

Innate-Type Allergy and Air Pollution

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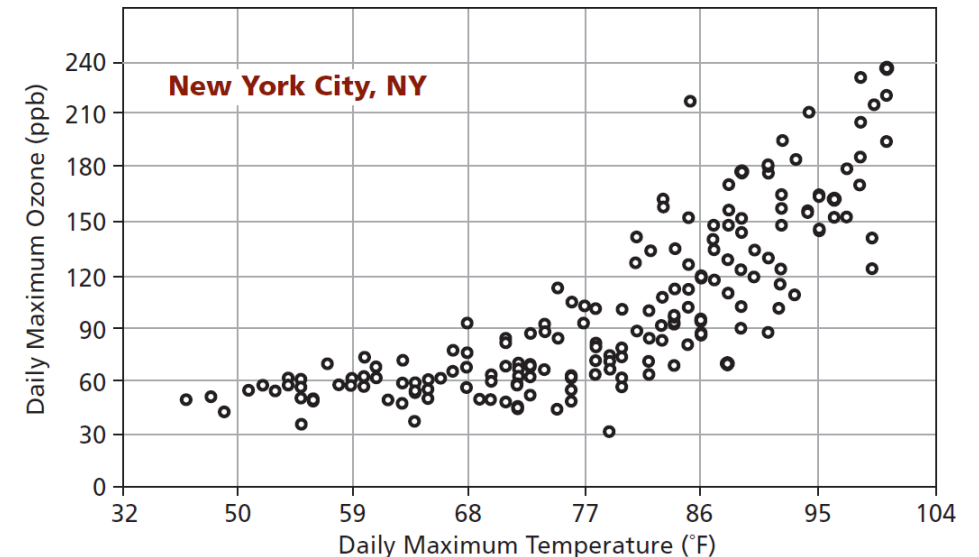
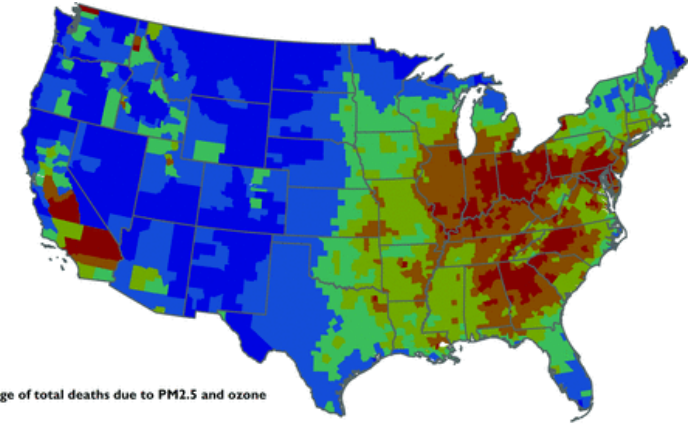


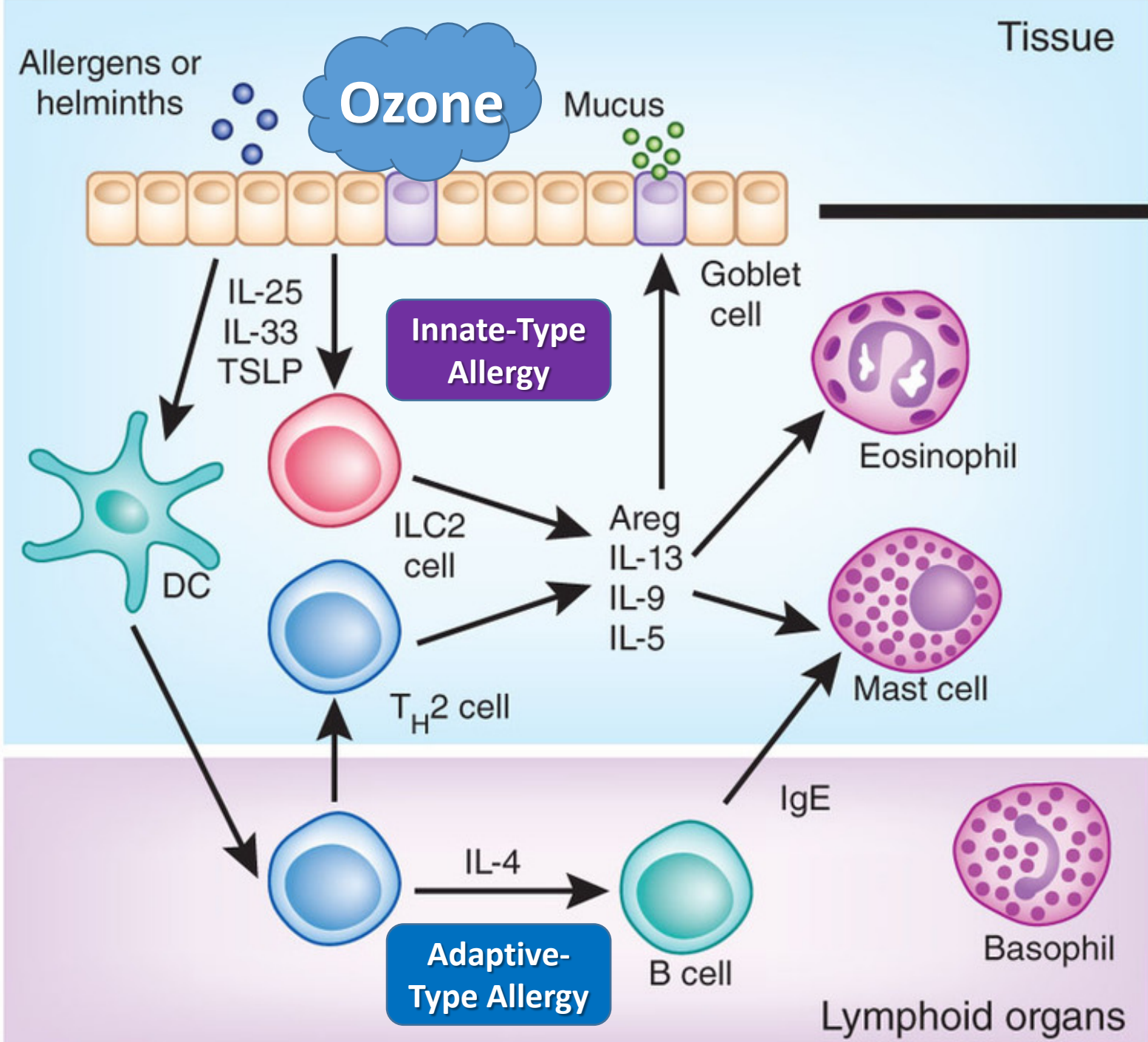
Outline for Talk

- Air pollution and allergic airway diseases (exacerbation and early onset)
- Innate- and adaptive-type allergy
- Ozone-induced type 2 airway immunity in mice
- Summary, future directions and questions

Air Pollution and Allergic Airway Disease

- Air pollution causes 3.3 million deaths/year worldwide
- 2.5 million disability-adjusted life years were attributable to ambient ozone (O₃) exposure alone in 2010
- Air pollutant exposure exacerbates pre-existing allergic rhinitis and asthma
- Ozone and airway allergy are predicted to increase with climate change
- **Do air pollutants contribute to the onset of airway allergy?**





Allergic inflammation

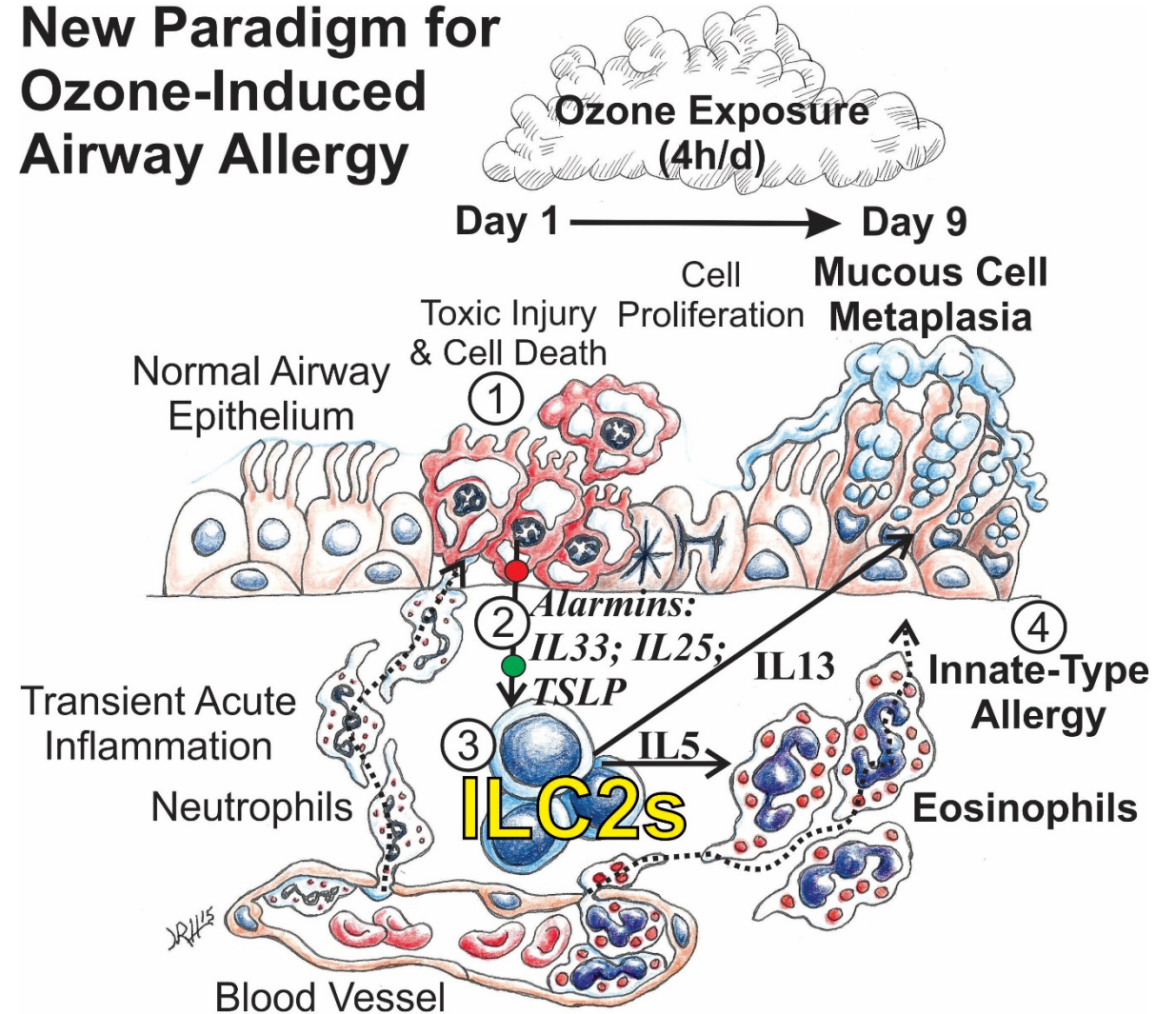
- Influx of eosinophils
- Mucous cell metaplasia
- Epithelial hyperplasia
- Ym1/2 proteins
- Airway Hyper-responsiveness

