Oxidized ultrafine particles induce the activation of the inflammasome in human peripheral blood mononuclear cells obtained from chronic obstructive pulmonary disease (COPD) patients


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Risk Factors and Chronic inflammation in COPD

Indoor and outdoor pollution

Smoking

Lung Chronic Inflammation

COPD (CHRONIC OBSTRUCTIVE PULMONARY DISEASE)

Disruption of lung parenchyma
Lung dysfunction
Smoking and Air Pollution are HIGH RISK FACTORS for CHRONIC LUNG INFLAMMATION:

• what is the role of hematopoietic cells?

Groups of Subjects

• Healthy non-smokers
• Healthy smokers
• COPD patients

PBMCs

Centrifugation
Blood
Ficol-Hypaque PLUS
granulocytes, erythrocytes

Pro-Caspase-1

Active Caspase-1

Pro-IL-1β

Pro-IL-18
Smokers are more susceptible to soot-mediated inflammation

PBMCs from healthy non-smokers and smokers

Cigarette smoke

UFPs

Active Caspase-1

Pro-IL-1β → Pro-IL-18 → INFLAMMATION

..BUT LATENT INFLAMMATION!!!

Nanoparticles in a premixed flame

**NOC**
Nanoparticles of Organic Carbon
dimension 2-5 nm
staked PAHs containing sub-units
with aliphatic/aromatic bonds
H/C >= 0.5

**Soot**
primary particles and chain like agglomerates
dimension primary particles 20-40 nm
dimension chain-like agglomerates > 100 nm
elemental carbon structures
H/C ~ 0.2 - 0.3

**Pyrolysis Soot**
chain-like agglomerates
dimension chain-like agglomerates > 100 nm
graphitic carbon structures
H/C ~ 0.1 - 0.05
Nanoparticles in a premixed flame

Soot agglomerates

Soot primary

NOC

Fuel/Air mixture

residence time
Combustion-generated UFPs induce the activation of the inflammasome

UFP tested: **SOOT E/DMF** of 20-30 nm diameter; Area (50-100pg/ml)=1.1E-4 - 2.2E-4 cm²/ml

Cytokine release: IL-18 and IL-33 release

One Way Anova followed by Tukey’s multiple comparison test
Combustion-generated UFPs induce the activation of the inflammasome

UFP tested: NOC-E and NOC-ED of 2-5 nm diameter; Area (50-100pg/ml)=1.0E-3 – 2.0E-3 cm²/ml
Cytokine release: IL-18 and IL-33 release

One Way Anova followed by Tukey’s multiple comparison test
...BUT

Soot/ethylene-dimethylfuran agglomerates DID NOT induce the activation of the inflammasome in COPD-derived PBMCs

A

B
Combustion-generated ultrafine particles increased oxidative stress

Pro-inflammatory Stimulus

Macrophages

Mitochondria

NLRP3 Inflammasome

Pro-inflammatory Stimulus

Pro-inflflammatory Stimulus

NLRP3

ASC

ASC

Pro-caspase 1

Caspase 1

IL-1β

IL-18

Pro-IL-1β

mtROS

release

A

NLRP3 expression

% NLRP3+ CD14+ cells

p<0.05

Non smoker

Smoker

COPD

B

mtROS release

% Mitoxo+ cells

p<0.005

p<0.005

Non-smoker

Smokers

COPD
Combustion-generated ultrafine particles increased oxidative stress in COPD-derived PBMCs

Soot-E and NOC

Environmental exposures

ROS

Immune Response

DNA base lesions

Pro-inflammatory chemokines/cytokines

Nucleus

OGG1-BER

8-oxoG base

OGG1-8-oxoG

Cytoplasm

OGG1

Transacting factors

Signaling

RNA pol II

8-OH-dG ng/ml

A

8-OH-dG levels

Non-smokers
Smokers
COPD

p<0.001

p<0.0001

p<0.05

p<0.01

p<0.001

8-OH-dG levels

B

OGG-1 expression

Non-smokers
Smokers
COPD

p<0.05

p<0.01

Ratio OGG1/b-actin

COPD

p<0.01

p<0.001

8-OH-dG levels

Combustion-generated ultrafine particles increased oxidative stress in COPD-derived PBMCs

Soot-E and NOC

Environmental exposures

ROS

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DNA base lesions

Pro-inflammatory chemokines/cytokines

Nucleus

OGG1-BER

8-oxoG base

OGG1-8-oxoG

Cytoplasm

OGG1

Transacting factors

Signaling

RNA pol II

8-OH-dG ng/ml

A

8-OH-dG levels

Non-smokers
Smokers
COPD

p<0.001

p<0.0001

p<0.05

p<0.01

p<0.001

8-OH-dG levels

B

OGG-1 expression

Non-smokers
Smokers
COPD

p<0.05

p<0.01

Ratio OGG1/b-actin

COPD

p<0.01

p<0.001

8-OH-dG levels
Combustion-generated ultrafine particles induced IL-18 release in an inflammasome-independent manner.

**Soot-E**

- COPD
- UFPs
- NLRP3

**Active Caspase-1**

**Y-vad**

**Inflammasome inhibitor**

**NO INFLAMMATION**

**A**

- CTR
- Soot-E 50pg
- Soot-E 100pg
- Y-Vad
- Y-Vad+Soot-E 50 pg
- Y-vad+Soot-E 100pg

**B**

- CTR
- Soot-E 50pg
- Soot-E 100pg
- Gly
- Gly+Soot-E 50 pg
- Gly+Soot-E 100pg
Combustion-generated ultrafine particles induced IL-18 release in an inflammasome-independent manner

NOC-E and NOC-ED

COPD
UFPs

NLRP3

Active Caspase-1

IL-33

IL-1a

IL-18

Y-vad

Gly

Inflammasome inhibitor

A

B

CTR
NOC-E 50pg
NOC-E 100pg
Y-Vad
Y-Vad+NOC-E 50 pg
Y-vad+NOC-E 100pg

CTR
NOC-E 50pg
NOC-E 100pg
Gly
Gly+NOC-E 50 pg
Gly+NOC-E 100pg

IL-18 pg/mL

0
2000
4000
6000

0
2000
4000
6000

Inflammasome inhibitor

NO INFLAMMATION
UFPs
SOOT-E and NOC
LOW CONCENTRATIONS (50-100pg/ml)
Higher superficial Area (cm²/ml)

Immune System Activation

IL-18, IL-33

LUNG INJURY

Cell transformation:
LUNG CANCER???

UFPs
Soot-E Agglomerates
Lower superficial Area (cm²/ml)

SMOKERS:
De Falco et al., Scientific Reports 2016
BUT NOT IN COPD

??? Work in progress
THANKS FOR YOUR ATTENTION!