Transgenerational susceptibility to asthma: Impact of maternal exposure to ultrafine particles during pregnancy

Djamal Achour

D. Achour[1], C. Grare[1], A. Rekbi[1], P. de Nadaï[2] and J-M. Lo Guidice[1]

1 Univ. Lille, CHU Lille, Institut Pasteur de Lille, ULR4483 - IMPECS, Lille, France
2 Univ. Lille, CNRS, CHU Lille, Institut Pasteur de Lille, U1019 - UMR9017 - CIIL, Lille, France

Our Team

IMPact de l'Environnement Chimique sur la Santé
Impact of the Chemical Environment on Health

Multi-disciplinary : toxicologists, epidemiologists and pharmacists

2 axis of research

Mechanisms of toxic action of aerocontaminants
analyze the pneumotoxicity of air contaminants and
identify biomarkers of effects.

Human Biomonitoring
Research and validation in humans of biomarkers of
exposure, impregnation and effects of aerocontaminants.

Air Pollution

- 4.2 million premature deaths (outdoor pollution); > 85% ➔ Fine particles[1]

- 238 000 premature deaths in EU : Fines particles[2]

1 : https://www.who.int/news-room/fact-sheets/detail/ambient-(outdoor)-air-quality-and-health
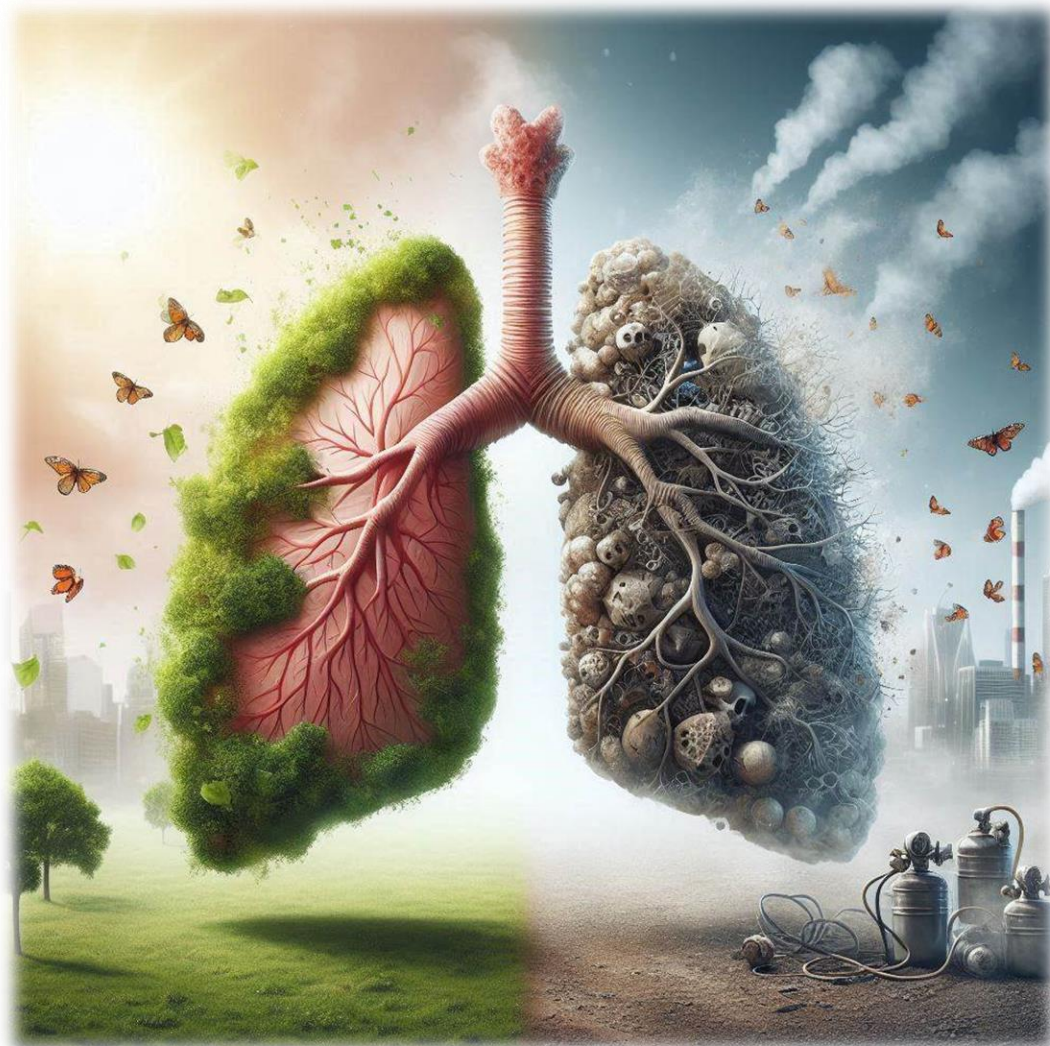2 : https://www.eea.europa.eu/fr/highlights/le-nombre-de-deces-prematures

Air Pollution

- 4.2 million premature deaths (outdoor pollution); > 85% ➔ Fine particles[1]

- 238 000 premature deaths in EU : Fines particles[2]

- Respiratory morbidity : Cancer, COPD, Fibrosis, **Asthma** …

Chronic
Obstructive
Pulmonary
Disease
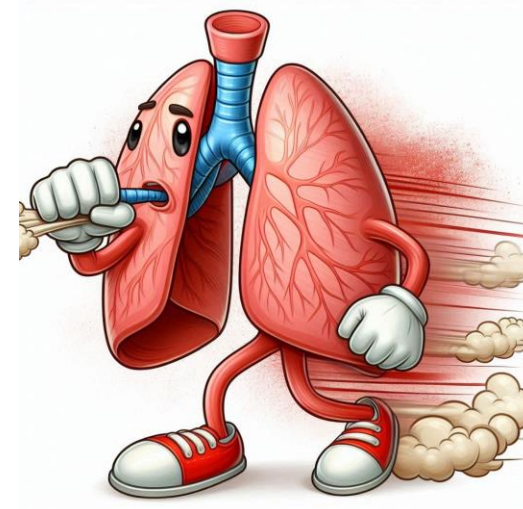
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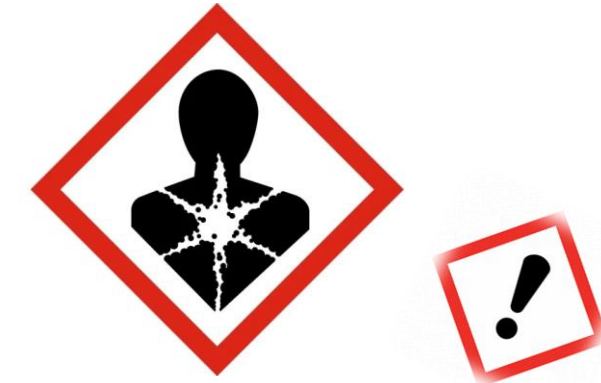2 : https://www.eea.europa.eu/fr/highlights/le-nombre-de-deces-prematures

Air Pollution

Chemical composition

Carbon species
- Elemental / organic carbon
- PAH, VOC...

Mineral fraction
- Mineral dust
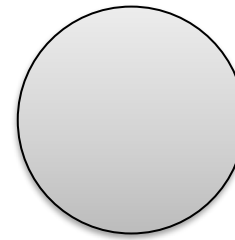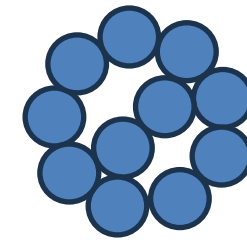- Ions
- Metals (Pb, Ni, Cd...)

Air Pollution

Chemical composition

Reactivity surface

Size

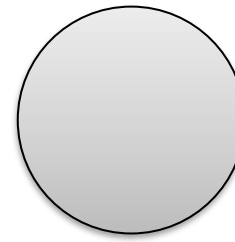PM$_{10}$ PM$_{2.5}$ PM$_{0.1}$

1 Coarse Particle (PM$_{10}$) = 1 000 000 UltraFine Particles (PM$_{0.1}$)

Air Pollution

Chemical composition
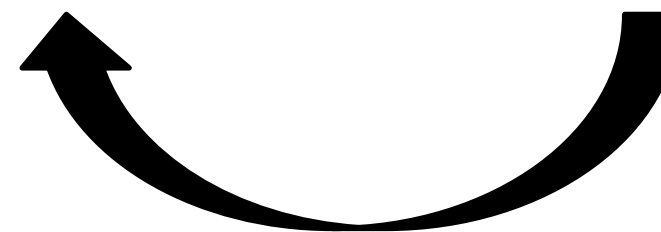
Reactivity surface
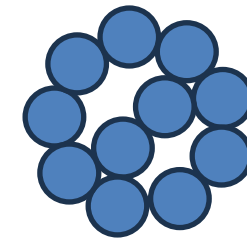
Size

PM_{10} $PM_{2.5}$ $PM_{0.1}$

1 Coarse Particle (PM_{10}) = 1 000 000 Ultra Fine Particles ($PM_{0.1}$)

Reactivity surface
X100

Air Pollution

Chemical composition
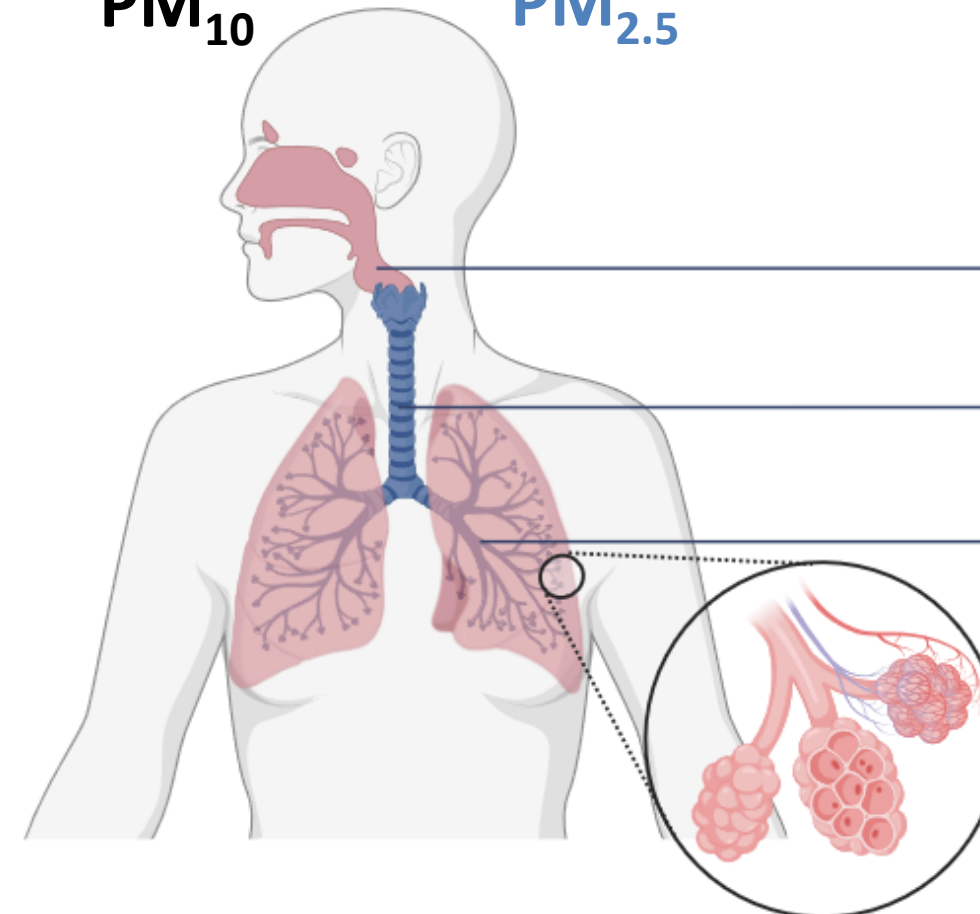
Size → Reactivity surface
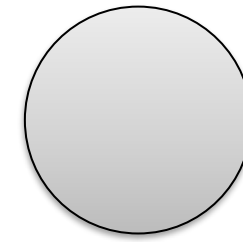
Size → Penetration

PM$_{10}$ PM$_{2.5}$ PM$_{0.1}$
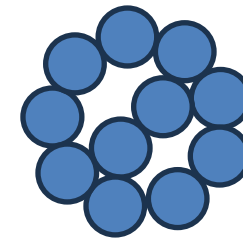
Nose and Throat
5 - 10 µm

trachea
3 - 5 µm

Bronchi & Bronchiole
2 - 3 µm & 1 - 2 µm

Alveoli
0.1 - 1 µm

Air Pollution and Asthma

Less mature lungs
Higher respiratory frequency

Asthma : Most common chronic disease in children
10% of european children

Pathophysiology of asthma

Normal airway	Asthmatic airway

- Airway inflammation
- Bronchial hyperactivity
- Respiratory tract remodeling

≠ Asthma

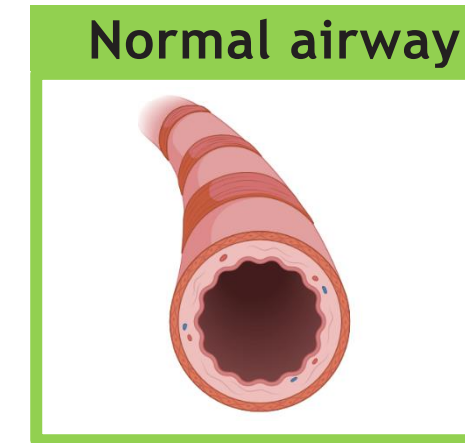| TH2 Asthma **Allergic** and eosinophilic	non-TH2 Asthma Neutrophilic...