Initiation and propagation of type 2 immune responses

Our Overarching Hypothesis

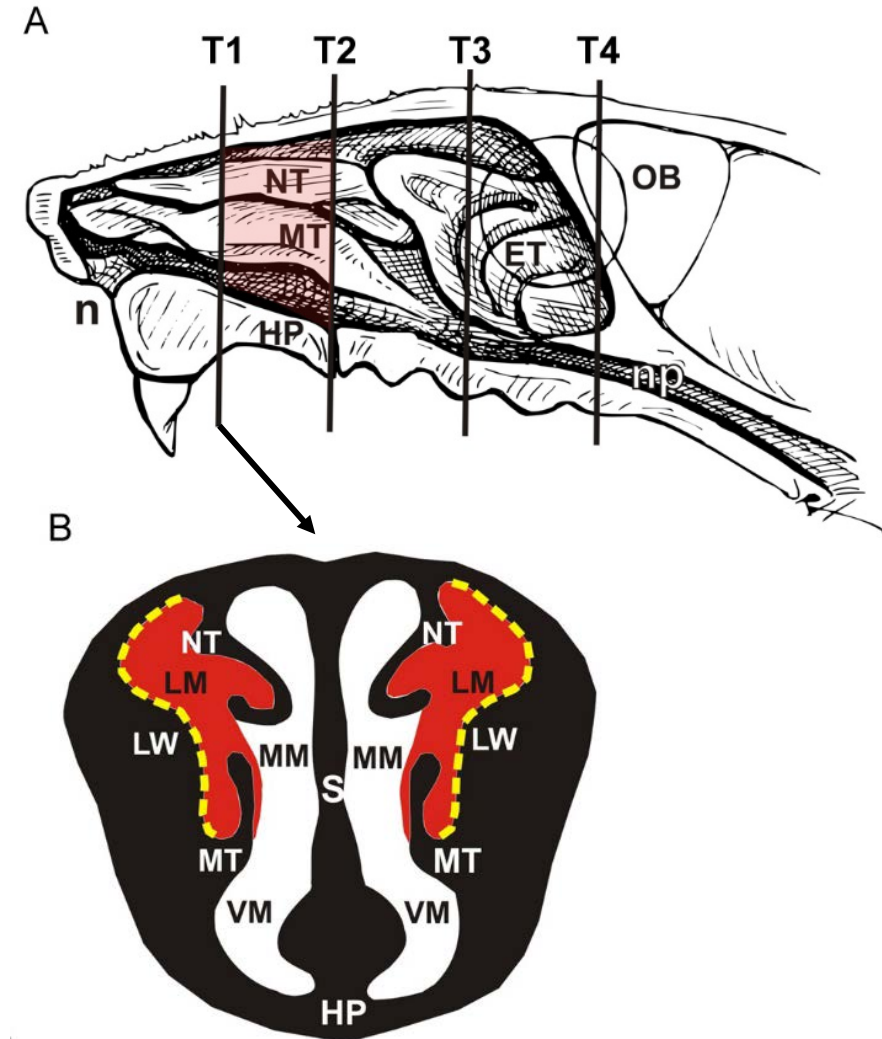
Repeated exposures
to ozone elicit
innate-type allergy
in the nose and lung.

New Paradigm for Ozone-Induced Airway Allergy

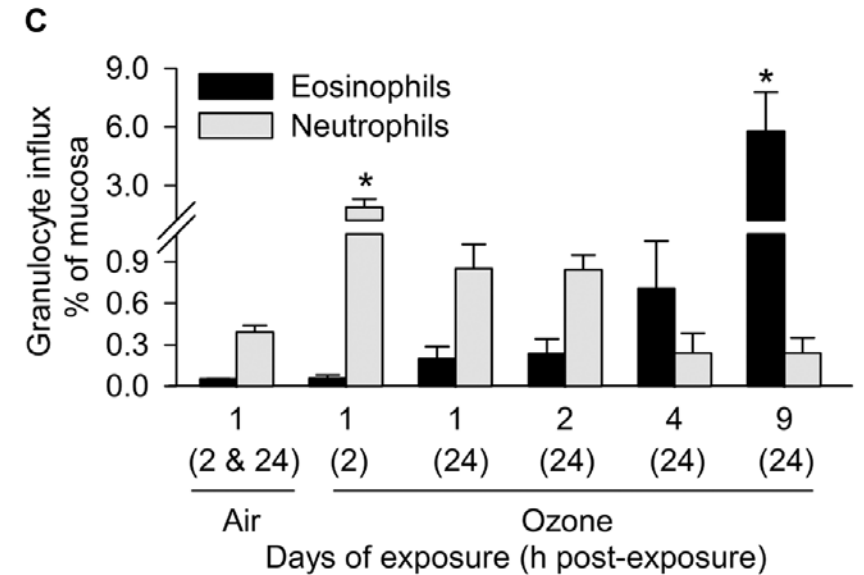
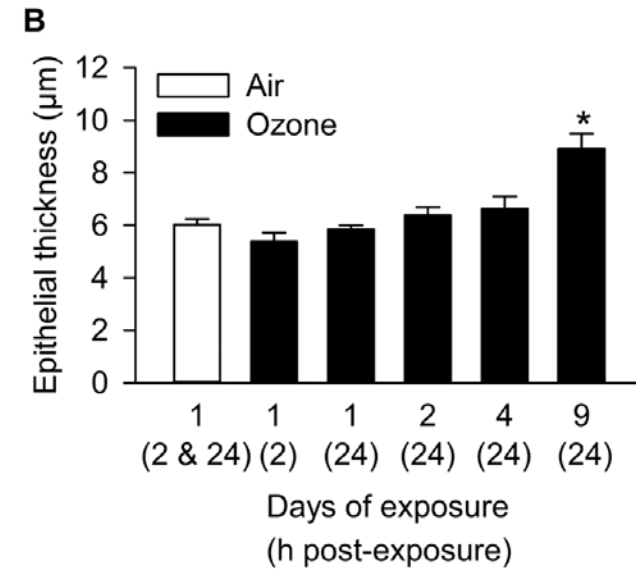
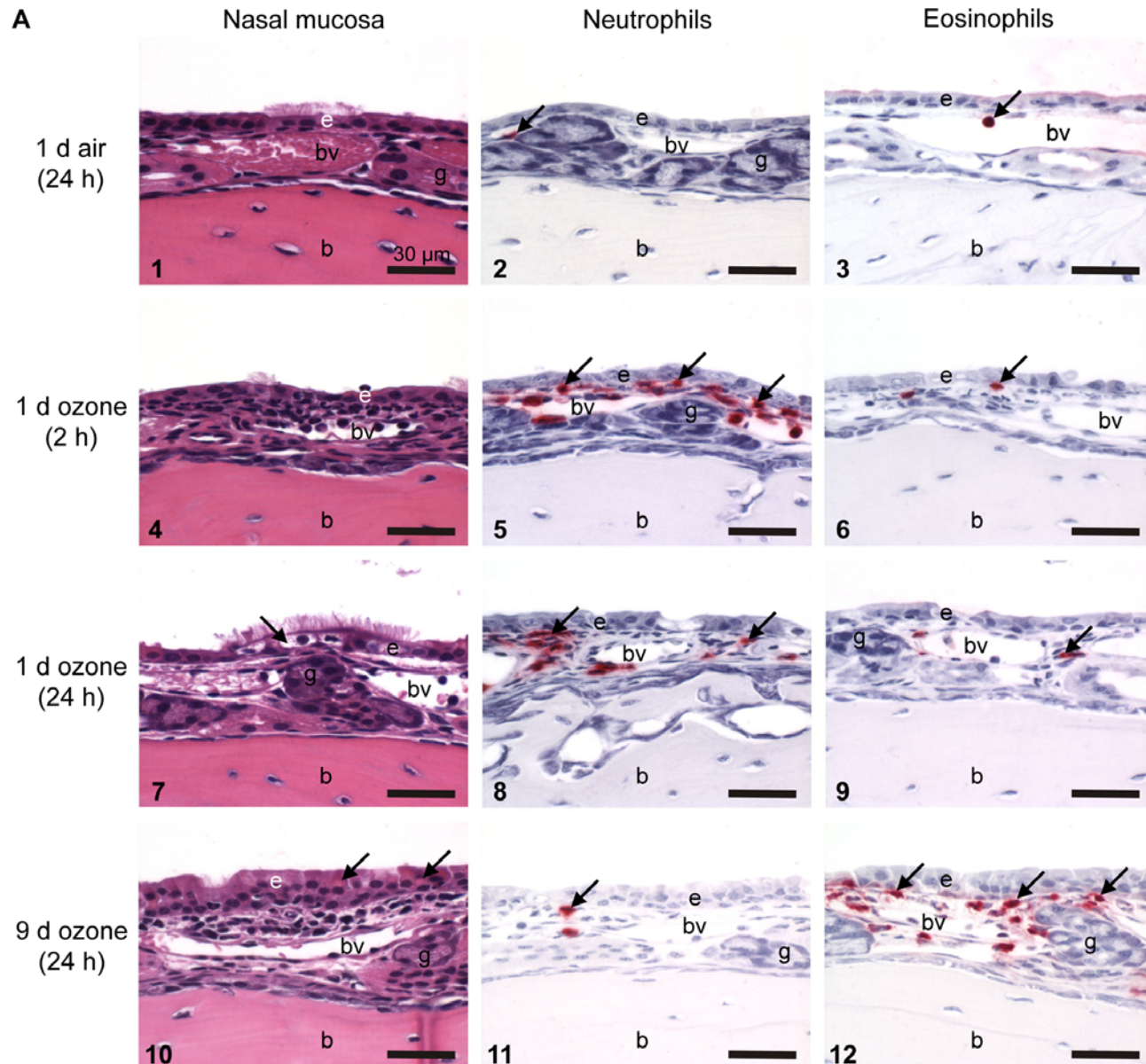


