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## Plan Overview

A Data Management Plan created using CORA.eiNa DMP

**Title:** ASPHIRE: AsseSsing the Pulmonary Hazards and molecular mechanisms of Nanoplastics from TyRe Wear Emissions

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### Project abstract:

#### SUMMARY

Airborne tyre wear nanoplastics (TW-NPLs) are an emerging environmental health threat with direct implications for public well-being. As a major component of urban particulate pollution, TW-NPLs are continuously inhaled by millions of people, yet their pulmonary effects, mechanisms of action, and long-term risks remain largely unknown. This knowledge gap is particularly concerning because TW-NPLs combine nanoscale size, allowing deep lung penetration, with a high load of toxic chemical additives. Understanding their biological behaviour is therefore essential to inform preventive public health measures and regulatory action.

ASPHIRE specifically addresses how TW-NPLs interact with lung cells and the pulmonary system, identifying the cellular and molecular mechanisms underlying toxicity and establishing causal links between molecular perturbations and adverse effects at the organism level. This knowledge will be essential for guiding mitigation strategies and regulatory measures

#### OBJECTIVES

1. To generate TW-NPL test materials that enable the study of inhaled TW-NPLs under conditions representative of real-world exposure
2. To define the cellular and molecular processes altered by long-term exposure to TW-NPLs using advanced New Approach Methodologies (NAMs), combined with validated effect biomarkers and multi-omics approaches.
3. To establish causal relationships and characterise the molecular mechanisms underlying pulmonary hazards induced by TW-NPLs exposure.

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# ASPHIRE: AssesSsing the Pulmonary Hazards and molecular mechanisms of Nanoplastics from TyRe Wear Emissions

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## 1. Data summary

### Provide a data summary

The ASPHIRE project aims to advance the evidence base on the pulmonary toxicity of tyre-wear nanoplastics (TW-NPLs) by elucidating the cellular and molecular mechanisms underlying adverse outcomes, thereby informing effective mitigation strategies and regulatory measures to safeguard public health.

### Purpose of the data

The data collected will support the following project objectives:

1. **Generation of TW-NPL test materials.** *Characterise various TW-NPLs physicochemical properties to understand their properties and cell internalisation behaviour.*
2. **Definition of the cellular and molecular processes altered by long-term exposure to TW-NPLs.** *Analyse the behaviour and fate of TW-NPLs using New Advanced Methodologies including the assessment of key biological effects, the analysis of omic alterations and functional and molecular assays to validate findings.*
3. **Establishment of causal relationships and characterisation to the molecular mechanisms.** *Integration of internal and external evidence using a weight-of-evidence approach and organization within an AOP framework.*

### Data collection

1. **Nanoplastics Characterisation**
  - o **Images:** TEM and SEM images measuring size, distribution, morphology, and shape.
  - o **Data:** DLS, NTA, FTIR, RAMAN and ICPMS measurements.
  - o **Origin:** Laboratory analyses of various TW-NPLs
2. **Nanoplastics Internalisation**
  - o **Images:** TEM, SEM, and sorter cytometry images measuring internalisation.
  - o **Data:** Measurements of internalisation, cell viability, and DNA damage.
  - o **Origin:** Laboratory experiments
3. **Advanced ALI Calu-3 model**
  - o **Images:** Barrier characterisation, internalisation, and bioaccumulation images.
  - o **Data:** TEER values, paracellular transport, internalisation, and bioaccumulation measurements.
  - o **Other:** Technical protocol of the advanced ALI Calu-3 system.
  - o **Origin:** Laboratory development and testing of the advanced ALI Calu-3 system.
4. **Key biological effects**
  - o **Data:** Genotoxic and oxidative DNA damage evaluation (comet assay), genomic instability (micronucleus assay), inflammatory response and immune alterations (cytokine arrays and ELISA), measurements of cell transformation (cell proliferation, morphology, anchorage-independent growth, migration, invasion), and stem cell function (sphere formation, colony-forming assays, flow cytometry).
  - o **Origin:** Long-term in vitro experiments on lung models.
5. **Omic alterations**
  - o **Data:** transcriptomics (transcriptomic analyses reads), epigenomics (epigenomic sequencing reads), and metabolomics.
  - o **Origin:** Long-term in vitro experiments on lung models.
6. **Integration of internal and external evidence**
  - o **Data:** internal relevant chemical, biological, and toxicological information on TW-NPLs health effects.
  - o **Other:** external relevant chemical, biological, and toxicological information on TW-NPLs health effects.
  - o **Origin:** experimental and omics for internal data. Public databases and existing literature for external data.
7. **AOP framework**
  - o **Other:** Proposal of new AOP following AOP-Wiki principles.
  - o **Format:** Conceptual framework document.
  - o **Origin:** Integration of experimental findings and AOP-Wiki principles

