

# Trans-generational effects of nano-TiO<sub>2</sub> with different surface properties on basil (*Ocimum basilicum*)

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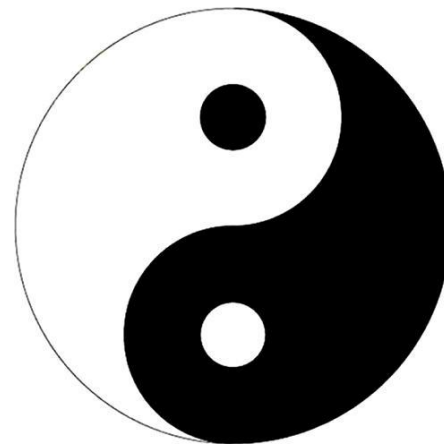


# Background – Taiji, nanotechnology, and plants

❖ **Properties:** Size, surface functionalization, surface area, band gap, electroconductivity, antibacterial, and among others.

❖ **Application:** Pigment, food additive, herbicide, skin care, glass, fertilizer, water treatment, medicine care, and among others.

(Nel. et al., 2013)



*Everything begins with two poles, the white is yang, and the black is yin.*

*----- I Ching*

❖ The fate, speciation, and translocation of nanomaterials (Gardea-Torresdey et al., 2014)

❖ Understanding of molecular and biochemical responses of plants to NMs stress (Ma et al., 2015)

❖ Long-term impacts in plant system (Servin and White, 2016)

# Background – Taiji of TiO<sub>2</sub> Nanoparticles

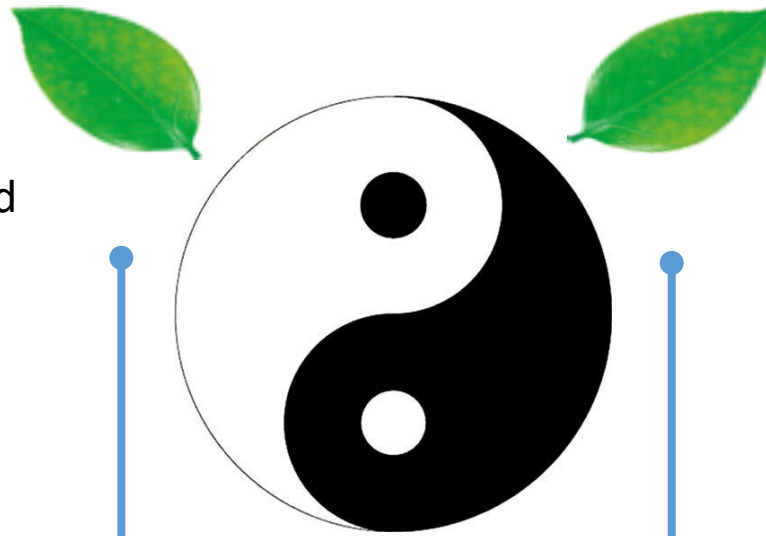
## Manufactured products

- ❖ **Food additive:** E171 (extracted from a natural white mineral)
- ❖ **Pigment:** P25 (mixture of anatase and rutile)
- ❖ **Skin care:** M212, M262 (rutile, with coatings)
- ❖ **Photocatalyst:** Fluka, A.R. (rutile)
- ❖ **Plastic, painting:** TR28, TR92, R1530 (rutile, with coatings)

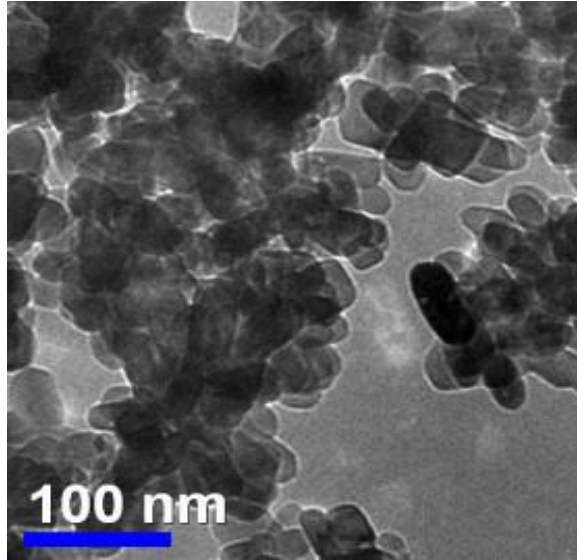
**Multi-generational effects**

- ❖ **Cucumber:** root-to-fruit translocation (Servin et al., 2013)
- ❖ **Lettuce:** paint-aged *n*-TiO<sub>2</sub>, shoot-to-root translocation (Larue et al., 2014)
- ❖ **Onion:** decrease mitotic index and increase chromosomal aberrations (Pakrashi et al., 2014)
- ❖ **Flax and Fennel:** enhance germination rate and root length (Clément et al., 2013)
- ❖ **Spinach:** increase enzymatic activities (Hong et al., 2005)

