ISIS@MACH ITALIA

UKRI Science and Technology Facilities Council
ISIS Neutron and Muon Source

# Experiment Proposal

Experiment number GP2023022

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|---|---|
| **Principal investigator (*)** | Miss Giulia Mugnaini, University of Florence, ITALY |
| **Co-investigator** | Dr Monica Tonelli, University of Florence, ITALY |
| **Co-investigator** | Professor Francesca Ridi, University of Florence & CSGI, ITALY |
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| **Experiment title** | Studies on the inner structure of biocomposite alginate/HNTs fibres |

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| **MRF Instrument** | **FIB-SEM GAIA 3** | **Days requested:** 1 |
| **Access Route** | Direct Access | **Previous GP Number:** No |
| **Science Areas** | Chemistry | **DOI:** No |
| **Sponsored Grant** | None | **Sponsor:** - |
| **Grant Title** | - | **Grant Number:** - |
| **Start Date** | - | **Finish Date:** - |
| **Similar Submission?** | - | |
| **Industrial Links** | - | |
| **Non-Technical Abstract** | In this study | |

**Publications** -

**ISIS neutron and muon source** **IM@IT E-platform:** Yes - ISIS Facility

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|---|---|---|
| **Instruments** | **IMAT** | **Days Requested:** Proposal will be submitted later |
| **Access Route** | Direct Access | **Previous RB Number:** No |
| **Science Areas** | | **DOI:** No |
| **Sponsored Grant** | None | **Sponsor:** |
| **Grant Title** | - | **Grant Number:** |
| **Start Date** | - | **Finish Date:** |
| **Similar Submission?** | | |
| **Industrial Links** | | |

# Sample record sheet

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|---|---|---|---|
| **Principal contact** | Miss Giulia Mugnaini, University of Florence, ITALY | | |
| **MRF Instrument** | **FIB-SEM GAIA 3** | **Days Requested:** 1 | |
| **Special requirements:** | | | |

## SAMPLE

| | | | |
|---|---|---|---|
| **Material** | Sodium Alginate, Calcium chloride, Halloysite nanotubes | - | - |
| **Formula** | $C_6H_8O_6$, $CaCl_2$, $Al_2Si_2O_5(OH)_4{\cdot}nH_2O$ | - | - |
| **Forms** | Solid | | |
| **Volume** | cc | | |
| **Weight** | 100 mg | | |
| **Container or substrate** | Plastic falcon | - | - |
| **Storage Requirements** | - | - | - |

## SAMPLE ENVIROMENT

| | | | |
|---|---|---|---|
| **Temperature Range** | Room temperature - K | - | - |
| **Pressure Range** | Room pressure - mbar | - | - |
| **Magnetic field range** | None - T | - | - |
| **Standard equipment** | None | - | - |
| **Special equipment** | No | - | - |

## SAFETY

| | | | |
|---|---|---|---|
| **Prep lab needed** | No | - | - |
| **Sample Prep Hazards** | No | - | - |
| **Special equip. reqs** | No | - | - |
| **Sensitivity to air** | No | - | - |
| **Sensitivity to vapour** | No | - | - |
| **Experiment Hazards** | No | - | - |
| **Equipment Hazards** | - | - | - |
| **Biological hazards** | No | - | - |
| **Radioactive Hazards** | No | - | - |
| **Additional Hazards** | - | - | - |
| **Additional Details** | - | - | - |
| **Sample will be** | Disposed of by instrument scientist | - | - |

**Science Case - Studies on the inner structure of biocomposite alginate/HNTs fibres**

## 1. Background and Context

The design of biocomposite materials with unique architectures is currently an attractive challenge in multiple research areas, including the agricultural field, thanks to the opportunity of combining the advantages of both natural polymers and inorganic nanofillers, resulting in a final material with remarkable and improved physico-chemical properties. Alginate (Alg) is a polysaccharide derived from marine brown algae, whose well-established applications are based on its ability to coordinate di- and trivalent cations (*e.g.* $Ca^{2+}$) according to the *egg-box* model, resulting in physical cross-linked hydrogels. Alginate biocomposite fibres with halloysite nanotubes (HNTs), which are naturally occurring aluminosilicate clays with a multi-walled tubular structure, are promising candidates as nanosized vehicles for the controlled-release of active molecules, owing to HNTs peculiar hollow structure. Furthermore, due to their elongated structure, HNTs can be assembled into anisotropic patterns through the application of an external force, *i.e.* the shear force applied during a extrusion protocol for fibre preparation. One of the major problems in the agricultural field is the overuse of toxic agrochemicals, such as fertilisers, pesticides and herbicides, which are associated with serious negative environmental impacts, including the contamination of the ecosystems. The design of agro-formulations based on the encapsulation of active compounds into nano-carriers is a promising, advanced and suitable way for the reduction of the negative side effects associated with the use of these compounds. As a result of the encapsulation, the toxic substances are, conveniently, locally released for long periods of time without harmful and unnecessary losses in the environment. In addition, this method could be useful for the delivery of micronutrients and genetic material for plant transformation. Aiming at the preparation of alginate fibres containing HNTs nano-carriers, the investigation of the structural organisation and mechanical and porous properties of the composite is particularly interesting towards the development of innovative multi-functional systems where biodegradable polymers can be used in outdoor applications, for example by wrapping these fibres around trees. The introduction of HNTs and their spatial organisation, indeed, strongly influences the mechanical properties of the fibres and consequently influence their possible ways of use. This research involves the work of different people, including a PostDoc, a researcher and a professor. The team contributes to the development of such systems with their complementary experiences on polymers, fibres preparation, and use of HNTs as nano-carriers for the slow delivery of active molecules in composites. Overall, the research is supported by a fellowship, and by The National Recovery and Resilience Plan of Italy "3A-Italy Circular and Sustainable Made in Italy" (PNRR, 3A-ITALY - B83C22004890007)

