

# CAN NON-ASBESTIFORM AMPHIBOLE FIBERS TRIGGER CARCINOGENESIS MECHANISMS?

Militello G.M.<sup>1</sup>, Gaggero L.<sup>1</sup>, Sanguineti E.<sup>1</sup>, Yus González A.<sup>1</sup> & La Maestra S.<sup>2</sup>

<sup>1</sup>Department of Earth, Environment and Life Sciences, University of Genoa, Italy

<sup>2</sup>Department of Health Sciences, University of Genoa, Italy





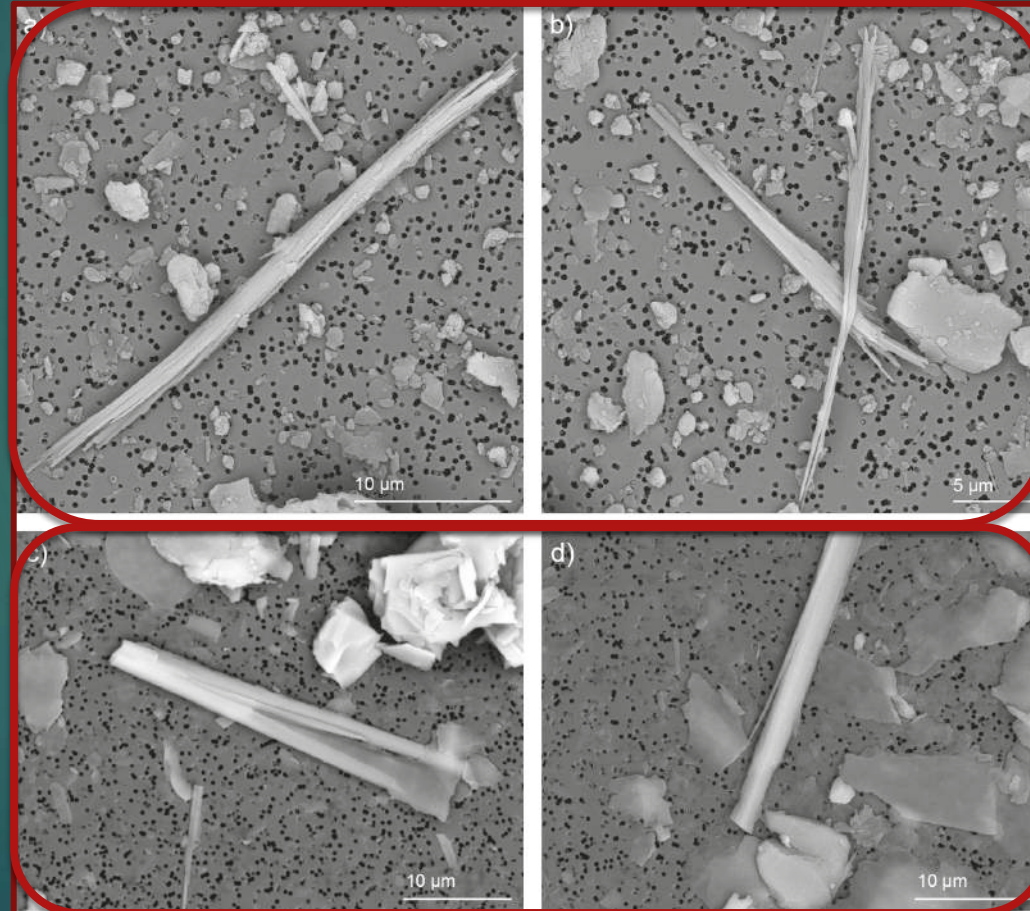
## AMPHIBOLES

World Health Organization (WHO)

Length (L) >5  $\mu\text{m}$ ; Width (W) <3  $\mu\text{m}$ ; L/W >3:1

Asbestiform  
and fibrous  
habit

Non-asbestiform amphiboles  
and serpentine minerals can  
split along **CLEAVAGE  
PLANES** to produce elongate  
particles with aspect ratio >3:1  
(and length > 5  $\mu\text{m}$ )