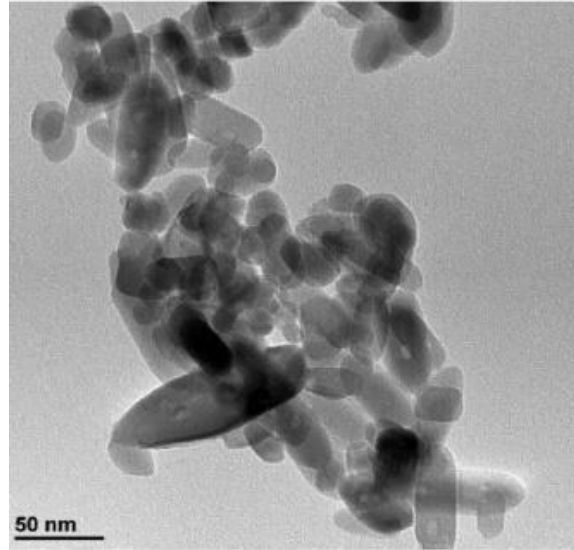
**The role of surface coating**



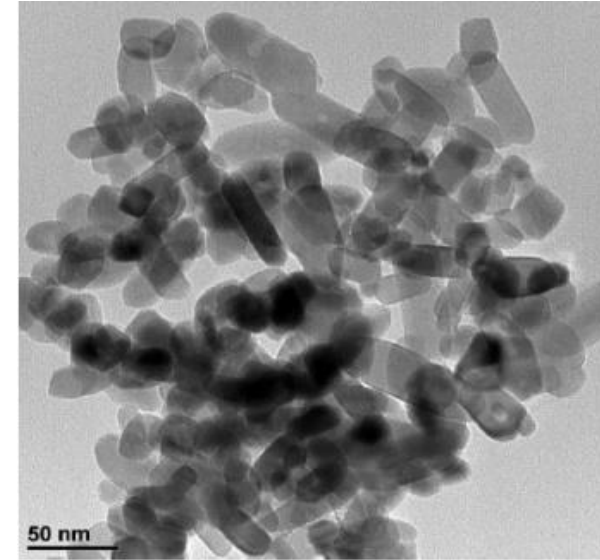
# Methodology – Characterization of nano-TiO<sub>2</sub>



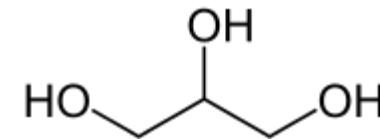
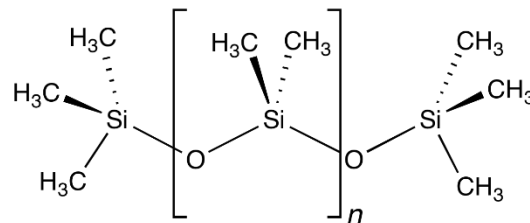
**Pristine: plain  
rutile nano-TiO<sub>2</sub>**



**Hydrophobic: M262,  
capped with Al<sub>2</sub>O<sub>3</sub> and  
encapsulated with  
dimethicone**



**Hydrophilic: M212,  
coated with Al<sub>2</sub>O<sub>3</sub> and  
encapsulated with  
glycerol**



# Methodology – Characterization of nano-TiO<sub>2</sub>

Properties	Nanoparticles		
	Pristine TiO <sub>2</sub>	Hydrophobic TiO <sub>2</sub>	Hydrophilic TiO <sub>2</sub>
Size (nm)	25-70	25-70	25-70
Crystal phase	Tetragonal, rutile	Tetragonal, rutile	Tetragonal, rutile
Surface area (m <sup>2</sup> /g)	20-40	47.6	55.7
Hydrodynamic size (nm)	341 ± 10	261 ± 5	282 ± 7
Zeta potential in DI water (mV)	-14.7 ± 0.5	27.0 ± 0.9	26.9 ± 0.5

Anatase and rutile mixture, P25  
(Previous studies)

**Rutile, preferential  
translocation  
in cucumber  
(Servin et al., 2012)**





# Background – Basil (*Ocimum basilicum*)



<http://www.precisionnutrition.com/wordpress/wp-content/uploads/2009/11/basil-bsp.jpg>



[https://en.wikipedia.org/wiki/Pesto#/media/File:Pasta\\_with\\_pesto.jpg](https://en.wikipedia.org/wiki/Pesto#/media/File:Pasta_with_pesto.jpg)



<http://www.bing.com/images/search?q=Basil&view=detailv2&id=8B54A65E2B0E7DD90372F5DF8E8D94958FF06F30&selectedIndex=7&ccid=D%2bvsykc&simid=608031507288426172&thid=OIP.M0febecc7291cf254480adfcfe94c044ao0&ajaxhist=0>