Air Pollution and Asthma

Less mature lungs
Higher respiratory frequency
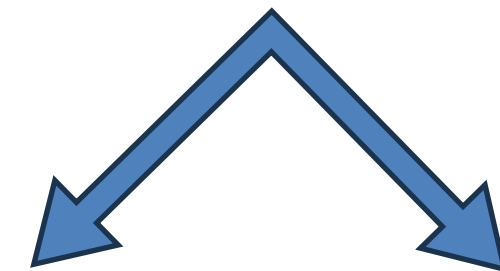
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Pathophysiology of asthma

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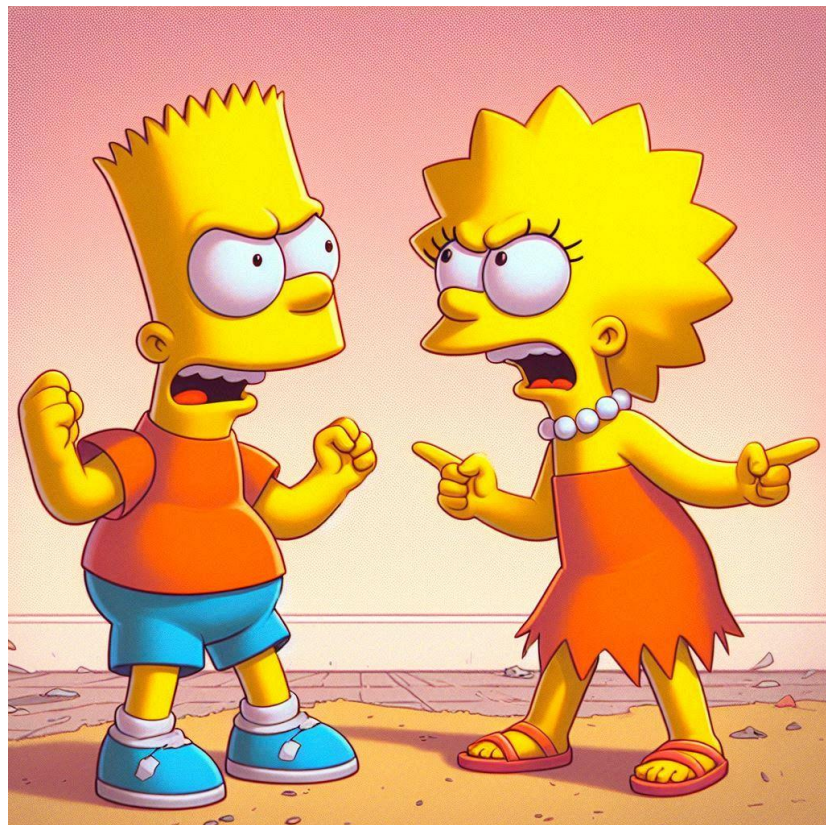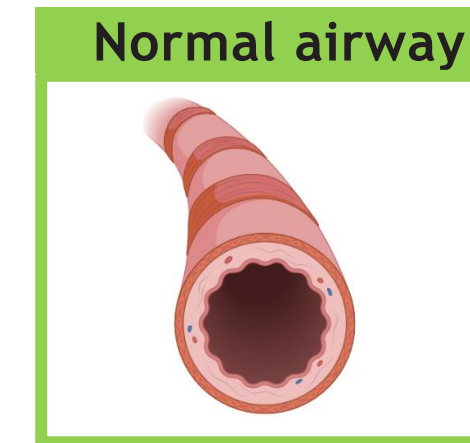
≠ Asthma

TH2 Asthma **Allergic** and eosinophilic	non-TH2 Asthma Neutrophilic…

Air Pollution and Asthma

Child cohort studies

Particulate pollution 🔗 Asthma

Khreis *et al.* 2017
Brauer *et al.* 2002
Morgenstern *et al.* 2007

PM **translocation** of from mother to **fetus**
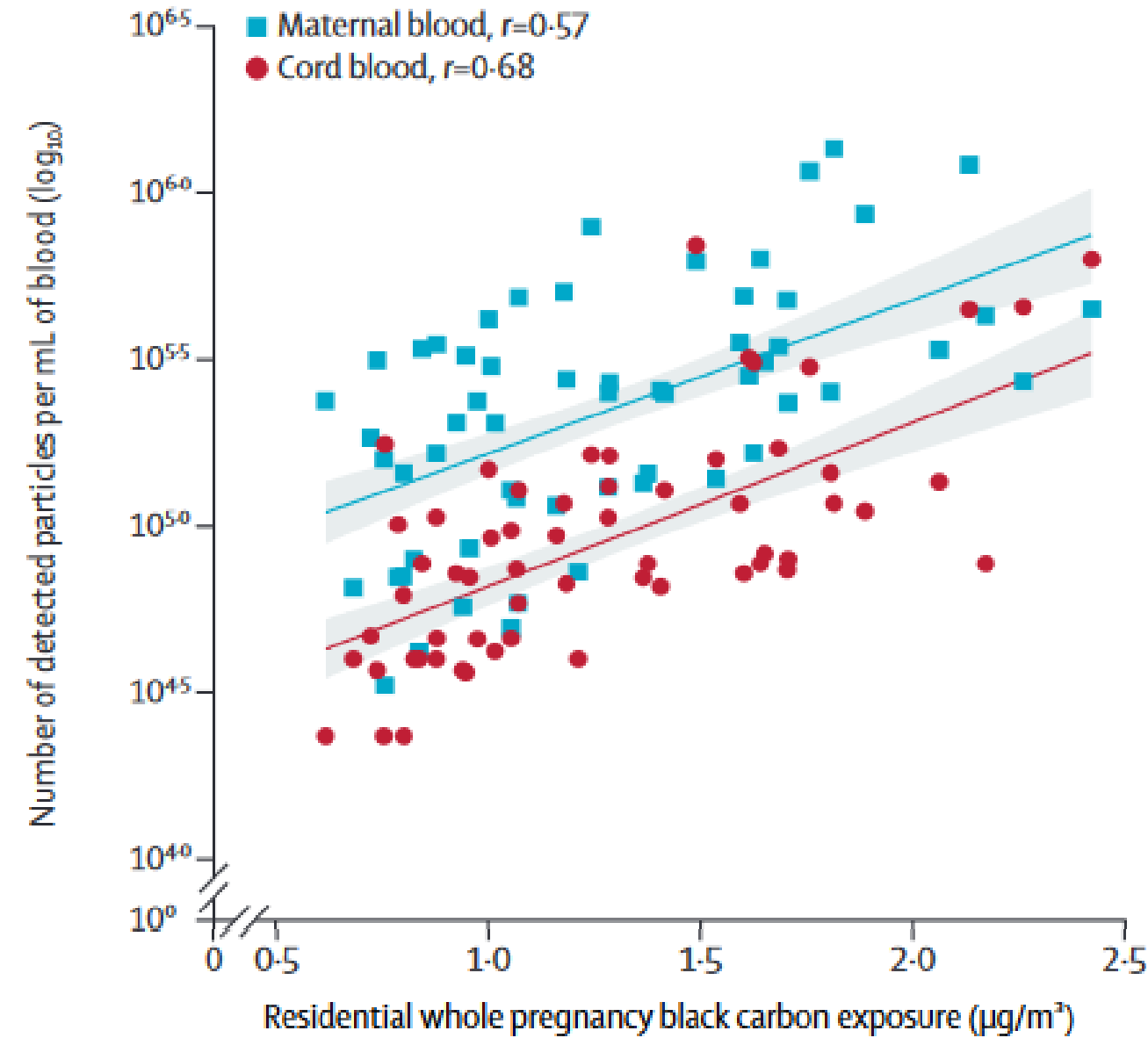
Bongaerts *et al.* 2022

Question

Does *in utero* exposure increase the risk of developing or exacerbating asthma?

Protocol

Sampling : High Volume Impactor Sampler
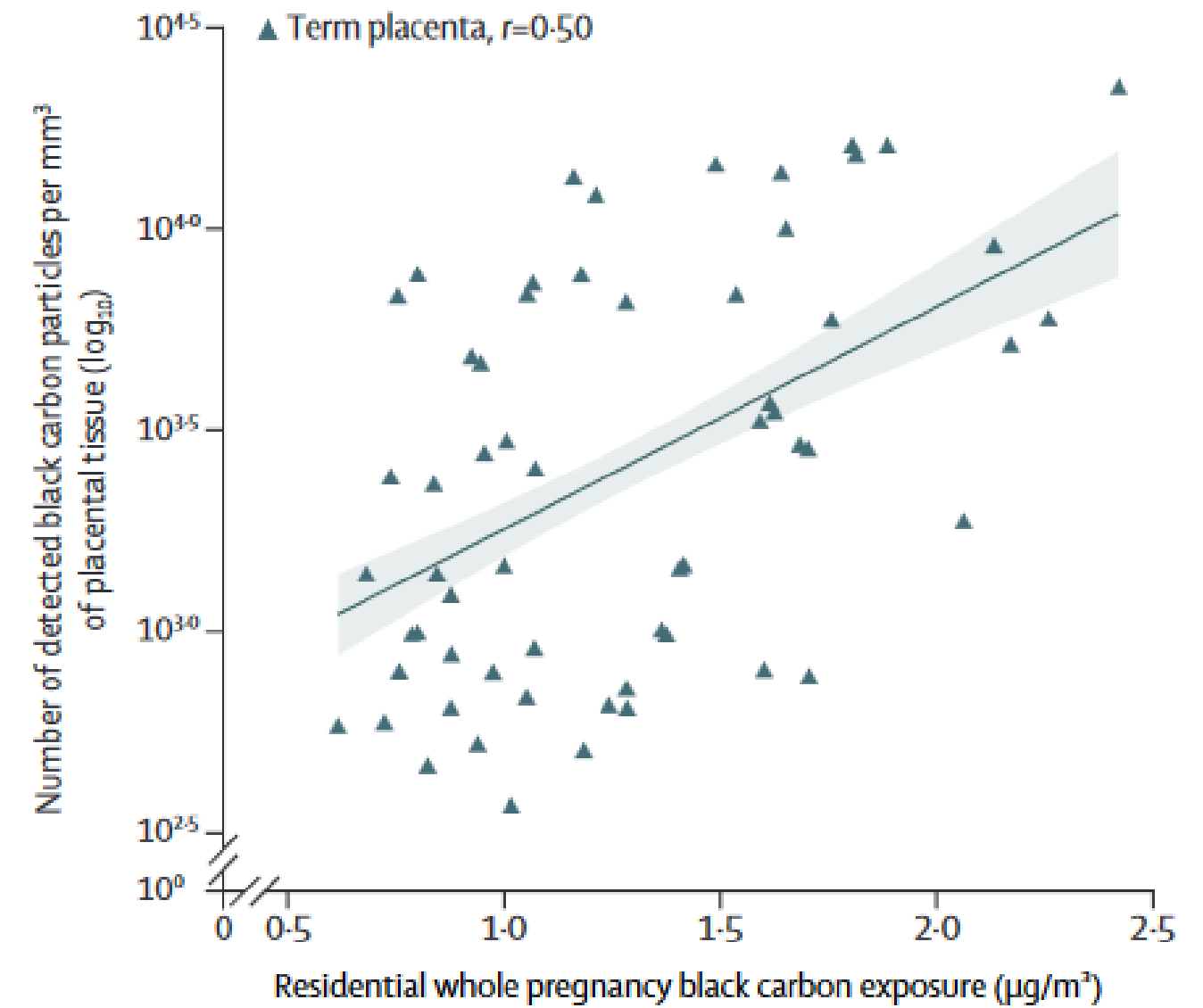HVIS

