CERM
Centro Risonanze Magnetiche
FIRENZE

CIRMMP
FIRENZE

SCIENTIFIC ANNUAL REPORT

2022

UNIVERSITÀ DEGLI STUDI FIRENZE

Contents

Foreword

The year 2022 was an important year for our Infrastructure. The new 1.2 GHz NMR instrument (28.2 T magnet), operative since Spring 2020, is benefitting our research as well as the research of our users. The significant improvement of the research performed at CERM/CIRMMP is detailed in the research activity session of this report: while still maintaining a high number of publications (74) their quality has significantly improved, with an average publication impact factor of about 10, that is almost doubled compared to 2021. This is due not only to the presence of excellent publications (***Chem. Rev., Nature, Nature Chemistry, Angew. Chemie, J. Am. Chem. Soc***…), but to a generalised quality improvement. Indeed, the publications with an impact factor above 5 are 57% of the total! The landscape of topics is dominated by structural and cellular biology, but material science, new NMR methods and metabolomics are also well represented.

Scientific research has always represented our strength, and it is attracting new users, who often become collaborators. Indeed, users find in our infrastructure not only an excellent NMR service but also the know-how to properly analyse the data and translate them into scientific results.

Despite the limitations due to Covid-19 pandemic that were still present in the first half of 2022, we were able to organise the Chianti Workshop "Opening New Doors on Magnetic resonance" (Principina Terra (GR, Italy) June 19th-24th 2022). This biannual meeting, which was delayed from 2020 to 2022 because of the pandemic, confirmed itself again as one of the major events in NMR and Magnetic Resonance of 2022. This was also the occasion to restart with meetings in presence and promote our infrastructure internationally.

In parallel, the role of CERM/CIRMMP in the European Research Infrastructure scenario was further reinforced. CERM/CIRMMP is the Italian centre (Instruct-IT) of Instruct-ERIC, an ESFRI Landmark. The key role of the Italian centre within Instruct-ERIC was strongly reaffirmed thanks to our strong involvement in most Instruct-ERIC activities, with a leading role in the Council and in the Executive Committee, as well as in the support to training, internationalisation, access and data management. The latter within EOSC-Life project, to enable the management, storage and reuse of data in the European Open Science Cloud (EOSC).

The activities of CERM/CIRMMP related to Instruct-ERIC were framed also within the canSERV and the ISIDORe projects, which coordinates the Biological and Medical European Research infrastructures (BMS RIs) to create platforms for access provision tackling cancer research and infectious disease outbreak, respectively.

At the national level, the activities of **Instruct-ITALIA**, the national consortium of infrastructures providing access to national users in structural biology, started in 2020, has rapidly increased, as detailed within this report. Instruct-ITALIA is a powerful tool for the Italian researchers who now have access to complementary techniques on different research fields: from NMR to Cryo-EM, to optical microscopy and X-ray techniques.

Through the recently funded NRRP project ITACA.SB we secured funds for potentiating the Italian center of Instruct-ERIC and for implementing new facilities in the country in order to provide the Italian Structural Biology community with methodologies to meet their needs for performing top-level research with the overall goal of strengthening the Italian role in Instruct-ERIC by ensuring the European user community high-end services at the Italian center and, boosting the exploitation of Instruct-ERIC resources in general.

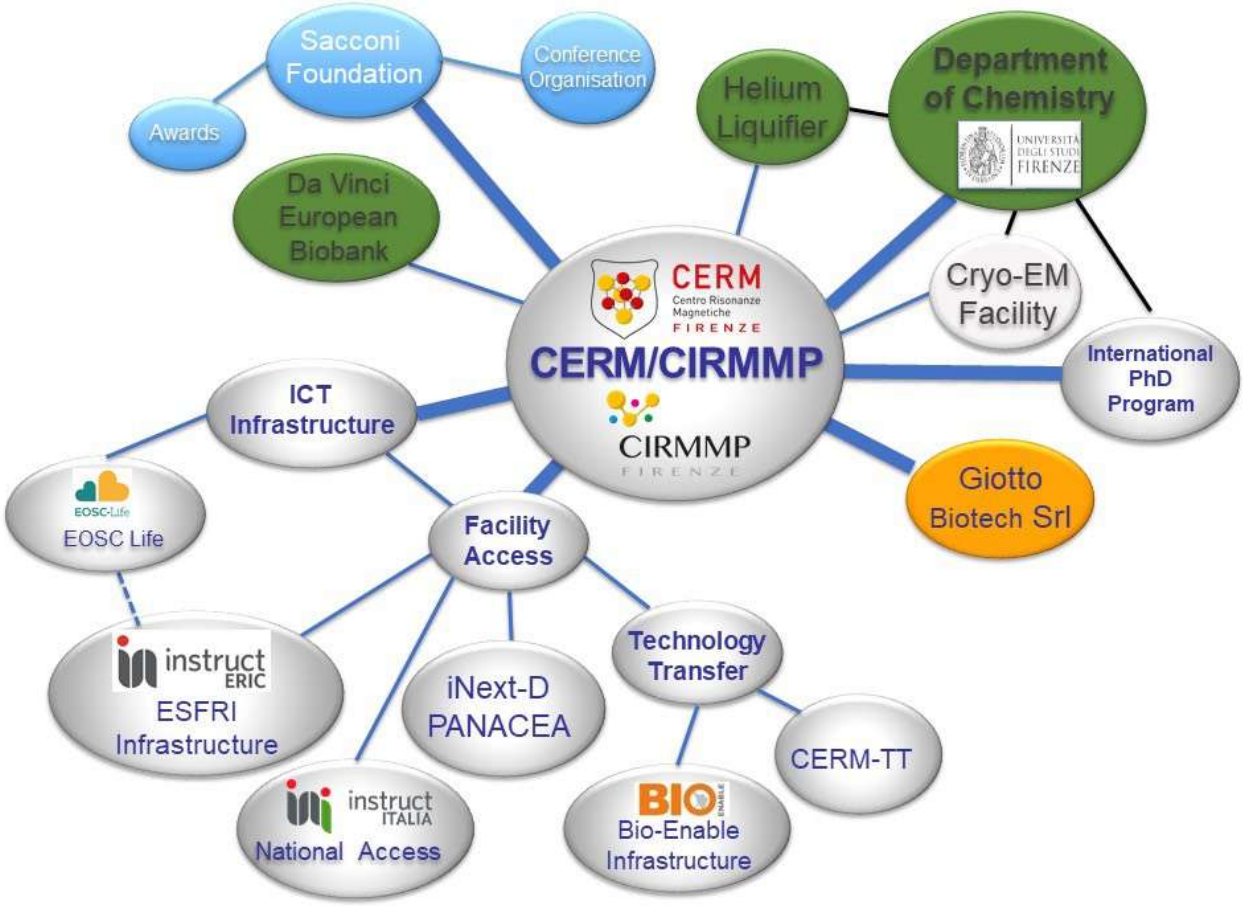
Figures

Also for 2022, the Italian Ministry of Education, University, and Research (MIUR) confirmed its support to the Italian centre of Instruct-ERIC within the International Action of the FOE funding. CERM/CIRMMP Investments and costs in 2022 amounted to € 3.185.000,00: € 300.000,00 towards training and education, € 1.840.000,00 for new equipment, and € 835.000,00 towards research activities. An additional € 210.000,00 covered operational costs.

The actual replacement value of the instrumentation at CERM/CIRMMP is close to € 58.000.000,00. In 2022, in addition to the faculty staff, the body of researchers included 22 PhD students, 16 postdoctoral scientists, and 16 undergraduate students.

We wish to thank all the people that contributed to make CERM/CIRMMP what it is today and who continue to drive it forward, and all the Institutions that provided their support.

Prof. Claudio Luchinat Prof. Lucia Banci


Sacconi Foundation
Conference Organisation
Awards
Helium Liquifier
Department of Chemistry
UNIVERSITÀ DEGLI STUDI FIRENZE
Da Vinci European Biobank
CERM
Centro Risonanze Magnetiche
FIRENZE
CERM/CIRMMP
CIRMMP
FIRENZE
Cryo-EM Facility
International PhD Program
ICT Infrastructure
Giotto Biotech Srl
EOSC-Life
EOSC Life
Facility Access
instruct ERIC
ESFRI Infrastructure
iNext-D PANACEA
Technology Transfer
CERM-TT
instruct ITALIA
National Access
BIO
Bio-Enable Infrastructure

Who we are

Introduction

CERM, Centre for Magnetic Resonance, is a ***scientific institution for research***, technology transfer and higher education of the University of Florence. It operates in synergy and collaboration with the Inter-University Consortium for Magnetic Resonance of MetalloProteins (CIRMMP) which includes three Italian Universities: Florence, Siena, and Bologna. CERM/CIRMMP is an ***infrastructure for Life Sciences*** with a particular focus on structural biology and specialisations in NMR spectroscopy, bioinformatics, molecular and cellular biology, novel drug and vaccine design, and metabolomics. Nevertheless it is open towards interfaces with other research fields, for example new material and biomaterial development, contrast agent and MRI techniques, and ICT technology.

Being a leading laboratory at both national and international level, CERM/CIRMMP receives funding from competitive project calls from the Tuscan Regional Government, the Italian Ministry of Higher Education and Research (MUR), and the European Commission (EC), as well as from private institutions.

The core technology at CERM/CIRMMP is NMR spectroscopy, and the onsite instrumentation is among the most advanced in the world. Since 1994 a European transnational access service, funded by EC, flanked the service provision at national level, that was already active since 1990. This long term expertise places CERM/CIRMMP at the top of the list among the European NMR Research Infrastructures in Life Sciences. CERM/CIRMMP actively stimulates interactions between private industry and public research institutions such as Universities, National Research Council (CNR) Institutes, and European counterparts, promoting synergistic activities such as collaborations and services to SMEs.

CERM/CIRMMP is the Italian Centre of Instruct-ERIC, which is the European research infrastructure in integrated structural biology defined in the European Strategy Forum on Research Infrastructures (ESFRI) Roadmap. The Italian centre of Instruct-ERIC, CERM/CIRMMP, is also included in the "*Roadmap Italiana delle Infrastrutture di Ricerca di interesse Pan-Europeo*" since 2010. In parallel, ***CERM/CIRMMP*** is also the core centre of the ***Instruct-ITALIA*** network, a new infrastructure to promote and to foster an integrated approach at the national level providing access to X-ray crystallography, NMR, Cryo-EM, as well as protein expression and crystallisation. ***Instruct-ITALIA*** has started its activity in early 2020, promoting a more effective interaction within Italian structural biologists, as well as at supporting access to the facilities of its national network.

Under the Next Generation EU scheme, Italy is receiving resources (NRRP Program), and through this scheme, CERM/CIRMMP scientists participated in several projects. Of high relevance and impact is the awarding to CERM/CIRMMP, together with a few CNR labora-

tories in Italy, of the project, named ITACA.SB, which allocated significant resources to reinforce the Italian Center of Instruct-ERIC and Integrated Structural Biology in Italy.

CERM/CIRMMP is also an e-infrastructure, participating in a European GRID-based platform, providing access to user-friendly platforms and CPU resources for a broad range of services for structural biology. These services leverage technologies created in the context of EOSC (European Open Science Cloud) development initiatives and are made available through the EOSC marketplace (for example, https://marketplace.eosc-portal.eu/services/eosc.wenmr.amber-based_portal_server_for_nmr_structures_amps-nmr).

CERM/CIRMMP also promoted the creation of the Da Vinci European Biobank, a "biobank of biological samples and biomolecular resources". CERM/CIRMMP has also developed a centre for research and technology transfer: CERM-TT, funded by the Tuscany Region. Finally, CERM/CIRMMP is coordinating the activities of Bio-Enable, a distributed Infrastructure promoting technology transfer to industry and funded by the Regional Government of Tuscany in the frame of POR FESR 2014-2020.

CERM/CIRMMP is located in the Scientific Campus ("Polo Scientifico") of the University of Florence in Sesto Fiorentino, an area just west of the city of Florence. The campus borders Florence International Airport and yet is a mere 15 minutes from the centre of Florence, world-renowned cradle of renaissance art and culture.

The Infrastructure

CERM/CIRMMP labs

The CERM/CIRMMP building covers an area of 3000 square metres hosting a number of laboratories, offices, and common rooms. The flagship of the Center is the impressive collection of NMR spectrometers which feature the largest magnetic field range in the world (from 1.2 GHz (installed in early 2020, is the first instrument in the world at this field) to the earth magnetic field) and ranks it among the best equipped laboratories in the world. The NMR labs are flanked by molecular and cellular biology laboratories that are optimised for NMR sample production. A complete list of the instruments available at CERM/CIRMMP is reported at pag. 37. In addition to the main building, further 500 square metres in adjacent buildings are available to CERM scientists and researchers scientifically associated to CERM/CIRMMP: laboratories at the Department of Chemistry Ugo Schiff and at GENEXPRESS; DA VINCI European Biobank; X-rays facilities; Helium liquefier. www.cerm.unifi.it

Instruct-ERIC

CERM/CIRMMP is an Instruct-ERIC Centre. Instruct-ERIC is the European research infrastructure in integrated structural biology, making cutting-edge technologies and high-end methods in a palette of tools for structural characterisation available to users.

Structural biology is one of the key approaches that contribute to the understanding of the molecular and cellular functions. The main experimental technologies are complementary, and increasingly link detailed atomic structure with cellular context. Structural biology is currently in the middle of a revolution enabled by significant advances in various technologies (direct electron detectors in EM, advances in synchrotron sources and detectors, XFELs, ultra-high field NMR, super-resolution cryo-light miscroscopy).