Aim 1: To determine the onset of ozone-induced eosinophilic rhinitis and nasal type 2 immunity

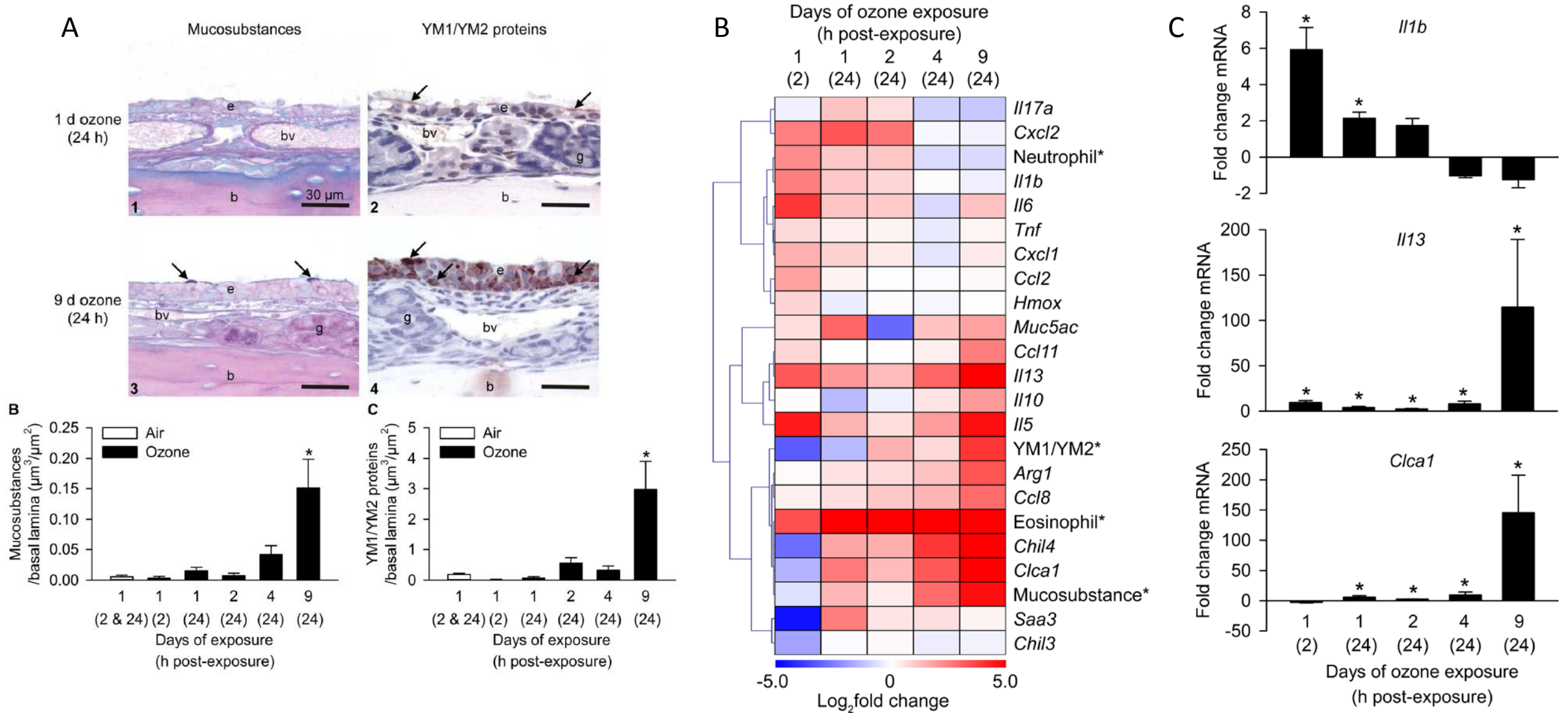
- C57BL/6 male mice
- 0 or 0.5 ppm ozone (4h/day) for 1, 2, 4 or 9 weekdays
- Nasal histopathology
- Immunohistochemistry and morphometric analysis
- qRT-PCR for relative mRNA expression of selected inflammatory cytokines and airway epithelial proteins