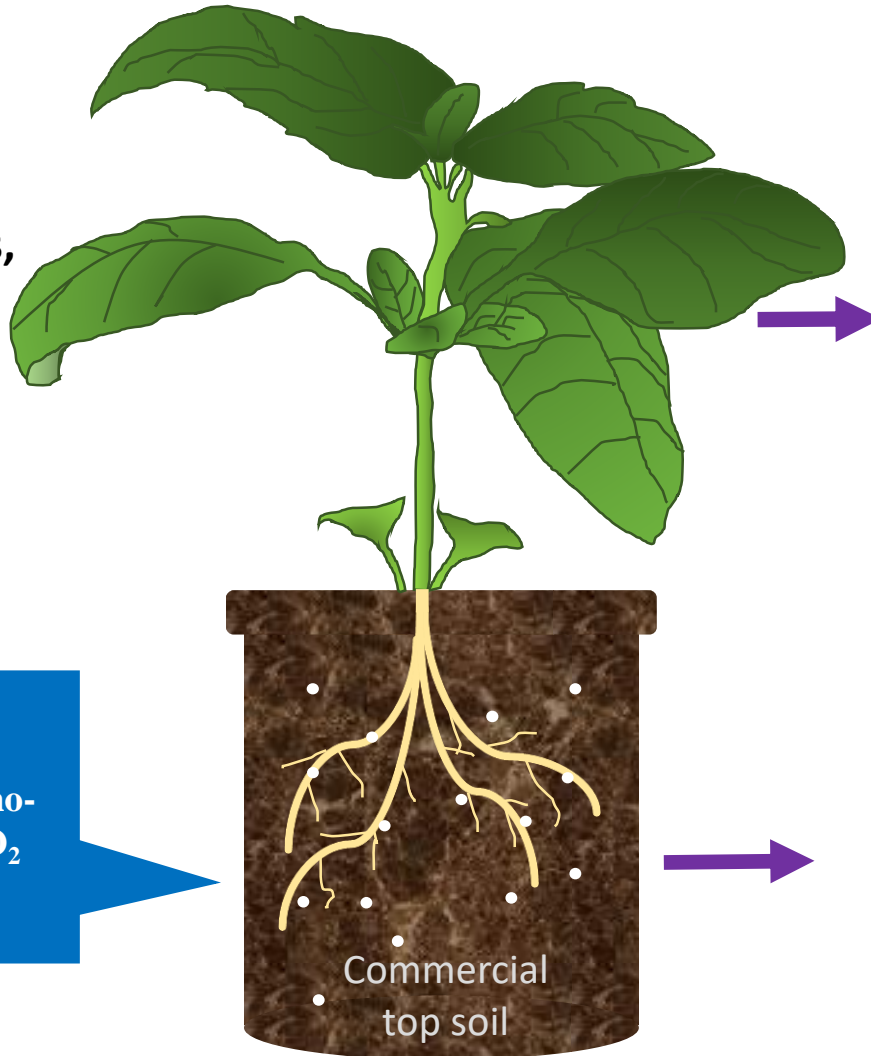
[https://en.wikipedia.org/wiki/Pho#/media/File:Pho\\_in\\_Saigon.jpg](https://en.wikipedia.org/wiki/Pho#/media/File:Pho_in_Saigon.jpg)

- 🌿 **A culinary herb prominently featured in European and Asian cuisine. → Fresh**
- 🌿 **Essential oil, medicinal value.**
- 🌿 **Shoots, roots and seeds.**