Floor 1
Plate

Floor 2
Plate

Floor 3
Filter

Pump
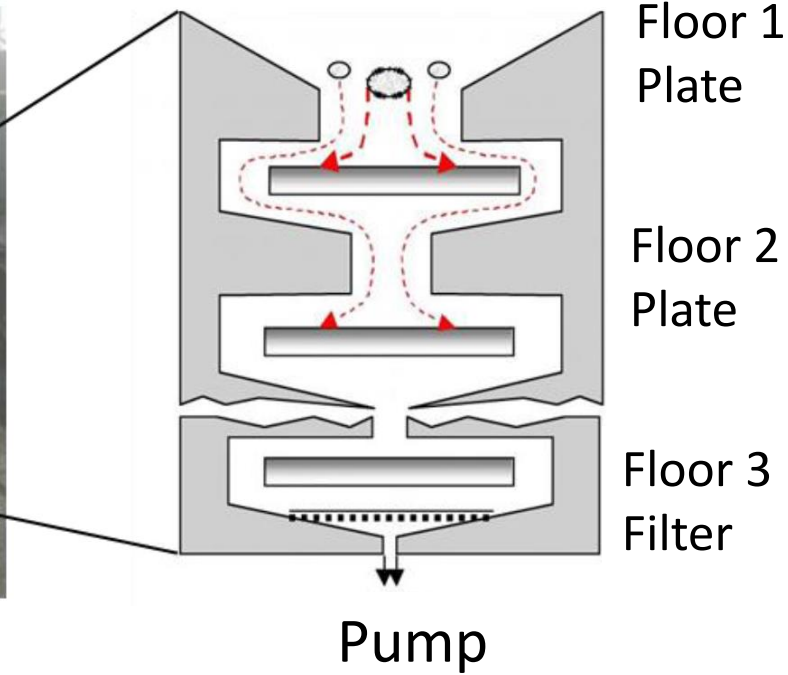
September 2013 – April 2014
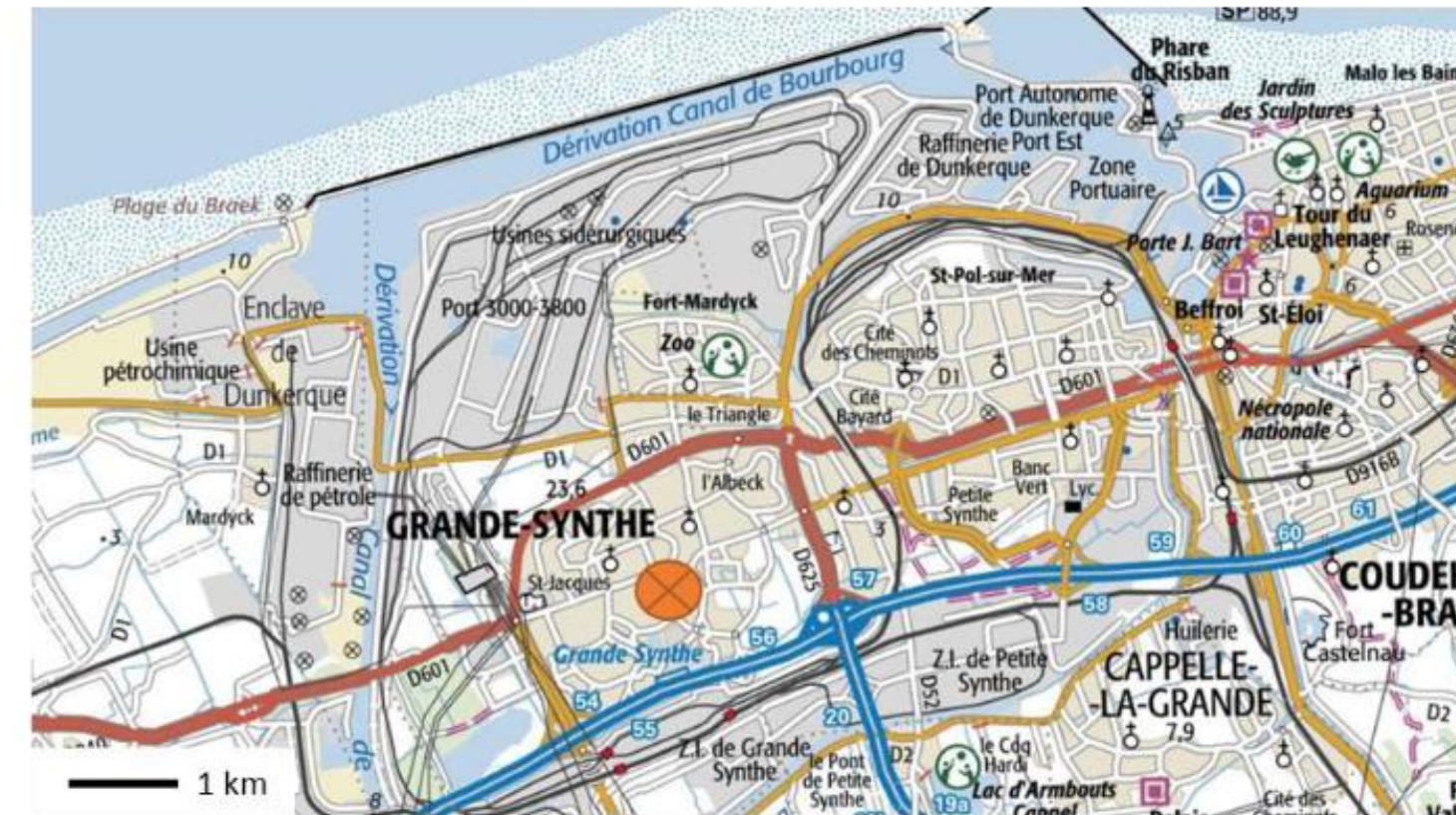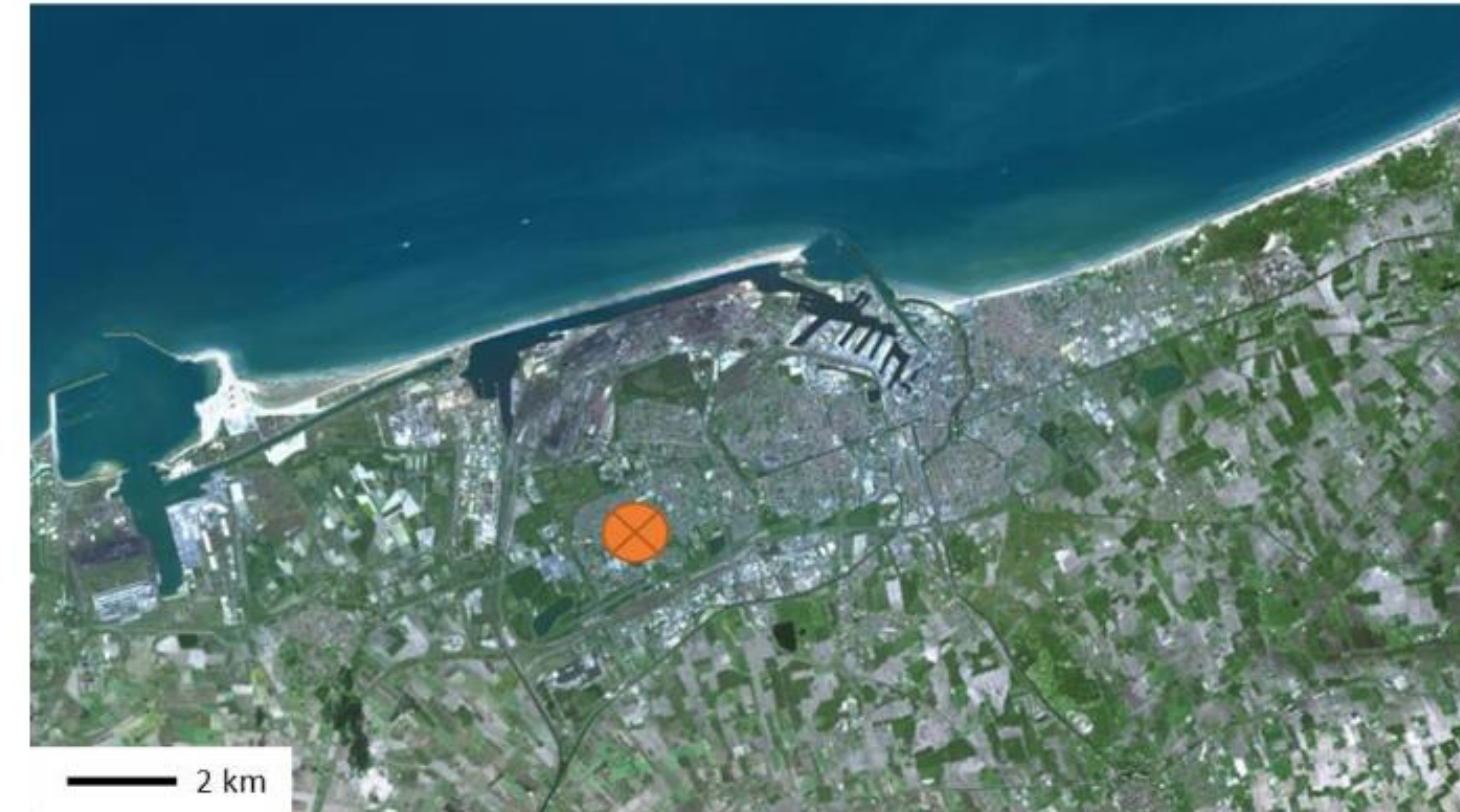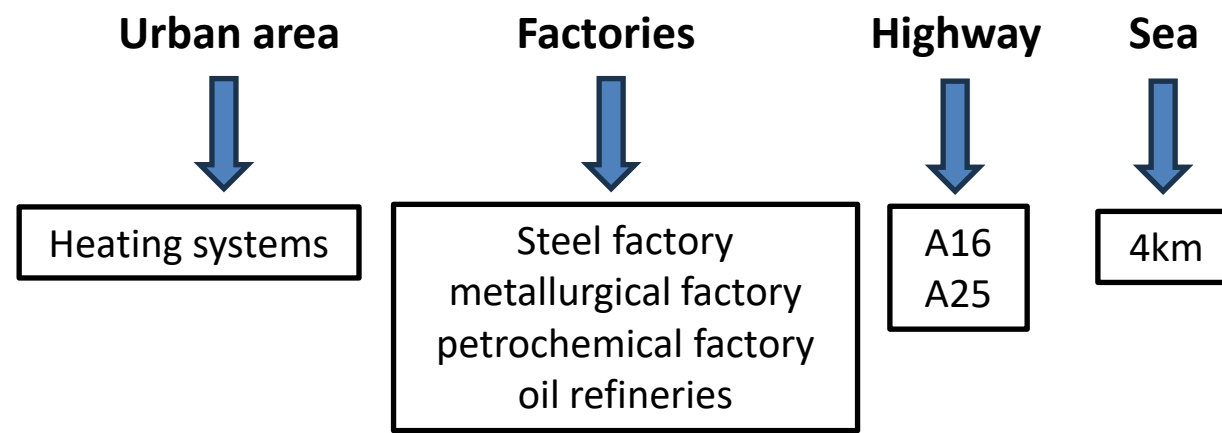
Dunkirk

Urban area	Factories	Highway	Sea
↓	↓	↓	↓
Heating systems	Steel factory metallurgical factory petrochemical factory oil refineries	A16 A25	4km

2 km

1 km

Protocol

1) Exposure of mothers 24 hours after mating

2) Birth of mice

3) Induction of asthma

4) Sacrifice the pups

UFP (30μg)

