Electronic cigarette induces pulmonary inflammation and COVID-ACE2 receptor regulation mediated by nAChR α7

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INTRODUCTION

- E-cigarette (e-cig) use is increasing rapidly over the years, but their acute effects on lung toxicity are largely unknown following inhalation of e-cig aerosols containing humectant propylene glycol (PG) alone and PG with nicotine.
- Prior reports have shown that acute e-cig exposure causes oxidative stress, lung inflammation, and impaired immune defense in the lung (against bacterial and viral infections).
- Here we are interested in understanding the sub-chronic effects of e-cig exposure induced inflammation and the correlation of e-cig vaping and COVID-19.

HYPOTHESIS

E-cig aerosol-induced pulmonary inflammation and ACE2 receptor dysregulation are both mediated by nAChRα7.

APPROACH

- **Sub-Chronic e-cig exposure:**

Wild-type (WT) and nAChRα7 knockout (KO) mice were exposed 2 hrs/day for 30 days to e-cig aerosol containing PG with or without nicotine (25mg/mL), using SCIREQ InExpose system. Mice were sacrificed post 24hr of last exposure, Broncho-alveolar lavage fluid (BALF) and lung tissues were collected for future analysis. E-cig (PG+nicotine) exposed mice showed higher serum cotinine levels (~400 ng/mL) compared to PG alone and air-exposed control (no detectable cotinine levels).

- **Differential cell and pro-inflammatory cytokine analysis:**

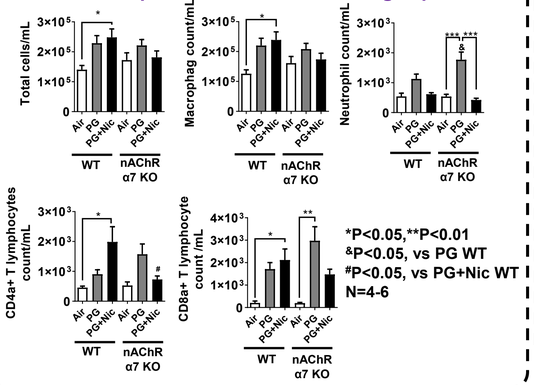
Inflammatory cell count and cytokine levels in BAL fluid were determined using flow cytometry and Luminex respectively.

- **Quantification of Inflammation/Covid-19 markers**:

Lungs were homogenized in RIPA buffer before protein quantification. The protein concentrations were measured by BCA kit, and equal amount of protein was used for Western blot. Both inflammation targets (NF-κB subunits: p50/p105) and Covid-19 markers (ACE2, Furin and TMPRSS2) were probed, and protein expression change folds were normalized by β-Actin.

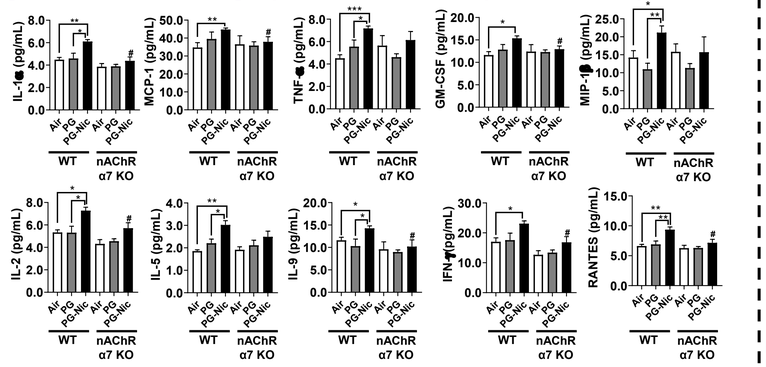
RESULTS

Figure 1. Up-regulated inflammatory cellular influx in BALF in response to sub-chronic e-cig exposure