# Previous project

Harvested at 65 days,  
flowering time

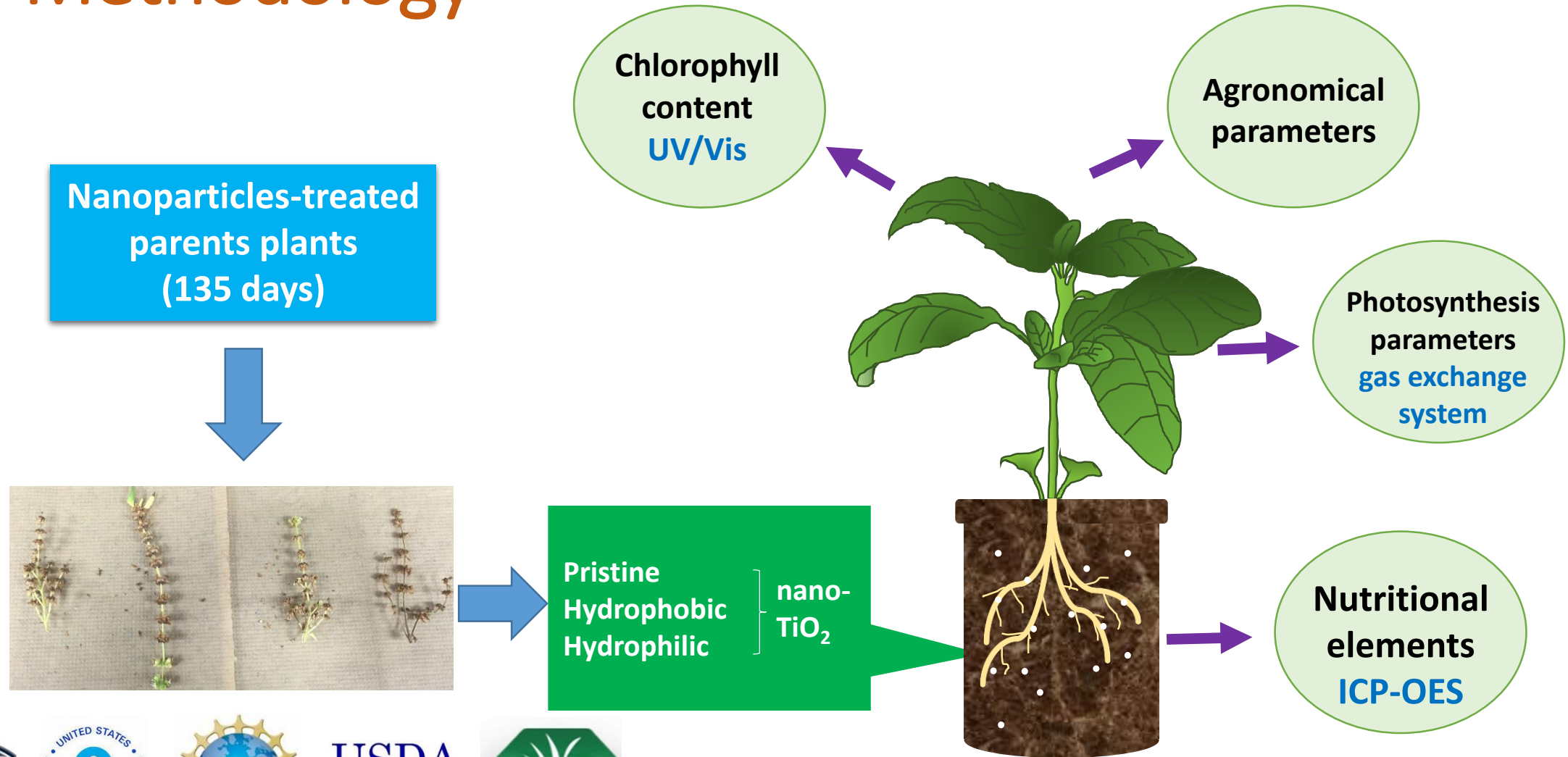


750 mg·kg <sup>-1</sup>		
Physiological and biochemical effects		
Pristine	↓	Biomass
		Reducing sugar
		CAT, APOX
Hydrophobic	↓	Germination rate
		Biomass
		Root length
		Starch
		CAT, APOX
Hydrophilic	↓	Germination rate
		Total sugar
		APOX

Ti accumulation in root (mg·kg <sup>-1</sup> )	
Control	15.3 ± 5.0 c
Pristine	86.1 ± 9.4 b
<b>Hydrophobic</b>	<b>160.4 ± 13.2 a</b>
Hydrophilic	113.9 ± 14.9 b

W. Tan et al.,  
Environmental Pollution,  
submitted.

# Methodology





# Experimental design

Type of NPs	Name	Seeds from parent plants (without, -; with +)	Same TiO <sub>2</sub> nanoparticles at 750 mg·kg <sup>-1</sup> (without, -; with +)
	Control-0	-	-
Pristine Hydrophobic Hydrophilic nano-TiO <sub>2</sub>	Control-750	-	+
	Treated-0	+	-
	Treated-750	+	+

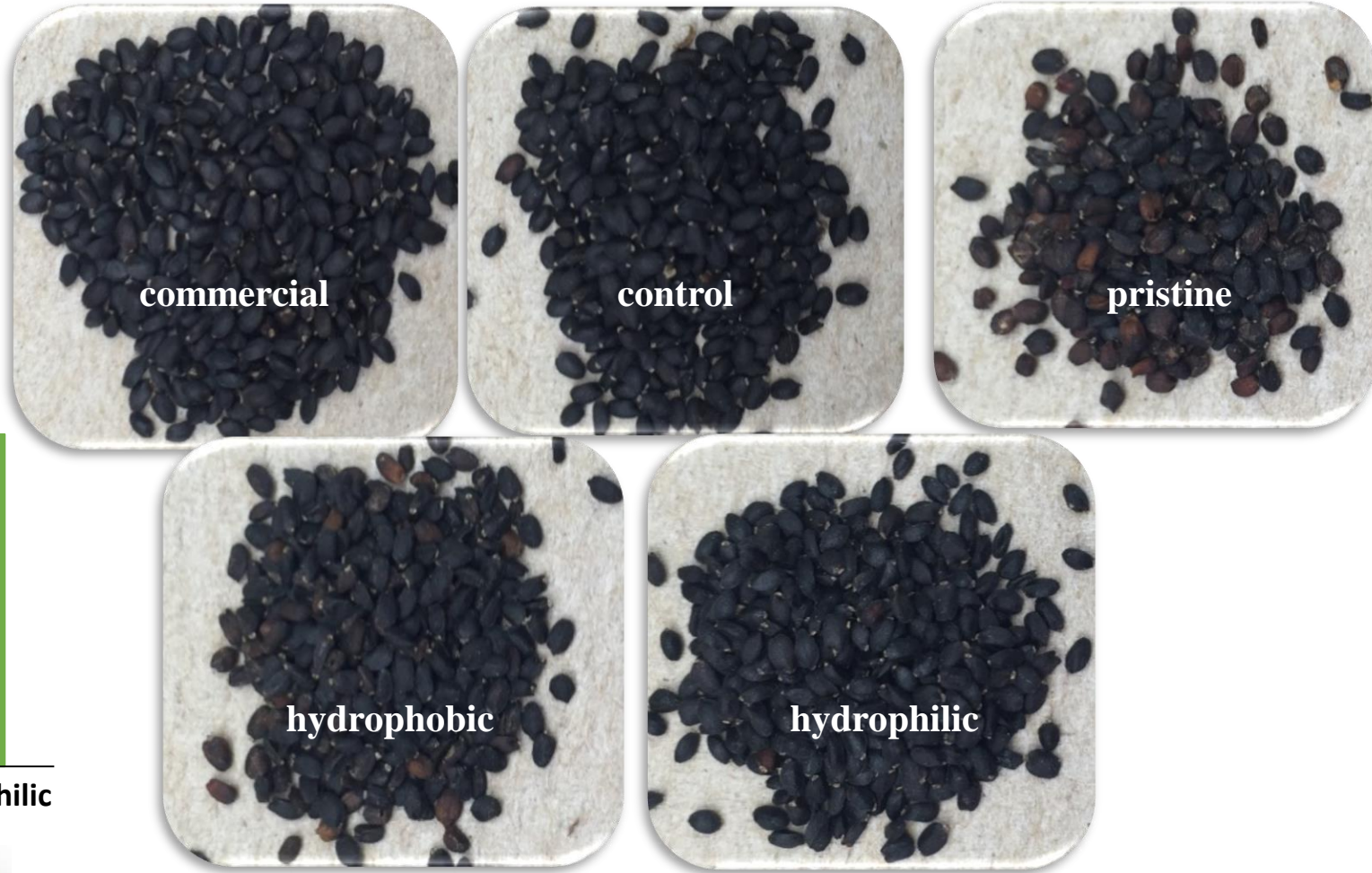
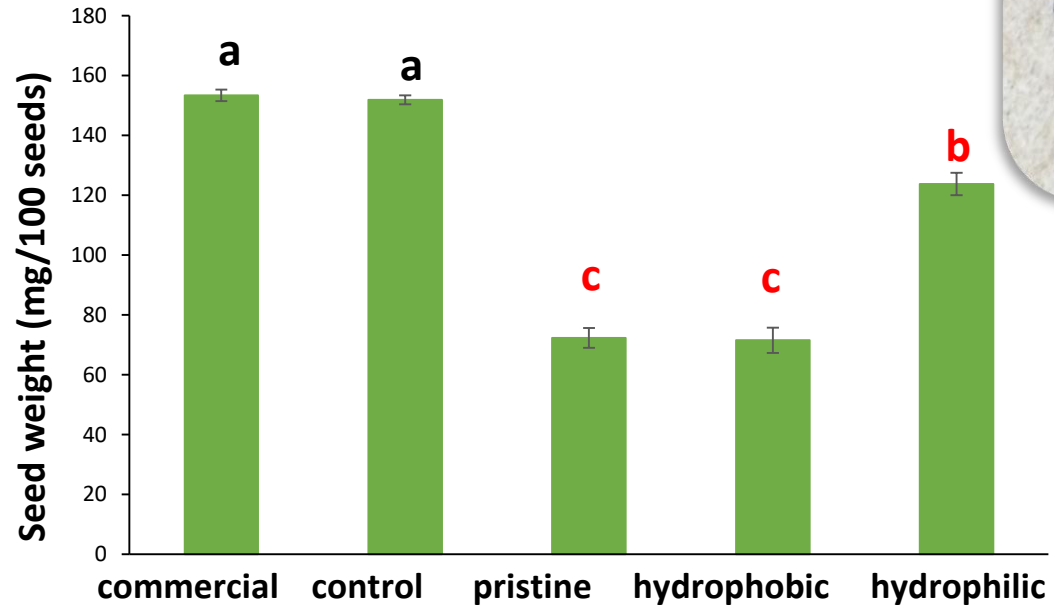
Effects caused  
Behaviors of second generation seeds