Control

Control   Df

UFP/PBS   UFP/Df

PBS/PBS   PBS/Df
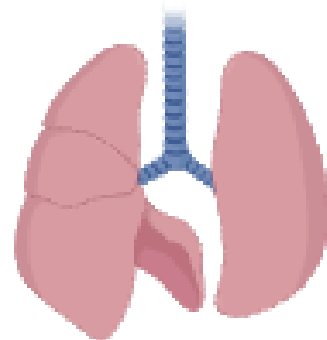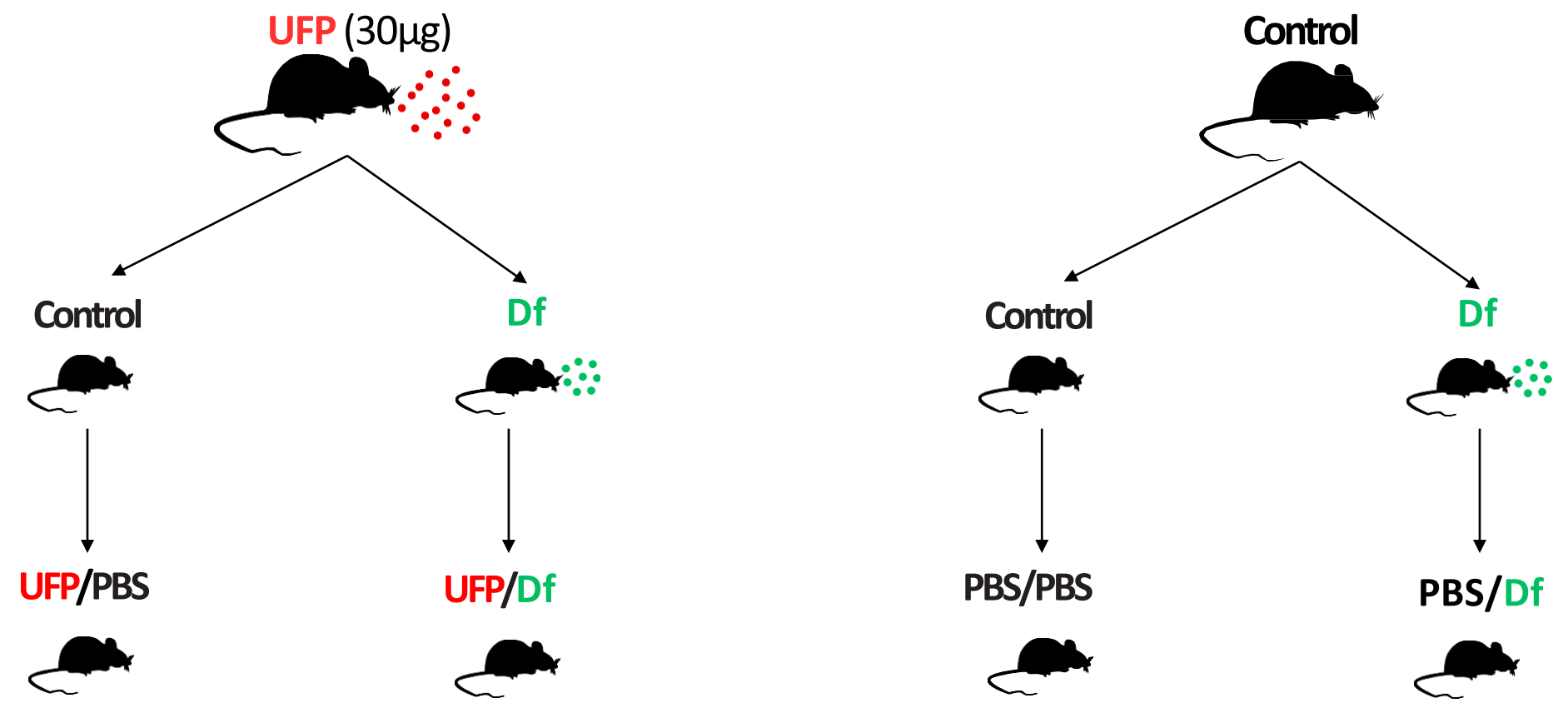
Lung
➢ **Histological analysis**
➢ **Cytokine quantification**
➢ Respiratory function
➢ Immune cell characterization
➢ Transcriptomic and epigenetic analyses

BAL
➢ **Cytological analysis**
➢ Cytokine quantification
➢ Immune cell characterization
➢ Transcriptomic and epigenetic analyses

Plasma
➢ Cytokine quantification
➢ Epigenetic analysis (miRNA)

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Results

Histological analysis
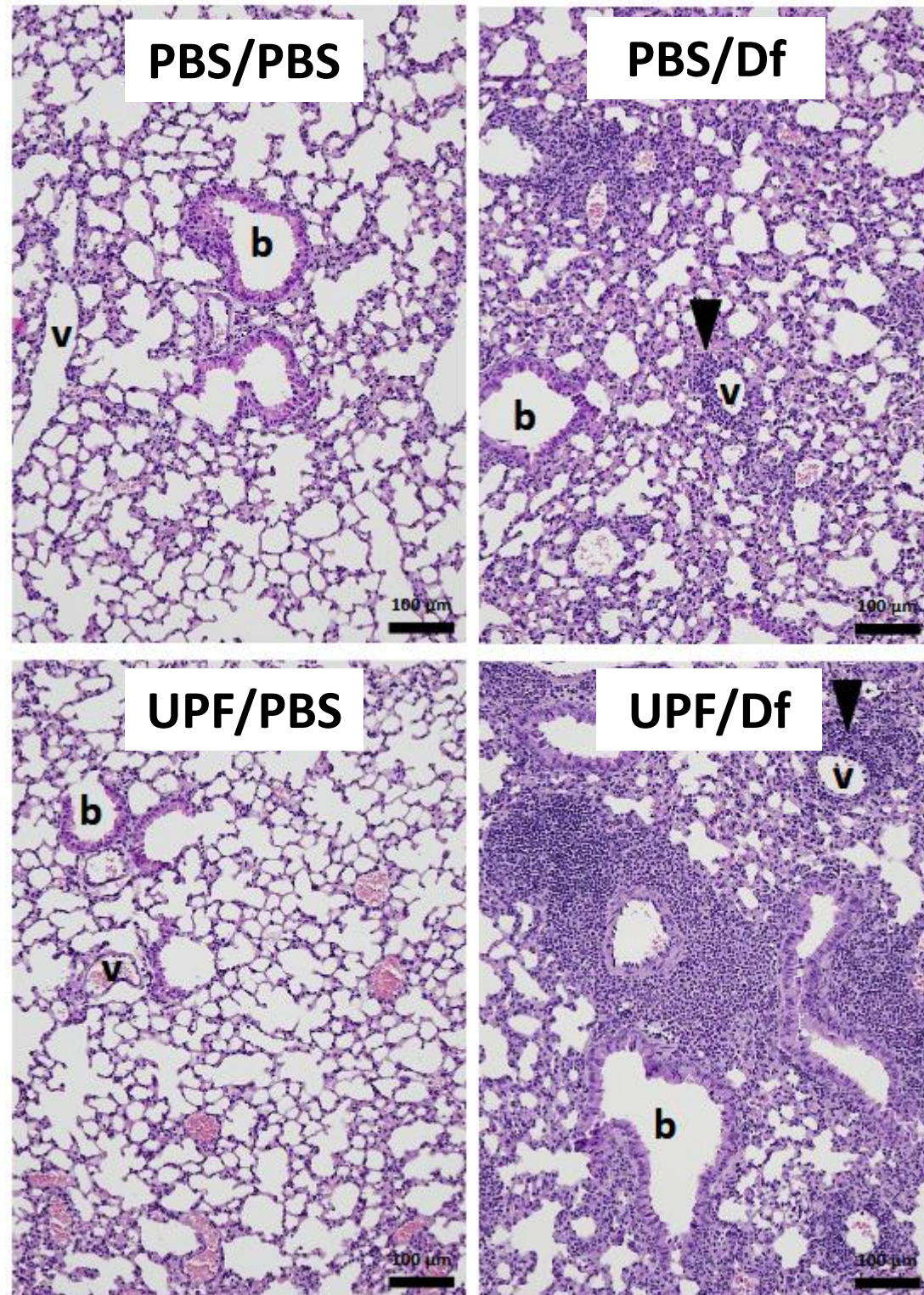
b : bronchi   v : blood vessels

Cumulative score :
- Bronchial lesions
- Bronchial content
- Bronchi Associated Lymphoid Tissue (BALT)
- Vasculitis
- Peri-vascular and peri-bronchial cuffing
- Interstitial pneumonia
- Emphysema
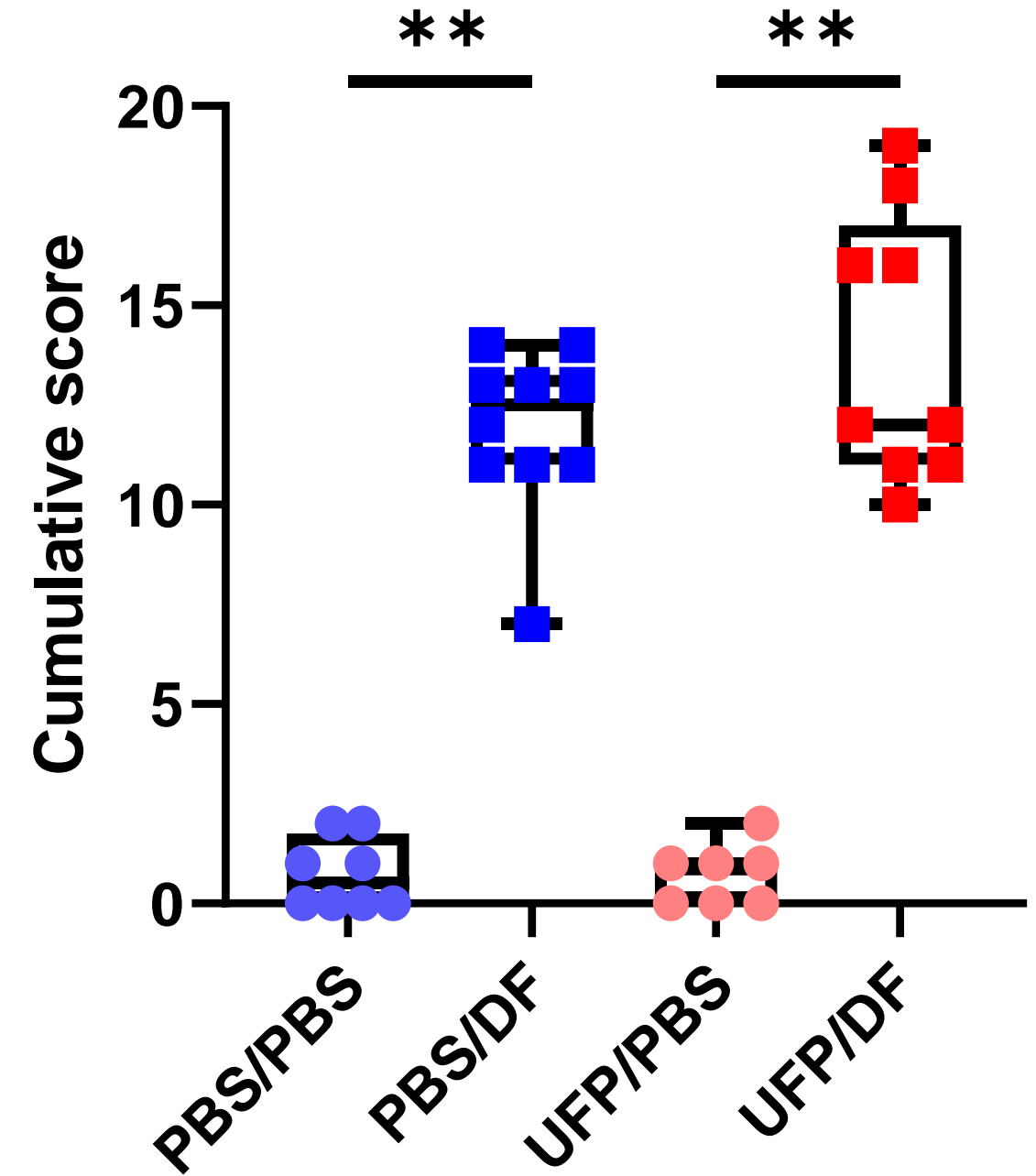
** P-Value < 0,01

