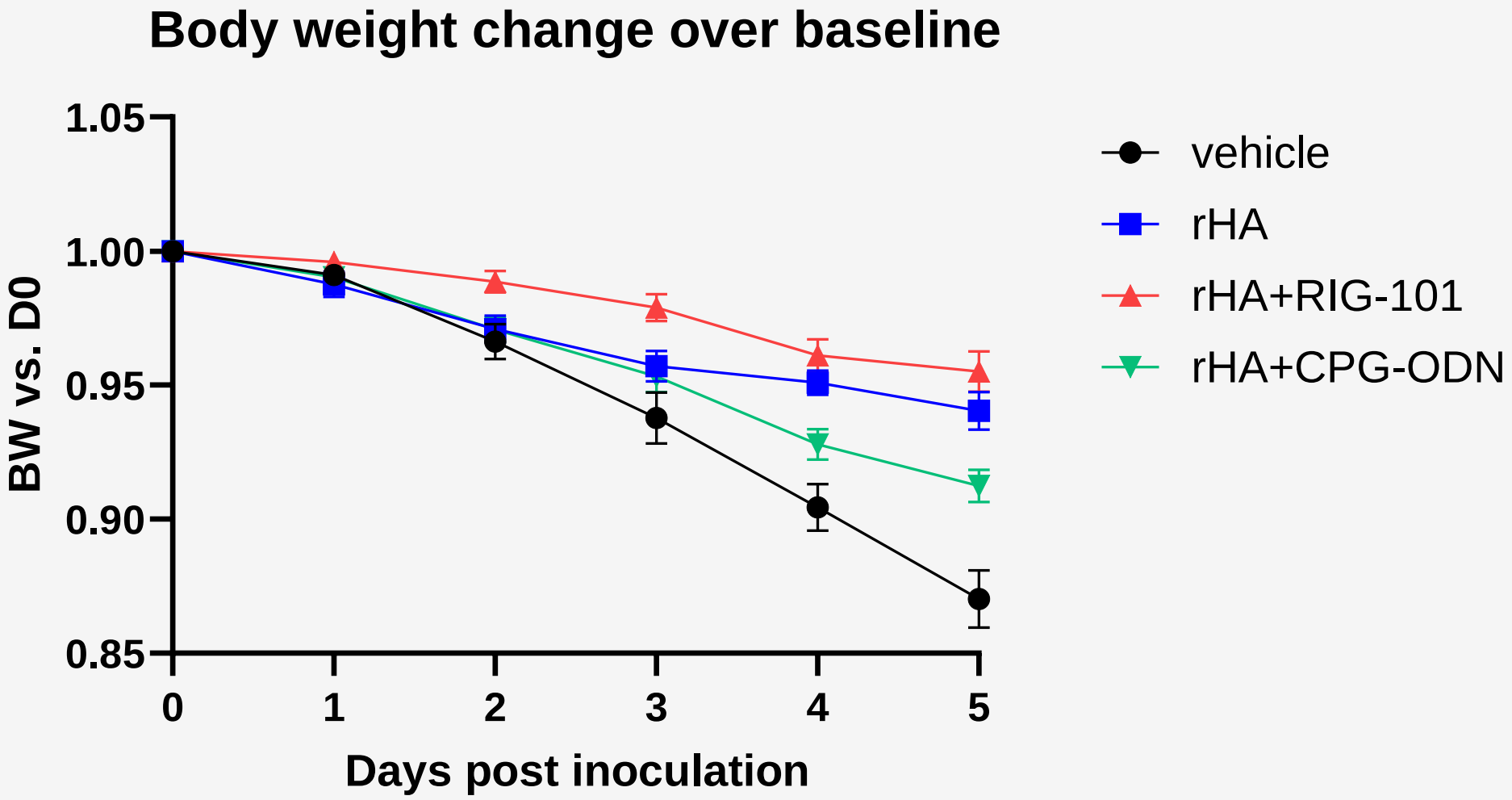


Intranasal treatment with RIG-101 NEED^{TM}, as an adjuvant, enhances H1N1 HA vaccination's *effects in vivo* in mice [inflammation]





Intranasal treatment with RIG-101 NEED™, as an adjuvant, enhances H1N1 HA vaccination's *effects in vivo* in mice [Body weight loss]



RIG101 enhances IgM and anti-virus antibody release from PBMCs primed sensitised with H1N1 rHA in co-culture of PBMCs and nasal epithelium *in vitro*