Instruct-ERIC buildups on a number Centres featuring the most advanced structural biology instrumentation and top-level expertise in the various methods. Instruct-ERIC offers a **single point of access** to both multiple techniques integrated at one Center or over various Centres, or to some Centres specialised in specific techniques. www.instruct-eric.eu

Instruct-ITALIA is the Italian Infrastructure for Integrated Structural Biology. It consists of a core of excellent research institutions and large centres that have a proven track record in structural biology and in service and expertise provision to users. Instruct-ITALIA aims to serve as a national consortium covering all main areas of structural biology research within Italy. https://www.cerm.unifi.it/instruct-it/

CERM TT

The CERM TT Competence Centre *dedicated to Ivano Bertini*, founder of CERM, was established in response to the request of the Tuscany Region to make available to the industries and production companies in Tuscany centres of technology transfer, innovation clusters with advanced equipment and skills to boost the economic growth of the region.

CERM TT strengthens and optimises the service offered by CERM/CIRMMP to the industry of the area: NMR instrumentation and advanced computing, a molecular biology laboratory for the production of proteins, scientific expertise and excellence, together with the maximum protection of industrial IP.

CERM TT performs analytical services and research and development (R&D) for companies. In particular it offers the following services:

- screening of drug candidates and drug-target interaction studies;
- smart design of drugs;
- analysis of pharmaceutical formulations.

Bio-Enable

BIO-ENABLE is a "distributed research infrastructure" led by CERM/CIRMMP and includes a few other Centres in Tuscany. BIO-ENABLE provides access to equipment and expertise to support industrial research and innovation. Tuscan companies operating in fields ranging from pharmaceuticals to biotechnology, from vaccines to biomaterials, from food to nanotechnology, can exploit the services of BIO-ENABLE in the development of their activities to be competitive at international level.

CERM leads the BIO-ENABLE consortium composed by:

- Magnetic Resonance Center (CERM/CIRMMP, coordinator)
- Institute of Neurosciences of the CNR – Pisa;
- BioRobotics Institute of Sant'Anna School of Advanced Studies - Pisa;
- Department of Medical Biotechnologies – University of Siena.

BIO-ENABLE can provide support at various levels and through different types of contracts: from simple access to instrumentation to specific types of advice, help and assistance to industrial research. BIO-ENABLE guarantees total confidentiality of the data collected at the various platforms, both during the course of the analysis and in the management and archiving of the data. www.bio-enable.it

Funded projects

CERM/CIRMMP cooperates at the international level with several universities, research institutions, and private industries with which it is involved in numerous research projects funded by the European Commission. Projects ongoing during 2022 are:

Remote NMR (R-NMR): Moving NMR infrastructures to remote access capabilities (HORIZON-CSA grant agreement n. 101058595, 01/07/2022-30/06/2025)

ISIDORe Integrated Services for Infectious Disease Outbreak Research. Grant agreement ID: 101046133 (01/02/2022-31/018/2025)

BeYond-COVID (BY-COVID) Grant agreement ID: 101046203 (1/10/2021-30/09/2024)

PANACEA "A Pan-European Solid-State NMR Infrastructure for Chemistry-Enabling Access", (H2020, contract n. 101008500, 01/09/2021-31/08/2025)

ITN "GLYTUNES – A multidisciplinary training network for the bioinspired development of glycomimetics tuning the Siglec-Sialoglycan axis" n. 956758 (01/03/2021-28/02/2025)

H2020 -INFRAIA iNEXT-Discovery - Structural Biology Research Infrastructures for Translational Research and Discovery (#871037) https://inext-discovery.eu

ITFoC Information Technology: The Future of Cancer Treatment https://itfoc.eu/

THE INFRASTRUCTURE

TRANSVAC2 - Improving and accelerating vaccine development in Europe

TIMB3 “Twin to Illuminate Metals in Biology and Bio-catalysis through Biospectroscopy” (H2020, contract n. 810856, 01/09/2018-31/08/2021)

EOSC-Life "Providing an open collaborative space for digital biology in Europe" (H2020, contract n. 824087, 01/03/2019-28/02/2023)

HIRES-MULTIDYN "Multiscale Dynamics with Ultrafast High-Resolution Re (H2020, contract n. 899683, 1/10/2020-30/09/2024)

EGI-ACE: Advanced Computing for EOSC (Horizon 2020 grant agreement n. 101017567, 01/01/2021-30/06/2023)

ITACA.SB: Potentiating the Italian Capacity for Structural Biology Services in Instruct-ERIC (Call MUR 3264 - M4/C2/L3.1.1 - ID Proposal IR0000009)

Italiadomani
PIANO NAZIONALE DI RIPRESA E RESILIENZA

Funded by the European Union
NextGenerationEU

NRRP and CERM/CIRMMP

The CERM/CIRMMP Infrastructure is also strongly involved in the ***National Recovery and Resilience Plan (NRRP)***, attending to several projects directly as infrastructure or with the direct involvement of its researchers. In particular *ITACA.SB* is an infrastructure project empowering the Structural Biology services offered by *Instruct-Italia.*

ITACA.SB: Potentiating the Italian Capacity for Structural Biology Services in Instruct-ERIC

The *ITACA.SB* project aimed at maintaining the excellence of NMR services of the Italian Centre of *Instruct-ERIC*, empowering and integrating the service capacity for protein production and biophysical characterisation (NMR, Crystallography,…), potentiating data management, and computational tools available for widening the exploitation of structural biology technologies. Furthermore, it promotes a reduction of the environmental impact of NMR structural biology activities at *Instruct IT*. Last but not least, *ITACA.SB* promotes outreach and networking for a stronger Italian SB community.

National Recovery and Resilience Plan

Call MUR 3264/2021 – M4/C2/L3.1.1

Applicant: Consiglio Nazionale delle Ricerche (CNR) **Co-Applicant:** Università degli Studi di Firenze

Starting date: 01.11.2022

Length: 30 months 40% of funds to South Italy infrastructures

Total amount: 17.977.617,89€

CERM@UniFi: 9.388.657,28€
CNR: 8.588.960,61€

Research Facilities involved in ITACA.SB: CERM/CIRMMP (Florence), IC: Institute of Crystallography (Bari, Caserta, Catania), IBPM: Institute of Molecular Biology and Pathology (Rome), ICB: Institute of Biomolecular Chemistry (Catania), IPCB: Institute for Polymers and Composite (Catania).

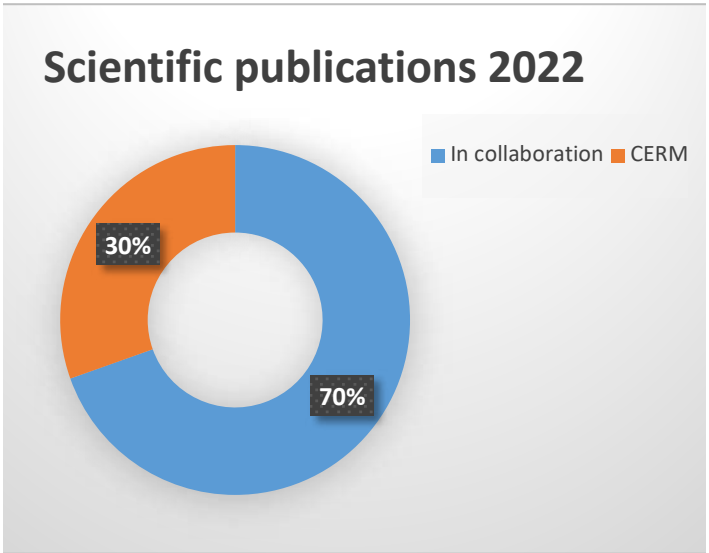
Other NRRP Projects with CERM/CIRMMP Researchers involved		
CN3 - Spoke 5	Pierattelli - Fragrai	
THE - Spoke 4	Cantini	Nanotechnologies for diagnosis and therapy
THE - Spoke 6	Rosato	Omics for cancer
THE - Spoke 8	Pierattelli - Felli - Parigi - Fragai - Allegrozzi	
PE8 - Spoke 2	Tenori - Vignoli	Improving the understanding of the biology of ageing
PE12 - Spoke 6	Felli	Mechanisms of neuronal cell degeneration and drug dependent reversal

Research Activities

Introduction

During 2022, a number of research projects have been carried out, either as an extension of the activities of previous years or as new projects. Most of these projects receive specific funding from national and/or European organisations.

NMR is the core technology of CERM, but year by year CERM research has been oriented more and more toward new applications and toward the integration with other techniques. This is one of the principles of the Integrated structural biology that underlies the Instruct-ERIC consortium, where CERM/CIRMMP is the Italian node. In the following pages it can be appreciated how much the present research in CERM/CIRMMP is spanning a wide range of applications, from the structural biology to the bioinformatics methods and Information Technology, from paramagnetic NMR methods to the development of new contrast agents for MRI, from the metabolomics and biomedicine to the development of new solid-state NMR methods for the characterisation of material surfaces and biomaterials.



In line with our mission to develop NMR as a technique and to integrate NMR with other techniques, most of our publications were done in collaboration with other research groups (70% of the overall number of publications). During 2022 we published 74 papers in international peer-reviewed journals, with several publication on very high impact factors journals (*Nature, Chemical Review, Nature Structural and Chemical Biology, J. Am. Chem. Soc.*,..).The average impact factor substantially improved from last year passing from 5.68 to 9.57, which is substantial jump in quality of the publications with 56% of the publications on journals with impact factors higher than 5. A complete list of publications is available at page 47.

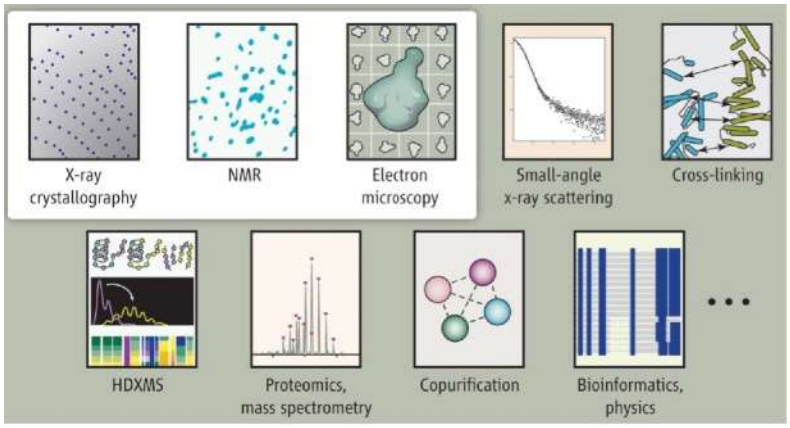
The interdisciplinary character of CERM/CIRMMP research projects, combined with the excellence of its instrumentation, constitutes a point of reference for the scientific community and for the cultural growth in the country, as demonstrated by the significant usage of the infrastructure by national scientists.

The Role of Solution NMR in Integrated Structural Biology

During the last decade structural biologists have elucidated the structure of ever larger protein complexes and biological machines. As these challenges became more ambitious, it has become necessary to use multiple experimental and computational methods, taking advantage of the different potential of each method. This approach is known as *'integrative structural biology'*. Early examples are the combined use of NMR and SAXS in structure determination of proteins, and of solid-state NMR and cryo-EM. For larger protein complexes the use of X-ray crystallography with cryo-EM has become popular. It is worth mentioning that mass spectrometry methods are now also part of the integrative toolkit so as bioinformatics, *in silico* molecular modelling and EPR.

Remarkably, NMR often plays an important role in these hybrid approaches. Of course, high-resolution NMR is unique in providing atomic-level structural information for biomolecules in solution. In addition, dynamic behaviour can be studied for an extremely wide range of time-scales.[1-4]

The major challenge of structural biology is understanding how biological machines function at the cellular level, within macromolecular complexes, or in a cellular pathway. It is not possible to understand dynamic processes and their coordination at a cellular level using a single technique. This task has become accessible through the integration of a number of approaches, spanning different resolution scales.



Integrated structural biology to unravel biological processes (10.1126/science.122856)

References:

(1) Abelein. A; Ciofi-Baffoni S. Mörman C, Kumar R, Giachetti A, Piccioli M, Biverstål H. ***J.Am.Chem Soc.*** 2022, *11*, 2571.

(2) Hricovíni, M; Pierattelli, R.; *et al.* ***Int. J. Mol. Sci..*** 2022, *23,* 14815.

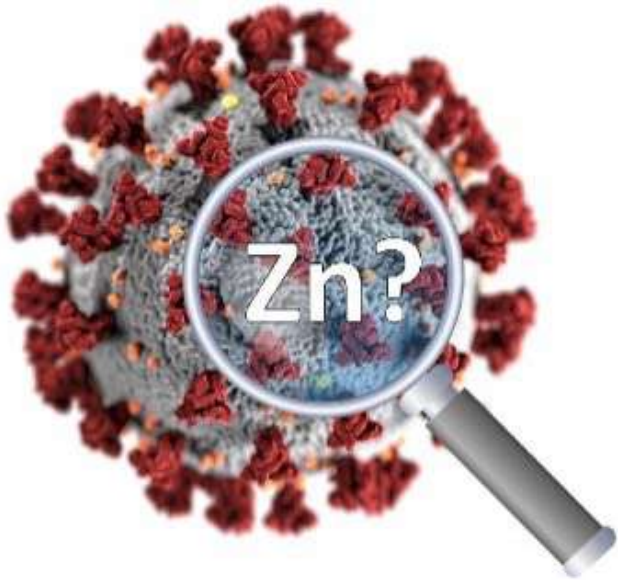
(3) Del Duca S, Puglia AM, Calderone V, Bazzicalupo M, Fani R ***Microorganisms*** 2022, *10,* 692

(4) Camponeschi F. ; Banci L. ***FEBS Lett*** 2022, *597,* 122-133.

