Part B-1

1. Excellence

1.1 Quality and pertinence of the project's research and innovation objectives (and the extent to which they are ambitious, and go beyond the state of the art)

Motivation: Active matter, denoting collections of actively moving particles, is an emerging subject of study at the interface between physics, chemistry and biology. Due to constant energy injection at single particle scale, active matter is constantly driven **out of equilibrium** and behaves very differently from well-understood equilibrium systems. Over the past three decades, we have seen rapid growth of interest and investigations into active matter systems across a broad range of length scales, from large animals to tissues and cells.[1–4] The ways they self-organize, as evidenced in bird flocks, fish schools and bacterial swarms, are exhibiting **new states of matter**, which are strictly prohibited in equilibrium systems. **In the presence of boundaries with complex geometries**, active matter exhibits **more exotic** phenomena including wall accumulation, motion rectification, ratchet effect and upstream swimming. These phenomena not only pose new challenges to fundamental biology and ecology, but also imply world-changing applications in therapeutics and robotics.[5,6]

State of the art: Due to their non-equilibrium nature, active matter show rich phenomena in complex environments. One the one hand, **confinement** influences transport[8], rheology[9–12], pressure[13], spatial distribution[14–16] and collective motion[17–28] of active matter. **Curved confining walls, which are ubiquitous in biological systems**, show rich and intriguing confinement effect on active matter. Inspired by the collective motion of confined driven filaments[18], Woodhouse and Goldstein constructed a theoretical model, demonstrating that the combination of circular confinement and activity allows for the emergence of stable self-organized rotational streaming.[21] Such circular confinement was then realized by emulsions and elastomer chambers, where single vortical flows were observed.[24,25,27] Liu et al. recently showed intriguing oscillatory dynamics in a similar geometry.[20] Ravnik and Yeomans studied the dynamics of active nematics under cylindrical confinement using simulation based on continuum equations. They showed that the collective vortical flows not only emerge along the cylinder axis (as shown by Woodhouse and others), but also within the plane of the cylinder.[23] Fily and co-workers developed a statistical theory for non-aligning, non-interacting active particles to study spatial distributions under strong confinement. They showed that in such confinement, particle concentrations at boundaries were proportional to the local curvature.[29] Nikola and co-workers showed, using particle-based theory and simulation, that not only the collective motion, but also the shear stress exerted by active particles on confining walls was wall-dependent.[13] These works show that curved confining walls alter the behavior and macroscopic properties of active matter. In particular, **the only key parameter of curved confinement, curvature**, has been shown to play an important role in particle spatial distributions[29] and collective motions[24]. On the other hand, complex-shape passive objects show intriguing persistent and directed motions in active baths, which can be used for the extraction of work.[30–33] Angelani and co-workers, using numerical simulations, showed that asymmetric gear-like objects spontaneously rotated in a directed way in active active baths, forming the concept of "micromotor" powered by active matter.[32] This idea was then realized in experiment in experimental[30,31] and theoretical grounds.[33]

The interplay between complex environments and active matter suggests **a possibility to control and engineer active matter** by carefully designing the confinement structures. While many exotic phenomena under confinement have been demonstrated, further understanding requires systematic investigations with well controlled model systems. Therefore, we propose the following project, which, we believe, is a crucial step to bridge the gap between fundamental studies and real-life applications of active matter.

Objectives: We seek to understand the interaction between complex geometries and the behavior of active matter, using a controllable model experimental system. Specifically, we want to (i) understand how confinement geometries influence the fluctuations of spherical particles. Systematic understanding will be gained by investigating the relevant dynamical properties of passive objects that are subject to various confining geometries. In addition to this simplest scenario, we also seek to understand the behavior of complex-shape objects in confined active bath. In particular, we seek to (ii) understand how confining wall curvature influences the fluctuations of complex-shape particles and to (iii) understand self-assembly of complex-shape passive particles in confined active baths. We expect to observe novel self-assembly principles in the presence of active matter under confinement.