## 2. Proposed experiments

In order to verify if the shear force applied during the extrusion process is enough to induce an alignment of the HNTs along the flow direction, the global distribution and orientation of the nanotubes inside the alginate matrix has to be investigated. In this view, FIB-SEM TESCAN GAIA 3 instrumentation available in the *ISIS@MACH ITALIA* infrastructure could be crucial, allowing for the direct visualisation of the inner part of a dry fibre section. The possibility of using ultramicrotomy as well as FIB for the sectioning of the composites will be considered, and the more suited technology will be used for the characterization of the fibres. These investigations could be complemented with neutron tomography analysis, to be subsequently performed using IMAT instrumentation available at ISIS Neutron and Muon Source, that could further provide a 3D reconstruction of HNTs-alginate biocomposite in its wet state, paying particular attention on the eventual formation of aggregates of HNTs: in fact, IMAT allows for resolving 60 μm structures (in high resolution mode), while HNTs aggregates could reach a size of few hundreds of microns (the actual size will be evaluated during the FIB-SEM experiments).

### 3. Summary of previous experimental proposals or characterisation

In this study hybrid fibres composed of calcium cross-linked alginate and halloysite nanotubes were prepared by means of extrusion technique. The nanotubes were incorporated and dispersed with different methods in the alginate solution to be injected into a calcium chloride bath for the preparation of the biocomposite. Different processing parameters were studied, such as the flow rate and calcium chloride concentration of the coagulation bath, to unravel the effect of these parameters on the morphology and dimensions of the obtained composite fibres. The injectability properties of alginate solutions enriched with HNTs were investigated by applying different loads on the plunger of a syringe and measuring the corresponding apparent flow rates (**Fig.1i**), demonstrating that all these formulations behave as a Newtonian fluid in the force range used in the preparation process. Through optical microscopy experiments, the average diameter of the composite fibres was determined and correlated with the processing parameters, demonstrating that at constant flow rate, an increase of the $CaCl_2$ concentration leads to an increase of the fibre average diameter (**Fig.1ii**); while no correlation between the flow rate and the average diameter was found. The effect of the preparation parameters was further investigated by means of scanning electron microscopy, confirming that $CaCl_2$ concentration has a strong effect on the biocomposite, affecting the surface morphology, whereas no specific effect of the flow rate was detected (**Fig.1iv**). In addition, the comparison between fibres prepared with and without HNTs suggests a certain alignment of the alginate chains and of the nanotubes in the direction of the flow (**Fig.1iii**).

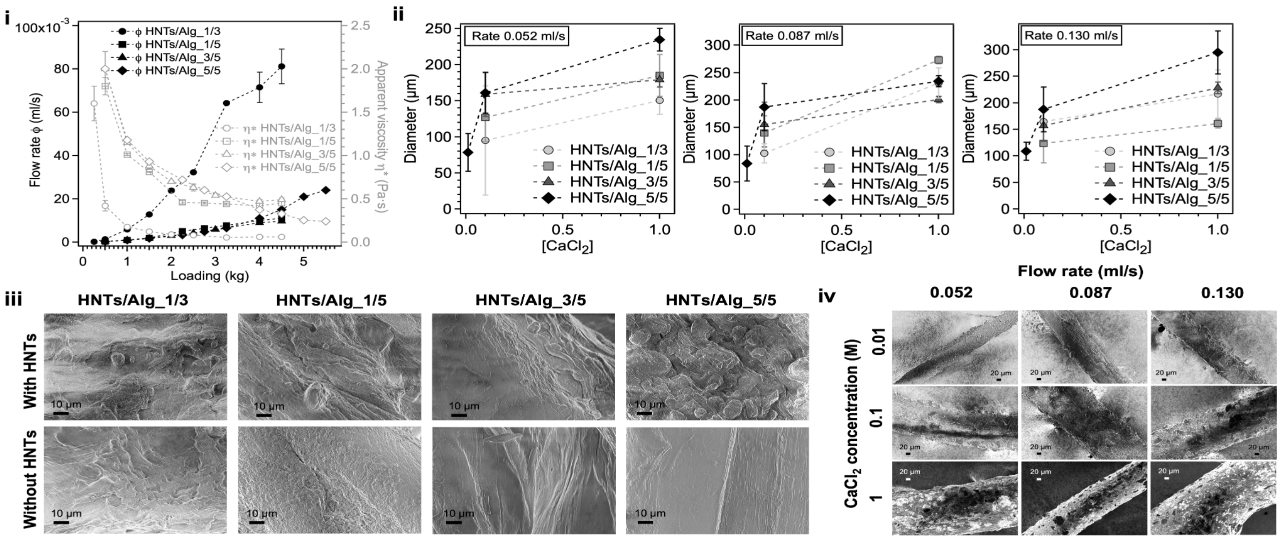


**Fig 1.** i) Flow rate and apparent viscosity as a function of the injection force; ii) Plot of the fibre's diameter as a function of the calcium chloride concentration at constant extrusion rate; iii) FE-SEM micrographs of the alginate fibres with and without HNTs (measurements were carried out on fibres prepared with the same processing parameters); iv) FE-SEM micrographs of the fibres prepared with formulation HNTs/Alg_5/5 at different flow rates and $CaCl_2$ concentrations.

### 4. Justification of experimental proposals request

The opportunity to use the FIB-SEM instrumentation will provide paramount information about the inner structure of the composite fibres, unravelling the dispersion of the nanotubes in the extruded biopolymer matrix. We plan on investigating three different biocomposites, prepared in different conditions and thus potentially displaying different distributions and orientations of the nanotubes. To this purpose, a working time of 1 day is expected to be needed.