Interaction of treated seeds and nano-TiO<sub>2</sub>



# Results - Seed production

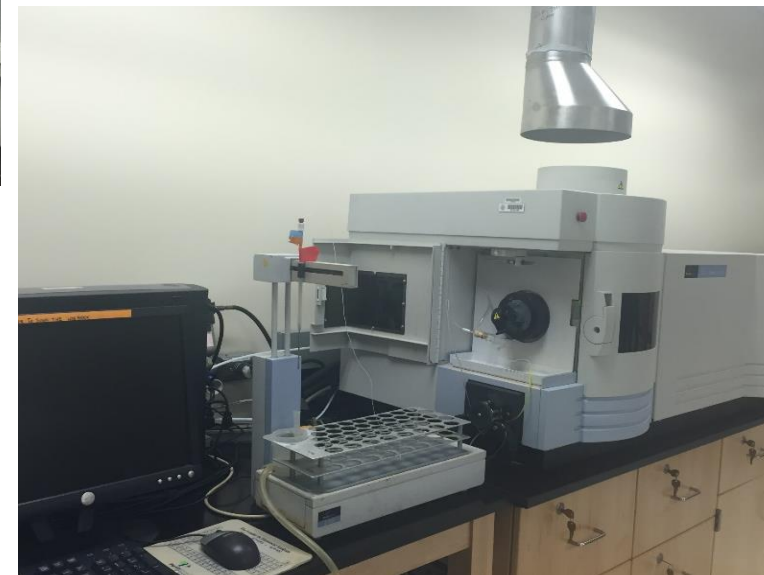
At 135 days, harvested seeds from control, or treated with pristine, hydrophobic, and hydrophilic nano-TiO<sub>2</sub> at 750 mg·kg<sup>-1</sup>



# Results - Nutritional elements



Dry tissue sample



2 ml HNO<sub>3</sub>,  
1 ml H<sub>2</sub>O<sub>2</sub>,  
5 ml H<sub>2</sub>SO<sub>4</sub>

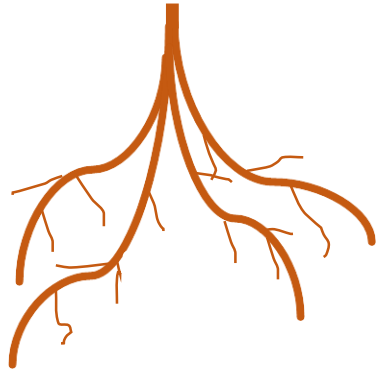
C. Larue et al., J Hazard Mater  
2014, 273, 17-26