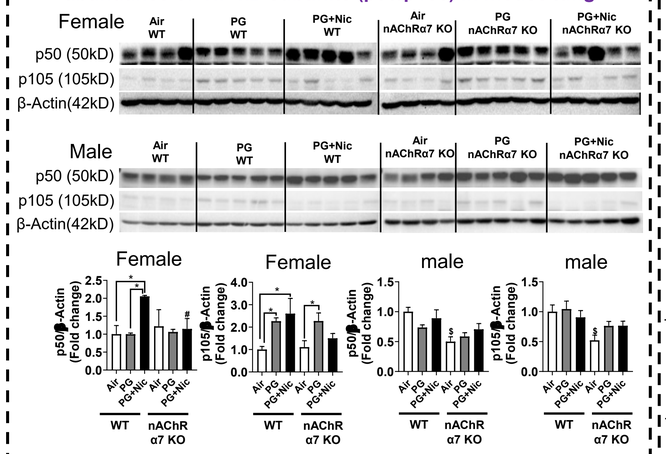
*P<0.05,**P<0.01
&P<0.05, vs PG WT
#P<0.05, vs PG+Nic WT
N=4-6

Figure 2. Sub-chronic e-cig exposure induced BALF pro-inflammatory mediators



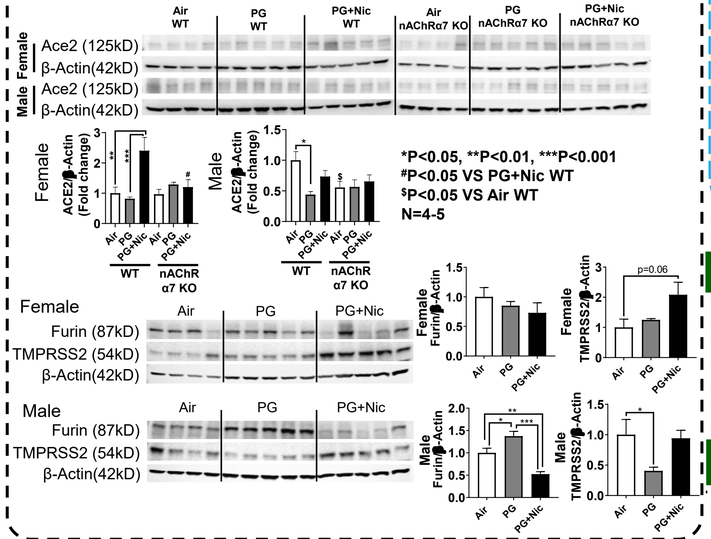
*P<0.05, **P<0.01, ***P<0.001; # P < 0.05 vs PG+Nic WT group; N=6-10

Figure 3. Sub-chronic e-cig exposure dysregulated protein abundance of NF-κB subunits (p50/p105) in mouse lung



*P<0.05,**P<0.01; $P<0.05, vs Air WT; #P<0.05, vs PG+Nic WT; N=4-5

Figure 4. Sub-chronic e-cig exposure altered ACE2, Furin and TMPRSS2 expression in mouse lungs with sex-based differences

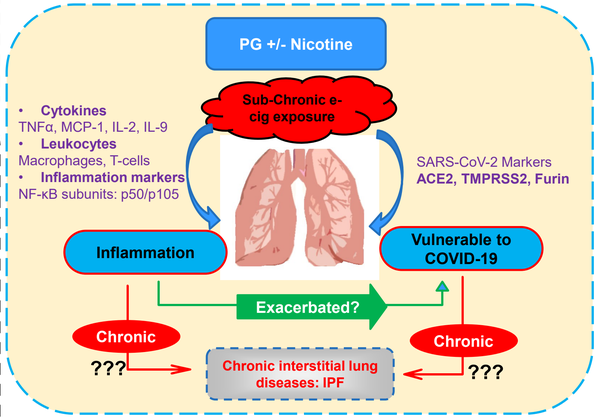


*P<0.05, **P<0.01, ***P<0.001
#P<0.05 VS PG+Nic WT
$P<0.05 VS Air WT
N=4-5

SUMMARY

- **Sub-chronic e-cig exposure induced pulmonary inflammation and up-regulated ACE2 expression.**
- **Both inflammation and ACE2 expression were mediated by nAChRα7.**
- **E-cig exposure increased the risk of SARS-CoV-2 infection in both male and female mice via different dysregulated proteases/ receptors.**

CONCLUSION



CONCLUSION

E-cig vaping has become a health concern and our research sheds new insights into how e-cig induced inflammation and COVID-19 receptor are mediated by nAChR alpha7 expression.

ACKNOWLEDGEMENT

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