Funded by
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INTEGRANO

MULTIDIMENSIONAL INTEGRATED QUANTITATIVE APPROACH TO ASSESS SAFETY AND SUSTAINABILITY OF NANOMATERIALS IN REAL CASE LIFE CYCLE SCENARIOS USING NANOSPECIFIC IMPACT CATEGORIES

WP3

Nano Tox and nano eco-tox data generation

3rd EC meeting

11/09/2024

Online

This project has received funding from the European Union's Horizon Europe research and innovation programme under GA No 101138414

WP3

Definition of case studies, data management and digital DST

Objectives

- To assess eco-toxicology of investigated composite NMs groups (T3.1)
- To provide (eco-)toxicological EE based on dose-response experimental outcomes (T3.1)
- To identify and correlate dimension, morphology and other p-chem features modulating impact on AOPs (T3.2)
- To assess exposure scenario and carry out studies on NMs persistence and bioaccumulation even in low concentration doses at prolonged simulated exposure (T3.1)

Tasks

Task 3.1	**Ecotoxicity: Fate and effects in biological and environmental relevant matrices** Leader: CNR; Partners: UNIMIB, BIU, UniTO.	**M6-M44**
Task 3.2	**Collecting toxicity data and filling gaps for an early identification of hazard potential. CFs for toxicological assessment by in-vitro advanced models** Leader: UNIMIB; Partners: CNR, AITEX, RoV, B4C, DRT, PRJ.	**M6-M44**

Work planned for the NEXT 6 MONTHS

PLANNING OF ACTIVITIES:

Task 3.1 - ECOTOXICITY	Collection of published papers reporting ecotoxic effects of metal and metal-oxide based NPs common to different CS. Extraction of relevant data such as p-chem info and ecotoxic endpoints (LC_{50}, EC_{50}..). The complete dataset will be the base for subsequent tests planning besides providing prediction toxicity based on published literature
Task 3.2 - TOXICITY	Collection of data from previous project working on Ag, TiO_2, CuO, ZnO, SiO_2 etc.. NPs possibly used in different CS. Extraction of relevant information such as p-chem (primary size, hydrodynamic size, z-potential…) and toxicity endpoint (EC_{50}, LC_{50}, LOAEL). Data will serve to define the gaps we need to fill to provide all the relevant hazard information. Data will serve to define the parameters (p-chem?) to screen if INTEGRANO NPs are the "same" of those applied in previous projects or new toxicological characterization is needed.

RECAP:

EC_{50} Effective concentration, i.e. the concentration of exposure that determine a 50% variation in the selected endpoint (viability of cells in culture, expression of a protein, alteration of morphology etc…) – this value is calculated from the available data.

LC_{50}, Lethal concentration, i.e. the concentration of exposure that determine the death of the 50% of the exposed population (relevant for ecotoxic experiments) – this value is calculated from the available data.

LOAEC, lowest observed adverse effect concentration, i.e. the lowest dose that determine a significant change in the biological response (statistical difference) – this value depends (is measured) on the tested concentration.