Computing for Integrative Structural Biology

Integrative structural biology combines data from multiple techniques to obtain a deeper understanding of complex biological systems.
To progress towards this goal, our work focused on providing thorough information on metal-binding systems through European databases and on the automation of tools for data analysis.

A cornerstone for the development of innovative computational techniques in structural biology is the availability of extensive data repositories that integrate structural information in the PDB providing biological context for proteins, to which we contribute our expertise on metal sites.[1] Information in biological databases is then exploited to implement knowledge discovery processes.[2]
In the respect of metal-analysis, we analysed the Zn-proteome of SARS-COV-2[3] and applied MetalPDB tools to shed light on the evolution of multi-heme proteins.[4]
Finally, we opened a new line of research with the aim of applying machine learning to the characterisation and analysis of metal sites in structures.[5,6]



We used an updated version of MetalPredator to search for zinc-binding proteins in the SARS-COV2 virus.

References:

(1) PDBe-KB consortium; Andreini, C.; Rosato, A.; Putignano, V. *et al.* ***Nucleic Acids Res.*** 2022, *50 (D1)*, D534-D542

(2) Bazayeva M, Laveglia V, Andreini C, Rosato A. ***J Inorg Biochem,*** 2022, *238,* 112025

(3) Andreini C, Arnesano F, Rosato A. ***Metallomics.*** 2022, *14,* mfac047.

(4) Soares R, Costa NL, Paquete CM, Andreini C, Louro RO. ***Mol. Biol. and Evol.*** 2022, *39,* msac139.

(5) Andreini C, Rosato A. ***Int J of Mol Sciences*** *23,* 7684

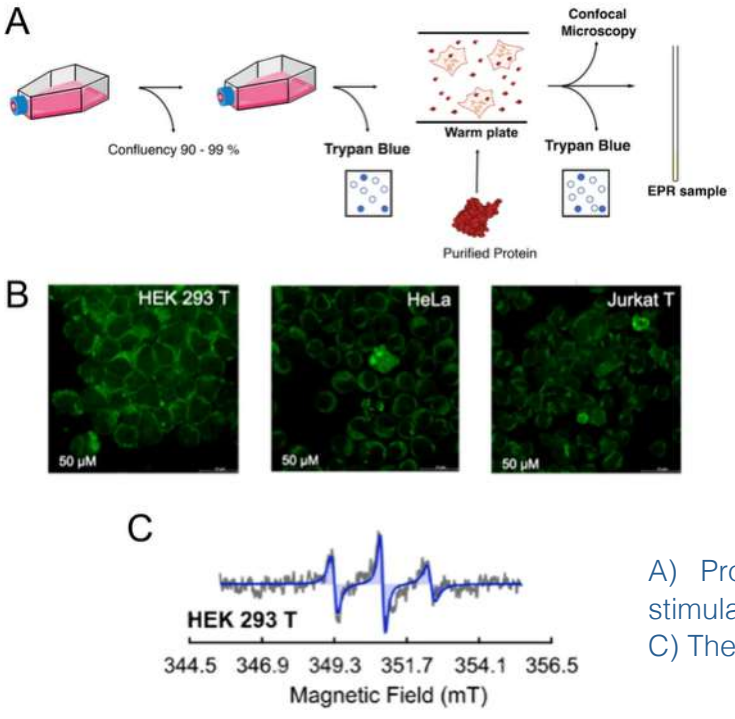
(6) Laveglia V, Giachetti A, Sala D, Andreini C, Rosato A. ***J Chem Inf and Mod.*** 2022, *62,* 2951-2960

In-cell NMR and EPR in Human Cells

In-cell NMR and EPR spectroscopies allow structural and functional characterisation of macromolecules inside living cells, making possible to study protein-protein and protein-ligand interactions in a highly physiological environment, in a non-destructive way.[1,2] These approaches can bridge the gap between *in vitro* techniques and cell-based assays, and thus have high potential in the development of new effective drugs against intracellular targets.[3]

At CERM, novel approaches are constantly being developed to further expand the range of applications of these methodologies, and to move towards even more physiological conditions. A novel method has been developed to deliver proteins in human cells for in-cell EPR applications. This method relies on mild thermal stimulation of human cultured cells, which stimulates a naturally-occurring cellular uptake mechanism. Spin-labelled proteins are successfully delivered to different cell lines (HEK293T, HeLa and Jurkat T lymphocytes) at sufficient concentration to enable EPR methods, such as double electron-electron resonance (DEER) measurements, for characterising protein conformations.[4]

CERM continues to pioneer the development of NMR and EPR methods to investigate proteins and small molecules in living human cells at atomic resolution. A novel approach has been developed to deliver spin-labelled proteins in human cells for characterisation by in-cell EPR spectroscopy. The method relies on a mild thermal stimulation treatment, and will provide a fast and effective alternative to existing protein delivery methods.



A) Protein delivery by thermal stimulation; B) GFP-treated cells; C) The in-cell EPR spectrum.

References:

(1) Luchinat, E.; Cremonini, M.; Banci, L. ***Chem. Rev.*** 2022, *122,* 9267–9306.

(2) Theillet, F.X.; Luchinat, E. ***Prog. Nucl. Magn. Reson. Spectrosc.*** 2022, *132-133,* 1-112.

(3) Luchinat, E.; Banci, L. ***Curr. Op. Struct. Biol.*** 2022, *74,* 102374.

(4) Torricella, F.; Barbieri, L.; Bazzurro, V; Diaspro, A.; Banci, L. ***Sci. Rep.*** 2022, *12,* 17190.

NMR Methods Fighting Covid-19 Pandemic

NMR has been used to evaluate different aspects of COVID-19: from the definition of the disease metabolomic fingerprint to the evaluation to the effects of vaccination on the metabolome/lipoproteome of healthy subjects. Application of NMR to assess the high order structure integrity of a monoclonal antibody for the treatment of the disease is also reported.

From the beginning of the pandemic, we used NMR profiling has been used to define the biochemical and metabolic alterations induced by COVID-19 disease. ^{1}H NMR spectra of serum samples of COVID-19 patients can be strongly discriminated from recovered subject with an accuracy >90%. The differences originate from significant alterations in the concentrations of several metabolites and a panel of lipoprotein components. We also observed that during the healing process, the metabolome and lipoproteome revert back to the "healthy" state with different rates, that is faster for metabolites than lipoproteins.[1]

NMR spectra of sera have been also used to define the changes induced by vaccination with Pfizer-BioNTech vaccine in a small group of healthy subjects. Vaccination does not induce any significant variation in the metabolome, whereas it causes changes at the level of lipoproteins. Interestingly, the effects are different for COVID-19-recovered subjects with respect to naïve subjects, suggesting that a previous infection reduces the vaccine modulation of the lipoproteome composition.[2]

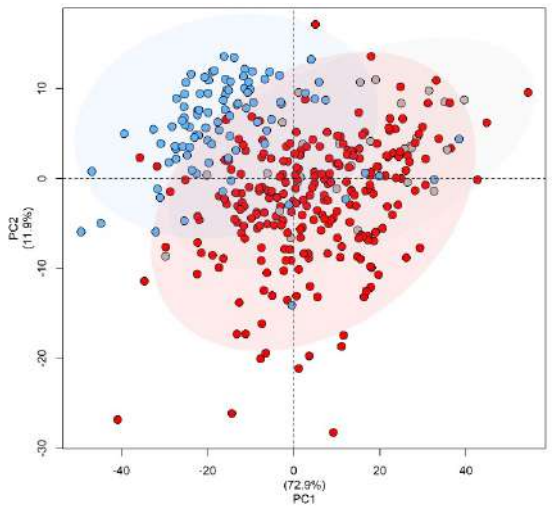
We also used NMR fingerprinting to characterise the higher-order structure of a potent human monoclonal antibody (mAb) for the COVID-19 treatment. The study confirmed that the methyl region of the 2D ^{1}H-^{13}C NMR spectrum can be used as a sensitive tool to control protein structural integrity and stability.

References:

(1)Ghini V; Meoni G; Pelagatti L; Celli T; Veneziani F; Petrucci F; Vannucchi V; Bertini L; Luchinat C; Landini G; Turano P. ***PLoS Pathog,*** 2022, *18,* e1010443

(2) Ghini V; Maggi L; Mazzoni A; Spinicci M; Zammarchi L; Bartoloni A; Annunziato F; Turano P. ***Front. Mol. Biosci.,***2022, *9*, 839809.

(3) Cantini F; Andreano E; Paciello I; Ghini V; Berti F; Rappuoli R; Banci L. ***Pharmaceutics*** 2022, *14,* 1981

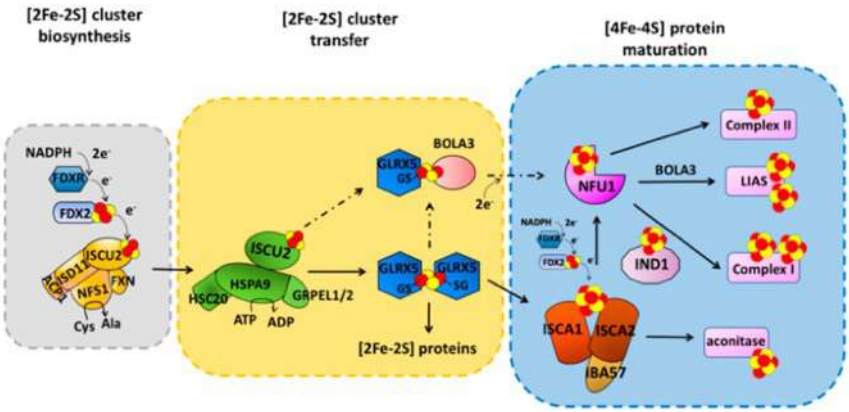


PCA analysis showing the discrimination between 1D ^{1}H NOESY spectra of plasma samples collected from COVID-19 patients (red dots) and recovered subjects (blue dots).

Molecular Mechanisms of Iron-Sulfur Protein Biogenesis in Humans

A mitochondrial iron-sulfur cluster assembly machinery is in charge of the maturation of mitochondrial Fe_4S_4 proteins. In humans, the last steps of the machinery are still not clearly identified. In 2022, we focused our attention on the role of ISCA1 and NFU1 in the maturation of human lipoyl synthase (LIAS). We show that the crucial actor is the C-domain of NFU1, which, by exploiting a protein-interaction affinity gradient increasing from ISCA1 to LIAS, drives the cluster to its final destination. Within this frame, we discuss how several iron-sulfur cluster-containing proteins activate, support and modulate the innate immune response to restrict viral infections, and how some of these proteins simultaneously support the replication of viruses.[1-5]

Iron-sulfur proteins are involved in fundamental cellular processes. Their biogenesis is a highly complex process present in all living cells. Several rare diseases and viral infections in humans are related to the function of iron-sulfur protein biogenesis. A picture of the molecular mechanisms at the basis of iron-sulfur protein biogenesis is fundamental to boost the development of human disease treatments.



The three steps of the mitochondrial iron-sulfur cluster assembly machinery required to mature mitochondrial Fe_4S_4 target proteins.

References:

(1) Honarmand Ebrahimi, K.; Ciofi-Baffoni, S. *et al.* ***Nat Chem.*** 2022, *14,* 253-266.

(2) Saudino, G.; Ciofi-Baffoni, S.; Banci, L. ***J Am Chem Soc.*** 2022, *144,* 5713-5717.

(3) Invernici, M.; Selvolini, G.; Silva, J.M. *et al.* ***Chem. Commun.*** 2022, *58,* 3533-3536.

(4) Camponeschi, F.; Ciofi-Baffoni, S.; Calderone, V.; Banci, L. ***Biomolecules*** 2022, *12,* 1009-1037.

(5) Ji, Y.; Wei, L.; Da, A. *et al.* ***Front Mol Biosci.*** 2022, *9,* 1032220.

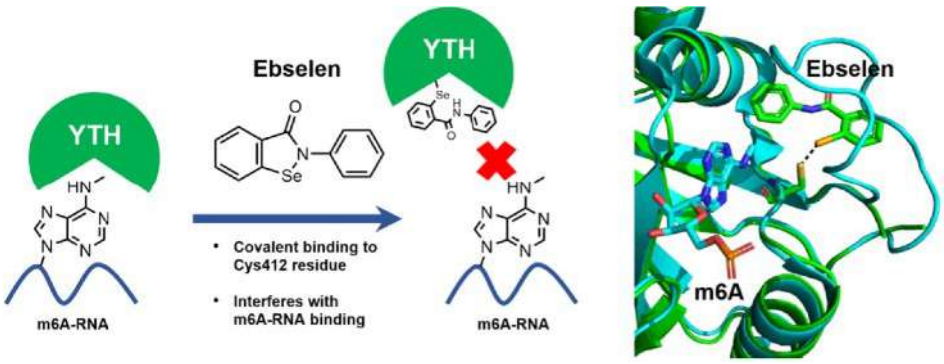
Proteins as Drugs and Drug Targets

RNA binding proteins and matrix metalloproteases are important pharmaceutical targets. The elucidation of the structural properties of the targets, the discovery of new ligands or the functionalisation of ligands to biomaterials open new avenues for the treatment of the related diseases.