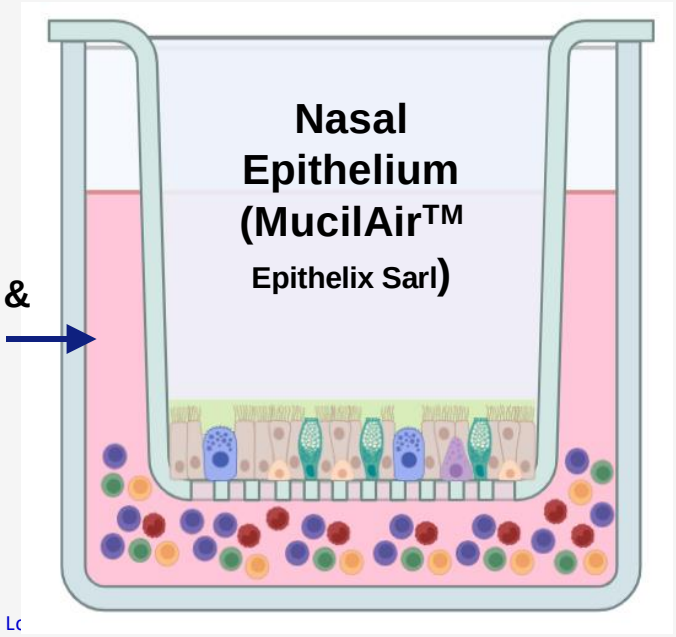
1. PBMC stimulation L-leucyl-L-leucine methyl ester (LLME)

Healthy Human PBMCs

2. PBMC Sensitisation

H1N1 rHA (10µg/ml) +CTB + CPG or +RIG-101

3. Co-culture and Collection

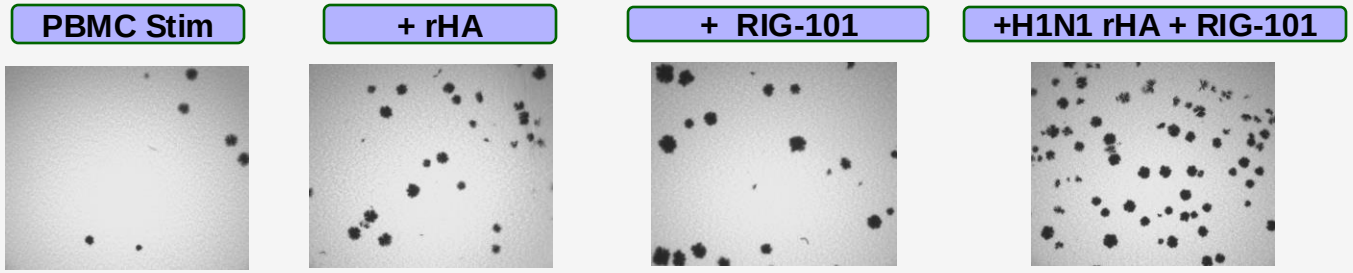


Stimulated & Sensitised PBMCs

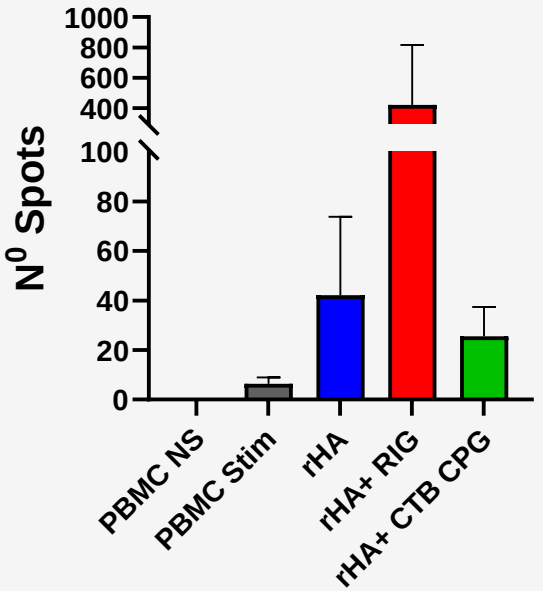
Day 4 :
Apical wash

Day 10 Collection:
Apical wash
Basal PBMCs

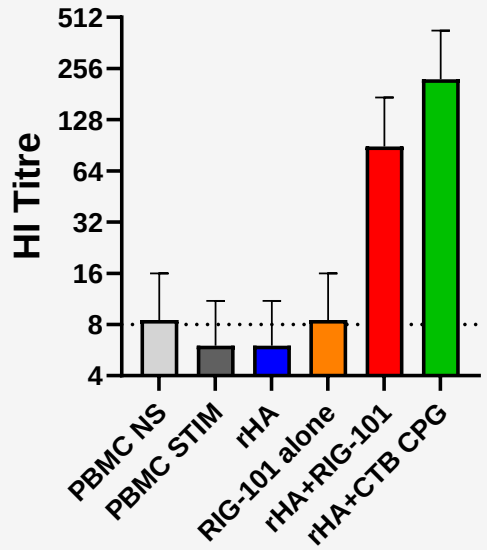
IgM release (ELISPOT)



IgM (ELISPOT)



Heamglutinin inhibition assay [virus neutrising Titre]



Summary

- **Addition of RIG-101 to H1N1 HA antigen was more effective on reduction of viral load and inflammation, and prevention of virus-induced body weight loss compared with HA antigen vaccination alone and CPG-ODN addition in mice**
- **In PBMC-nasal epithelium co-culture, RIG-101 clearly enhanced IgM production in LLME-treated PBMCs compared with H1N1 HA alone**

Conclusion

- **RIG-101, a RIG-I agonist, is promising adjuvant to boost mucosal vaccine effects against respiratory virus**

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Thank You

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