### Format of the data

Most of the data will be in ASCII (American Standard Code for Information Interchange) files, e.g. comma-separated variable (CSV) format, which can be imported into rich text files for word processing or spreadsheets. Where specialist software is used, information on free readers will be provided. Data will be produced in the following formats:

- Images: jpeg, odg, pdf, png, tiff, ptx
- Tables: xlsx, csv
- Text: docx, pdf, txt, fastq

### Expected size

The estimated size of the raw data may reach up to 1TB.

## 2. FAIR Data

### 2.1 Data findable (including metadata)

Data will be deposited and described in the open access repository CORA.RDR.

CORA.RDR is the repository of the Consorci de Serveis Universitaris de Catalunya (CSUC). This repository follows the Open Archives Initiative model, which allows interoperability with the OAI-PMH (Open Archive Initiative - Protocol for Metadata Harvesting) metadata transfer protocol. This protocol allows documents to be viewed from different platforms and collectors: Google Scholar, BASE, CORE, etc. This repository is OpenAIRE compliant and meets all the metadata requirements set by the European Commission.

CORA.RDR will provide a unique URL to access the document in the format <https://repository/record/1234>. The repository will assign DOIs.

**Omic data will be deposited in public repositories such as SRA (Sequence Read Archive, from National Center for Biotechnology Information).**

Files are structured in terms of project and publication characteristics with unique file names (id) for figures and files.

All open project outputs deposited in a repository will provide search terms along with their metadata. Keywords for open data will be selected from controlled vocabularies appropriate to the type of data.

The metadata standard used to describe the dataset will be the Dublin Core Schema, as it is a flexible and widely used standard, and is also the standard adopted by the European OpenAIRE repository.

Version control mechanisms should be established and documented before any data is collected or generated. In addition, all open data, publications and open source software will use DOI versioning. DOI versioning allows a dataset to be updated after it has been published and to cite either a specific version of a dataset or all versions of a dataset.

The manuscripts/documents/materials resulting from the Project's data will be stored in the institutional repositories (e.g. Dipòsit Digital de Documents, DDD, <https://ddd.uab.cat/>) which provides a unique URL to access the document with the format <https://ddd.uab.cat/record/XXXXXX> where XXXXXX is the identification code.

## 2.2 Data accessible

Data will be openly shared in a repository once it has been analysed and published.

In case it is decided to temporarily restrict the access to a particular data set, it will be possible to ask for individual access by contacting the principal investigator. The principal investigator will evaluate whether it is possible to grant access to the requested data.

Data will be made accessible to other group members and project partners in open access, and to the general public when there are no data protection issues through an established repository. Data will be stored in CSV, TSV or TXT to ensure flexibility for data reuse and to avoid format obsolescence as far as possible. CSV (comma separated values) files are used to store tabular data in plain text format. Most often the fields in this data are separated by commas but other delimiters can be used such as |. TSV (tab separated values) files are similar, but breaks are delimited by tabs. Both formats are widely supported and are often used to exchange data across multiple different computers and systems that support the format. Most modern spreadsheet packages can open CSV/TSV files for viewing. Although, to maintain formatting data, they can be later saved in a proprietary format like XLS (Microsoft Office Excel), ODS (Open Office spreadsheets) or numbers (Apple Mac), depending on the used software. TXT is a standard of unformatted text. It is recognized by any text editing or word processing program and can also be processed by most other software programs.

Images will be shared in PNG or PDF format since they are commonly used formats with widely distributed visualisation software.

Omics data will be fastq formatted, while metadata will be stored in text files (.csv).

When no embargo period applies and a data package related to a case study has been marked as public, it will be made openly available immediately, otherwise data will be made available when the embargo period finishes.

## 2.3 Data interoperable

The metadata standard used to describe the dataset will be the Dublin Core Schema, if any extensions are needed the Data Document Initiative will be included. Where possible, standard codes will be followed, for example country identification will be registered following the ISO 3166-1-alpha-2 codes and language identification will use the ISO 639-2 code.