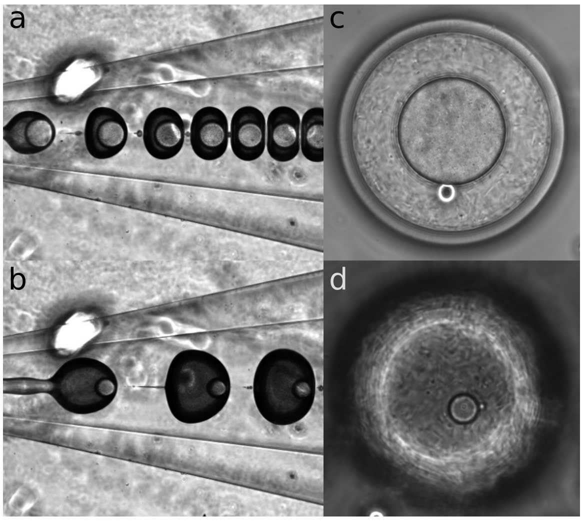

Figure 1: Double emulsions with swimming bacteria in the middle aqueous phase. (a, b) Double emulsions in a micro-capillary device, operating at different flow rates. (c, d) Collected double emulsions observed with a higher magnification.

Methodology: Emulsions offer controllable and high throughput experimental systems for studying confinement effect. Recent progress in **microcapillary device** allow us to produce double emulsions with well controlled compositions and dimensions.[34] To achieve the objectives, we will build a model experimental system based on (double) emulsions, to systematically study the interplay between complex environments and active matter.

To respond to objective (i) “understand how confinement geometries influence the fluctuations of spherical particles”, we will study the fluctuations of the inner droplets in oil/water/oil double emulsions with bacterial suspensions in its middle aqueous phase as the active bath (Fig.1). The double emulsion system naturally imposes the simplest form of curved confining walls: a spherical shell. The sizes of both outer and inner droplets can be controlled by operating the device at different flow rates, as demonstrated in Fig.1.

To respond to objective (ii) “understand how confining wall curvature influences the fluctuations of complex-shape particles”, we will use the emulsion systems as in (i), but replacing the inner droplets with complex-shape particles and study its fluctuations under different confining curvatures. The complex-shape particles will be fabricated using the **3D nano-printer** in ESPCI.

To respond to objective (iii) “understand self-assembly of complex-shape passive particles in confined active baths”, we will encapsulate multiple complex-shape particles into a droplet comprising active matter. We expect to observe novel self-assembly principles in the presence of active matter under confinement.

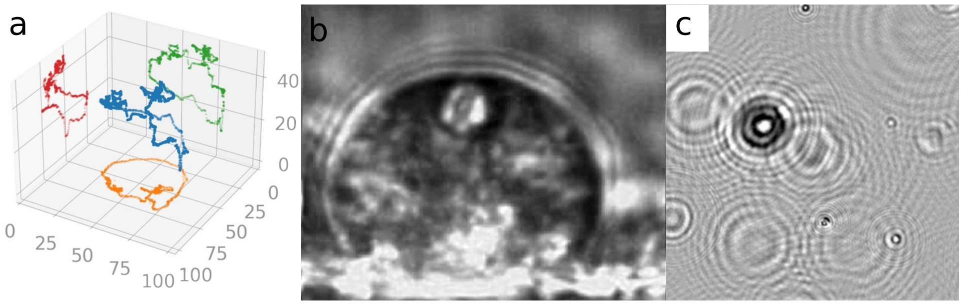

Figure 2: 3D imaging techniques. (a) 3D inner droplet trajectory obtained from 3D Lagrangian tracking system. (b) A side view of a double emulsion obtained from the tilted confocal microscope. (c) A holographic image.

The motions of the inner droplet and other objects confined in emulsions are inherently 3D. Therefore, **3D microscopy** is an experimental challenge in this project. There are two techniques for 3D microscopy hosted in ESPCI: (i) Lagrangian 3D Tracking System (preliminary tracks in Fig.2a) and (ii) tilted confocal microscope (Fig.2b). The former is developed by Prof. Eric Clement’s lab at

PMMH for studying how bacteria explore in 3D space. The need of this project will facilitate the development of the system for tracking more complex objects, in addition to bacteria. The latter is developed in the Gulliver lab. In addition, **holographic microscopy**, which I have worked with during PhD at the University of Minnesota in collaboration with Prof. Jiarong Hong, will also be a candidate method to achieve 3D imaging (Fig.2c).