ICP-OES

<b>Macro-elements</b>
Al, Ca, Mg, K, S, P
<b>Micro-elements</b>
Fe, Se, Zn, Cu, Mn, B, Mo, Ni, Co

# Results - Nutritional elements



Tissue	Treatment	Element
Pristine	Control-750	Zn
	Treated-0	Zn
	Treated-750	Mg, Zn
Hydrophobic	Control-750	Al, Co, Fe
	Treated-0	--
	Treated-750	--



Tissue	Treatment	Element
Pristine	Control-750	Ni
	Treated-0	--
	Treated-750	Fe
Hydrophobic	Control-750	--
	Treated-0	--
	Treated-750	Mg



# Results – Agronomical parameters

No significant difference in germinated seeds number, and root length

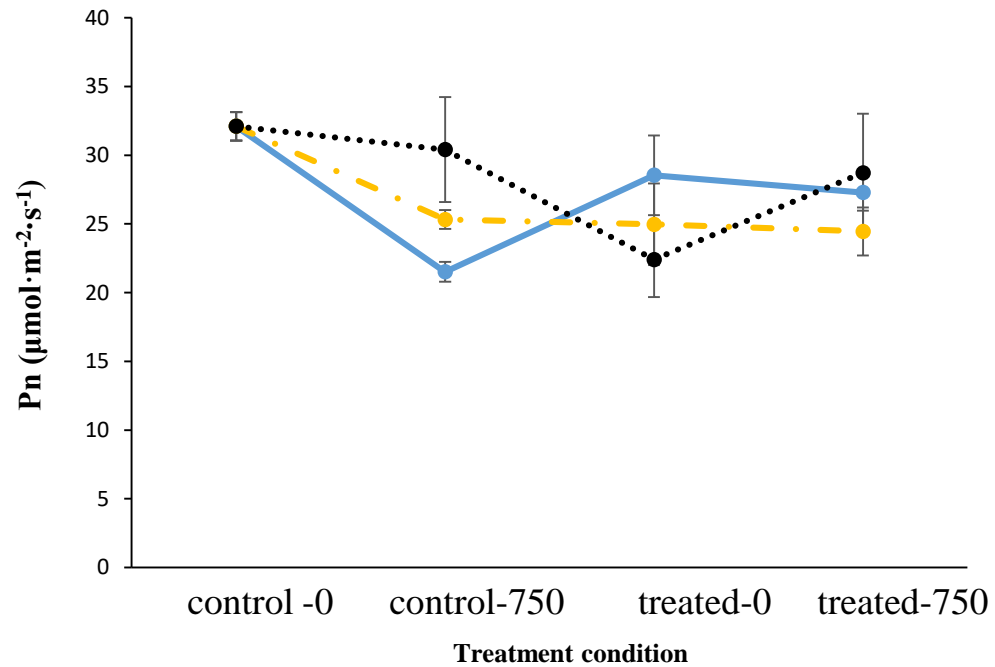
	Treatment	Pristine	Hydrophobic	Hydrophilic
Shoot length (cm)	Control-0	35.5 ± 2.2	35.5 ± 2.2 b	35.5 ± 2.2
	Control-750	38.0 ± 1.9	35.5 ± 1.5 b	37.1 ± 4.0
	Treated-0	39.4 ± 2.3	43.0 ± 1.6 a	35.6 ± 1.7
	Treated-750	39.3 ± 0.4	42.2 ± 0.6 a	35.5 ± 1.3
Biomass (%)	Control-0	9.5 ± 0.8	9.5 ± 0.8 b	9.5 ± 0.8
	Control-750	11.0 ± 1.5	13.4 ± 1.6 ab	12.0 ± 1.5
	Treated-0	10.7 ± 1.0	13.2 ± 1.3 ab	12.3 ± 0.6
	Treated-750	9.9 ± 1.0	13.8 ± 0.5 a	12.1 ± 1.5