Acinetobacter baumannii is a Gram-negative pathogen, known to acquire resistance to antibiotics used in the clinic. The RNA-binding proteome of this bacterium is poorly characterised, in particular for what concerns the proteins containing RNA Recognition Motif (RRM). The research activity of CERM/CIRMMP led to the identification and the structural characterisation of a protein containing a RRM domain that is a new potential drug target.[1,2]

The scientists of CERM/CIRMMP have also characterised another RNA binding protein that is a potential pharmaceutical target (YTHDF) involved in the regulation of the fate of target mRNAs. In particular, the binding of an organo-selenium compound (ebselen) to this protein has been investigated by NMR proving the high affinity of the ligand for the target protein.[3]

In a different line of research, the activity of CERM/CIRMMP resulted in the development of a unique material, obtained by the functionalisation of hyaluronic acid with an inhibitor of matrix metalloproteases, with potential application in the treatment of dry eyes syndrome.[4]



The binding of ebselen to YTHDF protein interfere with the binding of mRNA to the protein.

References:

(1) vAssoni, G.; La Pietra, V.; Digilio, *et al.* ***Ad. Drug Deliv. Rev.*** 2022, *181,* 114088.

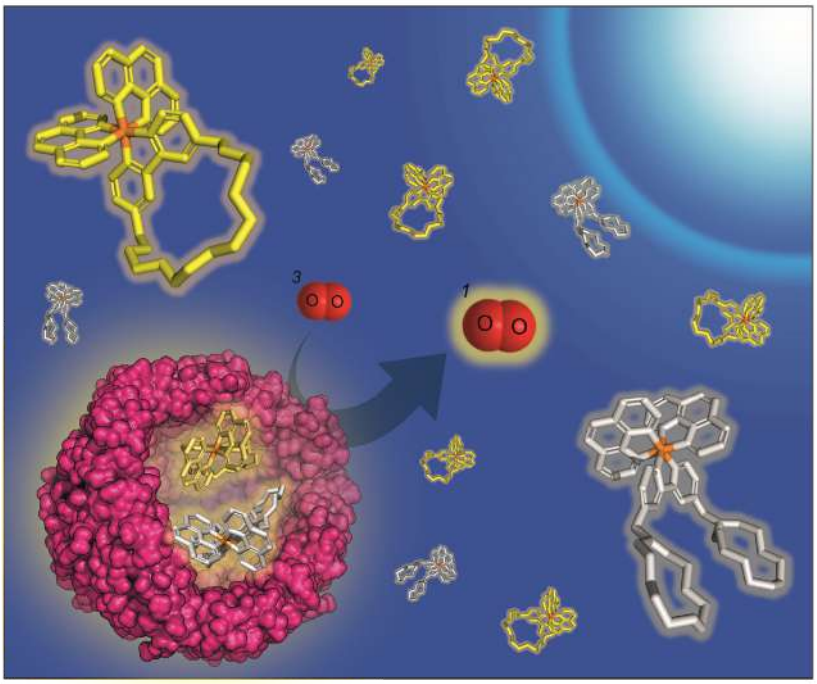
(2) Ciani, C.; Pérez-Ràfols, A.; Bonomo, I. *et al.* ***Biomolecules*** 2022, *12,* 922.

(3) Micaelli, M.; Dalle Vedove, A.; Cerofolini, L. *et al.* ***ACS Pharmacol Transl Sci*** 2022, *5* (10), 872–891.

(4) Burgalassi, S.; Fragai, M.; Francesconi, O.; Cerofolini, L.; Monti, D.; Leone, G.; Lamponi, S.; Greco, G.; Magnani, A.; Nativi, C. ***ACS Macro Lett*** 2022, *11,* 1190–1194.

The Human Ferritin Cage as a Drug Nanocarrier

Two Ru(II) polypyridyl complexes have been successfully encapsulated into human H–ferritin. The resulting nanocomposites are highly luminescent, display great stability and preserve the native shell-core structure of the protein. The singlet oxygen sensitising properties of the Ru(II)-complexes are largely maintained in their encapsulated form. Ru(II)-ferritin nanocomposites are exclusively internalised by cancer cells expressing the Transferrin receptor 1, via receptor-mediated endocytosis of H–type ferritin. Light-activation leads to a marked dose-dependent cytotoxic effect uniquely against cancer cells.[1]



Encapsulation into ferritin of ruthenium(II) photosensitisers that can be light-activated to produce the highly cytotoxic singlet oxygen ($^1O_2$).

The ferritin nanocage is suitable for the targeted delivery of drugs towards cells overexpressing specific receptors. Here, we report about the use of recombinant human H–ferritin as a carrier for Ru(II)-based photosensitisers, demonstrating its potential as a platform for the tumour-targeted delivery of agents for photodynamic therapy.

References:

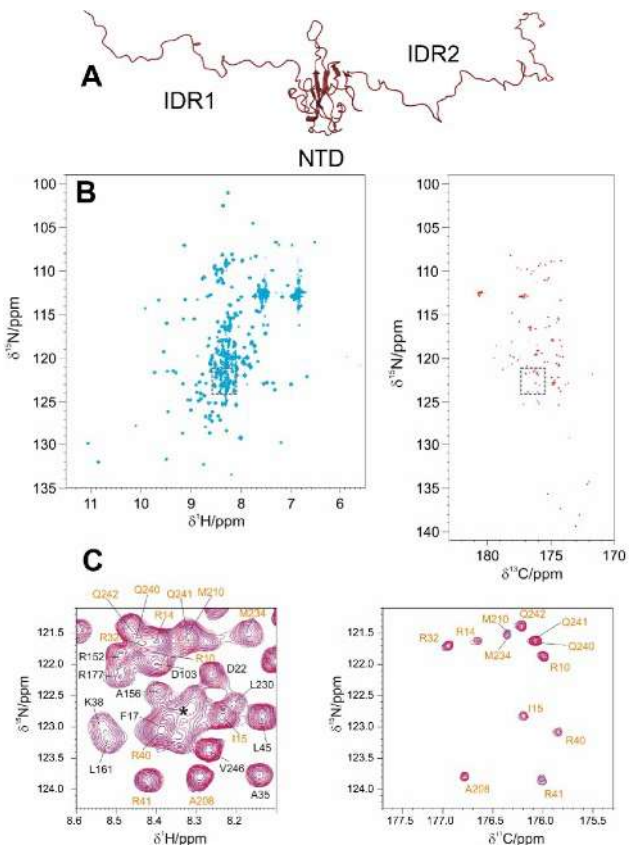
(1) Conti, L.; Ciambellotti, S.; Giacomazzo, G.E.; Ghini, V.; Cosottini, L.; Puliti, E.; Severi, M.; Fratini, E.; Cencetti, F.; Bruni, P.; Valtancoli, B.; Giorgi, C.; Turano, P. ***Inorg. Chem. Front.*** 2022, *9,* 1070-1081.

Intrinsically Disordered Proteins by NMR

The highly flexible intrinsically disordered regions of the nucleocapsid protein from SARS-CoV-2 modulate the interaction with RNA and with enoxaparin, a natural polyanion used to cure Covid19 patients. These intriguing results were obtained through the suite of NMR experiments based on ^{13}C direct detection.

The SARS-COoV-2 Nucleocapsid protein has not been considered so far as a potential drug target because of the structural and dynamic heterogeneity conferred by its flexible linkers connecting the two globular domains. However, they have been found to be crucial for the function of the protein, involved in packaging the genomic RNA.

The suite of ^{13}C detected NMR experiments developed at CERM to focus on intrinsically disordered protein regions was crucial to characterise their properties. A construct comprising the RNA binging domain and the two flanking disordered regions (1-248) was investigated. Multiple receiver NMR experiments allowed to acquire simultaneously a 2D CON, reporting information about the flexible regions and 2D HN spectra showing both the IDRs and the globular domain. These experiments revealed an important role played by the flexible linkers in the interaction with RNA as well as with enoxaparin, one of the drugs used to cure severely ill Covid 19 patients.[1-3]



The figure reports the result of the mr_CON/HN experiment (doi: 10.1016/j.bpj.2019.05.017, Panel B and C) acquired on the NTR construct (Panel A). Panel C shows an enlargement of the superimposed spectra in absence (blue) and with the addition of 0.1 equivalents of heparin (magenta).

References:

(1) Felli IC, Pierattelli R. ***Chem. Rev.*** 2022, *25,* 9468-9496

(2) Schiavina M, Pontoriero L, Tagliaferro G, Pierattelli R, Felli IC. ***Biomolecules*** 2022, *12,* 1302.

(3) Pontoriero L, Schiavina M, Korn SM, Schlundt A, Pierattelli R, Felli IC. ***Biomolecules*** 2022, *12,* 929.

Solid-state NMR in Structural Biology

Antibiotics acting through new biological mechanisms are important to combat antimicrobial resistance. Teixobactin (TXB) and its derivatives represent a new class of antibiotics, with almost none detected resistance. Solid-state NMR experiments clarified their specific mode of action with atomic detail. TXB acts binding to the widely conserved pyrophosphate group in Lipid-II, a precursor of the bacterial cell wall that is not present in eucaryote, and gives highly specificity to the antimicrobial activity. ^{1}H-^{31}P ssNMR 2D correlation spectra on membrane Lipid-II samples clearly show the TXB interaction and identify the specific residues involved. This work, developed in collaboration with the Markus Weingarth group (Utrecht University) in the framework of the iNEXT project, also evidence that TXB forms small fibril-like aggregates that destroy the bacterial membrane, amplifying the antimicrobial activity.[1]

In other examples, at CERM we used solid-state NMR to identify control the status of the proteins embedded in silica matrix,[2] and the pattern of the residues of a pharmaceutical target (the programmed cell death ligand 1, PD-L1) involved in the interaction with a new multispecific biological drug[3,4]. This epitope mapping on the target proteins provides key information to improve the affinity and to monitor the manufacturing process and drug stability.[3,4]

Solid state NMR (ssNMR) is one of the most powerful technique to characterise not soluble biomolecules and membrane proteins. In the example here reported we shows how ssNMR made it possible to understand the action mode of teixobactin, a new antibacterial molecule, unraveling the interaction with the cell membrane and the binding to cell wall precursors.

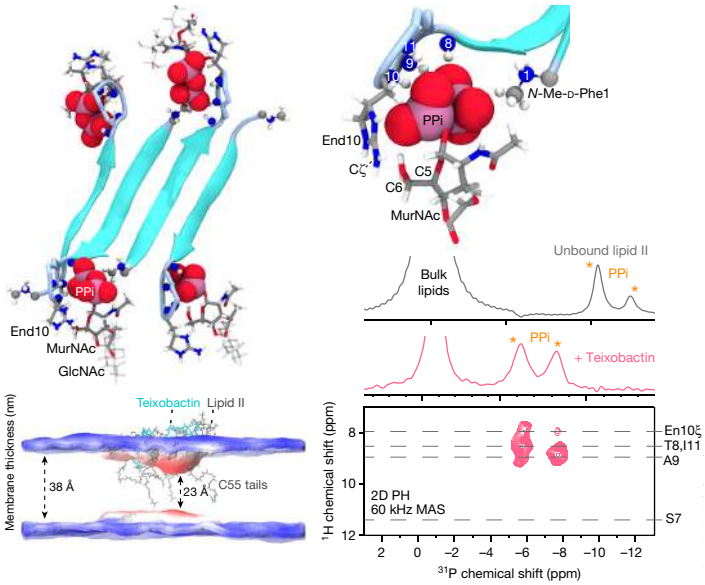
References:

(1) Shukla, R.; *et al.* ***Nature,*** 2022, *608,* 390–396. doi.org/10.1038/s41586-022-05019-y.

(2) Bruno, F. *et al.* ***Langumir*** 2022, *38,* 8030–8037

(3) Rizzo D, *et al. J. Am. Chem. Soc.* **2022**, *144,* 10006–10016.

(4) Cerofolini L, *et al. Solid State Nucl Magn Reson.* **2022,** *122,* 101828.



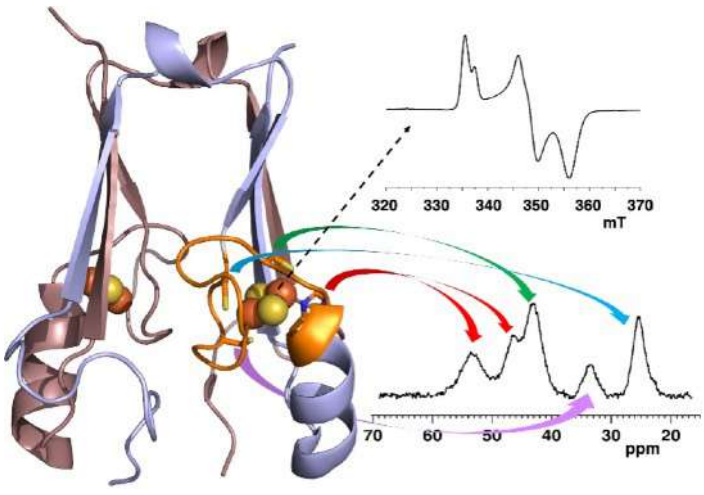
Action mode of the Teixobactin fibrils (ribbons) interacting with the pyrophosphate (red) (top figures). This interaction is clearly demonstrated by 2D ^{1}H-^{31}P spectra (bottom right). TXB fibrils binds bacteria membrane thinning the membrane layer.

NMR of Paramagnetic Systems

NMR of paramagnetic molecules is a highly versatile methodology, which provides information at different levels, from long range constraints to local information about electronic structure.