Expected outcome: Combining the microfluidic techniques and the advanced 3D microscopy, we expect to generate a complete set of fluctuation data (trajectories) of passive oil droplets in confined active bath of various confining curvature and active particle concentration. An automated 3D tracking system, incorporating artificial intelligence technology, will also be demonstrated. Fluctuations of non-spherical passive particles will be compared with spherical particles. Self-assembly of non-spherical particles in active matter will be demonstrated. These experimental data are expected to facilitate further development of active matter models, and eventually close the gap between theory and application.

1.2 Soundness of the proposed methodology (including interdisciplinary approaches, consideration of the gender dimension and other diversity aspects if relevant for the research project, and the quality of open science practices, including sharing and management of research outputs and engagement of citizens, civil society and end users, where appropriate)

Overall methodology: Our proposed research aims to systematically study the dynamics of particles in swimming bacterial bath under different confining curvature. Taking advantage of the microfluidic **double emulsion** experimental system, we can access **a wide range of the key parameter – curvature**. To overcome the key experimental challenge, **3D imaging**, we have three candidate methods (3D tracking, tilted confocal and holography) ready to be tested. These two advantages will enable us to systematically study how curvature influences the fluctuations of an active bath. Furthermore, **3D nano-printer** hosted in ESPCI enables me to produce micro-particles with arbitrary shapes. Taken together these techniques, the objectives of the proposed research can be achieved.

Integration of disciplines: This project is inherently interdisciplinary, since it requires combining expertise in: i) biochemistry and biophysics to produce bacterial suspensions with controlled properties, ii) hydrodynamics and statistical physics to study their collective motion, iii) microfluidics and surface chemistry to produce complex confining environments, iv) topology and geometry to analyze the patterns emerging in these constrained environments. I have a solid physical and biological background, as well as experimental skills to carry out this project. In addition, my experience in studying active matter under confinement ensures that I am able bring together the expertise from me and the suggested host to achieve the objectives of the proposed project.

Gender dimension and other diversity aspects: ?

Open science practices: Host softwares and algorithms and GitHub. Host data on open data repositories, like HEPData. Publish milestone works on pre-print database, like arXiv. Youtube videos.

Research data management: Data will be organized and uploaded to open repositories every month with a report describing the purpose and statistics of them. A separate document will be maintained to account for the status and evolution of the whole data set over time.

1.3 Quality of the supervision, training and of the two-way transfer of knowledge between the researcher and the host

Qualifications and experience of the supervisors (Projects? Awards? Publications? Collaborators? Former students, number and careers?

Prof. Eric Clement and his group are world leading experimentalists on the study of active fluids, with special expertise in the behavior of active particles in complex environments. He has been the co-PI of **ANR project BacFlow (2015-2020)**, studying *Hydrodynamic transport and dispersion of bacterial suspensions: from the micro-hydrodynamic scale up to porous media*. He has also been the co-PI of **Joint Research Program (PRC) CNRS-Royal Society (2017-2019)**, focusing on *Macroscopic and Microscopic properties of active matter*. He has published more than 115 papers in international journals. Prof. Clement has developed many close collaborations with researchers with on an international level. Current collaborators include, for example, Prof. Rodrigo Soto and Maria-Luisa Cordero (Universitad de Chile) and Prof. Jasna Brujic (New York University). **#level of experience in training/supervising.**

Prof. Anke Lindner is an internationally acknowledged specialist in the interactions between anisotropic particles and flow. She has been the leader of many projects, including the ongoing PaDyFlow (particle dynamics in flow of complex suspensions) that was selected for a consolidator ERC grant (2016-2021). She is also part of the collaborative ITN project CALIPER starting in September 2019. She has been awarded the “Maurice Couette” award and CNRS silver medal for her study of complex fluids in flow. Former students of hers now work as research scientists at companies such as “Total”, “Arkema” and small start-up companies as “DNA script” and “Aratinga Bio”. Many of them pursue a career in academia and hold positions as assistant professors at Montpellier University, Sorbonne University, Perdue University or University of Dortmund. She has published 63 primary research papers, 1 book chapter and 1 patent which is currently pending. To complement her experimental research Prof. Lindner has developed many close collaborations, acknowledged by common publications, with theoreticians on an international level. Ongoing collaborations comprise for example Mike Shelley, Courant Institute, and Simons Foundation New York, USA, David Saintillan (UCSD, USA), Howard Stone, Princeton University, USA and François Gallaire, EPFL, Switzerland. Her knowledge will be valuable, especially when we study the motion of non-spherical particles in active baths.