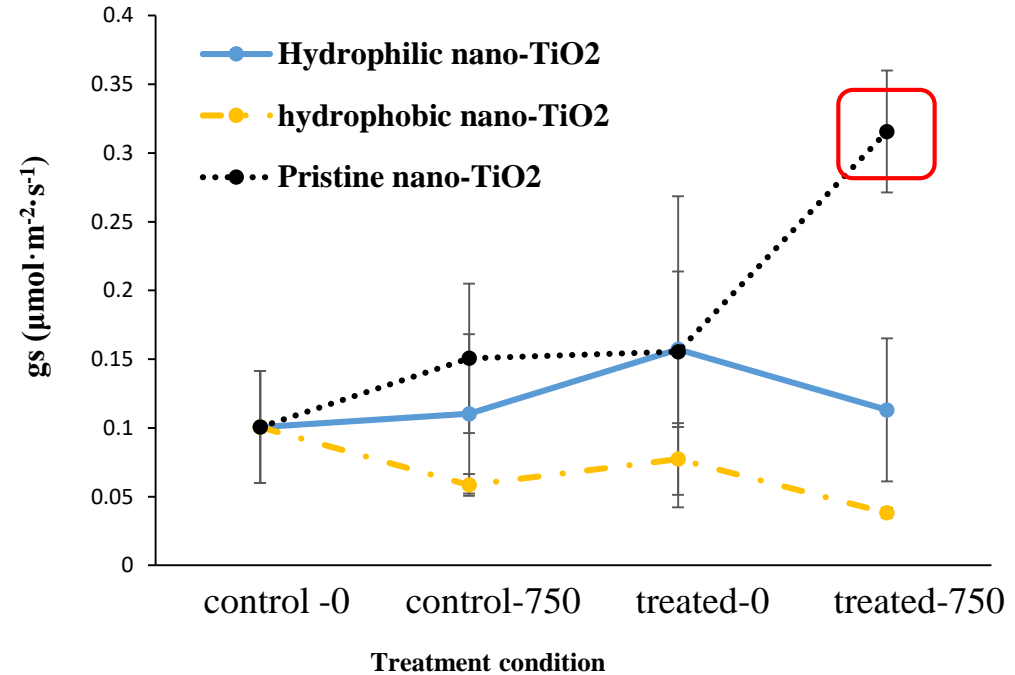


# Results - Photosynthetic parameters

Photosynthetic rate (Pn)

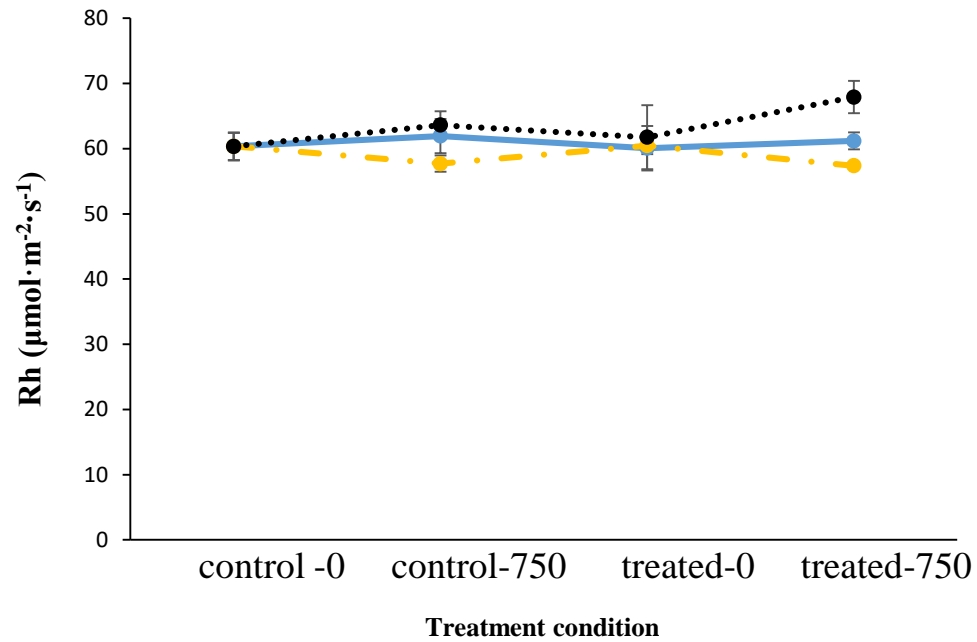


Stomatal conductance (gs)

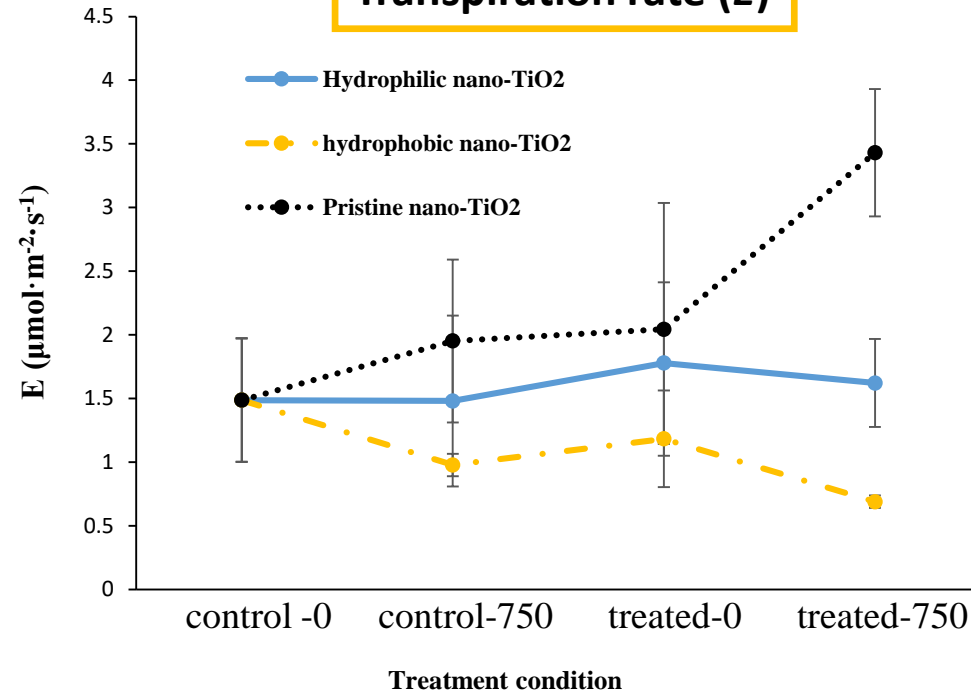


# Results - Photosynthetic parameters

Relative humidity ( $R_h$ )