Acicular amphiboles and  
cleavage fragments (that  
meet the same criteria) are  
**NOT UNIVOCALLY  
REGULATED** as asbestos

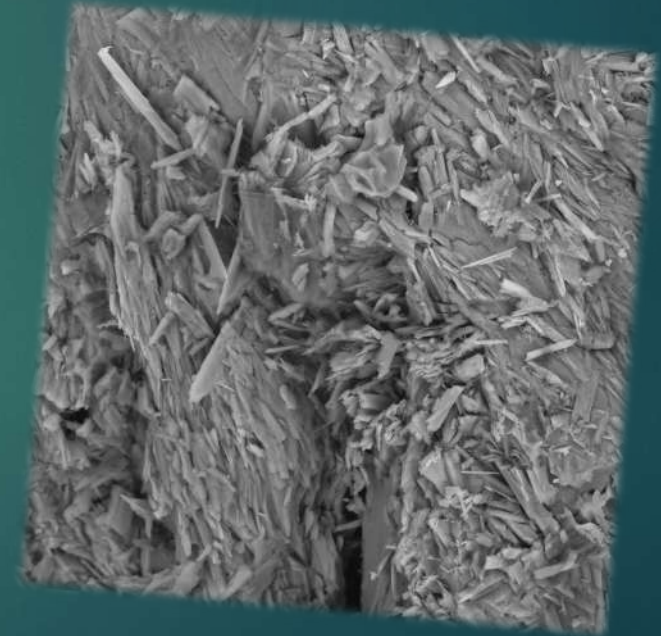
Non-asbestiform  
and fibrous habit

## BIOASSAY: FACTORS CONDITIONING PATHOGENICITY

Ultrafine asbestos fibers, especially amphibole fibers, can overcome the pleural lung barrier...

- Breathability or bioavailability
- Biodynamic behavior
- Bioresistance
- **DIMENSIONS (FIBROUS SHAPE)**
- Fiber surface reactivity

**WHAT ABOUT NON-ASBESTIFORM AMPHIBOLES?**





## STARTING MATERIALS FOR BIOASSAYS

### Sample with asbestiform amphiboles

**F3** Amphibole vein in serpentinite  
(Northern Apennines)

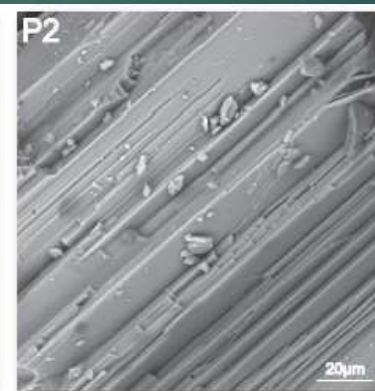


### Samples with Non-asbestiform amphiboles

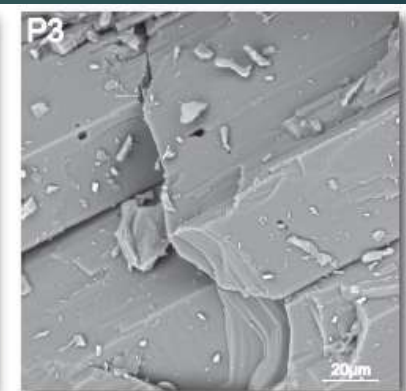
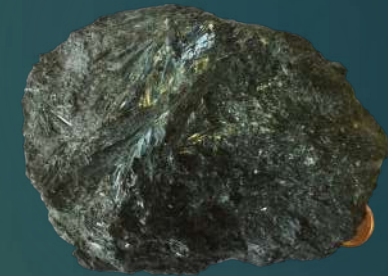
**P1** Metagabbro  
(Northern Apennines)