Paramagnetic NMR provides a wealth of experimental restraints that can be used to investigate structure and dynamics of biological macromolecules,[1] as in iron-sulfur cluster proteins.[2] NMR can also be used to probe the magnetic properties of single ion magnets at room temperature, complementing traditional magnetochemical measurements. We did so for a dysprosium(III) complex with a magnetic susceptibility anisotropy 4 times larger than lanthanoid binding tags (LBTs) used in structural biology.[3] This connection can allow for predicting how to improve LBT for structural biology studies.[4] In collaboration with the group of Frank Neese (MPI Muelheim) we continue working on the correct Spin Hamiltonian treatment for quantum chemical calculations, now providing the definitive proof that the magnetic susceptibility (for instance from *ab initio* calculation) can be used for calculating pseudocontact shifts even outside of the point dipole approximation.[5]

Specifically on iron-sulfur (Fe-S) proteins, we investigated mitoNEET, a protein involved in several cellular processes and diseases, but with a still unknown function. Paramagnetic NMR and EPR spectroscopies were used to characterise the first coordination sphere of mitoNEET [2Fe-2S] clusters, providing molecular fingerprints useful to investigate the role of [2Fe-2S] clusters in health and disease.[6]



The NMR assignment of the first coordination sphere of mitoNEET provides a fingerprint helpful to address functional aspects related to the cellular role of the protein.

References:

(1) Parigi G, Ravera E, Luchinat C, ***Curr. Opin. Struct. Biol.*** 2022, *74,* 102386

(2) Trinidade IB, Coelho A, Cantini F, Piccioli M, Louro RO, ***J. Inorg. Biochem.,*** 2022, *243,* 111871

(3) De Sousa Santana F, Perfetti M, Briganti M, Sacco F, Poneti G, Ravera E, Soares J, Sessoli R, ***Chem Sci,*** 2022, *13,* 5860

(4) Ravera E, Gigli L, Fiorucci L, Luchinat C, Parigi G, ***Phys. Chem. Chem. Phys.,*** 2022, *24,* 17397-17416

(5) Lang L, Ravera E, Parigi G, Luchinat C, Neese F, ***J. Chem. Phys.,*** 2022, *156, 15,* 154115

(6) Camponeschi F, Piccioli M, Banci L, ***Molecules,*** 2022, *27,* 8218.

Field Cycling Relaxometry

Paramagnetic complexes are largely employed in magnetic resonance imaging (MRI) for their ability to increase the image contrast. The amount of contrast agent to be injected can be lowered by increasing its relaxivity. This result can be achieved through the modulation of the reorientation time of the paramagnetic complex, by binding it to macromolecules. Asparaginase is a large tetrameric protein assembly, currently used against acute lymphoblastic leukemia. A gadolinium(III)-DOTA derivative was conjugated to asparaginase and its relaxation properties were investigated in order to assess its efficiency. We observed a very large increase in the relaxivity of the paramagnetic protein with respect to the small gadolinium chelate.[1]

Hydrogel nanoparticles composed of chitosan and hyaluronic acid and incorporating Gd-complexes were also characterised through ^{1}H NMR relaxometry. Interestingly, the macrocyclic complex $[Gd(DOTP)]^{5-}$, lacking metal-bound water molecules, showed a very large relaxivity; a careful analysis of the relaxation data emphasised the fundamental role of second sphere water molecules with strong and long-lived hydrogen bonding interactions with the complex.[2,3]

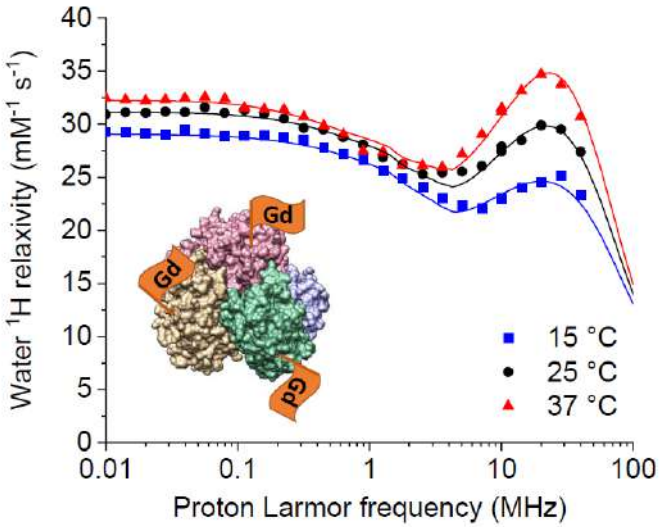
Field cycling relaxometry provides access to structural and dynamic parameters on which nuclear relaxation depends. It represents a precious tool for the optimisation of contrast agents for MRI and for investigating how dynamic processing present in nanoparticles influence the proton relaxation efficiency.

References:

(1) Carniato, F.; Ricci, M.; Tei, L.; Garello, F.; Terreno, E.; Ravera, E.; Parigi, G.; Luchinat, C.; Botta, M. ***Inorg. Chem.*** *2022, 61, 13,* 5380-5387.

(2) Licciardi, G.; Rizzo, D.; Salobehaj, M.; Massai, L.; Geri, A.; Messori, L.; Ravera, E.; Fragai, M.; Parigi, G. ***Bioconj. Chem.*** 2022, *33,* 2411-2419.

(3) Licciardi, G.; Rizzo, D.; Ravera, E.; Fragai, M.; Parigi, G.; Luchinat, C. ***NMR Biomed.*** 2022, *35,* e4623.



^{1}H relaxivity profiles of GdDOTA-conjugated asparaginase II at 15, 25 and 37 °C. Solid lines are the best fit profiles obtained with the Florence NMRD program.

Solid-state Methods and DNP for Materials

Solid-state NMR (ssNMR) is the method of choice to characterise advanced materials, especially when they are amorphous or composed by several phases. We report here applications to the advanced characterisation of supported inorganic catalyst, functioanlised Graphene Oxides and stubborn plastic waste.

Taking profit of the two orders of magnitude of sensitivity enhancement of the DNP technique we applied solid-state NMR to characterise the surface of an Iridium supported catalyst (figure below). Combining ^{13}C-{^{15}N} and ^{29}Si-{^{15}N}-REDOR and ^{13}C-INADEQUATE experiments we determined the structure of the catalytic Ir-active species. Furthermore, we discovered new multiple ligand species not expected for these systems. This characterisation adds new hypothesis in the activity of these catalysts.[1]

New DNP methods were also developed, increasing the efficiency at ultrafast-MAS[2] and at temperature >200 K[3].

Functionalised graphene oxide (GO) are attracting a considerable interest for they peculiar catalytic, and antimicrobial properties[2] and the easiness of its preparation. Saccaride bound GO show a strong antimicrobial property also able to disgregate bacteria biofilms.[4] GOs has also important catalytic properties, for example in the synthesis of 2,3-disubstituted quinolines in three-component Povarov reactions of anilines, aldehydes and electron-enriched enol ethers.[5] ssNMR makes possible to easily characterise all these compounds, controlling the synthetic processes and investigating the real active sites on GO surface.[4,5]

As well, we applied solid-state NMR (^{1}H-^{13}C HETCOR experiments) in the characterisation of stubborn plastic waste, in order to understand its composition and the suitable treatment to recover this material.[6]

References:

(1) Jabbour R, *et al..J Am Chem Soc.* **2022,** *144,* 10270-10281

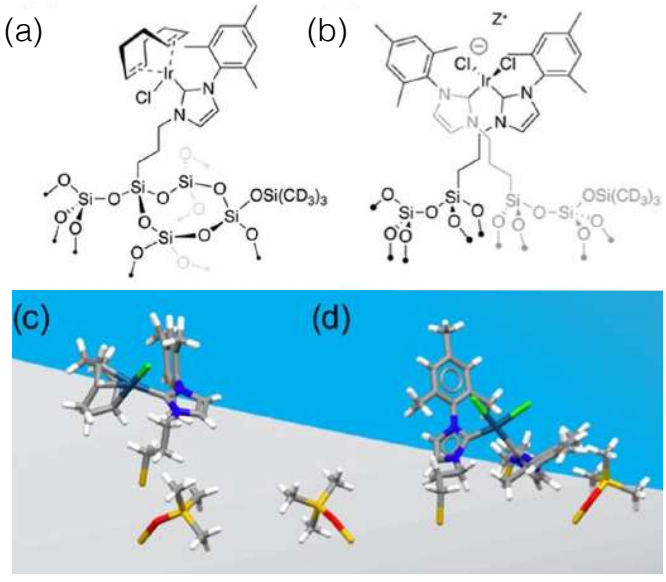
(2) Berruyer, P. *et al.* ***J. Phys. Chem. C*** 2022, *126,* 7564–7570.

(3) Kausik, M. *et al.* ***Phys. Chem. Chem. Phys.***, 2022, *24,* 12167–12175

(4) Tricomi J, *et al. Nanoscale,* ***2022,*** *14,* 10190–10199.

(5) Gallorini R, *et al. ACS Omega* **2022**. *7*, 37757–37763.

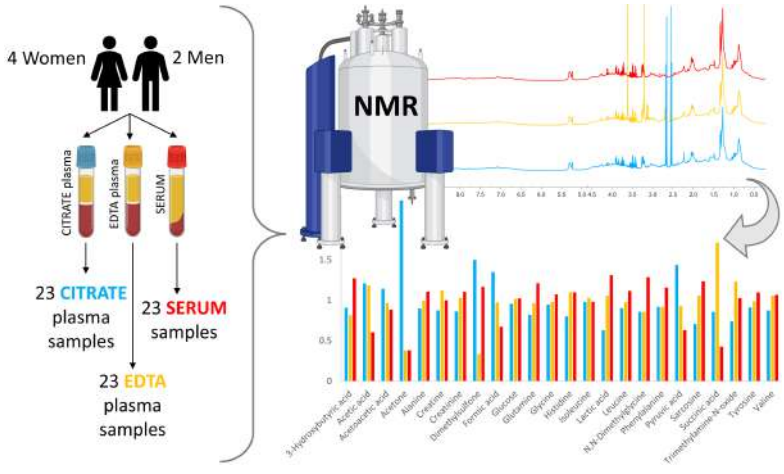
(6) Caputo, S.; *et al. RSC Advances,* **2022,** *12*, 15834-15847.



Silica supported Ir-catalyst studied with DNP ssNMR. In addition to the expected specie (a), we observed an unexpected bischelated species (b). REDOR experiments provide structural data to determine the structures of both catalytic species (c) and (d), respectively.

Metabolomics in Biomedicine

During 2022 the NMR-based metabolomics group at CERM have been largely focused on the metabolomic definition of ageing. We used serum/plasma metabolomics to investigate causes associate to healthy and diseased ageing, and the metabolic alterations associated to the onset and evolution of Parkinson's disease (PD).[1–4] Moreover, changes in the metabolome in light of age, sex and Rh and ABO blood systems were studied.[1,3–5] We also explored the potential of NMR-based metabolomics to study cerebro- and cardiovascular pathological conditions in elderly/adult individuals with two aims: characterising the underlying mechanism of cardiovascular risk in patients undergoing bariatric surgery,[6] and using metabolomics to predict prognosis of patients diagnosed with heart failure[7] and after stroke.[8]
Another thread of interest concerns cell metabolomics,[9] focusing on the mechanism of action of anticancer metallodrugs. In particular, we have analysed the effect of established Pt(II) drugs on the A2780 ovarian cancer cell lines, highlighting common mechanistic features.[10]
From the methodological point of view, along with continued activities regarding the definition of optimal pre-analytical procedures,[11-12] we have successfully applied chemometrics approaches usually employed in metabolomics to study other "omics" data.[13-14]



Variations of levels of NMR-detectable metabolites in three different blood derivatives: citrate plasma (light blue), EDTA plasma (yellow) and serum (red).

Metabolomics offers a comprehensive, dynamic, and accurate picture of a cellular model, a biofluid, an organ, or an organism at a molecular level. Thus, it is an invaluable instrument to obtain information on diseases' underlying biochemistry, to diagnose and to prognosticate pathological conditions.

References:

(1) G. Meoni, *et al.*, ***NPJ Parkinsons Dis*** 2022, *8,* 14.

(2) G. M. Dimitri, *et al.*, ***Appl. Sci.-Basel*** 2022, *12,* 8954.

(3) F. Di Cesare, *et al.*, ***J. Gerontol. A Biol. Sci. Med. Sci.*** 2022, *77,* 918–926

(4) F. Di Cesare, *et al.*, ***GeroScience*** 2022, *44,* 1109-1128.

(5) F. Di Cesare, *et al.*, ***J. Proteome Res.*** 2022, *21,* 2655–2663.

(6) L. Castagneto-Gissey, *et al.*, ***eBioMedicine*** 2022, 76.

(7) A. Vignoli, *et al.*, ***Front.Cardiovasc. Med.*** 2022, *9*, 851905.

(8) C. Licari, *et al.*, ***J. Proteome Res.*** 2023, *22,* 16–25.

(9) C. Bernacchioni, *et al.*, ***Cells*** 2022, 11.

(10) V. Ghini, *et al.*, ***Dalton Trans.*** 2022, *51,* 12512–12523.

(11) V. Ghini, *et al.*, ***New Biotech.*** 2022, *68*, 37–47.

(12) A. Vignoli, *et al.*, ***J. Proteome Res.*** 2022, *21*, 1061–1072.