The vocabulary used in the data will be as standard as possible to ensure its comprehension by non-experts in the field.

Both CORA.RDR and DDD follow the Open Archives Initiative model, which enables interoperability with the OAI-PMH (Open Archives Initiative - Protocol for Metadata Harvesting) transmission protocol. Documents will be accessible through various platforms and aggregators, including Google Scholar, BASE, CORE, Europeana, Hispana, WorldCat, OpenAIRE, OpenDOAR, RECOLECTA, and ROAR.

## 2.4 Data reusable

The data will be made permanently available for re-use via the institutional repository. Each archived dataset will have its own permanent repository ID and will be easily accessible.

We anticipate that most of the data generated will be made available without restriction, with only those datasets subject to IPR and confidentiality issues being restricted. Where access to data is restricted, agreements will be made based on the individual data sets. Requests for the use of the data by externals will be approved by the project principal investigators.

The data quality will be ensured by different measures. These include validation of the data through experimental replicates, technical replicates, concordance with data obtained from complementary experimental models and techniques, and intra-/inter- laboratory comparisons.

The data collected in this project will be made available for re-use after the field seasons and publications have been completed. Attribution-Non-Commercial license (By-Nc) will be used. Data may be embargoed for reasons of competitive advantage, including the completion of a PhD thesis, in which case an embargo period will be maintained.

## 3. Allocation of resources

### Explain the allocation of resources

There are no additional costs associated with the mechanisms described to make the database FAIR and sustainable.

CORA.RDR and other institutional mechanisms of data preservation are covered by the budget of Universitat Autònoma de Barcelona and Consejo Superior de Investigaciones Científicas.

## 4. Data security

### Address data recovery as well as secure storage and transfer of sensitive data

During the development of the project, access to the data will be exclusively for internal use by the members of the team. To follow regulations and security procedures according to EU standards, files will be stored in the UAB cloud-based Microsoft SharePoint service, guaranteeing security through login access, regular automated backups and GDPR compliance. Larger data files produced will be stored on the institutional servers. The server ensures that the regulations and security procedures are compiled following the EU standards. The building and room will be secured 24 hours a day.

## 5. Ethical aspects

### To be covered in the context of the ethics review, ethics section of DoA and ethics deliverables. Include references and related technical aspects if not covered by the former

All the activities carried out by the project comply with ethical principles and relevant national, EU and international legislation, for example the Charter of Fundamental Rights of the European Union and the European Convention on Human Rights.

## 6. Other issue

### Refer to other national/funder/sectorial/departmental procedures for data management that you are using (if any)

As part of the Universitat Autònoma de Barcelona's commitment to FAIR and open data, all research active staff (postdoctoral fellows, doctoral students) are expected to prepare DMPs for their own data, according to the University's Research Data Management Policy.

This includes quantitative and qualitative statements, raw data from measurements and derived data - either cleaned or extracted from a researcher's primary dataset or derived from an existing

source.

## **7. Further support in developing your DMP**

### **Further support in developing your DMP**

This DMP has been created using "eiNa DMP" (<https://dmp.csuc.cat>), following the "Plan Estatal (in english)" template proposed by the Agencia Estatal de Investigación. This Deliverable is a living document and will be updated when necessary.

## Planned Research Outputs

### Image - "Nanoplastics characterisation images"

Images from TEM and SEM measuring size, distribution, morphology and shape from the studied nanoplastics within the initial screening.

### Dataset - "Nanoplastics characterisation data"

DLS, NTA, FTIR, RAMAN and ICPMS measurements for the studied nanoplastics within the initial screening

### Image - "Nanoplastics internalisation images"

Images from TEM, SEM and sorter cytometry measuring internalisation of studied nanoplastics within the initial screening

### Dataset - "Initial screening of nanoplastics internalisation, cytotoxicity and genotoxicity"

Measurements of internalisation, cell viability and DNA damage for the studied nanoplastics within the initial screening

### Technical protocol - "Advanced ALI Calu-3 model"

Technical protocol of an advanced version of the ALI Calu-3 system.

### Dataset - "Studies from advanced ALI Calu-3 model"

Measurement of TEER (trans epithelial electrical resistance) values, paracellular transport, internalisation and bioaccumulation from tyre wear nanoplastics.

### Image - "Images from advanced ALI Calu-3 model"

Images from barrier characterisation and tyre wear nanoplastics internalisation and bioaccumulation.