Cytological Analysis

Number of cells in BAL

Eosinophils

Macrophages

Neutrophils

Lymphocytes
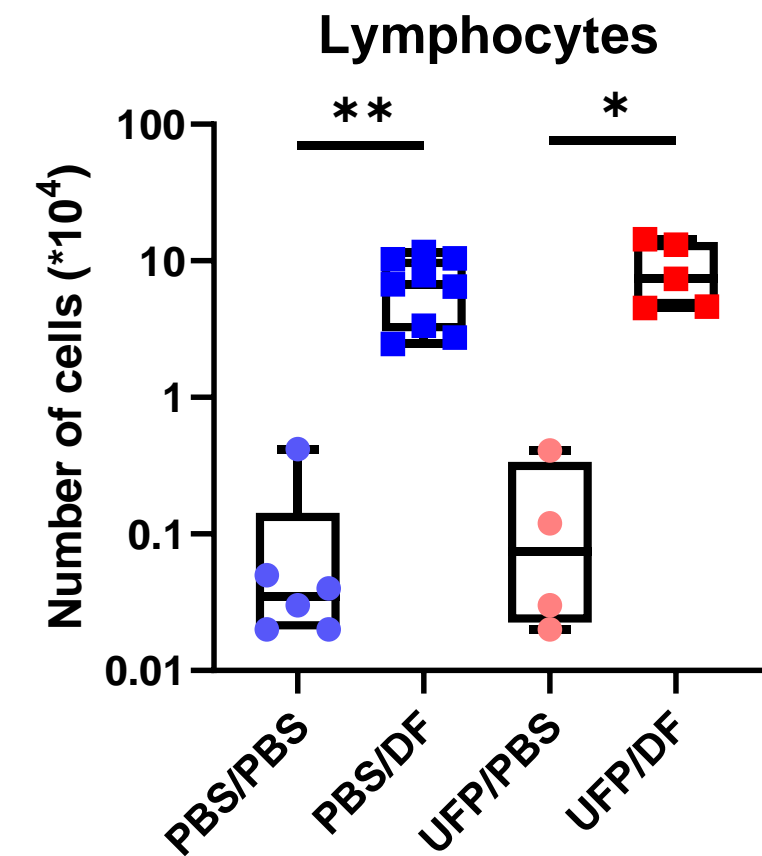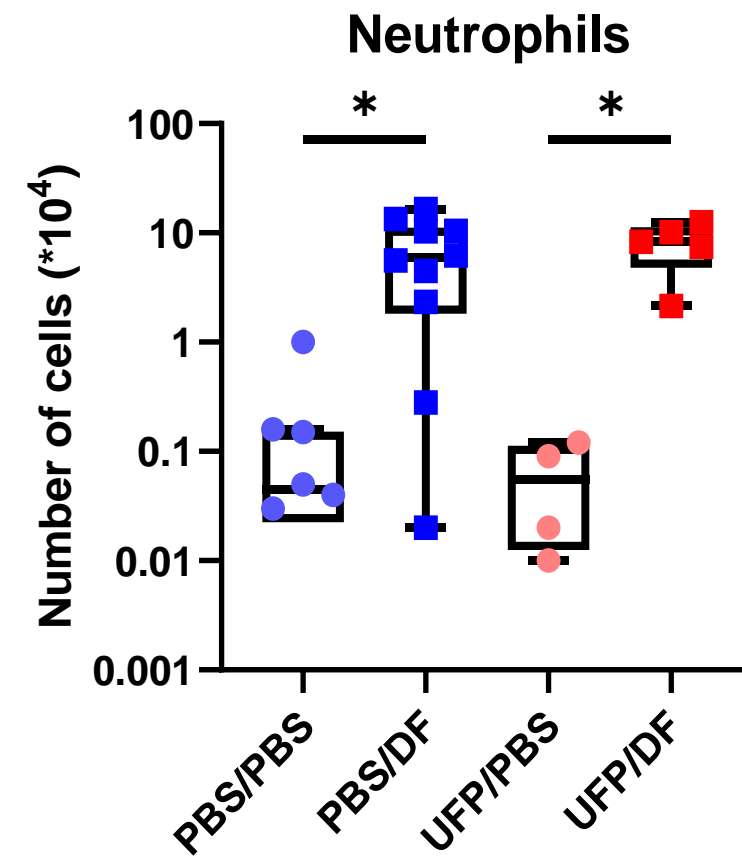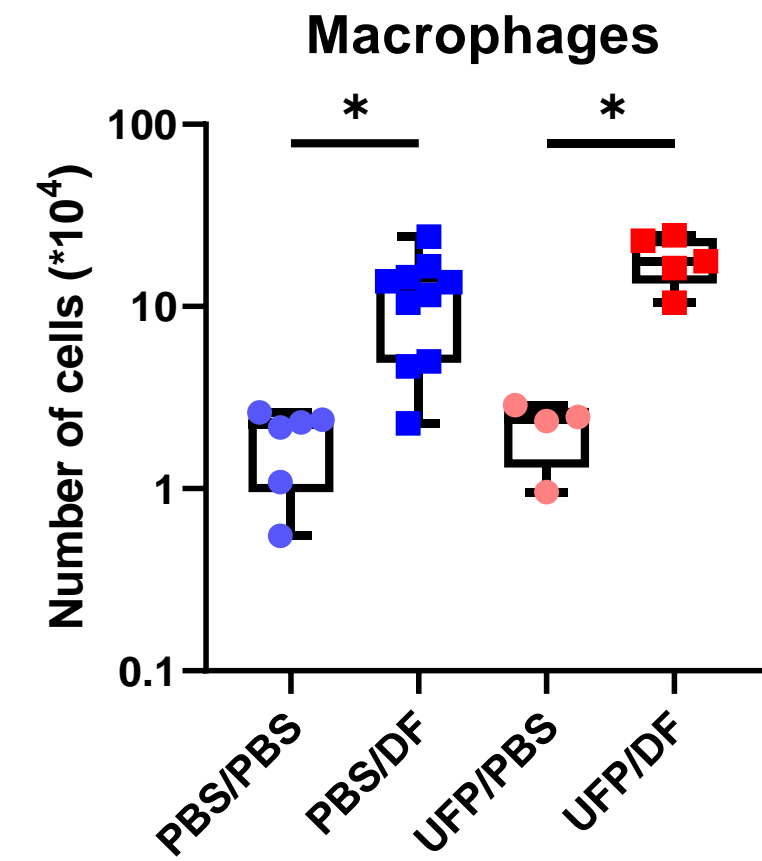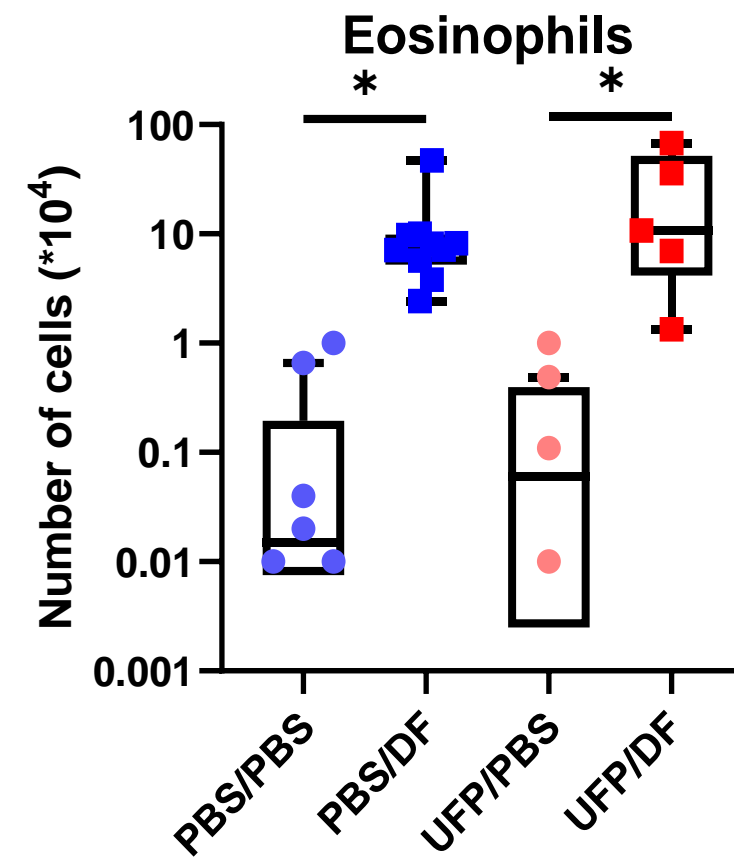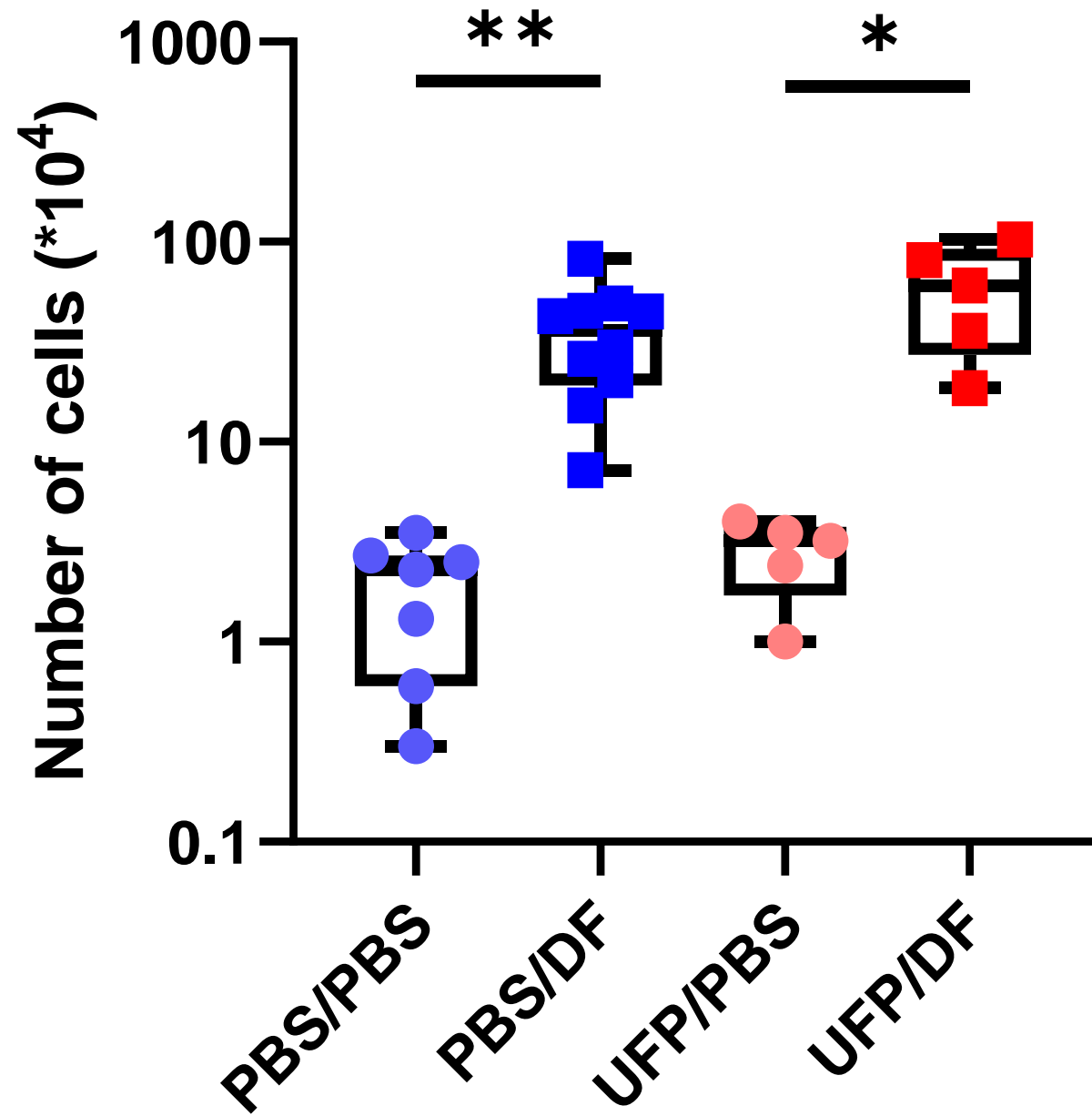
*p-Value < 0,05
**p-Value < 0,01