Nano Material	Type	Other forms available*	NF Provider	Project 1	Project 2	Commercial property	Commercial property 2	NPs / NEPs	Form	Life cycle stage	Test category	Core Conc (g/L)	Doping conc (M)	Z-average nm (water)	Dev.st Z-aver nm (water)	Z-average t0 nm (MQ)	Dev.st Z.averT0 nm	Z-average t0 nm (Medium)
AgHECs2.8				ASINA		antibacterial	antiviral	NP	colloidal	synthesis	p-chem characteratior	5,117	0,14	170,8	0,6			
AgHECs2.8				ASINA		antibacterial	antiviral	NP	colloidal	synthesis	in vitro assay	5,117	0,14	170,8	0,6			
AgHECs5.5				ASINA		antibacterial	antiviral	NP	colloidal	synthesis	p-chem characteration							
AgHECs5.5				ASINA		antibacterial	antiviral	NP	colloidal	synthesis	in vitro assay							
AgHECs6.4				ASINA		antibacterial	antiviral	NP	colloidal	synthesis	p-chem characteratior	5,19	0,32	329	12			
AgHECs6.4				ASINA		antibacterial	antiviral	NP	colloidal	synthesis	p-chem characteratior	5,19	0,32	329	12			
AgHECs6.4				ASINA		antibacterial	antiviral	NP	colloidal	synthesis	in vitro assay	5,19	0,32	329	12			
AgHECs6.4				ASINA		antibacterial	antiviral	NP	colloidal	synthesis	in vitro assay	5,19	0,32	329	12			
AgHECp				ASINA		antibacterial	antiviral	NP	powder	synthesis	p-chem characteration					304,89	34,89	150,29
AgHECp				ASINA		antibacterial	antiviral	NP	powder	synthesis	p-chem characteration					304,89	34,89	304,6
AgHECp				ASINA		antibacterial	antiviral	NP	powder	synthesis	in vitro assay					304,89	34,89	150,29
AgHECp				ASINA		antibacterial	antiviral	NP	powder	synthesis	in vitro assay					304,89	34,89	304,6
AgCUR				ASINA		antibacterial	antiviral	NP	colloidal	synthesis	p-chem characteratior	5,8	0,009	114	1	91,27	5,79	
AgCUR				ASINA		antibacterial	antiviral	NP	colloidal	synthesis	in vitro assay	5,8	0,009	114	1	91,27	5,79	
AgCUR_A				ASINA		antibacterial	antiviral	NP	colloidal	synthesis	p-chem characteratior	6,02	0,004	2921	475			
AgCUR_A				ASINA		antibacterial	antiviral	NP	colloidal	synthesis	in vitro assay	6,02	0,004	2921	475			
AgCUR_B				ASINA		antibacterial	antiviral	NP	colloidal	synthesis	p-chem characteratior	5,78	0,018	93	0,8			
AgCUR_B				ASINA		antibacterial	antiviral	NP	colloidal	synthesis	in vitro assay	5,78	0,018	93	0,8			
Ag				ASINA		reference		NP	powder	synthesis	p-chem characteration					270,6	53,18	
Ag				ASINA		reference		NP	powder	synthesis	p-chem characteration					270,6	53,18	
Ag				ASINA		reference		NP	powder	synthesis	in vitro assay					270,6	53,18	328,71
Ag				ASINA		reference		NP	powder	synthesis	in vitro assay					270,6	53,18	188,22
AgPVP				ASINA		reference		NP	powder	synthesis	p-chem characteration					695,91	617,49	545,96
AgPVP				ASINA		reference		NP	powder	synthesis	p-chem characteration					695,91	617,49	255,85
AgPVP				ASINA		reference		NP	powder	synthesis	in vitro assay					695,91	617,49	545,96
AgPVP				ASINA		reference		NP	powder	synthesis	in vitro assay					695,91	617,49	255,85
SiO2-TiO2	core-shell	CS1, CS2, CS3	CENTI	PROTECT		antibacterial		NP	powder	synthesis	p-chem characteration					297	4,5	
SiO2-TiO2	core-shell	CS1, CS2, CS3	CENTI	PROTECT		anitbacterial		NP	powder	synthesis	p-chem characteration					297	4,5	
SiO2-TiO2	core-shell	CS1, CS2, CS3	CENTI	PROTECT		anitbacterial		NP	powder	synthesis	p-chem characteration					297	4,5	

Nano Material	PdI (water)	Err_PdI (water)	PdI T0 (MQ)	Err_PdI T0 (MQ)	PdI T0 (Medium)	Err_PdI T0 (Medium)	Medium type	Z-average tech	Z-pot (water)	Z-pot t0 (MQ)	Err_Z-pot t0 (MQ)	Z-pot tech
AgHECs2.8	0,27	0,006						DLS	12,2			DLS
AgHECs2.8	0,27	0,006					DMEM 1% FBS	DLS	12,2			DLS
AgHECs5.5								DLS				DLS
AgHECs5.5							DMEM 1% FBS	DLS				DLS
AgHECs6.4	0,404	0,062						DLS	21,1			DLS
AgHECs6.4	0,404	0,062						DLS	21,1			DLS
AgHECs6.4	0,404	0,062					DMEM 1% FBS	DLS	21,1			DLS
AgHECs6.4	0,404	0,062					FET Water	DLS	21,1			DLS
AgHECp			0,37	0,07				DLS		11	1,07	DLS
AgHECp			0,37	0,07				DLS		11	1,07	DLS
AgHECp			0,37	0,07	0,35	0,06	DMEM 1% FBS	DLS		11	1,07	DLS
AgHECp			0,37	0,07	0,29	0,04	FET water	DLS		11	1,07	DLS
AgCUR	0,389	0,009						DLS	-35	-34,13	1,59	DLS
AgCUR	0,389	0,009					DMEM 1% FBS	DLS	-35	-34,13	1,59	DLS
AgCUR_A	1	0						DLS	-36,8			DLS
AgCUR_A	1	0					DMEM 1% FBS	DLS	-38,8			DLS
AgCUR_B	0,39	0,008						DLS	-47			DLS
AgCUR_B	0,39	0,008					DMEM 1% FBS	DLS	-47			DLS
Ag			0,45	0,004				DLS		-23,58	4,84	DLS
Ag			0,45	0,004				DLS		-23,58	4,84	DLS
Ag			0,45	0,004	0,37	0,13	DMEM 1% FBS	DLS		23,58	4,84	DLS
Ag			0,45	0,004	0,44	0,06	FET Water	DLS		23,58	4,84	DLS
AgPVP			0,7	0,26				DLS		-11,7	4,89	DLS
AgPVP			0,7	0,26				DLS		-11,7	4,89	DLS
AgPVP			0,7	0,26	0,43	0,06	DMEM 1% FBS	DLS		-11,7	4,89	DLS
AgPVP			0,7	0,26	0,5	0,08	FET water	DLS		-11,7	4,89	DLS
SiO2-TiO2			0,068					DLS		-39		DLS
SiO2-TiO2			0,068					DLS		-39		DLS