**P2** Dolomites  
(Swiss Alps)

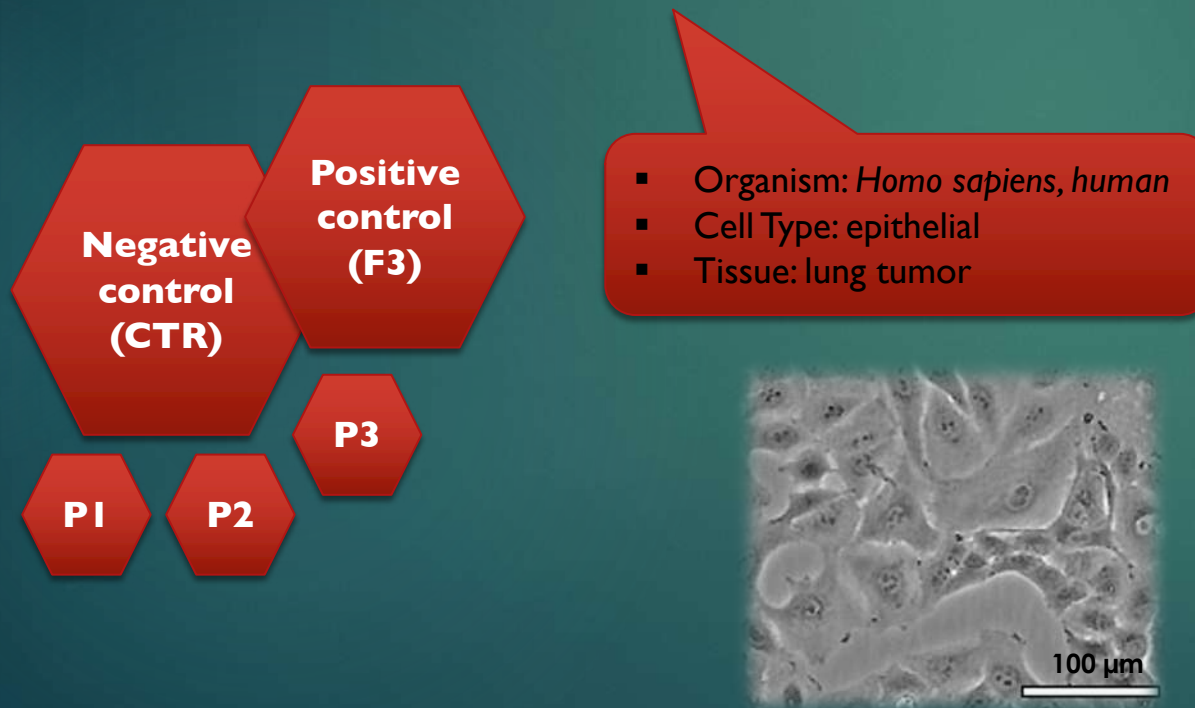


**P3** Amphibolite  
(Ligurian Alps)



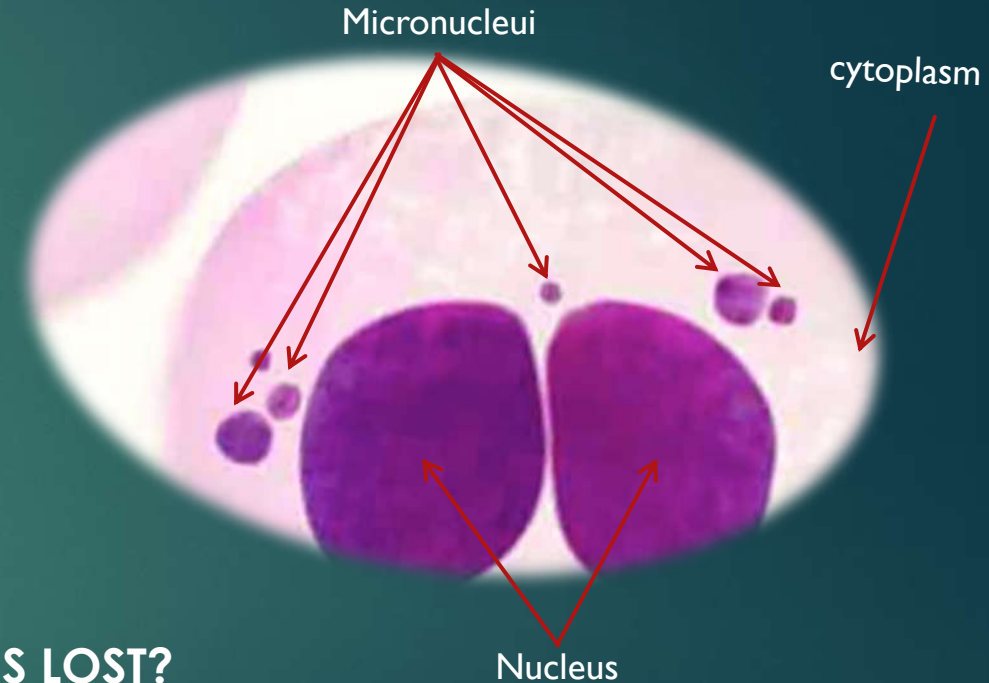
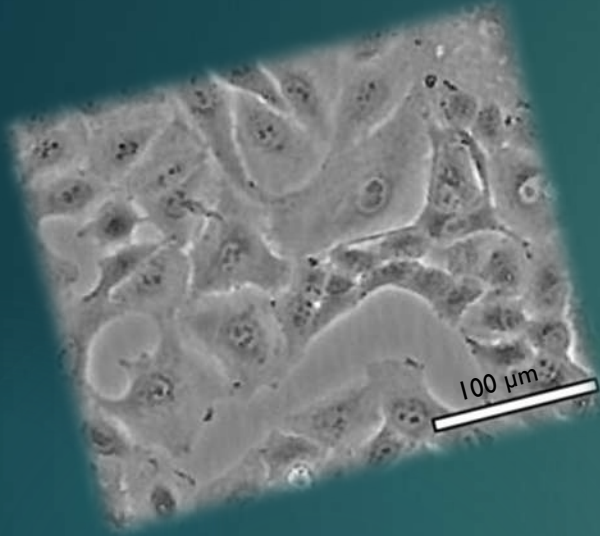
## IN VITRO TESTS

- ① Grinding and sieving of samples;
- ② Five separate well of A549 cells were seeded and treated after 24 h.
- ③ A concentration of  $15.62 \mu\text{g}/\text{cm}^2$  of each sample was inoculated into the cultured wells and allowed to interact for 24 h;
- ④ Evaluation of the frequency micronucleated cells.



## WHAT IS A MICRONUCLEUS?

Loss of genetic material (Chromosomal aberrations) due to both mechanical action and oxidative stress induced by fiber exposure.