Cytokine quantification

Protein lung extracts

Panel of cytokines :

- IFN-γ
- IL-1β
- IL-2
- IL-4
- IL-5
- IL-6
- IL-9
- IL-10
- IL-12p70
- IL-15

- IL-16
- IL-17A
- IL-17C
- IL-17E
- IL-17A/F
- IL-17F
- IL-21
- IL-22
- IL-23
- IL-27

- IL-31
- IL-33
- IP-10
- KC/GRO
- MCP-1
- MIP-1α
- MIP-2
- MIP-3α
- TNF-α

Cytokine quantification

Protein lung extracts

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- TNF-α

Cytokine quantification

Cytokine quantification

Non-TH2 Asthma

Conc IL1b

Conc of IL6

Conc of IL17A

Conc of IL21

Conc of KC/Gro
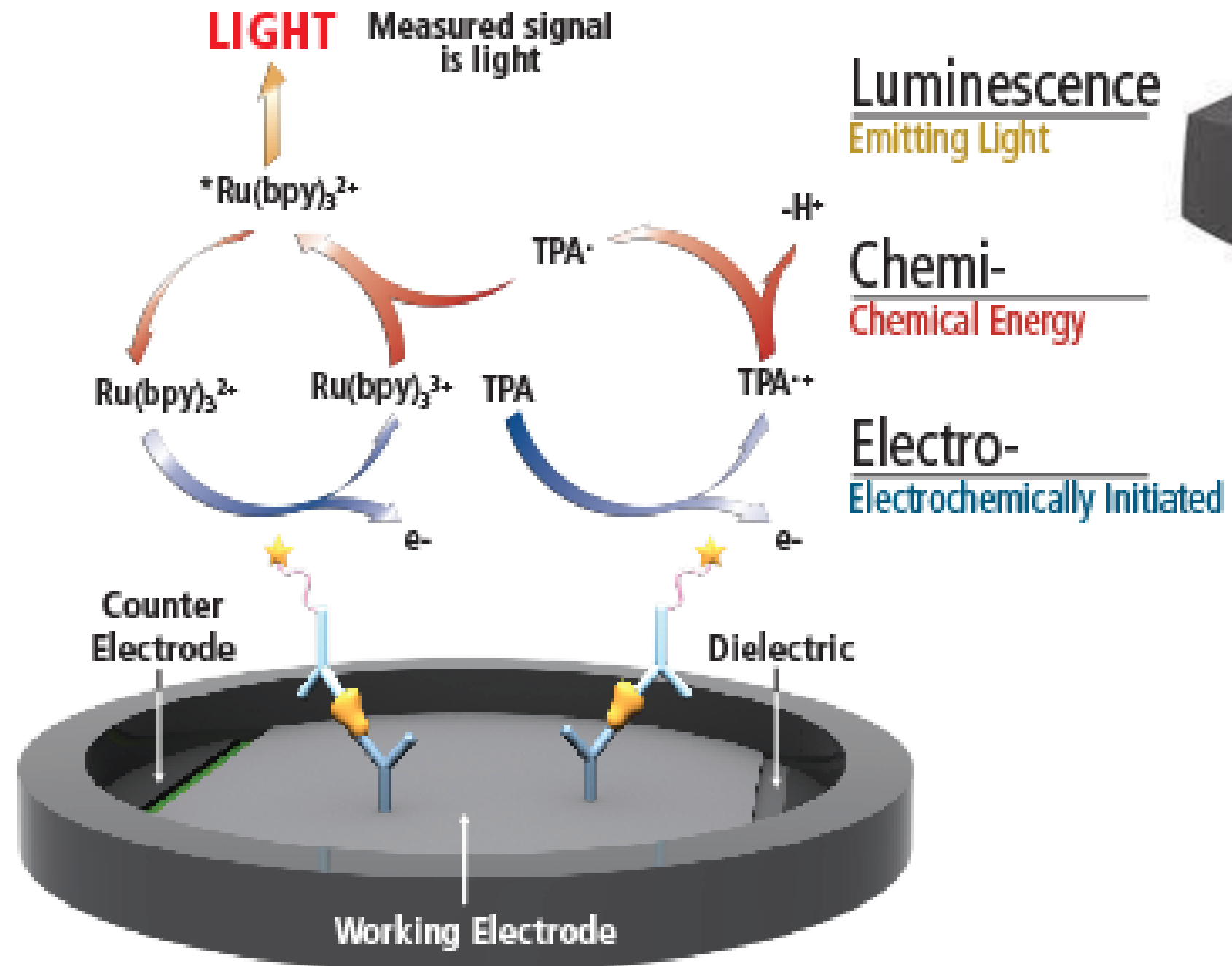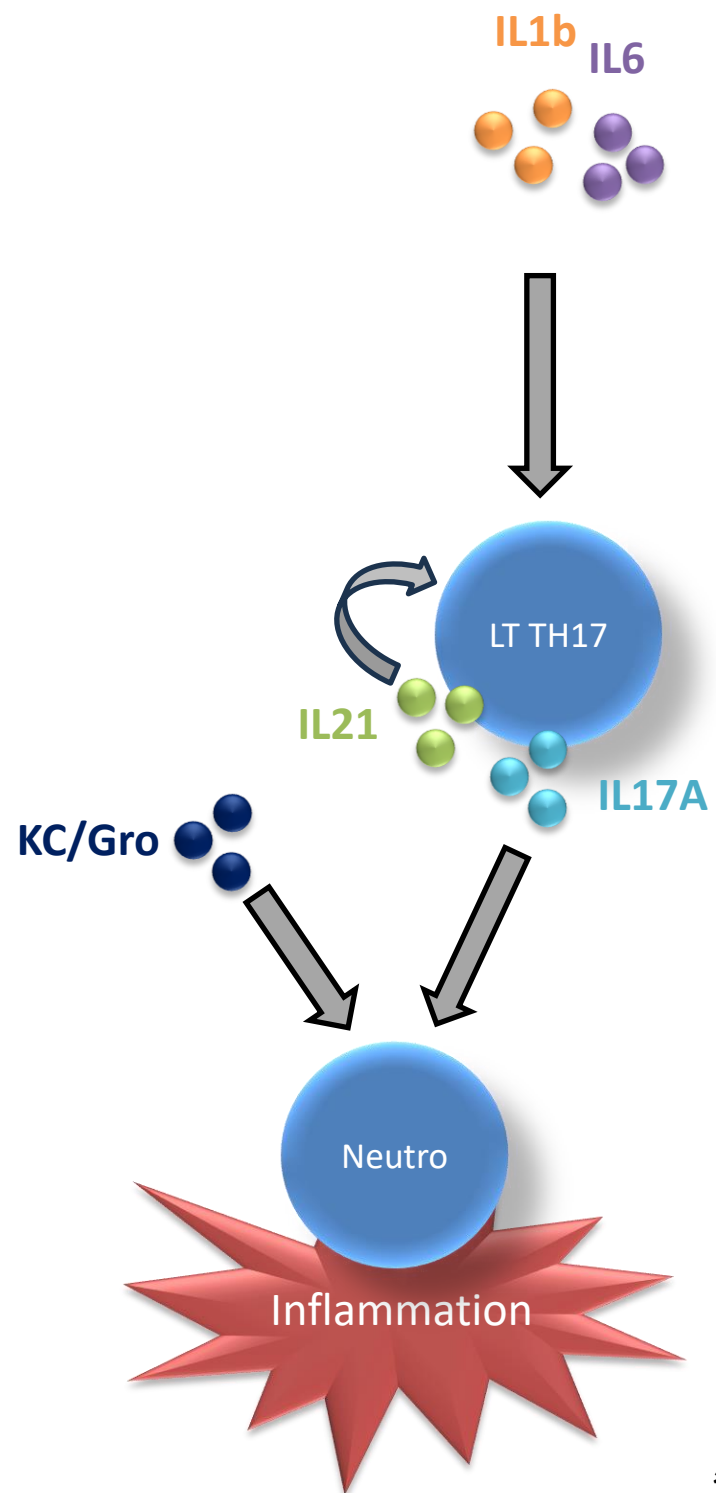
*p-Value < 0,05
**p-Value < 0,01
***p-Value < 0,001
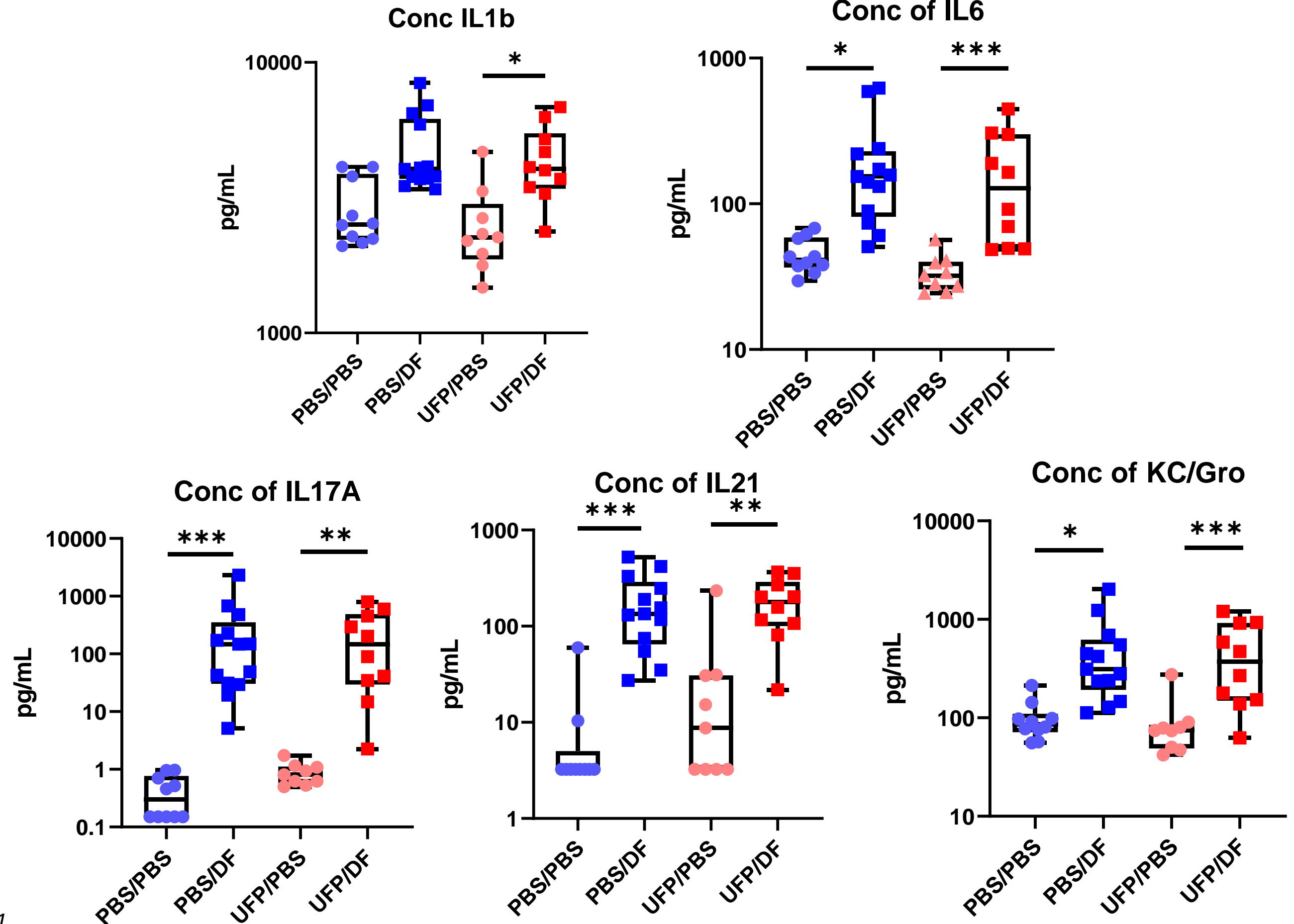
14

Conclusion

Preliminary work

Induction of allergic asthma in mice: inflammation after exposure of mice to allergen

Inflammatory reaction too strong ➔ mask the effect of the UFPs?

Conclusion

Preliminary work

Induction of allergic asthma in mice: inflammation after exposure of mice to allergen

Inflammatory reaction too strong ➔ mask the effect of the UFPs?