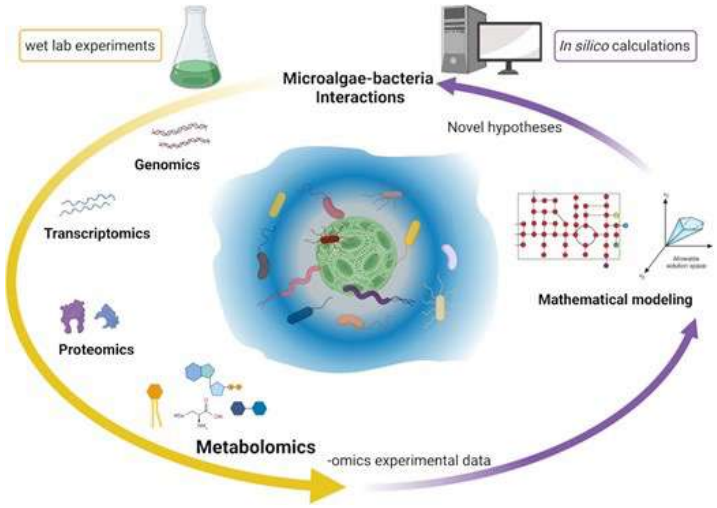
(13) A. Vignoli, L. Tenori, C. Luchinat, ***Metallomics*** 2022.

(14) M. M. Zinga, *et al.*, ***Front Mol Biosci*** 2022, *9,* 1070394.

Other Applications of Metabolomics

Metabolomics encompasses various areas of basic and applied research. It is also utilized in the food and beverage industry for quality assessment and to understand the impact of processing.
Metabolomics is employed in the study of animal-derived products, aiming to enhance consumer awareness and for veterinary medicine purposes. Furthermore, exploring microorganisms and plants can help in the identification of novel metabolites with ecological significance.-

Even though conventional quality, safety, and authenticity control in food is based on targeted strategies, high-resolution ^{1}H-NMR, performed under well-defined instrumental specifications, offers several advantages, including producing an extremely reproducible food fingerprint and fully quantitative data just from single experiment.
In 2022, the CERM metabolomic group applied ^{1}H-NMR spectroscopy in the characterisation of metabolites in different foods. Analysing milk, we arrived to recognise the product from cows with subclinical and clinical mastitis.[1] In another work on cow milk we distinguishing between intensive and extensive farming conditions.[2]
In the winemaking sector, metabolomics has been employed to study the wine production using different types of containers, from steel tanks to the older yet resurgent earthenware amphorae.[3]
New applications of metabolomics are emerging in combination with the other *-omics* techniques for example in the field of ecology. In fact, metabolomics is a valuable tool for describing interactions between microorganisms, plants, and algae, which could aid in the identification of new molecules with ecological relevance[4].



The two major routes to study the multifaced microalgal–bacterial interactions: the *-omics* (genomics, transcriptomics, proteomics, and metabolomics), and the computational tools.

References:

(1)Bobbo T., *et al.*, ***J. Dairy Sci.*** 2022, *105,* 535-548.

(2) Niero G., *et al.*, ***J. Dairy Sci.*** 2022, *105,* 9702-9712.

(3) Martellini T., *et al.*, ***Flavour Frag.. J.,*** 2022, *37* 219-233.

(4) Daly G., *et al.*, ***FEMS Microbiol. Rev.***, 2022, *46*, uac020.

National and Transnational access

Instruct-ERIC ESFRI Infrastructure – European and National NMR Research Infrastructure

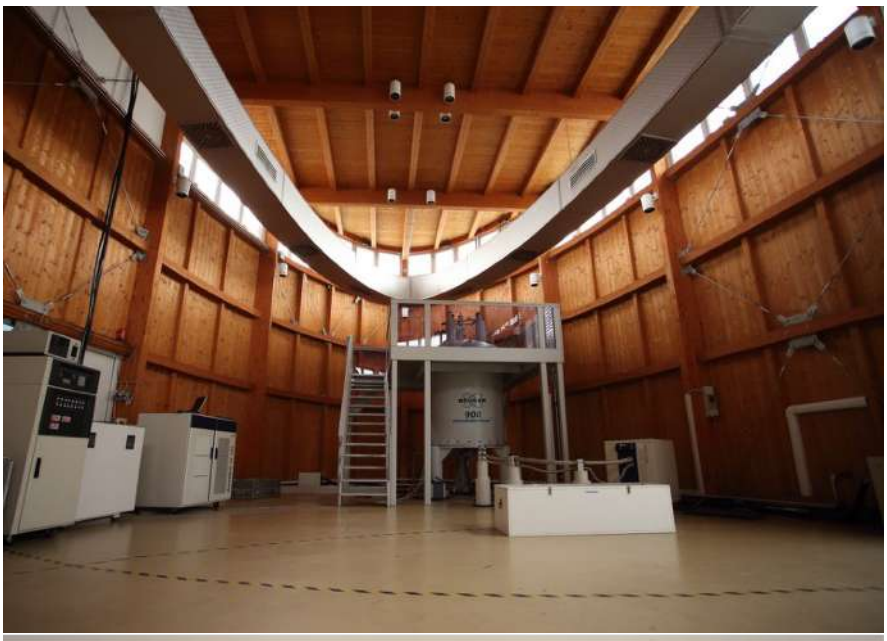
CERM/CIRMMP is the key centre for application and development of NMR spectroscopy within Instruct-ERIC, an ESFRI infrastructure operative since 2012.

Instruct-ERIC provides access to unique instrumentation in a variety of different structural techniques (see pages 9). This innovative approach allows for a description of biological cells at the molecular level, in order to understand how living organisms function in normal and pathological conditions and to design drugs and vaccines. The possibility of access to Instruct-ERIC represents a unique opportunity for researchers, both at the national and European level, to strengthen the innovation capacity of the research performed.

Since 2021 the project PANACEA (https://panacea-nmr.eu/) has started. PANACEA is a consortium funded by the HORIZON2020 program to offer European researchers access to advanced Solid-State NMR instruments for the investigation of chemical and pharmaceutical solid compounds, as well as organic and inorganic materials. The platform is open to scientists and industrial partners with or without previous experience in solid-state NMR.

In addition, CERM/CIRMMP continues to provide access to its instrumentation to all national users whose research is outside the Instruct-ERIC scope, provided their research project matches quality criteria in terms of scientific interest, excellence and feasibility. CERM/CIRMMP is promoting the development of a national platform Instruct-ITALIA to favour the development of a consortium of infrastructures in structural and cellular biology for national access service.

In all cases, access is granted on the basis of peer-review of the received proposals, and after a feasibility check by the staff scientists of the receiving infrastructure. Technical assistance is provided for the acquisition of the data. Scientific collaborations are welcome but not required. The uniqueness of access provision at CERM/CIRMMP infrastructure lies in the wide number of available NMR instruments, the variety of the NMR equipment (probes, automatic sample changers,…) and the exceptional expertise of the scientific and technical stuff, which represents an ideal environment for NMR research, especially in the field of structural and functional characterisation of biological systems. The description of the NMR instrumentation made available under the above mentioned access projects at CERM/CIRMMP is reported in the dedicate paragraph at page 37. Notably, in 2020 we have installed the first world 1.2 GHz instrument operative since April 2020, and its contribution to research is already visible in the research session.
Molecular biology and cellular biology labs are also strategic for the users needs to prepare and/or optimise the large variety of samples for structural characterisation, together with oth-



er biophysical equipments for EPR, CD, UV-vis, stopped-flow measurements, manual and automated crystallisation facilities and X-ray diffractometry. Users can also access other university infrastructures available in the campus, such as those of mass spectrometry, Raman resonance, and non-linear spectroscopies.

CERM/CIRMMP also provides access to its computational e-infrastructure, which includes a cluster for the more intensive calculations, with 16 blades harbouring a total of 80 CPU cores. Ten servers are used to host services from web pages to databases and to enable access to the European Grid. A number of graphic stations are available for interactive NMR data analysis.

During 2022 we recorded 431 days of access to the NMR spectrometers. A more detailed analysis shows that 253 days NMR access were provided to academic users via Instruct-ERIC, Instruct-ITALIA and iNEXT-Discovery, 44 via PANACEA and 134 days to industry users, either as services or through formal collaborations.

Beside NMR access provision, the infrastructure provided access to protein production services via Instruct-ERIC and to other structural biology techniques via Instruct-ITALIA.

Worth to mention the implementation of a platform for the management of NMR access (https://amp.cerm.unifi.it/) improving data findability and experiment reproducibility and, thanks to new in-house LIMS, track of all the experiments performed and allows long-term data storage.

Collaborations with Industries

CERM/CIRMMP has a long tradition in collaborations with industries: from simply providing access and service to its instrumentation, to establishing a more collaborative activity in research projects or to the participation as partners in international project calls. This number does not include the access provided to industrial partners through collaborative projects.

We warmly thank the following companies for stimulating interactions:

Bracco SpA

Bruker BioSpin

Dompé Pharmaceutical

Italmatch Chemicals

Glaxo Smith Kline

Giotto Biotech Srl

Merck

COLLABORATION WITH INDUSTRIES

Menarini Srl

Valagro S.p.a.

Abiogen S.p.a.

Infineum

Danger and Safety

Buona Steve Jones

INOTREM, control innate immunity

Probiotical S.p.a.

COLLABORATION WITH INDUSTRIES

Indena S.p.a.

Stelar S.r.l.

Extra Byte S.r.l

A special acknowledgment to Gruppo SAPIO Srl,
official supplier of all the cryogenic

Flanking Institutions

Da Vinci European BioBank

The Da Vinci European Biobank (daVEB) is handled by CsaVRI (Centro Servizi Di Ateneo Per la Valorizzazione della Ricerca e la Gestione dell'Incubatore) and it is certified ISO9001:2015. It is a research biobank that stores human biospecimens (plasma, serum, urine, tissues, cells) and bacterial expression vectors at cryogenic temperatures (Mechanical freezers for storage at –80 °C, equipped with auxiliary LN_2 cooling system and tanks for cryopreservation in nitrogen vapour phase at –150 °C, with automatic nitrogen supply).

Thanks to the involvement of scientific and technical staff in the management of daVEB, CERM has established connections with the ESFRI European Biobank Infrastructure BBMRI, which are reinforced by the metabolomics research activities of CERM.

The interaction between daVEB and CERM is strategic and synergistic. Scientific collaborations in the metabolomic field contribute to the development of SOPs validated by NMR and to the enrichment of the biobank in terms of type and number of samples. daVEB currently houses a collection of unique samples (biofluids, tissues and DNA) of growing importance by number in the following areas: COVID-19, melanoma, rare skin diseases, diseases of the genital-urinary cancer, cardio-circulatory diseases, digestive diseases, breast cancer, non-Hodgkin's lymphoma, diseases associated with the ageing. On the other hand, the biobank acts as a support to the metabolomics research via NMR carried out at CERM by providing a storage service of samples and the associated data, following protocols in accordance with international standards.

The daVEB is a partner of the RISE project (Competence center-RISE Network infrastructure for industrial research and incubation for advanced services to innovative companies), coordinated by CSAVRI; PAR-FAS funding of Regione Toscana It operates as an infrastructure to support experimental development activities and provision of services, with open access to private companies.

https://www.unifi.it/vp-11370-da-vinci-european-biobank.html

Giotto Biotech Srl

Giotto Biotech Srl Giotto Biotech S.r.l. is a SME founded in 2011 as a spin-off of CERM that aims at contributing to the biomedical sciences by providing enabling products and services, with a particular focus on complementary technologies in the field of NMR. Giotto Biotech provides a full range of compounds and custom manufacturing to supply research needs in the field of biomedical sciences, consulting and services. The company is active in various fields, including

protein production and isotope labelling, organic synthesis, services for NMR, and information technology. The services include NMR metabolomics and statistical analysis.

In 2022 Giotto Biotech has been involved in several research projects funded at the European or National level (FLAG-ERA-ITFoC, Information Technology: Future of Cancer Treatment; ITN EC RNAct, Enabling proteins with RNA recognition motifs for synthetic biology and bio-analytics; ITN GLYTUNES, A multidisciplinary training network for the bio-inspired development of glycomimetics tuning the Siglec-Sialoglycan axis; SENSOGM, Development of biophotonic sensors for environmental determination of GMOs, funded by the Tuscany Region; RESPIRA - ROVER AND UAS FOR REMOTE AIR MONITORING, funded by ARTES 4.0 and MISE; NMR metabolomics: the Made in Italy revolution for food certification, Financed by Fondazione CR Firenze). Giotto Biotech research activity is carried out in synergy with CERM scientists. As an outcome of this collaboration, in 2022 Giotto Biotech and CERM researchers co-authored three scientific publications.

http://www.giottobiotech.com/

Fondazione Luigi Sacconi

The Luigi Sacconi Foundation was established in 1996 to honour the memory of Prof. Luigi Sacconi, who was a prominent figure in Chemistry and founder of the General and Inorganic Chemistry School in Florence where many international scientists have been educated.

Its aim is to promote scientific research in the molecular sciences at the local, national and international levels. Particular attention is addressed to chemistry, in its implications and applications concerning health, quality of life, environment, energy, and technological and industrial development. For this purpose, the Luigi Sacconi Foundation collects documents and publications, promotes seminars, courses and meetings and other activities supporting the exchange of scientific knowledge, subsidises the activity of Italian and foreign researchers, and establishes awards.

The Sacconi Medal Lecturer 2022 has been awarded to Prof. Matthias Beller scientific director of the Leibniz Institute for Catalysis (LIKAT), Rostock, Germany. The award ceremony took place on 9 September 2022 during the XLVIII National Congress of Inorganic Chemistry held in Pisa.