Prof. Teresa Lopez-Leon is an expert in studying the effect of confinement and curvature on the properties of liquid crystals. She has been the leader of XX project. She has been awarded XX award for her contribution in XX. She has published XX research papers in international journals. She has developed collaborations with XX and XX. She has advised XX PhD students and XX postdocs. She has the technique of producing double emulsions readily available for the proposed research. Her knowledge on confinement effect in general will provide new perspectives to understand the behavior of confined active matter.

Two-way transfer of knowledge between the researcher and the host organisation

There is a great potential for transfer of knowledge between me, the host laboratories at the PMMH and Gulliver labs in ESPCI.

On the one hand, the hosts have knowledge on active matter, complex fluids and microfluidics, which forms an ideal combination of knowledge and techniques that are necessary for conducting the proposed research, and will be a great opportunity for me to acquire new knowledge and to carry out an interdisciplinary and impactful study.

On the other hand, my knowledge of studying the diffusion, rheology and collective motions of bacterial suspensions, combined with working experience of microfluidics, advanced microscopy and image analysis together aligns perfectly with the required skill set for carrying out the proposed research. Additionally, I can share with the host a mutant strain of *E. coli* whose motility can be controlled by light. I genetically engineered the mutant strain during my PhD at the University of Minnesota, and I believe it can lead to a more versatile experimental system and more concrete outcome of this research project. I will also share holographic imaging technique with the host lab, which can be a candidate method for 3D imaging.

Planned training activities for the researcher (scientific aspects, management organisation, horizontal and key transferable skills...)

#meeting with supervisors #seminars, journal clubs and conferences #...

1.4 Quality and appropriateness of the researcher's professional experience, competences and skills

I obtained bachelor's degree from Tsinghua University in 2014 studying *Self-assembly of Janus nanoparticles* and PhD degree in 2021 with my thesis entitled *Novel properties and collective phenomena of active fluids*. During these investigations, I have acquired expertise of various experimental techniques, including confocal microscopy, microfluidics, genetic engineering, particle synthesis and image analysis, as well as experience in numerical simulation. These studies have led to 7 publications in international peer reviewed journals, with additional papers under preparation.

The proposed research topic matches well with my experience and skill set. I will be able to quickly adapt my expertise to the new work environment. In the mean time, the supervision from world leading scientists and being able to use the state-of-the-art equipments will benefit my career development enormously.

2. Impact

2.1 Credibility of the measures to enhance the career perspectives and employability of the researcher and contribution to his/her skills development

4 aspects: technical, mentoring, research and project management

I aim to become an independent researcher in academia to advance the fundamental understanding of active matter, which has potential to overcome challenges in therapeutics and robotics. By working with the host groups at PMMH and Gulliver in ESPCI on investigating into cutting-edge problems in the interdisciplinary field of active matter, my career perspectives and employability will be enhanced in several aspects:

- Improve my technical skills in specific methods
- Improve my communication skill by presenting my research to audience of different background and expertise
- Learn to define and manage a research project by setting objectives, planning and making adjustments to the objectives and plans
- Improve mentoring skills by working together with junior students

2.2 Suitability and quality of the measures to maximise expected outcomes and impacts, as set out in the dissemination and exploitation plan, including communication activities

- conferences (scientific groups); outreach activities (public);

2.3. The magnitude and importance of the project's contribution to the expected scientific, societal and economic impacts

Expected scientific impact: #we demonstrate an interdisciplinary approach, bringing together active matter, 3D imaging and microfluidics, to construct a well-controlled and biomimetic confined environment to study the confinement effect on the self-organization and complex interactions of active matter and particles #we create new knowledge on how confining curvature influence the behavior of active matter #we reinforce the 3D Lagrangian tracking system by incorporating AI technologies

Magnitude: how many people are impacted? Need to survey how many active matter research groups there are in the world?

Economical/technological and societal impact: this research has the potential of being applied in therapeutics and robotics, which would have economical/technological and societal impact. However, due to its fundamental nature, direct foreseeable impact is hard to claim.