Nano Material	Specific model	Target	Viability (EC50 ug/ml)	Viability LOAEC (ug/ml)	note	Viability test	IL8 (EC50)	IL8 LOAEC	IL6 (EC50)	IL6 LOAEC	Inflammation test	EC50 Ox Resp (ug/mL)	LOAEC Ox Resp (ug/mL)	OR test	other tests value
AgHECs2.8	none														
AgHECs2.8	A549	lung	91,53	10		Alamar Blue	na	na			ELISA	na	na	DCFH ROS	
AgHECs5.5	none														
AgHECs5.5	A549	lung	na	20		Alamar Blue	52,32	100			ELISA	na	100	DCFH ROS	
AgHECs6.4	none														
AgHECs6.4	none														
AgHECs6.4	A549	lung	7,29	4		Alamar Blue	na	na			ELISA	na	na	DCFH ROS	
AgHECs6.4	zebrafish embr	environment				OECD 236	na	na			ELISA	na	na	DCFH ROS	
AgHECp	none														
AgHECp	none														
AgHECp	A549	lung	57,05	100	Max conc teste	Alamar Blue	21,69	50			ELISA	na	100	DCFH ROS	
AgHECp	zebrafish embr	environment				OECD 236									
AgCUR	none														
AgCUR	A549	lung	na	50		Alamar Blue	na	na			ELISA	na	100	DCFH ROS	
AgCUR_A	none														
AgCUR_A	A549	lung	5,65	10		Alamar Blue	na	na			ELISA	na	na	DCFH ROS	
AgCUR_B	none														
AgCUR_B	A549	lung	na	100	Max conc teste	Alamar Blue	na	na			ELISA	na	na	DCFH ROS	
Ag	none														
Ag	none														
Ag	A549	lung	na			Alamar Blue	na	na			ELISA	42,74	50	DCFH ROS	
Ag	A549	lung	na			Alamar Blue	na	na			ELISA	42,74	50	DCFH ROS	
AgPVP	none														
AgPVP	none														
AgPVP	A549	lung	na			Alamar Blue	na	na			ELISA	49,33	50	DCFH ROS	
AgPVP	zebrafish embr	environmen	na			OECD 236	na	na			ELISA	49,33	50	DCFH ROS	