Contribution to "*Chianti Workshop Opening New Doors for Magnetic Resonance* "which took place in Principina Terra (Grosseto) June 19 to 24, 2022;

Cooperation in the organisation of the "12th International Copper Meeting", held in Sorrento (Naples) September 18 to 23, 2022.

FLANKING INSTITUTIONS

Collaboration in the organisation of the IUPAC Day "The legacy of Alessandro Volta" and the great challenges for humanity: the past and the future of electrochemistry", held in Como on October 24, 2022.

http://www.cerm.unifi.it/fondazione

Instrumentation

Solution and Solid-State NMR Spectrometers

In 2020, the first 1.2 GHz NMR instrument operating at 28.2 T has been installed at CERM. This instrument is currently operating with solution TCI and TXO cryoprobes. All NMR instruments are state-of-the-art, digital spectrometers equipped with a variety of cryo-probes, as well as of specific probes covering a broad range of frequencies and of observable nuclei. In addition, all the standard pulse sequences for spectroscopic, structural, dynamical, and functional characterisation, tailored pulse sequences for structural determination of high molecular weight proteins and paramagnetic systems are implemented. ^{13}C direct-detection solution protocols for "protonless" NMR experiments and structural characterisation of biomolecules, including unfolded or partially unfolded ones, are developed and updated. Pulse sequences and experiment setup have been implemented for the detection and characterisation of paramagnetic systems, and in this field CERM has been pioneer since decades. For this reason, the 400 MHz instrument is equipped with a special 3mm High Power probe designed for the investigation of paramagnetic systems. Solid-state MAS probes cover almost all the presently achievable MAS frequencies, from a few hundred of Hz to *ultra-fast* MAS regime, and since 2017 we have a new 0.7mm CP MAS probe spinning up to 111 kHz. Special protocols and devices are available for solid state experiments both for biological and inorganic material characterisation. Set-up and pulse sequences for ***in-cell*** NMR experiments are also implemented.

INSTRUMENTATION

B_0 Field (T)	^{1}H Larmor Frequency (Bore)	Probe heads
28.2	1200 MHz (NB 54 mm)	TCI Cryo 3 mm solution ($^{1}H/^{13}C/^{15}N$ with ^{2}H decoupling) TXO Cryo 5 mm solution ($^{1}H/^{13}C/^{15}N$ with ^{2}H decoupling)
22.3	950 MHz (NB 54 mm)	TCI Cryo 5 mm solution ($^{1}H/^{13}C/^{15}N$ with ^{2}H decoupling)
21.1	900 MHz (NB 54 mm)	2x TCI Cryo 5 mm solution ($^{1}H/^{13}C/^{15}N$ with ^{2}H decoupling) TXI RT 5 mm solution ($^{1}H/^{13}C/^{15}N$ with ^{2}H decoupling)
20.0	850 MHz (WB 89 mm)	3.2 mm CP MAS DVT $^{15}N/^{13}C/^{1}H$ 1.3 mm CP MAS ^{1}H-$^{19}F/BB/^{15}N$ 0.7 mm CP MAS $^{1}H/^{13}C/^{15}N$
18.8	800 MHz (NB 54 mm)	TXI RT 5 mm solution($^{1}H/^{13}C/^{15}N$ with ^{2}H decoupling) QXI RT 5 mm solution($^{1}H/^{13}C/^{15}N/^{31}P$ with ^{2}H decoupling) ^{1}H-Selective High Power RT (prototype) 3.2 mm CP MAS DVT Low-E $^{15}N/^{13}C/^{1}H$ 1.3 mm CP MAS ^{1}H-^{19}F/BB-X/BB-Y 1.3 mm CP MAS $^{1}H/^{13}C/^{15}N$
16.4	700* MHz (NB 54 mm)	TCI Cryo 5 mm solution($^{1}H/^{13}C/^{15}N$ with ^{2}H decoupling) TXI RT 5 mm solution($^{1}H/^{13}C/^{15}N$ with ^{2}H decoupling)
16.4	700 MHz (NB 54 mm)	TXO Cryo 5 mm solution($^{13}C/^{15}N/^{1}H$ with ^{2}H decoupling) TXO RT 5 mm solution($^{13}C/^{15}N/^{1}H$ with ^{2}H decoupling) TXI RT 5 mm solution($^{1}H/^{13}C/^{15}N$ with ^{2}H decoupling)
16.4	700 MHz (WB 89 mm)	3.2 mm CP MAS $^{15}N/^{13}C/^{1}H$ 4.0 mm CP MAS $^{15}N/^{13}C/^{1}H$
14.1	600 MHz (NB 54 mm)	2 x TXI RT 5 mm solution($^{1}H/^{13}C/^{15}N$ with ^{2}H decoupling) HR-MAS 4.0mm ($^{1}H/^{13}C/^{15}N$ with ^{2}H decoupling) ^{1}H - Selective High Power RT, 5 mm solution ^{1}H - Selective RT, 5 mm solution BBI RT 5 mm solution BBO RT 5 mm solution BBO RT 10 mm solution / BB RT -Low-γ -10 mm solution
14.1	600** MHz (NB 54 mm)	TXI RT 5 mm solution ($^{1}H/^{13}C/^{15}N$ with ^{2}H decoupling)

INSTRUMENTATION

11.7	500 MHz (NB 54 mm)	TCI Cryo 5 mm solution(^{1}H/^{13}C/^{15}N) TXI RT 5 mm solution (^{1}H/^{13}C/^{15}N) TBO RT 5 mm solution (^{1}H/^{31}P/BB) BBI RT 5 mm solution
9.4	400* MHz (NB 54 mm)	BBO RT 5 mm solution BBI RT 5 mm solution (^{1}H/BB) BBI RT 3 mm solution (^{1}H/BB) ^{1}H-Selective High Power 5 mm solution
0.33-1.25	EPR	X and Q Band cavities, X (9.43 GHz) Q-Band (35 GHz)
0.00024-1	Fast Field Cycling Relax-ometer	0.01-45 MHz 10 mm solution tubes

* With sample changer ** Standardized for metabolomics: equipped with the SampleJet robotic and refrigerated charger, along with dedicated routines for the analysis of biofluids through the Bruker IVDr platform

X-ray Crystallography

CERM/CIRMMP is equipped with standard crystallization facilities and with an automated nano-dispensing device (Mosquito, TTP Labtech). Furthermore, it has full access to the Interdepartmental Crystallography Centre of the University of Florence (CRIST, https://www.crist.unifi.it), equipped, among other instruments, with two sealed-tube diffractometers. The most recent one is a Bruker D8 Venture with double microsource (Cu and Mo) bearing a Photon III Pixel Array detector and the older one is an Xcalibur PX Ultra (Oxford Diffraction) equipped with a 165mm CCD detector for routine in-house data collections. Both diffractometers are equipped with a liquid nitrogen cryosystem. Regular access to synchrotron beam time slots in European facilities is also available. Biological and Biophysical Facilities and Services

Molecular and Cellular Biology

CERM/CIRMMP is equipped with state-of-the-art facilities for gene cloning and protein expression and purification. Large scale protein expression in prokaryotes and yeast is available through the use of fermenters. Different isotope labelling schemes, including specific labelling schemes oriented to NMR characterisation, can be achieved through the use of auxotrophic strains. Fully equipped facilities for protein purification are available, including last-generation instruments for streamlined purification (ÄKTA pure chromatography system) and equipment for protein purification.

A dedicated modern glove box, equipped for protein purification and reconstitution in anaerobic environment is also available as support for the bimolecular Lab.

A mammalian expression lab for in-cell NMR is also equipped with modern instrumentation.

EPR

9.43 GHz (X-Band, continuous wave, Elexsys E 580E) and 35 GHz (Q-Band, pulsed, Elexsys E 580E) instrument.

Multi Angle/Dynamic Light Scattering

Instrument for measurements on batch samples or on in-flow samples (FPLC coupling).

Isothermal Calorimetry (ITC)

ITC device to measure thermodynamical parameters in micro-samples. The instrument is fully equipped for studying protein-ligand and protein-protein thermodynamical parameters.

Optical Spectroscopy

Absorption/Fluorescence Spectrophotometer operating from 1000 to 200 nm, ***Circular Dichroism*** (CD) spectrometer operating form 1200 to 200 nm (Near-IR, Visible, UV) to derive infor-

mation on the proteins secondary structure or protein-metal interaction, and stopped-flow spectrophotometer are available in the infrastructure.

Computational Structural Biology Tools

CERM/CIRMMP provides integrated databases and software for genome browsing, metal binding analysis, structure calculation with/without paramagnetic restraints, sequence exploration, domain organisation, protein complex analysis.

Access to programs for NMR data processing and structural calculations is also provided via web.

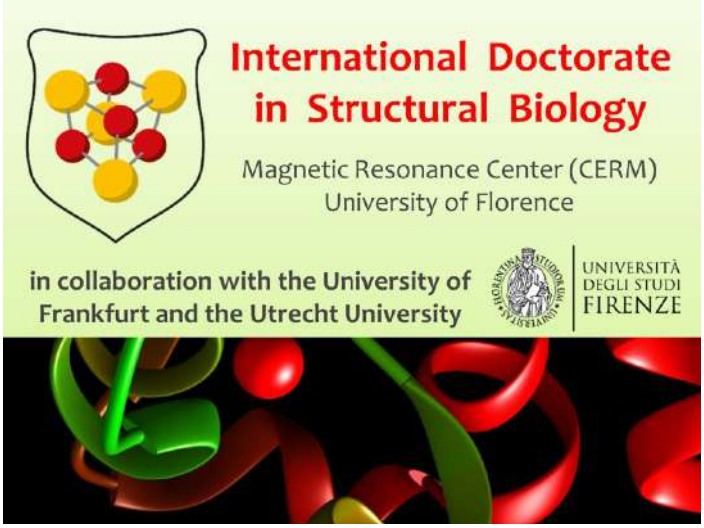
Electronic infrastructure (e-infrastructure)

The grid and cloud-based services of CERM/CIRMMP are part of the WeNMR thematic services (https://www.egi.eu/case-study/wenmr/), which have been developed throughout a variety of collaborative European projects, the most recent ones being the EOSC-Hub and EGI-ACE initiative. Services for structural biology are also a crucial component of technological development ongoing in the context of the European Open Science Cloud (EOSC). In particular, the EOSC-Lie project has provided a framework to create curated software pipelines spanning all aspects from data processing to the deposition of final results (a.k.a. scientific workflows), using standardised approaches and management systems. This enables our structural biology workflows be deposited in public repositories and be reused also by other NMR centres on their own computing infrastructure. The WeNMR thematic services provide application-level services specific to different cases in Structural Biology, with a main focus on NMR-based tools. Those services are supported thanks to the strong commitment of providers giving access to grid, cloud and data storage computing resources, through a Service Level Agreement signed with the EGI Federation. The user community served by the WeNMR services encompasses over 12000 registered users over the years from more than 95 different countries. Among recently added services, there are pipelines for data analysis in fragment screening campaigns, which will be exploited in conjunction with EOSC services and other European projects.
CERM/CIRMMP maintains a node of the European Grid Initiative. The available hardware comprises two clusters with 80 and 1024 CPU-cores respectively, and four TB of shared storage. A cluster with six Nvidia Tesla K20 GPGPU cards is also available.

Training & Education

International Doctorate in Structural Biology

The **International PhD course in Structural Biology** is a research doctorate of the *University of Florence* hosted at CERM that runs in collaboration with the *Frankfurt and Utrecht Universities*. The scientific fields cover most of the molecular aspects of life sciences.



The main objective of the International PhD course in Structural Biology is the training of research doctors at the forefront of the knowledge in modern methodologies in molecular and structural biology, biotechnology and systems biology. It provides both theoretical and hands-on training in structural techniques applied to biological macromolecules in solution and in the crystalline state, as well as in non-crystalline materials such as fibrils or amyloid, and to biological macromolecules in their cellular environment. It also provides state-of-the-art training in molecular biology for the expression of isotope-enriched recombinant proteins and specifically those for NMR studies. Finally, it offers top level ICT training thanks to the well-established expertise and the exploitation of the e-infrastructure. Bioinformatics, biostatistics and NMR-metabolomics training is offered as well.

The scientific themes covered by the PhD course are:

1. **NMR spectroscopy** (in solution and in the solid state) and X-ray crystallography aimed at studying structure, function and dynamics in biological macromolecules and protein-protein adducts;

2. **Molecular and cellular biology techniques** for the production of proteins, DNA and bacterial and prokaryotic cell growth;

3. **Drug and vaccine development**, through rational design techniques and structural characterisa-

tion of biological drugs;

4. **Bioinformatics** to understand the structure-function relationship in biomolecules and in particular in metalloproteins through the large scale analysis of databases;
5. ***In cell* NMR** studies, by which molecular pathways and cell import-export mechanisms are investigated;
6. **Metabolomics** studies, in which the individual metabolic fingerprints are related to disease states and fingerprints are utilised to provide early diagnosis or even identification of pre-disease states.

The added value of this PhD course is in the development of a *transnational educational project,* able to form PhDs at the forefront regarding the scientific formation, knowledge and development of research and technology, capable to consider multi-disciplinary, transnational cooperation and mobility as primary needs, and to evaluate collaborative projects as a requirement for high quality research. The doctoral program also relies on Faculty members who, in addition to scientists from CERM, include professors from other departments of the University of Florence and from the Universities of Frankfurt, Utrecht, Oxford and Lyon, all top places for structural biology.

Full-time attendance is mandatory, as is commitment to research activities. In addition to seminars and courses, students are asked to provide research seminars as a basic tool for their own training. Every PhD student is encouraged to liaise with foreign universities and take part in teaching and research training as well as in internships abroad.