Nasal epithelial thickness and granulocytes with increasing days of exposure

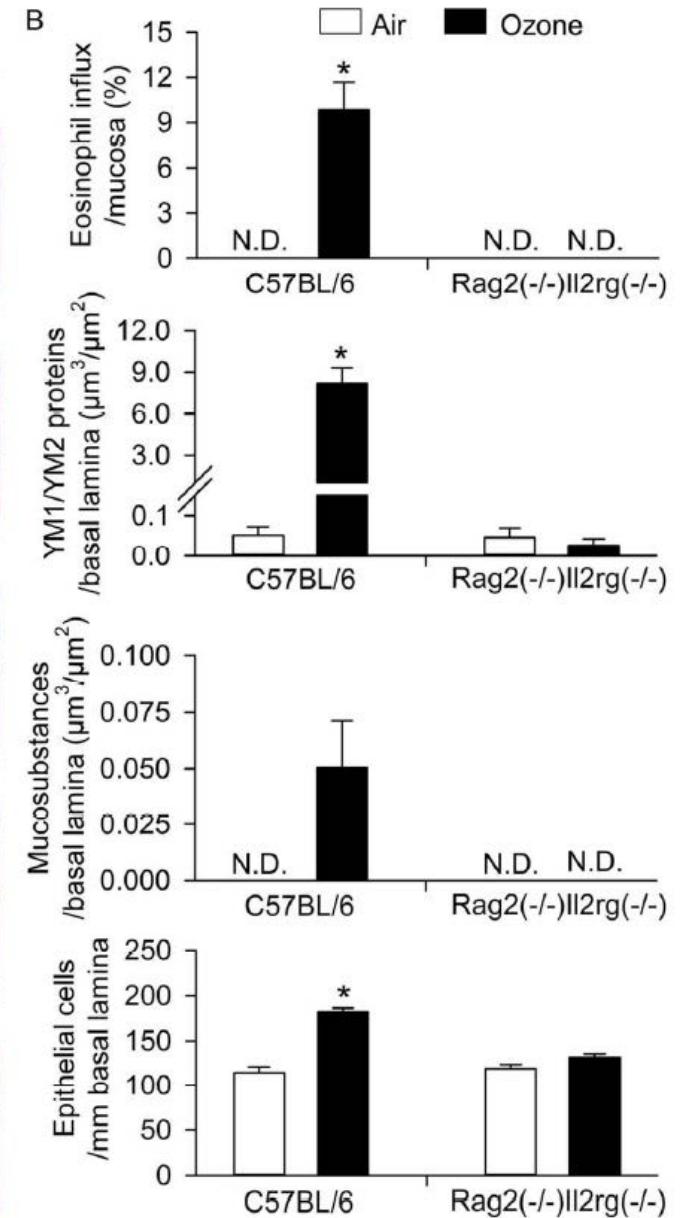
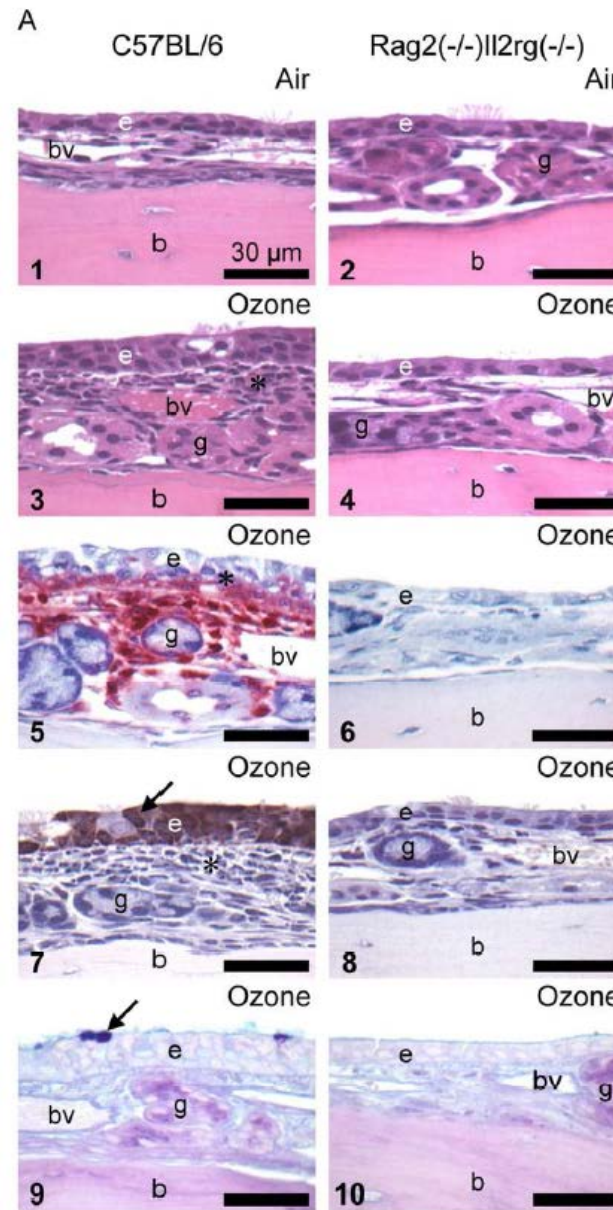
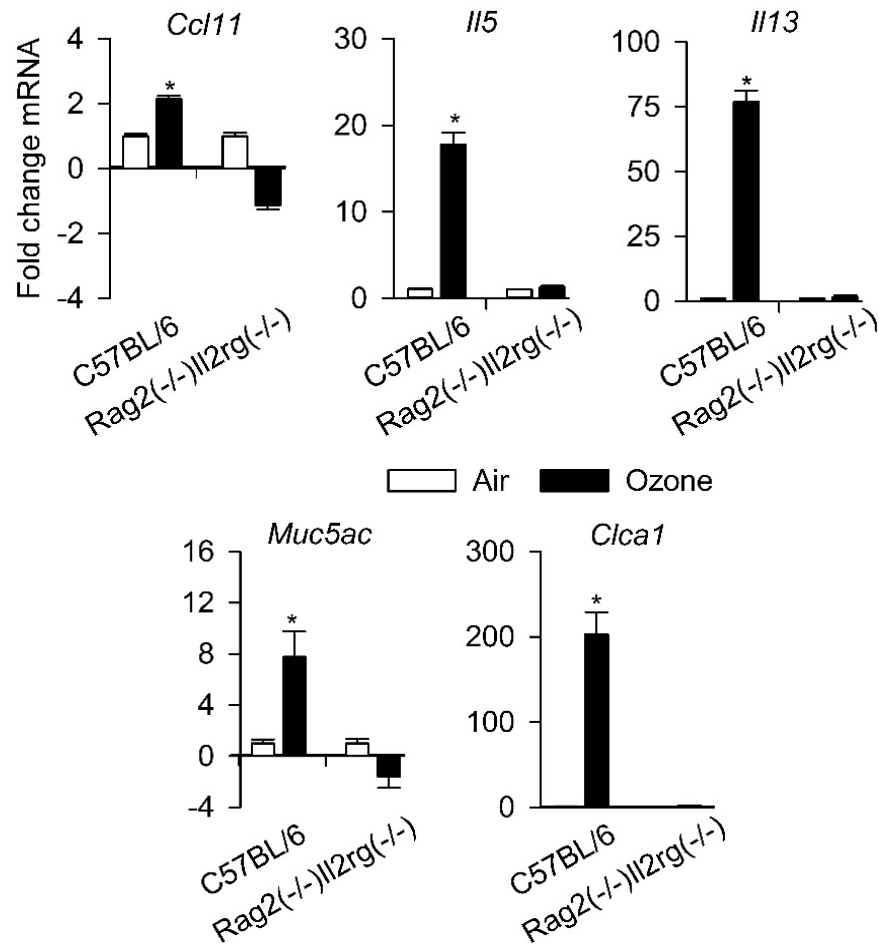


Nasal epithelial protein and mRNA expressions with increasing days of exposure



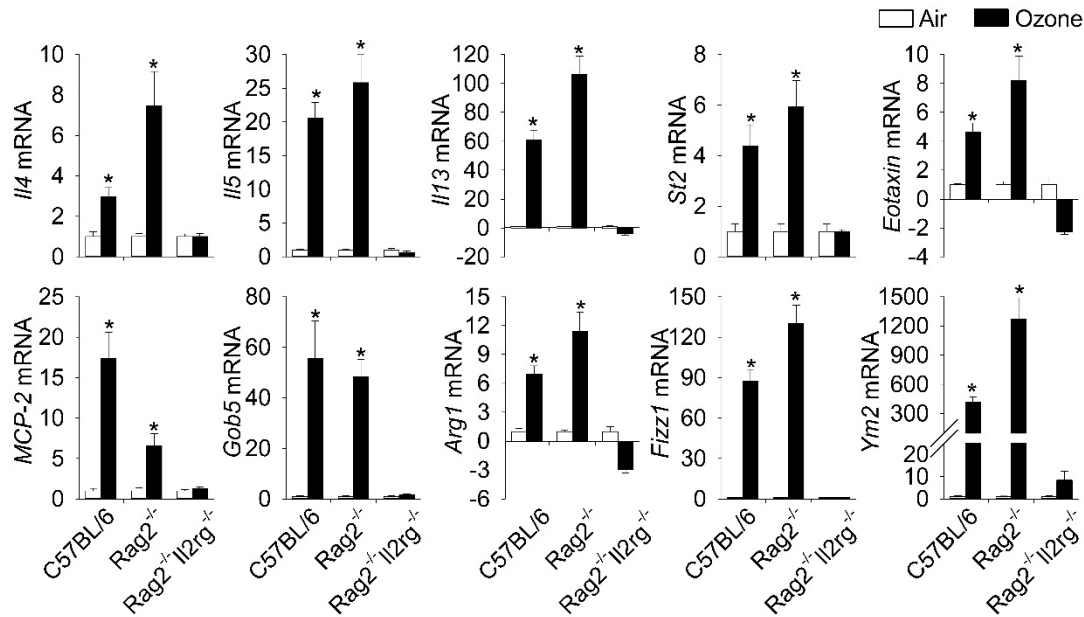
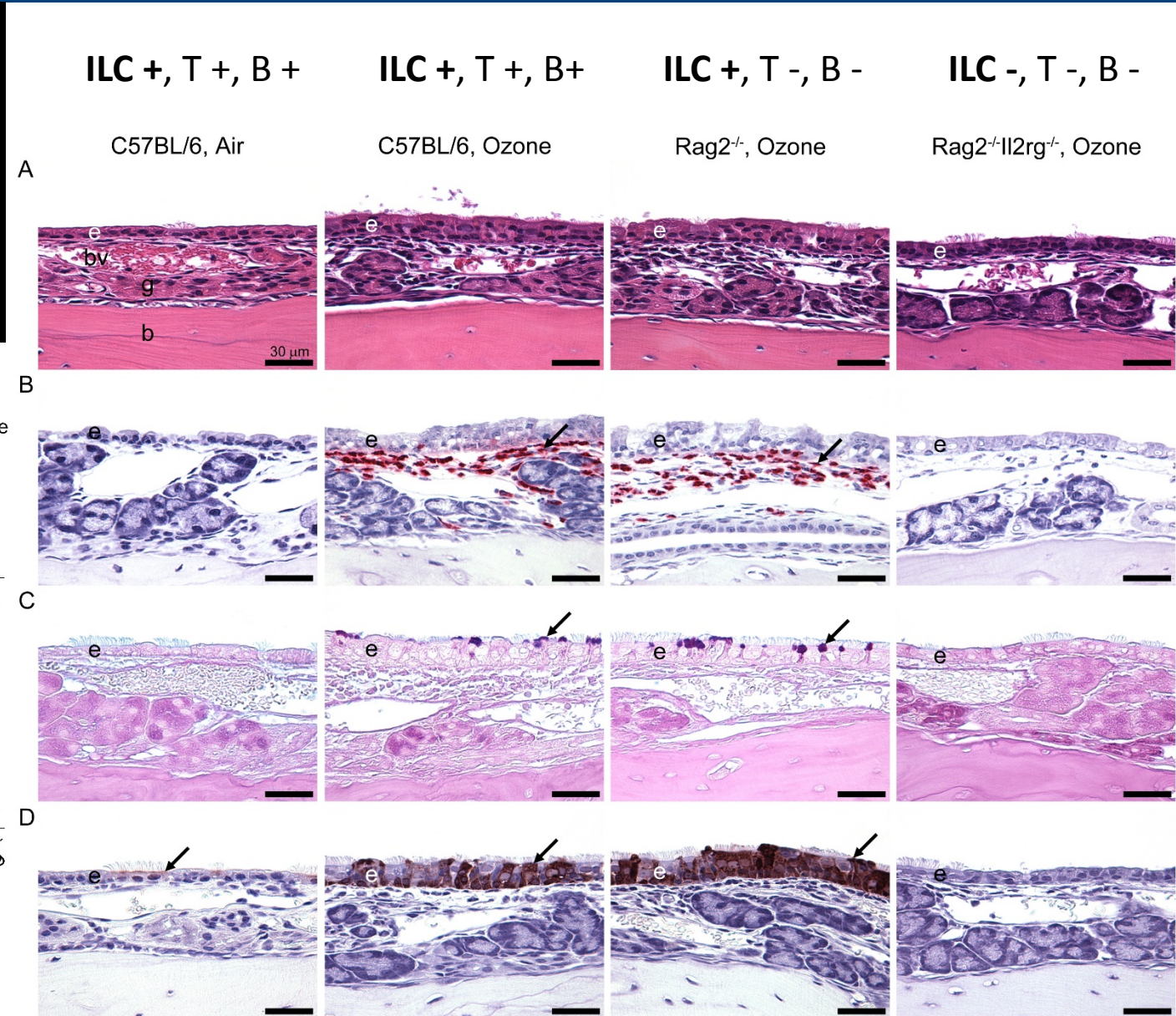
Aim 2: To determine the role of lymphoid cells in ozone-induced eosinophilic rhinitis and nasal type 2 immunity