Perspective

More samples ➔ male and female

Analyses in progress : respiratory function, gene expression, epigenetic modifications

Thank you for your attention

Djamal Achour

djamal.achour@univ-lille.fr

https://impecs.univ-lille.fr/

Air Pollution

Reactivity surface

PM$_{10}$ **PM$_{2.5}$**   **PM$_{0.1}$**
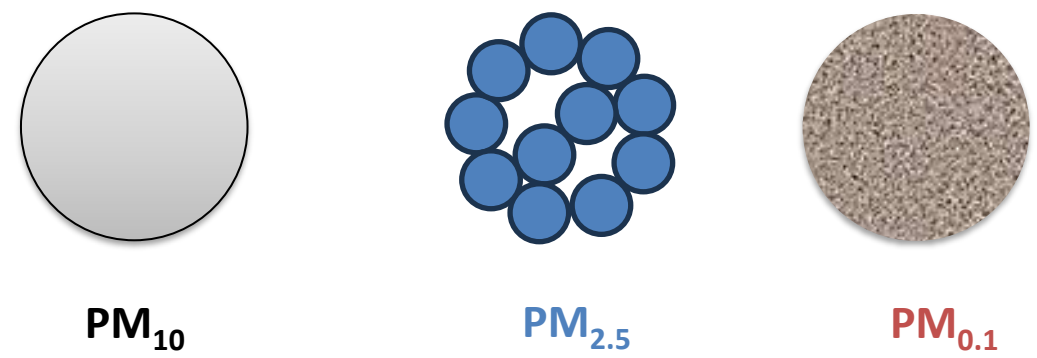
Size	CP	FP	UFP
Volume (µm3)	524	8	0,0005
Nn of particule for same volume	1	64	1 000 000
Reactivity surface (µm²)	314	1257	31420

X100

CP : Coarse Particles
FP : Fine Particles
UFP : Ultra Fine Particles

Appendix - Protocol

Particles

Sampling: High flow cascade impactor
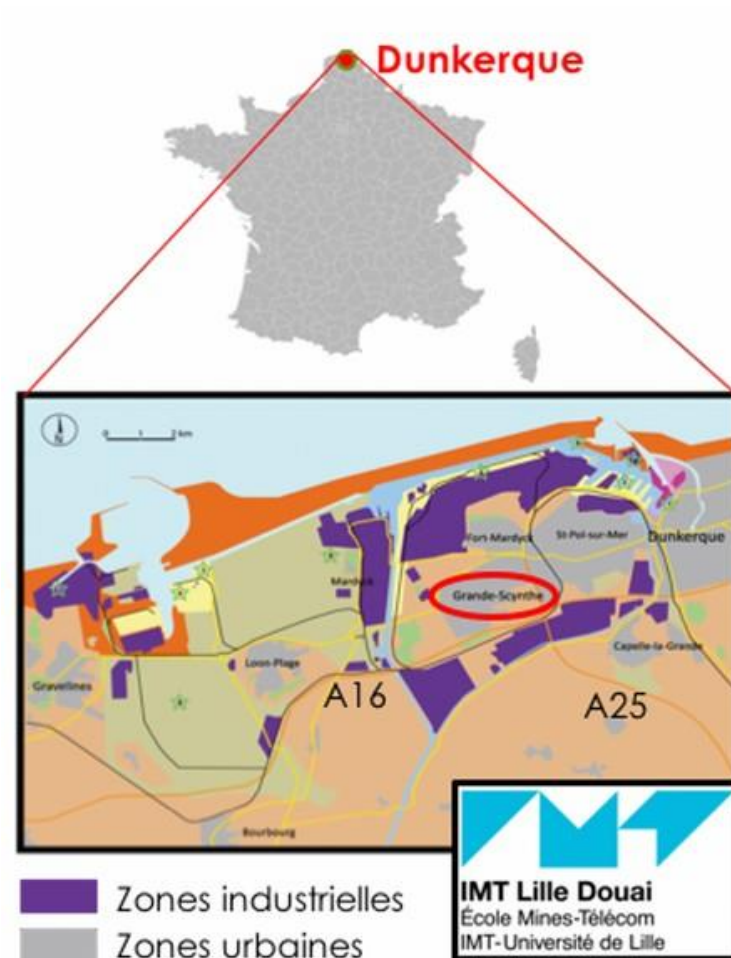
Mice

C57BL/6
9 to 12 weeks

Administration

Intranasal instillation

- Exposure of pregnant mice

Sensitisation of offspring to mite extracts

Appendix - Particles characterization

PAH	Acrony	UFP (ng/mg)
Fluoranthene	(FLA)	4,3
Pyrene	(PYR)	3,7
Benzo(c)phenanthrene	(BcPHE)	0,2
Benzo(a)anthracene	(BaA)	4,2
Chrysene	(CHR)	2,8
5-Methylchrysene	(5MCHR)	3,4
Benzo(e)pyrene	(BeP)	0,8
Benzo(b)fluoranthene	(BbF)	8,0
Benzo(j)fluoranthene	(BjF)	9,9
Benzo(k)fluoranthene	(BkF)	4,6
Benzo(a)pyrene	(BaP)	5,3
Dibenzo(a,l)pyrene	(DalP)	0,4
Dibenzo(a,h)anthracene	(DahA)	1,5
Benzo(g,h,i)perylene	(BghiP)	12,5
Indeno(1,2,3-c,d)pyrene	(IP)	12,1
Dibenzo(a,e)pyrene	(DaeP)	2,7
Anthanthrene	(ANTH)	3,3
Coronene	(COR)	4,5

| Element (µg/g)	UFP	Origin
Ba	64.4	Other
Be	0.3	Other
Co	9.3	Other
Sn	111.1	Other
Si	7568.1	Other
As	64.8	Combustion of traffic, heating, coal
Cd	19.3	Combustion of traffic, heating, coal
Cs	8.9	Combustion of traffic, heating, coal
Pb	541.9	Combustion of traffic, heating, coal
Rb	44.1	Combustion of traffic, heating, coal
Sb	49.3	Combustion of traffic, heating, coal
Ti	5.7	Combustion of traffic, heating, coal
K	30368	Combustion of traffic, heating, coal
Mo	36.7	Heavy fuel oil
Ni	199.7	Heavy fuel oil
V	196.3	Heavy fuel oil
Ce	6.3	Floor resuspension
Cu	425.2	Floor resuspension
Sr	49.3	Floor resuspension
Zn	2460.7	Floor resuspension
Al	3621.1	Floor resuspension

Element (µg/g)	UFP	Origin
Mn	582.2	Steel industry Marin
Ca	11857.4	Steel industry Marin
Fe	10267	Steel industry Marin
Mg	6344.8	Steel industry Marin
Na	283147	Steel industry Marin
La	3.1	Industrial zone
Cr	120.7	Industrial zone

Appendix - Cytokines

IFN-g	Pro-inflammaroty
IL-2	Ambivalent role
IL-4	Lymphocyte B Differenciation
IL-5	Eosinophile recruitement
IL-6	Pro-inflammaroty
IL-8	Pro-inflammaroty
IL-10	Anti-inflammatory
IL-13	Mucus secretion
IL-17A	Stimulate IL-6 and IL-8
IL-17E/IL25	stimulates the production of IL-4/IL-5/IL-13
IL-33	Pro-inflammaroty
CCL2	Monocyte and macrophage recruitment
CCL20	Lymphocyte T Cells and antigen-presenting recruitment
IL1RL1	IL-33 Receptor
VGEFA	stimulates the production of IL-4/IL-5/IL-13

Appendix - Histopathology

Lung lesions were scored following 7 parameters :