### Dataset - "Key biological effect: cell transformation"

Measurements of cell proliferation, morphology, anchorage independent growth, migration, invasion and stem cell function (sphere formation, colony-forming assays and flow cytometry) from long-term *in vitro* experiments on lung models.

### Dataset - "Omic alterations: epigenomics"

Reads from epigenomic sequencing from long-term *in vitro* experiments on lung models.

### Dataset - "Omic alterations: transcriptomics"

Reads from transcriptomic analyses from long-term *in vitro* experiments on lung models.

### Workflow - "Pipeline for transcriptomic analysis"

Reviewed pipeline for the analysis of transcriptomic sequencing.

### AOP proposal - "AOP framework"

Framework for a new AOP following the AOP-wiki principles

### Presentation - "Documentation from communication and dissemination"

Presentations, posters and abstracts from participation in conferences and congresses.

### Workflow - "Pipeline for epigenomic analysis"

Reviewed pipeline for the analysis of epigenomic sequencing.

### Dataset - "Key biological effects"

Measurements of genotoxic and oxidative DNA damage (comet assay), genomic instability (micronucleus assay), and inflammatory and immune alterations (cytokine arrays and ELISA) from long-term *in vitro* experiments on lung models.

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## Planned research output details

Title	Type	Anticipated release date	Initial access level	Intended repository(ies)	Anticipated file size	License	Metadata standard(s)	May contain sensitive data?	May contain PII?
Nanoplastics characterisation images	Image	Unspecified	Embargoed	CORA. Repositori de Dades de Recerca		Creative Commons Attribution Non Commercial 4.0 International	None specified	No	No
Nanoplastics characterisation data	Dataset	Unspecified	Embargoed	CORA. Repositori de Dades de Recerca		Creative Commons Attribution Non Commercial 4.0 International	None specified	No	No
Nanoplastics internalisation images	Image	Unspecified	Embargoed	CORA. Repositori de Dades de Recerca		Creative Commons Attribution Non Commercial 4.0 International	None specified	No	No
Initial screening of nanoplastics internalisation, ...	Dataset	Unspecified	Embargoed	CORA. Repositori de Dades de Recerca		Creative Commons Attribution Non Commercial 4.0 International	None specified	No	No
Advanced ALI Calu-3 model	Technical protocol	Unspecified	Embargoed	CORA. Repositori de Dades de Recerca		Creative Commons Attribution Non Commercial 4.0 International	None specified	No	No
Studies from advanced ALI Calu-3 model	Dataset	Unspecified	Embargoed	CORA. Repositori de Dades de Recerca		Creative Commons Attribution Non Commercial 4.0 International	None specified	No	No
Images from advanced ALI Calu-3 model	Image	Unspecified	Embargoed	CORA. Repositori de Dades de Recerca		Creative Commons Attribution Non Commercial 4.0 International	None specified	No	No
Key biological effect: cell transformation	Dataset	Unspecified	Embargoed	CORA. Repositori de Dades de Recerca		Creative Commons Attribution Non Commercial 4.0 International	None specified	No	No
Omic alterations: epigenomics	Dataset	Unspecified	Embargoed	Sequence Read Archive		Creative Commons Attribution Non Commercial 4.0 International	None specified	No	No
Omic alterations: transcriptomics	Dataset	Unspecified	Embargoed	Sequence Read Archive Gene Expression Omnibus		Creative Commons Attribution Non Commercial 4.0 International	None specified	No	No
Pipeline for transcriptomic analysis	Workflow	Unspecified	Open	GitHub		Creative Commons Attribution Non Commercial 4.0 International	None specified	No	No
AOP framework	AOP proposal	Unspecified	Embargoed	CORA. Repositori de Dades de Recerca		Creative Commons Attribution Non Commercial 4.0 International	None specified	No	No
Documentation from communication and dissemination	Presentation	Unspecified	Open	Dipòsit Digital de Documents de la UAB		Creative Commons Attribution Non Commercial 4.0 International	None specified	No	No
Pipeline for epigenomic analysis	Workflow	Unspecified	Open	GitHub		Creative Commons Attribution Non Commercial 4.0 International	None specified	No	No
Key biological effects	Dataset	Unspecified	Embargoed	CORA. Repositori de Dades de Recerca		Creative Commons Attribution Non Commercial 4.0 International	None specified	No	No