- Lymphoid cell-deficient Rag2(-/-)IL2rg (-/-) and Lymphoid cell-sufficient C57BL/6 mice
- 0 or 0.5 ppm ozone (4h/day) for 9 days

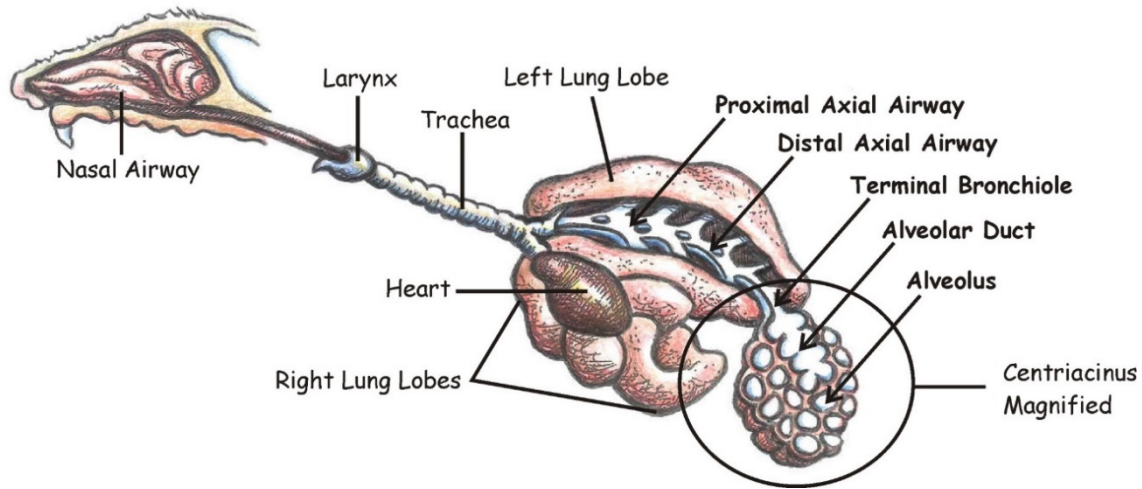


Aim 3: Determine the role of ILCs in ozone-induced eosinophilic rhinitis and nasal type 2 immunity

- Lymphoid cell-deficient Rag2(-/-) IL2rg(-/-), lymphoid cell-sufficient C57BL/6 mice, ILC-sufficient and T & B cell-deficient Rag2(-/-) mice
- 0 or 0.8 ppm ozone (4h/day) for 9 days



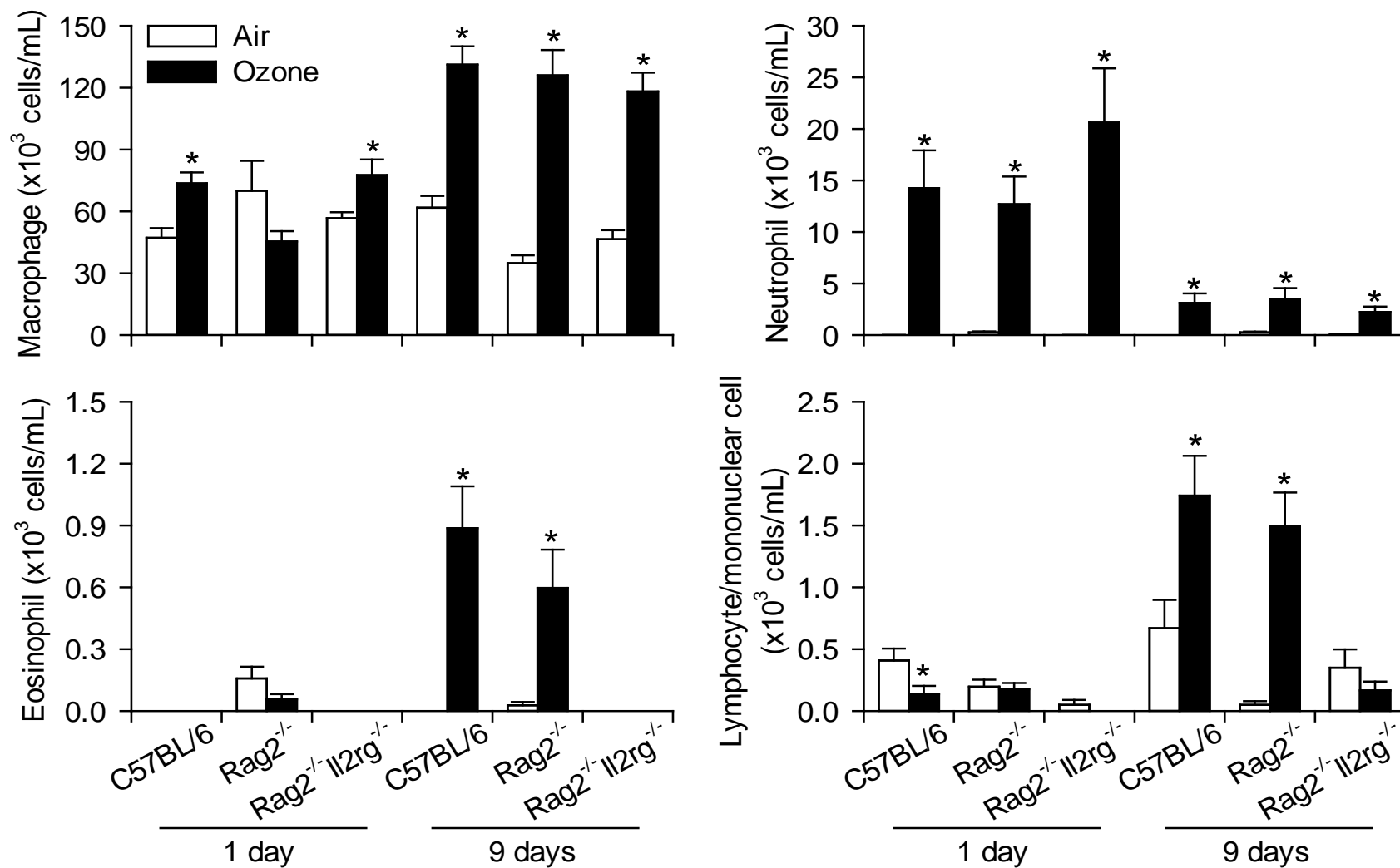
Aim 4: Determine if repeated exposure to ozone induces innate-type allergic airway responses in the lung



