INNOVATIVE STUDY PROTOCOL TO SUBSTANTIATE THE ANTIPOLLUTION CLAIM
COMPONENTS OF AIR POLLUTION THAT AFFECT THE SKIN AND THEIR MECHANISM OF ACTION

- INDUSTRIES
- VEHICLES EXHAUSTS
- CIGARETTES

- Nano Particle
- HEAVY METALS
- PM$_{2.5}$
- PM$_{10}$
- PAH HAH
- SO$_2$ NO$_2$
- O$_3$
Why the anti-pollution Claim?

PM2.5: WHO Limit: 25µg/m³

OZONE: WHO Limit: 100µg/m³

According to Dr. E Tanzi (Clinical Prof of Derm at the George Washington University Medical Centre): UV is the first skin precursor followed closely by Pollution.
Mechanism of action of pollutants

Air pollution and skin diseases: Adverse effects of airborne particulate matter on various skin diseases.
Kim KE et al., life sciences 2016

Effect of pollution on the skin

- Ageing of the skin
- Increase in the number of age spots
- Hyperpigmentation
- Increase in skin inflammation
- Roughen the skin (less smooth)
In vitro and ex vivo assays to evaluate the effects of pollution on the skin


**in vitro and ex vivo** assays to evaluate the effects of Pollutants on the skin

Monitoring the efficacy of test items for the anti-pollution claim

**Antioxidant capacity: oxidative stress (Reactive Oxygen Species (ROS))**

Study system: monolayer cell cultures or skin explants
Inducer: Cigarette smoke, ozone, infrared, UVR, blue light

**Anti-Inflammatory potential (IL-6)**

Study system: monolayer cell cultures or skin explants
Inducer: Cigarette smoke, ozone, infrared, UVR, blue light

**Anti-Aging potential (MMP-1)**

Study system: monolayer cell cultures or skin explants
Inducer: Cigarette smoke, ozone, infrared, UVR, blue light

**Collagen integrity (total-P1NP)**

Study system: monolayer cell cultures or skin explants
Inducer: Cigarette smoke, ozone, infrared, UVR, blue light

**Elastin integrity**

Study system: monolayer cell cultures or skin explants
Inducer: Cigarette smoke, ozone, infrared, UVR, blue light
In vivo tests to evaluate the effects of pollution on the skin
Evaluation of the anti-pollution effects: Laboratory exposure
Evaluation of the anti-pollution effects: Laboratory exposure

Options:
1. Ozone
2. Ambient Dust (NIST 1649b)
3. Ozone + Ambient Dust
4. Ozone+ UV (or Blue Light)
5. Ambient Dust+UV (or Blue Light)
6. Ozone+ Ambient Dust + UV (or Blue Light)

More than 18 protocols exists
1. Evaluation of the protective effect over 1 day
2. Evaluation of the protective effect over 5 days
3. Evaluation of the protective and repairing effects (5+5 days)
4. We can tests a Maximum of 3 products simultaneously for protective effect and a maximum of 2 product for protective +repairing
Evaluation of the anti-pollution effects: Laboratory exposure

- Treated versus Non Treated (Placebo or Reference)
- Exposure on the upper back
- Single or Multiple Exposure Protocol
- Sampling by Swabbing and Tape Stripping

Parameters to be analysed:
- Squalene (LC/MS)
- MDA (By GC/MS)
- Carbonylated Proteins (Confocal Microscopy)
• Test to be carried out in New Delhi or Brazil

• Split Face Design

• Evaluation at D0-D28-D56

• Parameters Evaluated:
  - Biochemical Analysis (Squalene, Carbonylated Proteins, MDA...)
  - Biophysical Analysis
    - Skin Elasticity
    - Skin Hydration
    - Skin Barrier Function
    - Skin Pigmentation, and Homogeneity
    - Skin Micro-relief

- Self Evaluation Question
Evaluation of the anti-pollution effects: *in vivo* test

Help reduce the detrimental impact of pollutant and regenerates the skin

Cosmetic acting as a Barrier function

Cosmetic removing pollutants from the skin
Evaluation of the anti-pollution effects: Barriers & Cleansing

- Treated versus non treated (or vehicle/comparator)
- Exposition to pollutant
- Cleaning of investigational Zone
- High Resolution Photography followed by Image Analysis for the quantification of Particule on the skin

www.cidp-cro.com
Evaluation of the anti-pollution effect: HAIR

<table>
<thead>
<tr>
<th>Condition</th>
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<tbody>
<tr>
<td>UNPATTERNED HAIR LOSS</td>
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<tr>
<td>DRYNESS OF SCALP</td>
</tr>
<tr>
<td>SCALP IRRITATION</td>
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<tr>
<td>DANDRUFF</td>
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<tr>
<td>EXCESSIVE SEBUM SECRETION</td>
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<tr>
<td>PREMATURE GRAY HAIR</td>
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<tr>
<td>DRY FRIZZY HAIR</td>
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Mechanism of action of pollutants on hair

- Pollutants
- ROS
- Inflammation
  - Hair decoloration
  - Melanin oxidation
  - Hair peroxidation
  - Tryptophan degradation

Hair follicle
Hair shaft

- Pollutants
- ROS

HAIR FOLLICLE
HAIR SHAFT

- Inflammation
- Melanin oxidation
- Hair peroxidation
- Tryptophan degradation
**ex vivo** assays to evaluate the effect of Ozone on the hair: oxidation

**Effect of pollution on hair follicles**

**Antioxidant capacity: oxidative stress (Reactive Oxygen Species (ROS))**

Study system: hair follicle  
Inducer: Cigarette smoke, ozone, infrared, UVR, blue light

**Anti-Inflammatory potential (IL-6)**

Study system: hair follicle  
Inducer: Cigarette smoke, ozone, infrared, UVR, blue light

**Effect of pollution on hair shaft**

**Melanin Content Assay**

Study system: tresses of natural hair  
Inducer: Cigarette smoke, ozone, infrared, UVR, blue light

**Lipid Peroxidation**

Study system: tresses of natural hair  
Inducer: Cigarette smoke, ozone, infrared, UVR, blue light

**Tryptophan degradation**

Study system: tresses of natural hair  
Inducer: Cigarette smoke, ozone, infrared, UVR, blue light
What is our proposal?

- in vitro Evaluation

- Short Term *in vivo* Tests with the Biochemical Evaluation of Biomarkers

- Long Term In use test with Biochemical and Biophysical Evaluation to Objectively Substantiate the “Anti-pollution” Claim
THANK YOU

QUESTION

A STRONG FOOTHOLD ON EACH CONTINENT