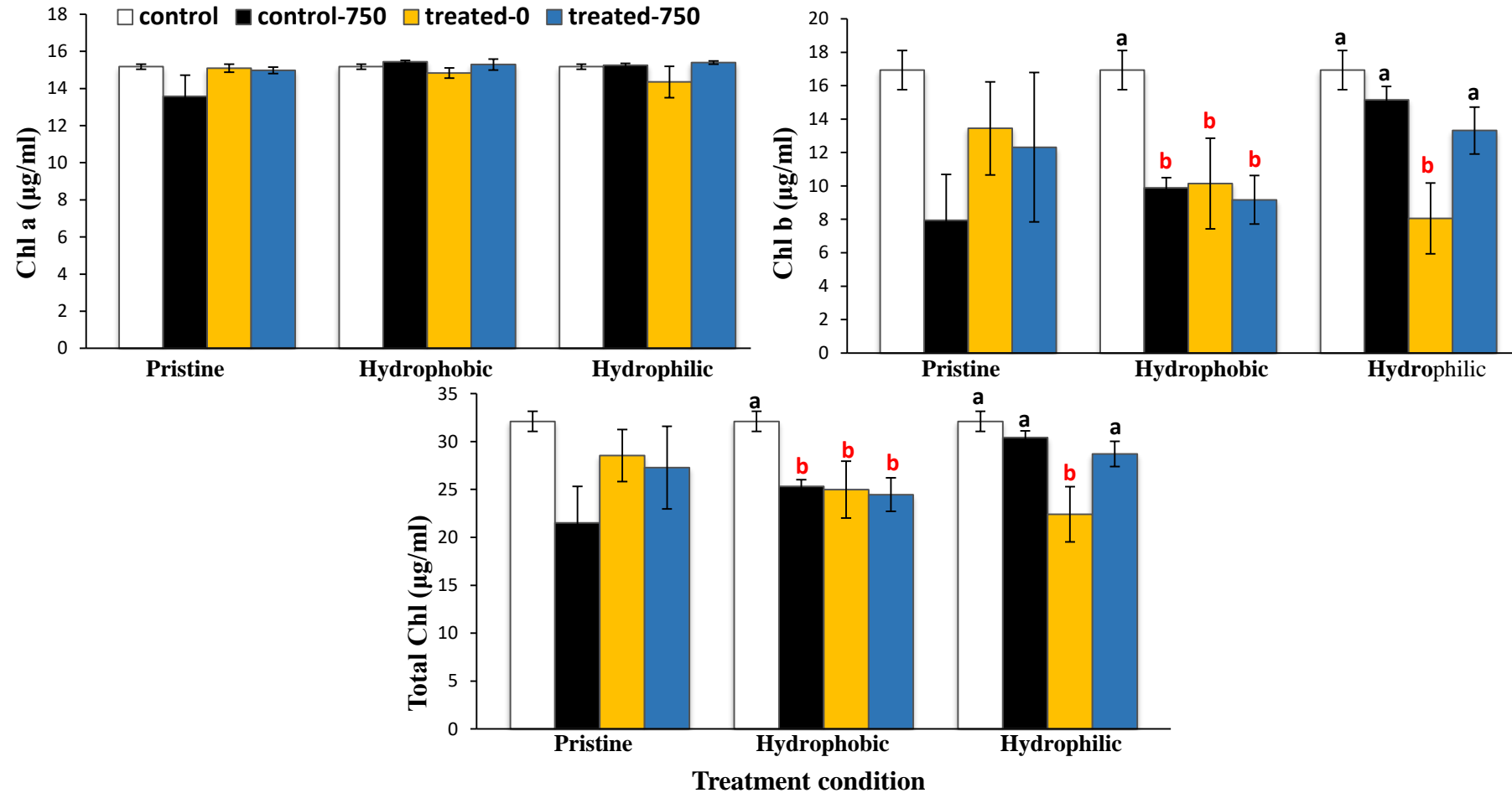


Transpiration rate ( $E$ )





# Results - Chlorophyll content



# Conclusion - Transgenerational effect

## Effects by NPs



- ❖ Hydrophobic → lower chlorophyll b and total chlorophyll content
- ❖ Pristine → higher Mg, Zn in root and Ni in shoot

## 2<sup>nd</sup>-gen seeds



- ❖ Hydrophobic → longer shoot length.
- ❖ Hydrophobic, hydrophilic → lower chlorophyll b and total chlorophyll content

## Interaction of 2<sup>nd</sup>-gen seeds and NPs



- ❖ Hydrophobic → higher biomass and shoot length; lower chlorophyll b and total chlorophyll content
- ❖ Pristine → increase of stomatal conductance.



# Conclusion – Surface property

Pristine



- ❖ Treated-750: a significant increase of stomatal conductance
- ❖ Treated-0: lower chlorophyll b content
- ❖ increase of Mg, Zn, Fe, and Ni.

Hydrophobic



- ❖ Treated-750: higher biomass and shoot length
- ❖ Control-750: lower chlorophyll b content
- ❖ increase of Al, Co, Fe, Zn, Mg

Hydrophilic



- ❖ Control-750: lower chlorophyll b content





# Acknowledgements



Go, miners!