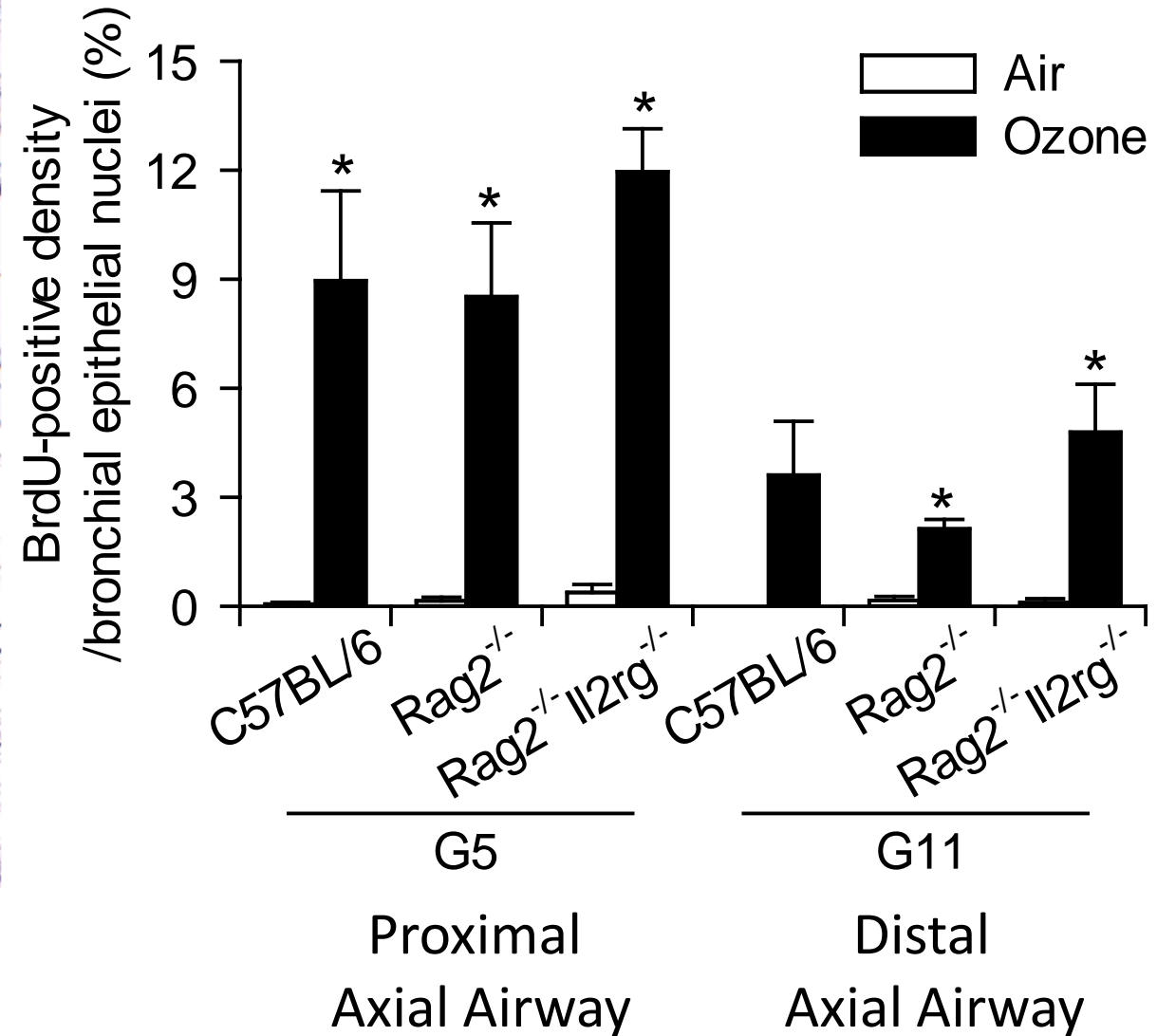
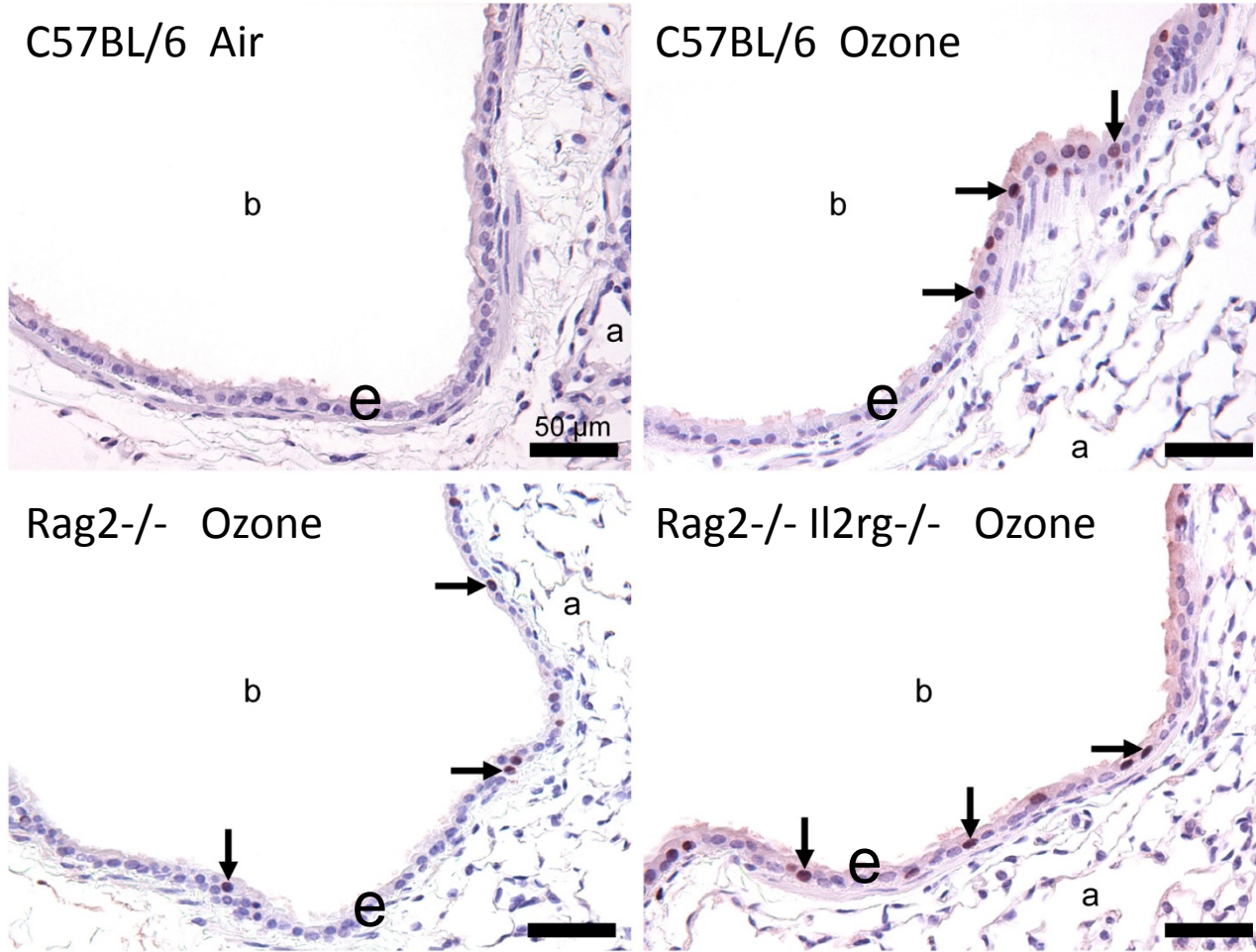
- Lymphoid cell-deficient Rag2(-/-)IL2rg (-/-), lymphoid cell-sufficient C57BL/6 mice, ILC-sufficient and T & B cell-deficient Rag2(-/-) mice
- 0 or 0.8 ppm ozone (4h/day) for 1 or 9 days
- Bronchoalveolar lavage fluid analysis for cells and cytokines
- Pulmonary histopathology
- Morphometric analysis
- qRT-PCR for mRNA expression of inflammatory cytokines and epithelial proteins