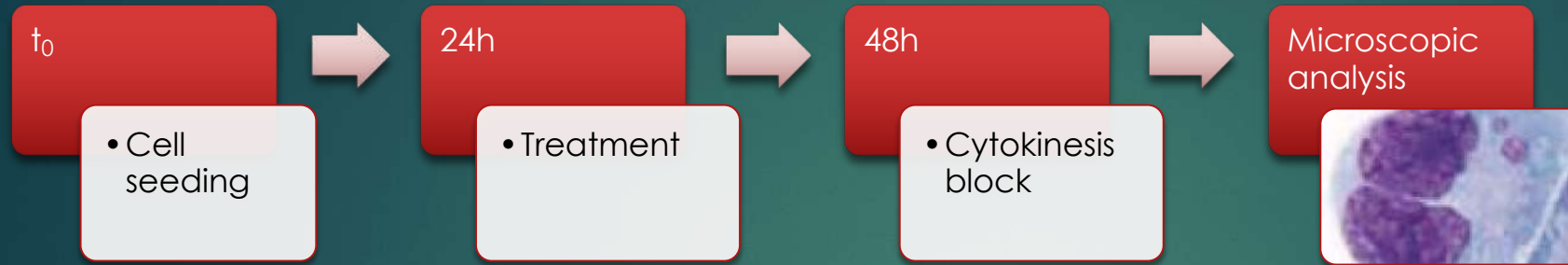


## WHAT HAPPENS WHEN GENETIC MATERIAL IS LOST?

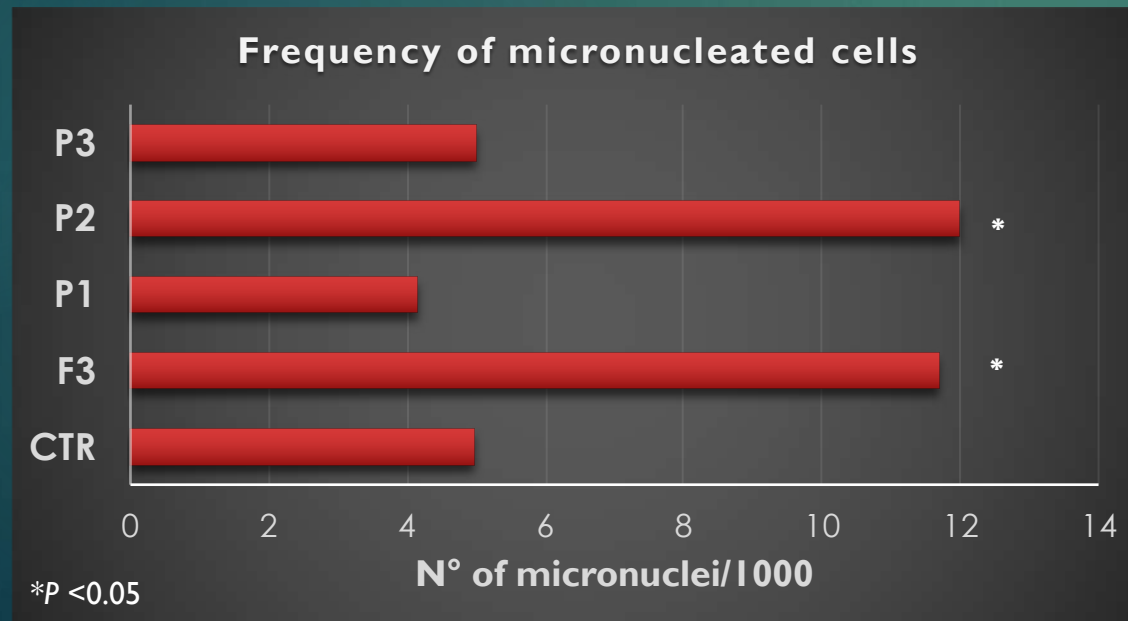
Death (**NECROSIS** or  
**APOPTOSIS**)

Cancer cells (**Process of  
Carcinogenesis**)