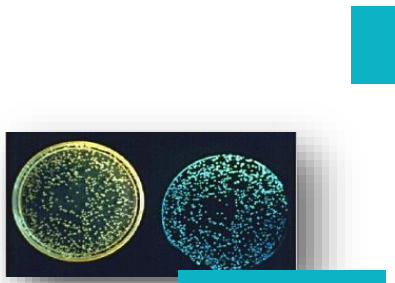
Task 3.1

Ecotoxicity: Fate and effects in biological and environmental relevant matrices

Aquatic organisms as biological models

Standard (i.e. UNI EN ISO, ISO, ASTM) and innovative methods

Ecosafety of innovative nano materials/products



Bacteria

Microalgae

Crustaceans

Rotifers

Echinoderms

Molluscs

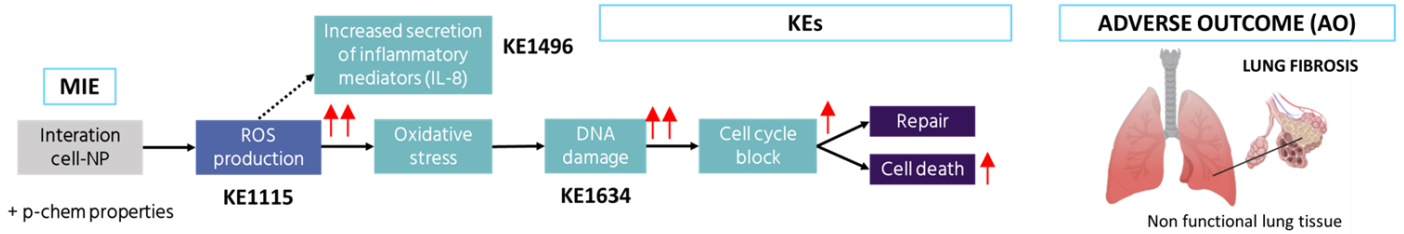
Cnidarians

Task 3.2

Collecting toxicity data and filling gaps for an early identification of hazard potential. CFs for toxicological assessment by in-vitro advanced models

Main objectives:

- Biological characterization of the effects of NMs according to relevant AOPs
- Exposure and hazard of the NMs for humans and environmental organisms by using OECD and ISO methodologies
- Implementation of mechanistic toxicology studies according to SSbD.
- Hazard assessment to derive relevant dose-response or concentration-response curves also by NAMs
- Correlation between hazard endpoints and p-chem of the NPs



- **Important** to have an update of this Gannt to be aware of what to expect and when… also to be able to define delays in delivery and of expected results from WP3.

					WHO		March				April				May					June			
							3-9	10-16	17-23	24-30	31-6	7-13	14-20	21-27	28-4	5-11	12-18	19-25	26-1	2-8	9-15	16-22	23-29
		Activity / Taks	**% done**	***comments***	from	to	**W10**	**W11**	**W12**	**W13**	**W14**	**W15**	**W16**	**W17**	**W18**	**W19**	**W20**	**W21**	**W22**	**W23**	**W24**	**W25**	**W26**
CS1		AgHEC																					
		AgCUR																					
		SiO2																					
		Bio-SiO2																					
		Bio-SiO2@TiO2																					
		Egyptian Blue																					
		ZnO																					
		CuO																					
CS2		CSF-Sil1-Rust		*Samples available by end of May - characterisation depends on partners and repairs*																			
		CSF-Sil1-Rice		*Samples available by end of May - characterisation depends on partners and repairs*																			
		CSF-Sil1-Rust-Rice		*Samples available by end of May - characterisation depends on partners and repairs*																			
		bio-SiO2@TiO2																					
CS3		Diatomite		*Samples already in use*																			
		Gas Beton		*Samples already in use*																			
		Bio-SiO2		*Samples already in use*																			
CS4		Egyptian Blue (μ_m)		*The sample will be characterised with SEM-EDS and VIL (Visible Induced Luminescence) by mid June*																			
		Egyptian Blue (n_m)																					
		Bio-Egyptian Blue (μ_m)		*Using silica from rice husks - delivery of EB product not yet known*																			
		Bio-Egyptian Blue (n_m)		*Using silica from rice husks - delivery of EB product not yet known*																			
		AgHEC																					
		CA fibre synthesis																					
CS5		C-dots																					

Where we are

- Completion of the dataset of already available data, almost done (both for Ecotox e Tox) end of September should be ready.
- Sharing of the dataset for identification of the gaps in Ecotox-Tox data, from October
- Identification of the main gaps to be filled also with novel NPs (viability, inflammation, oxidative response, genotox???)
- Continue discussing with case studies leaders to understand the timing of delivering of the materials, the number of materials expected to be “new”, the kind of information (p-chem) will be available, and the hazard information expected to be relevant for the specific material and CS (human, environment, both)

Deliverables

Del.	Title	Lead Beneficiary	Diss. Level	Due Month	Date
D3.1	Quantitative assessment of Ecotoxicity endpoints for indication of suitable nano-specific EE for marine and terrestrial environment eco-toxicity ICs	CNR	PU	44	August 2027
D3.2	Relevant dose-response functions for the NMs in their different LC stages, for human and environmental toxicology outcomes supporting the definition of nanospecific EE for human and environmental toxicity ICs	UNIMIB	PU	44	August 2027