Ozone-induced eosinophils only in ILC-sufficient mice exposed for 9 days

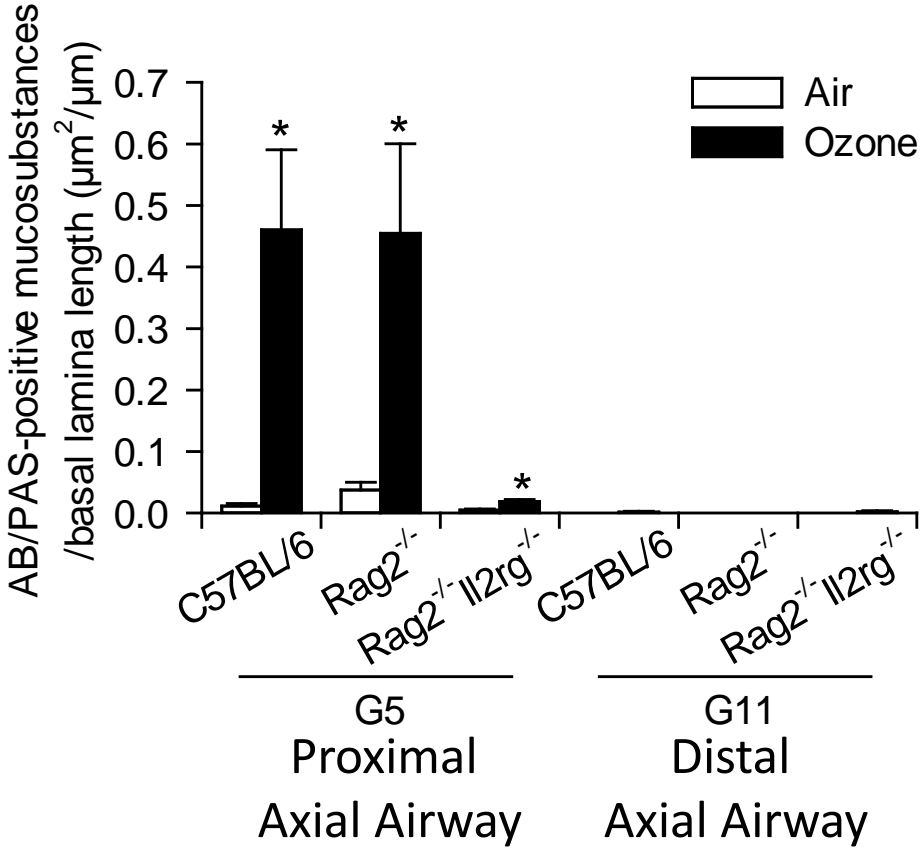
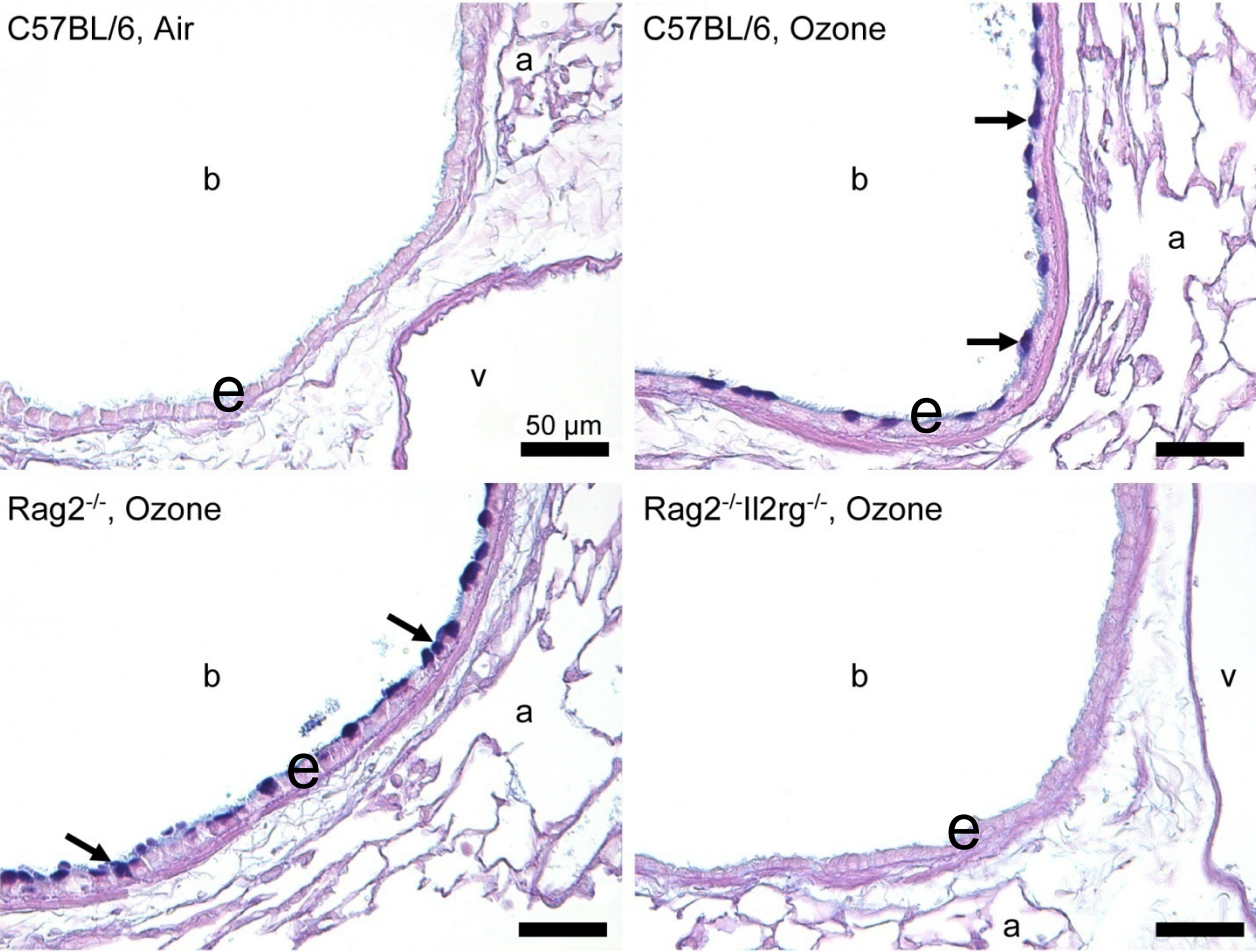
Inflammatory Cells in Bronchoalveolar Lavage Fluid



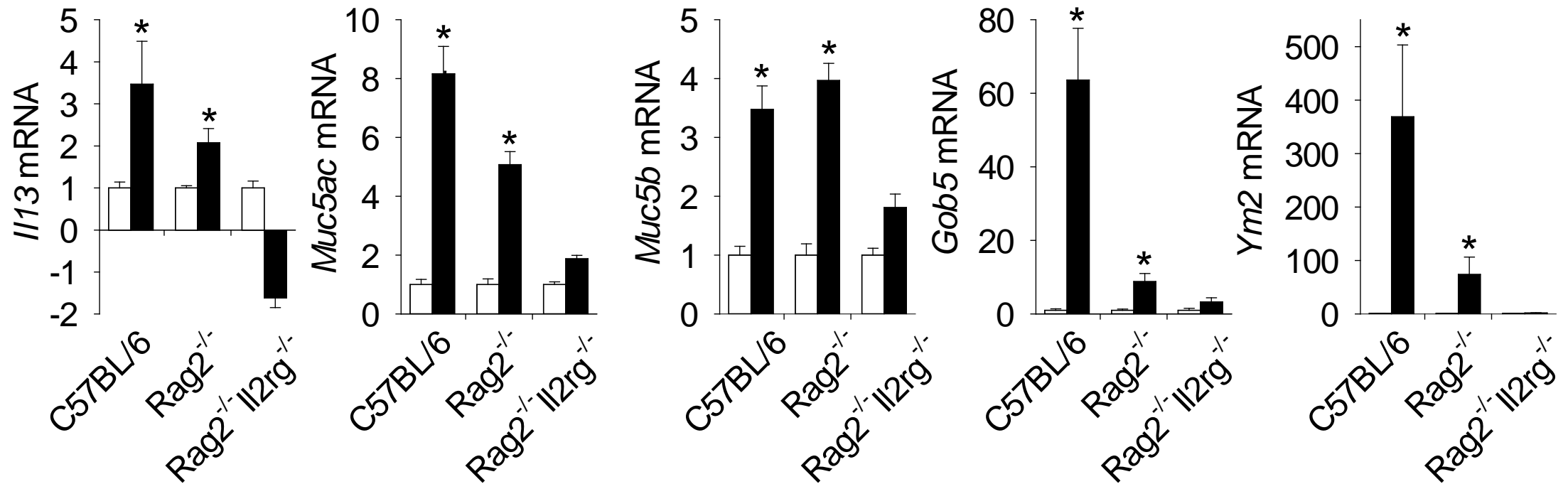
Bronchiolar epithelial injury and cell proliferation after 1 day of ozone exposure in both ILC-sufficient and -deficient mice



Mucous cell metaplasia in bronchiolar epithelium after 9 days of ozone exposure in ILC-sufficient mice, but not in ILC-deficient mice



Mucus- and type 2 immune-related mRNA overexpression in the lungs of ILC-sufficient mice after 9 days of ozone exposure



Summary

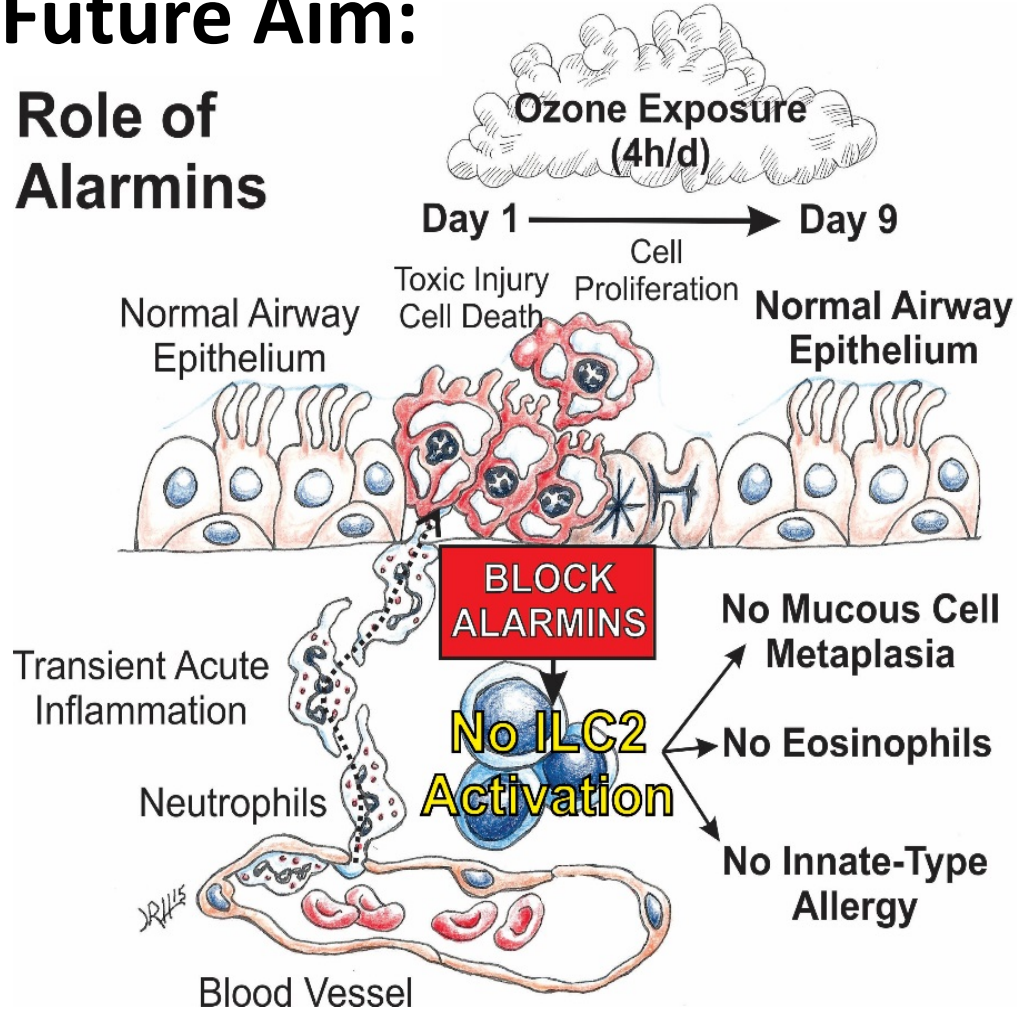
Mouse Strain	T & B cells	ILCs	O3-induced lesions
C57BL/6	+	+	+
Rag2(-/-)	-	+	+
Rag2(-/-)Il2rg(-/-)	-	-	-