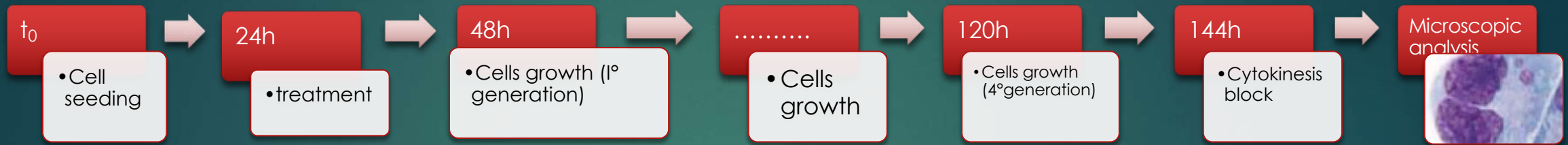




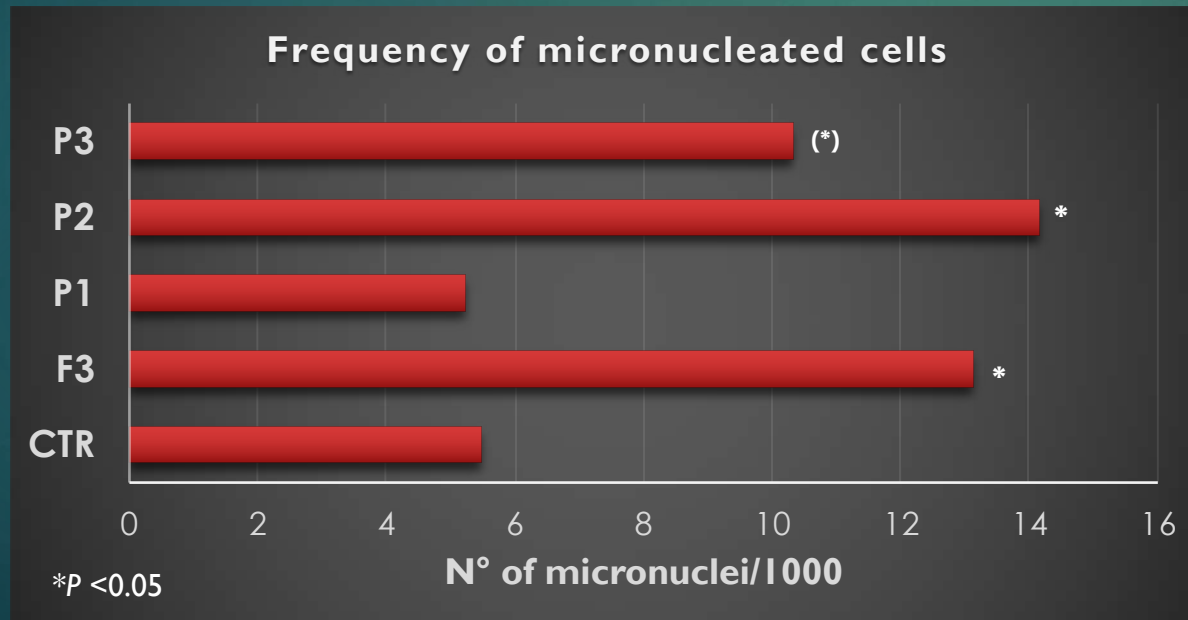
**I° GENERATION:** Chromosomal aberrations in A549 cells treated with  $15.62 \mu\text{g}/\text{cm}^2$  of each sample for 24h



$$\frac{\text{N° of micronuclei}}{[\text{monocellular cells} + (\text{binucleate cells} \times 2)]} \times 1000$$



#### 4<sup>o</sup> GENERATION: Long-term effect on DNA after 96h treatment



$$\frac{\text{N° of micronuclei}}{[\text{monocellular cells} + (\text{binucleate cells} \times 2)]} \times 1000$$



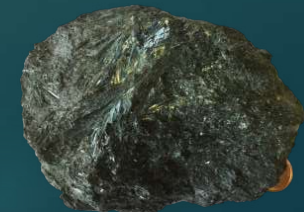
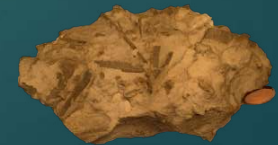
## FIRT RESULT ON NON-ASBESTIFORM AMPHIBOLES TOXICITY

Preliminary results have shown an increase of chromosomal aberrations that can also be trigger by  
exposition to NON-ASBESTIFORM AMPHIBOLES

| A549 Cells                |             |                                 |             |                                 |
|---------------------------|-------------|---------------------------------|-------------|---------------------------------|
| Samples                   | Micronuclei | Cytotoxicity<br>(1° generation) | Micronuclei | Cytotoxicity<br>(4° generation) |
| Negative control<br>(CTR) | 6           |                                 | 7           |                                 |
| Positive control<br>(F3)  | 16          | *                               | 20          | *                               |
| P1                        | 5           | -                               | 7           | -                               |
| P2                        | 14          | -                               | 20          | *                               |
| P3                        | 6           | -                               | 16          | (*)                             |

### WORK IN PROGRESS

transformation efficiency  
in no-tumoral cells is  
presently investigated



**THANK FOR  
YOUR KIND  
ATTENTION!**