- **Bronchial lesions** ➔ 0=no lesion; 1=individual cell necrosis; 2=focal denudation; 3=segmental total denudation
- **Bronchial content** ➔ 0=empty lumen; 1=presence of a small amount of isolated material; 2=partial obliteration by clumps of material; 3=complete segmental lumen obliteration
- **Bronchi Associated Lymphoid Tissue (BALT) :** 0= no BALT; 1= peri-bronchial limited collection of mononuclear cells; 2= well-formed follicles
- **Vasculitis** ➔ 0=no lesion; 1=leukostasis; 2=focal wall damages (including leukocytoclasis); 3=transmural vessel wall alteration and/or vascular lumen obliteration).
- **Peri-vascular and peri-bronchial cuffing** ➔ 0=no lesion; 1=discontinuous focal inflammatory cell infiltration; 2=continuous circumferential inflammatory cell infiltration; 3=coalescing inflammatory cell infiltration between bronchi and vessels)
- **Interstitial pneumonia** ➔ 0=no lesion; 1=focal alveolar wall thickening; 2=extensive alveolar wall thickening; 3=lung parenchyma focal consolidation
- **Emphysema** ➔ 0=no lesion; 1= alveolar lumen enlargement; 2= extensive distal airspace enlargement and alveolar wall destruction; 3=presence of small "bullae" corresponding to an air-filled space in the parenchyma
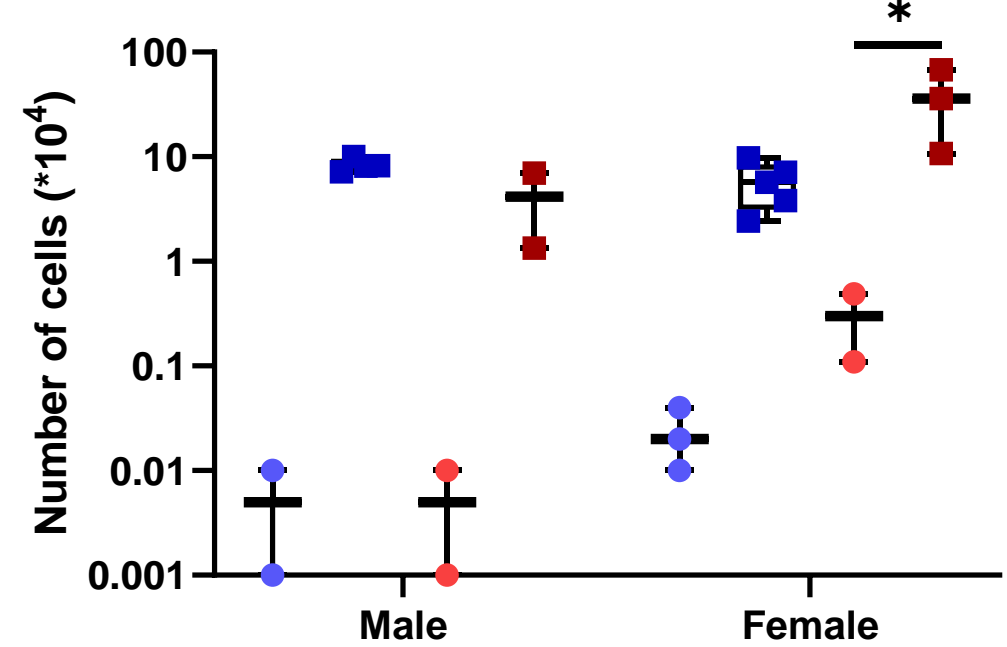
A cumulative composite score, ranging from 0 to 20
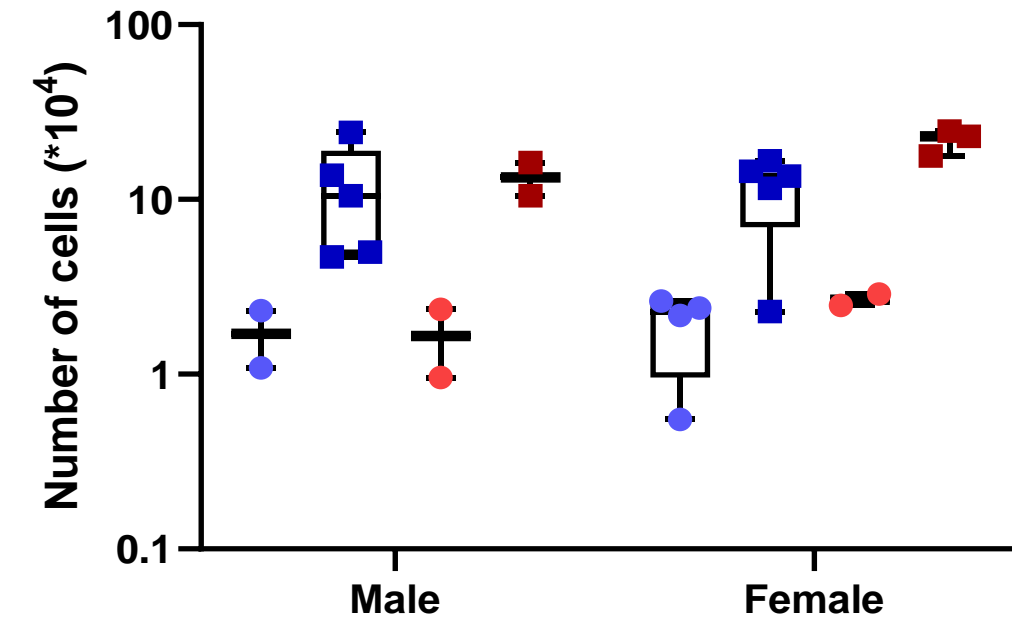
Histological analysis
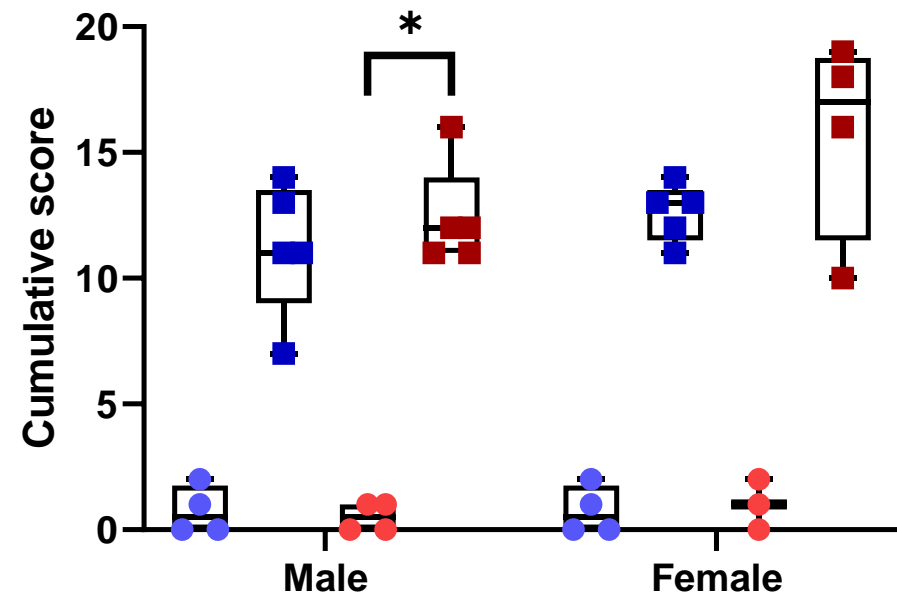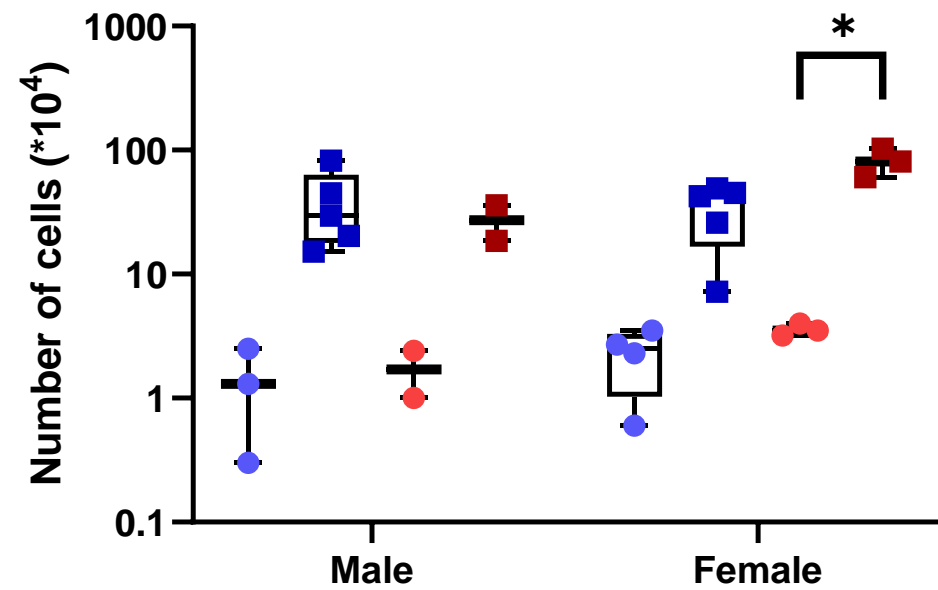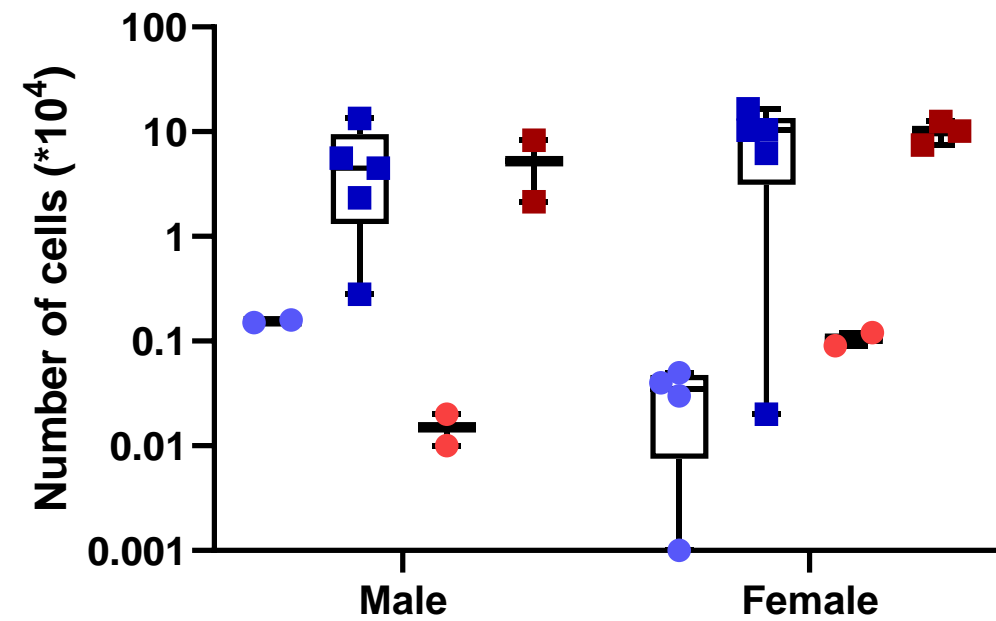
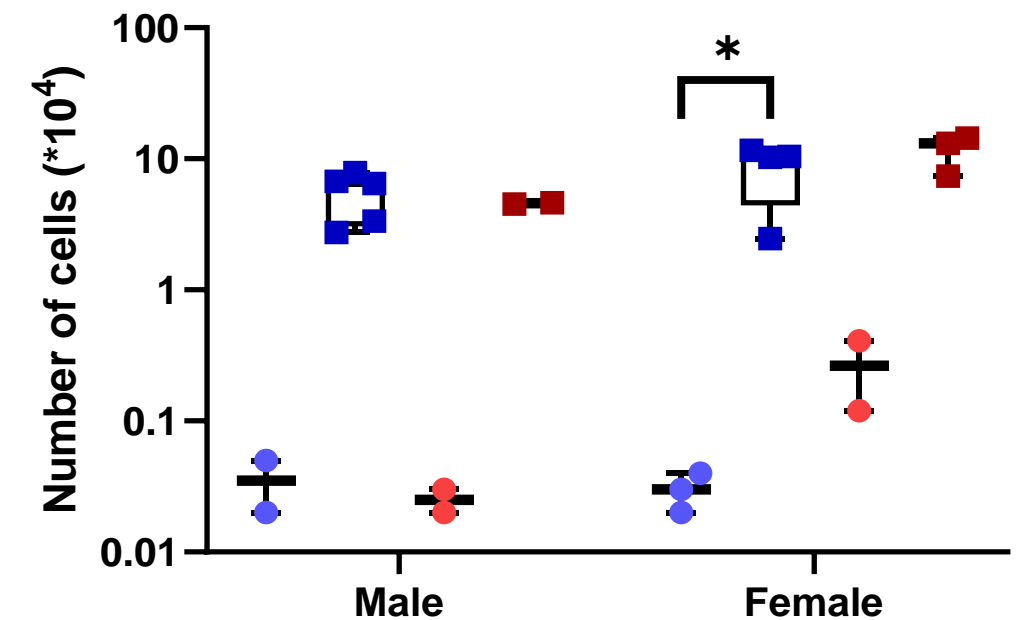Number of eosinophils cells in BAL

Number of Macrophages cells in BAL

Total number of cells in BAL

Number of neutophils cells in BAL

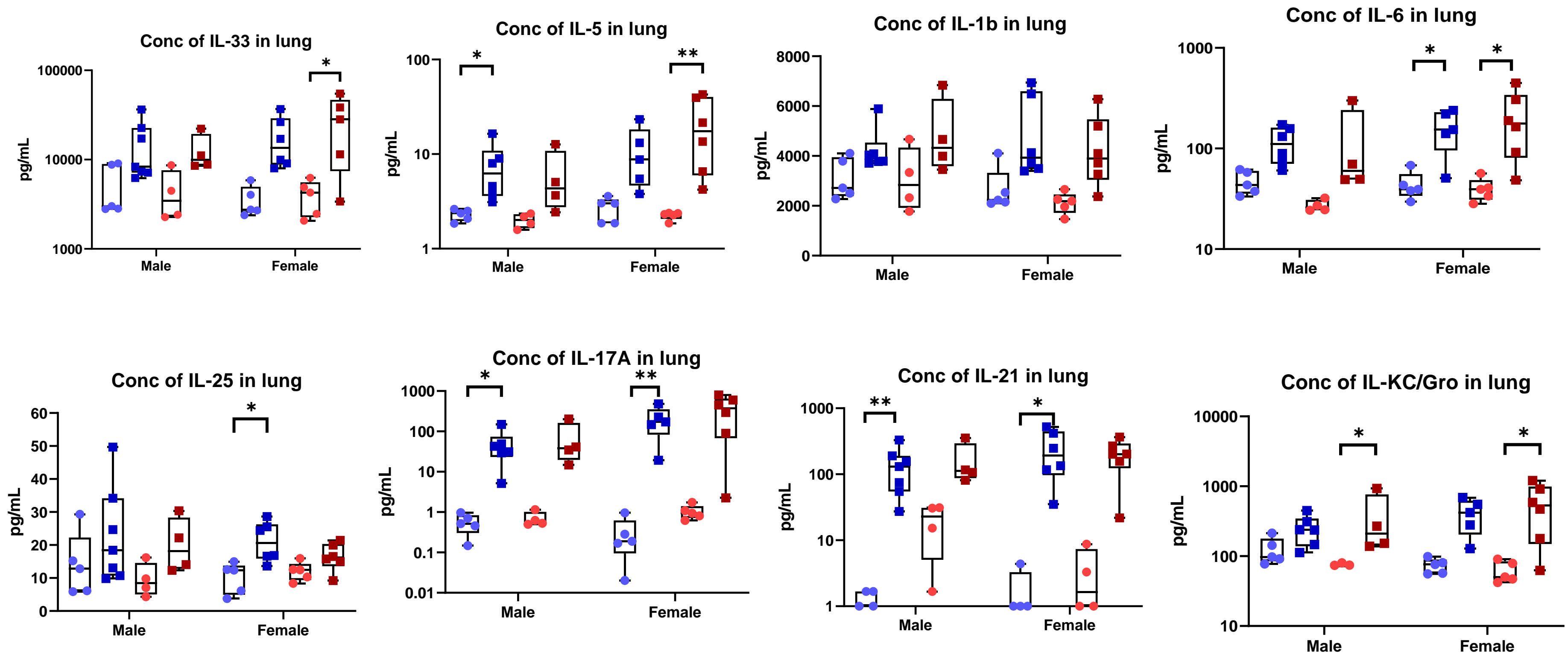Number of Lymphocytes cells in BAL

* P-Value < 0,05

Appendix - Cytokines quantification

Conc of IL-33 in lung

Conc of IL-5 in lung

Conc of IL-1b in lung

Conc of IL-6 in lung

Conc of IL-25 in lung

Conc of IL-17A in lung

Conc of IL-21 in lung

Conc of IL-KC/Gro in lung

* P-Value < 0,05