3. Quality and Efficiency of the Implementation

3.1 Quality and effectiveness of the work plan, assessment of risks and appropriateness of the effort assigned to work packages

Milestones	Description	Month
M1	Set up protocols of double emulsion generation, 3D imaging and data analysis	6
M2	Data acquisition and analysis	14
M3	Fabrication of anisotropic particles	17
M4	Acquiring self-assembly data of anisotropic particles in active bath	21

Work package number	1	Start month	1	End month	8
Work package title	Dilute temperature				
Objectives	Measure the "effective temperature" of confined dilute active bath				
Description	- Demonstrate the capabilities of the proposed methods: producing double emulsions with microfluidic device, recording 3D motion of inner droplets with Lagrangian 3D tracking system and confocal microscopy - Set up an experimental protocol - Collect inner droplet trajectory data in various curvatures - Write programs to extract the trajectories from images				
Methodology					
Deliverables	Report on specific activities or results, data management plans, ethics or security requirements				Month
	D1.1	Set up experimental protocol			2
	D1.2	Collect and analyze trajectory data			5
	D1.3	Conference – APS DFD meeting USA, November 2022			8

Work package number	2	Start month	7	End month	15
Work package title	Concentrated temperature				
Objectives	Measure the "effective temperature" of confined concentrated active bath				
Description	- Measure effective temperature at a range of different bacterial concentrations, spanning from random swimming to collective motion, at fixed geometry - Collect inner droplet trajectory data in various geometries and concentrations to complete a "phase diagram"				
Methodology					
Deliverables	Report on specific activities or results, data management plans, ethics or security requirements				Month
	D2.1	Make double emulsions with dense bacterial shell			9
	D2.2	APS March meeting USA, March 2023			12
	D2.3	Write towards a manuscript			15

Work package number	3	Start month	15	End month	24
Work package title	Complex-shape object				

Objectives	Measure the dynamics of complex-shape particles in active bath		
Description	- Fabricate complex-shape particles using 3D nano-printer - Measure fluctuations of single complex-shape particles in active emulsion - Investigate self-assembly of complex-shape particles in active emulsion		
Methodology			
Deliverables	Report on specific activities or results, data management plans, ethics or security requirements		Month
	D3.1	Fabrication of complex particles using 3D printer	18
	D3.2	APS DFD Meeting, USA 2023	20
	D3.3	Emulsions of bacterial suspensions with single particle	22
	D3.4	Emulsions of multiple particle	23
	D3.5	APS March Meeting, USA 2024	24

The work plan of the project is outlined in the Gantt chart below (April 2022 – April 2024):

Month	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24
WP1																								
WP2																								
WP3																								
Management																								
Milestone						M1								M2			M3				M4			
Deliverable		D1.1			D1.2			D1.3	D2.1			D2.2			D2.3			D3.1		D3.2		D3.3	D3.4	D3.5
Secondment																								
Conference								DFD				APS								DFD				APS
Seminar																								
Outreach																								

3.2 Quality and capacity of the host institutions and participating organisations, including hosting arrangements

The host institution ESPCI in Paris is a prestigious institute of research and higher education in science and engineering, with five Nobel Prize winners associated with the School - Pierre Curie, Marie Curie, Frédéric Joliot-Curie, Pierre-Gilles de Gennes, and Georges Charpak. It hosts 9 high level laboratories with many world class researchers focusing on active matter, the topic I am pursuing. It also has a strong international scientific committee that is composed of eminent, internationally recognized figures from the academic world, research and industry. As a champion for scientific excellence, ESPCI Paris is actively working to take science and the love of science beyond the labs and lecture halls. Inspiring an appetite for knowledge by fostering curiosity and interest in science and experimentation is increasingly vital today, in light of an obvious lack of interest in the western world for scientific studies and careers. A renewed interest in science today will contribute to tomorrow's knowledge-based economy. The seven ESPCI Paris auditoriums are the venue for a variety of lectures, ranging from the Experimental Lectures series with unique lab-bench demonstrations to the "Les Chantiers du Savoir" series which tackles more challenging topics. The School campus also hosts popular events for the general public all year round, including the Sciences on the Seine Festival, Fête de la Science, Researchers' Night, and Science Académie. The host labs have regular activities that can help me integrate in the team and the institution, such seminars, lab trips, meetings at various levels. The labs are also equipped with top-level infrastructure and instruments that are essential for carrying out the proposed research. The ESPCI Paris also provides its employee with services and benefits from the CNAS (National Social Action Committee), services to improve the material and moral conditions of employees. The ESPCI Paris being a funder member of the PSL University, I will have access to the full scope of PSL services (student and campus services, events, sports, culture, etc).

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