Repeated exposures to ozone elicit innate-type allergy in the nose and lung of mice, that is likely to be dependent on type 2 cytokine-producing innate lymphoid cells. This suggests a new paradigm for the epidemiologic association of air pollution and allergic airway diseases.

Future Studies, Acknowledgments and Questions

Future Aim:

Role of Alarmins



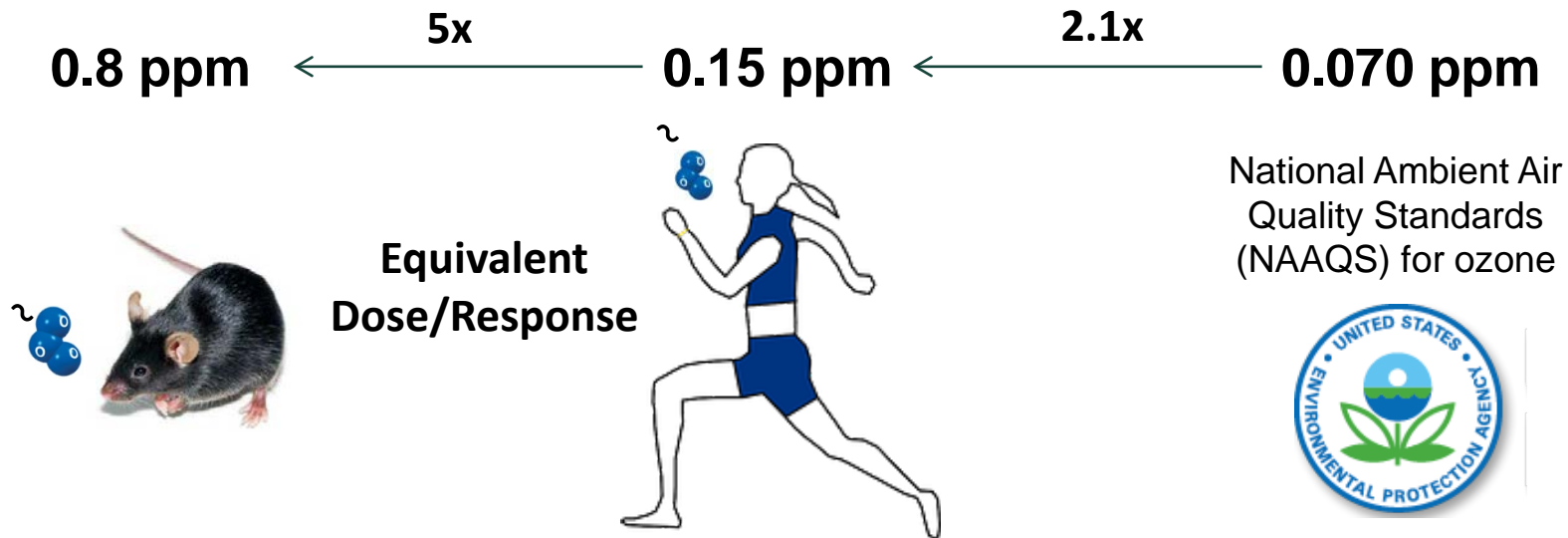
Kazuyoshi Kumagai, Chee Bing Ong, Daven Jackson-Humbles, Ryan Lewandowski, Nick Buglac, Phil Brook, Ning Li, and James Wagner

Questions?

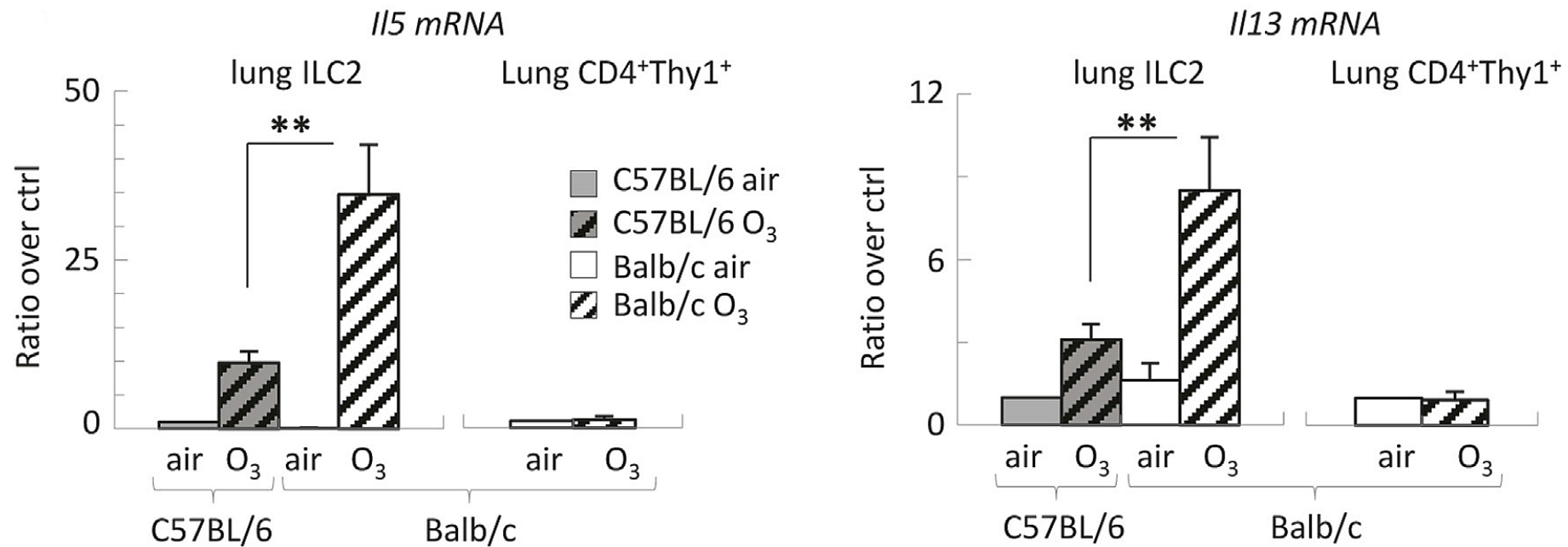
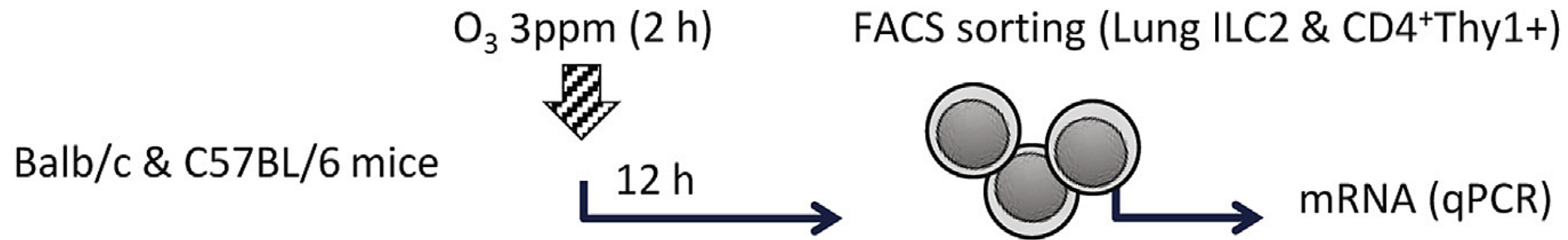


Rationale for Mouse Exposures to Ozone

- Based on previous studies in ozone-exposed rodents and humans (Hatch et al. 2013, 1994), the respiratory dose/response to **0.8 ppm ozone in mice** is equivalent to **0.15 ppm in exercising humans** (people are 5x more sensitive to ozone than mice).
- Concentration of 0.15 ppm is approximately twice as much as that of the current U.S. 8 h NAAQS for ozone (**0.070 ppm**).



Recent findings by others: Single high ozone exposure, ILC2s, Eosinophilic Inflammation, Balb/c versus C57BL/6 mice



Recent findings by others: Single high ozone exposure, ILC2s, AHR, Balb/c mice