Post-Doctorate

CERM/CIRMMP hosts a number of post doctoral researchers. Some of them are former PhD students who remain at CERM after the end of the PhD, others come from all over the world for performing research projects and being trained in the methodologies in which CERM/CIRMMP excels. There are also several short- or long-term visitors coming from Italian and foreign universities.

CERM/CIRMMP Organisation

SCIENTIFIC BOARD OF CERM

Prof. Lucia Banci - CERM Director
Prof. Claudia Andreini
Prof. Marco Fragai
Prof. Moreno Lelli
Prof. Paola Turano
Prof. Antonio Rosato

CIRMMP BOARD OF DIRECTORS

University of Florence
Prof. Claudio Luchinat (*President*)
Prof. Lucia Banci

University of Bologna
Prof. Stefano Ciurli
Prof. Francesco Capozzi

University of Siena
Prof. Daniela Valensin
Prof. Cecilia Pozzi

Dr. Francesca Morelli – *Director*

FACULTY MEMBERS

Prof. Claudia Andreini
Prof. Lucia Banci
Dr. Silvia Ciambellotti
Prof. Vito Calderone
Prof. Francesca Cantini
Prof. Simone Ciofi-Baffoni
Prof. Isabella Caterina Felli
Prof. Marco Fragai
Prof. Moreno Lelli
Prof. Claudio Luchinat
Dr. Gaia Meoni
Prof. Giacomo Parigi
Prof. Mario Piccioli
Prof. Roberta Pierattelli
Prof. Enrico Ravera
Prof. Antonio Rosato
Dr. Leonardo Tenori
Prof. Paola Turano

CIRMMP SCIENTIFIC BOARD

Dr. Francesca Morelli – *Director*
Prof. Lucia Banci
Prof. Claudio Luchinat
University of Siena
Prof. Daniela Valensin
Prof. Stefano Mangani
University of Bologna
Prof. Francesco Capozzi
Prof. Stefano Ciurli

ASSOCIATE FACULTY MEMBERS

University of Florence
Prof. Alessandro Mordini
Prof. Cristina Nativi
University of Siena
Prof. Daniela Valensin
University of Bologna
Dr. Elena Babini
Prof. Francesco Capozzi
Prof. Stefano Ciurli
Dr. Barbara Zambelli
Dr. Enrico Luchinat

CERM/CIRMMP ORGANISATION

Personnel

POST DOCTORAL FELLOWS

Argyriou Aikaterini

Letizia Barbieri

Francesca Camponeschi

Linda Cerofolini

Simona Del Giudice

Veronica Ghini

Andrea Giachetti

Tatiana Kozyreva

Vincenzo Laveglia

Francesca Lucarelli

Josè Pedro Malanho da Silva

Tommaso Martelli

Carlo Mengucci

Valentina Pecchioli

Marco Schiavina

Alessia Vignoli

PhD STUDENTS

Milana Bazayeva (XXXVI)

Lorenzo Bracaglia (XXXVII)

Lucrezia Cossottini (XXXVII)

Francesca Di Cesare (XXXV)

Deborah Grifagni (XXXV)

Francesco Milanesi (XXXV)

Giulia Licciardi (XXXVI)

Anna Perez I Rafols (XXXV)

Bach Tung Lan Pham (XXXVI)

Letizia Pontoriero (XXXV)

Maria Salobehaj (XXXVI)

Francesco Torricella (XXXV)

Valentina Vitali (XXXVI)

Beatrice Bargagna (XXXVII)

Letizia Fiorucci (XXXVII)

Valentina Monaci (XXXVII)

Lorenzo Niccoli (XXXVII)

Francesca Sacco (XXXVII)

Giulia Roxana Gheorghita (XXXVII)

Luis David Padilla Cortés (XXXVII)

Francesco Bruno (XXXVII PON DM 1061/2021)

Naomi Anna Consoli (XXXVII PON DM 1061/2021)

UNDERGRADUATE STUDENTS

Tommaso Attucci

Tessa Bolognesi

Azzurra Costantino

Michela Dal Pozzolo

Alessia De Santis

Matilde Ercolano

CERM/CIRMMP ORGANISATION

Mirko Fantoni

Gabriele Olivieri

Andrea Orsetti

Gabriele Pampalon

Lapo Querci

Leonardo Querci

Maria Anna Rodella

Angela Sofia Tino

Walter Vieri

Stefano Zineddu

TECHNICIANS

Marco Allegrozzi

Fabio Calogiuri

Rebecca Del Conte

Leonardo Gonnelli

Massimo Lucci

Enrico Morelli

Cristina Mescalchin

PROJECT MANAGER

Francesca Morelli

ADMINISTRATIVE SUPPORT

Isabella Barbaro

Francesca Di Gloria

Milena Moazzi

Laura Norfini

Lisa Orlando

GIOTTO SCIENTISTS

Mercia Ferreira de Sousa

Tommaso Martelli

Tatiana Kozyreva

Laura Bassani

Stefano Cardelli

Visiting Scientists at CERM

Pasquale Russomanno - PhD Student

University of Naples Federico II, Italy

Anaísa Coelho - PhD Student

University of Lisboa, Portugal

Marta Tiemblo Marin - PhD Student

University of Naples Federico II, Italy

Helen Liu - PhD Student

Berkeley - University of California

Prof. Liliana Quintanar

Centro de Investigación y de Estudios Avanzados (Cinvestav) - Mexico

List of publications

1. Luchinat E, Cremonini M, Banci L. Radio Signals from Live Cells: The Coming of Age of In-Cell Solution NMR, ***Chem. Rev.***, *2022*, *122*, 9267–9306, doi: 10.1021/acs.chemrev.1c00790. (IF 72.087).

2. Felli IC, Pierattelli R. 13C Direct Detected NMR for Challenging Systems, ***Chem. Rev.***, 2022, *122*, 9468–9496, doi: 10.1021/acs.chemrev.1c00871. (IF 72.087)

3. Shukla R, Lavore F, Maity S, Derks MGN, JonesCR, Vermeulen BJA, Melcrová A, Morris MA, Becker LM, Wang X, Kumar R, Medeiros-Silva J, van Beekveld RAM, Bonvin AMJJ, Lorent JH, Lelli M, Nowick JS, MacGillavry HD, Peoples AJ, Spoering AL, Ling LL, Hughes DE, Roos WH, Breukink E, Lewis K, Weingarth M. Teixobactin kills bacteria by a two-pronged attack on the cell envelope. ***Nature***, 2022, *608*, 390–396, doi: 10.1038/s41586-022-05019-y. (IF 69.504)

4. Honarmand Ebrahimi K, Ciofi-Baffoni S, Hagedoorn PL, Nicolet Y, Le Brun NE, Hagen WR, Armstrong FA. Iron-sulfur clusters as inhibitors and catalysts of viral replication. ***Nat. Chem*** 2022, 14, doi: 10.1038/s41557-021-00882-0. (IF 24.274)

5. Varadi M, Armastrong D, Carazo JM, Andreini C, Rosato A, et al. (PDBe-KB Consortium). PDBe-KB: collaboratively defining the biological context of structural data, ***Nucleic Acids Research***, 2022, *50*, D534-D542, doi: 10.1093/nar/gkab988. (IF 19.160)

6. Saudino G, Ciofi-Baffoni S, Banci L. Protein-Interaction Affinity Gradient Drives [4Fe-4S] Cluster Insertion in Human Lipoyl Synthase, ***J. Am. Chem. Soc.*** 2022, *144*, 5713-5717, doi: 10.1021/jacs.1c13626. (IF 16.383).

7. Abelein A, Ciofi-Baffoni S, Mörman C, Kumar R, Giachetti A, Piccioli M, Biverstål. H. Molecular Structure of Cu(II)-Bound Amyloid-β Monomer Implicated in Inhibition of Peptide Self-Assembly in Alzheimer's Disease. ***J. Am. Chem. Soc.*** 2022, *11*, 2571-2584, doi: 10.1021/jacsau.2c00438. (IF 16.383).

8. Rizzo D, Cerofolini L, Giuntini S, Iozzino L, Pergola C, Sacco F, Palmese A, Ravera E, Luchinat C, Baroni F, Fragai M. Epitope Mapping and Binding Assessment by Solid-State NMR Provide a Way for the Development of Biologics under the Quality by Design Paradigm, ***J. Am. Chem. Soc.*** 2022, *144*, 10006-10016. doi: 10.1021/jacs.2c03232. (IF 16.383)

9. Jabbour R, Renom-Carrasco M, Chan KW, Völker L, Berruyer P, Wang Z, Widdifield CM, Lelli M, Gajan D, Copéret C, Thieuleux C, Lesage A. Multiple Surface Site Three-Dimensional Structure Determination of a Supported Molecular Catalyst**. *J. Am. Chem. Soc.*** 2022, *144*, 10270-10281, doi: 10.1021/jacs.2c01013. (IF 16.383)

10. Assoni G, La Pietra V., Digilio R, Ciani C, Licata NV, Micaelli M, Facen E, Tomaszewska W, Cerofolini L, Pérez-Ràfols A, Varela-Rey M, Fragai M, Woodhoo A, Marinelli L, Arosio D, Bonomo I, Provenzani A, Seneci P. HuR-targeted agents: An insight into medicinal chemistry, biophysical, computational studies and pharmacological effects on cancer models. ***Adv Drug Deliv Rev.***, 2022, *181*, 114088, doi: 10.1016/j.addr.2021.114088. (IF 15.47)

11. Berg H, Wirtz Martin MA, Altincekic N, Alshamleh I, Kaur Bains J, Blechar J, Ceylan B, de Jesus V, Dhamotharan K, Fuks C, Gande SL, Hargittay B, Hohmann KF, Hutchison MT, Marianne Korn S, Krishnathas R, Kutz F, Linhard V, Matzel T, Meiser N, Niesteruk A, Pyper DJ, Schulte L, Trucks S, Azzaoui K, Blommers MJJ, Gadiya Y, Karki R, Zaliani A, Gribbon P, da Silva Almeida M, Dinis Anobom C, Bula AL, Bütikofer M, Putinhon Caruso Í, Caterina Felli I, Da Poian AT, Cardoso de Amorim G, Fourkiotis NK, Gallo A, Ghosh D, Gomes-Neto F, Gorbatyuk O, Hao B, Kurauskas V, Lecoq L, Li Y, Cunha Mebus-Antunes N, Mompeán M, Cristtina Neves-Martins T, Ninot-Pedrosa M, Pinheiro AS, Pontoriero L, Pustovalova Y, Riek R, Robertson AJ, Jose Abi Saad M, Treviño MÁ, Tsika AC, Almeida FCL, Bax A, Henzler-Wildman K, Hoch JC, Jaudzems K, Laurents DV, Orts J, Pierattelli R, Spyroulias GA, Duchardt-Ferner E, Ferner J, Fürtig B, Hengesbach M, Löhr F, Qureshi N, Richter C, Saxena K, Schlundt A, Sreeramulu S, Wacker A, Weigand JE, Wirmer-Bartoschek J, Wöhnert J, Schwalbe H. Comprehensive Fragment Screening of the SARS-CoV-2 Proteome Explores Novel Chemical Space for Drug Development, ***Angew. Chem. Int. Ed.*** 2022, *14*;61(46):e202205858, doi: 10.1002/anie.202205858. (IF 15.336)

12. Daly G, Ghini V, Adessi A, Fondi M, Buchan A, Viti C. Towards a mechanistic understanding of microalgae-bacteria interactions: integration of metabolomic analysis and computational models. ***FEMS Microbiol Rev***, 2022 46(5):fuac020 doi: 10.1093/femsre/fuac020 (IF 15.177)

13. Castagneto-Gissey L, Angelini G, Mingrone G, Cavaretta E, Tenori L, Licari C. The early reduction of left ventricular mass after sleeve gastrectomy depends on the fall of branched-chain amino acid circulating levels. ***eBioMedicine***, 2022, *76*, 103864, doi: 10.1016/j.ebiom.2022.103864. (IF 11.205)

14. De Sousa Santana F, Perfetti M, Briganti M, Sacco F, Poneti G, Ravera E, Soares J, Sessoli R. A textbook Single Molecule Magnet outperforming current pseudocontact shift agents. ***Chem. Sci.,*** 2022, *13*, 5860, doi: 10.1039/d2sc01619b. (IF 9.969)

15. Theillet FX, Luchinat E. In-cell NMR: Why and how? ***Prog Nucl Magn Reson Spectrosc,*** 2022, *132-133*:1-112, doi: 10.1016/j.pnmrs.2022.04.002. (IF 9.447).

16. Meoni G, Tenori L, Schade S, Licari C, Pirazzini C, Bacalini MG, Garagnani P, Turano P; PROPAG-AGEING Consortium, Trenkwalder C, Franceschi C, Mollenhauer B, Luchinat C. Metabolite and lipoprotein profiles reveal sex-related oxidative stress imbalance in de

novo drug-naive Parkinson's disease patients, ***NPJ Parkinsons Dis.***, 2022; *8*, 14 doi: 10.1038/s41531-021-00274-8. (IF. 9.304).

17. Soares R, Costa NL, Paquete CM, Andreini C, Louro RO. A New Paradigm of Multiheme Cytochrome Evolution by Grafting and Pruning Protein Modules ***Mol. Biol. and Evol.*** 2022*, 39*, msac139. doi: 10.1093/molbev/msac139.(IF 8.800)

18. Tricomi J, Cacaci M, Biagiotti G, Caselli L, Niccoli L, Torelli R, Gabbani A, Di Vito M, Pineider F, Severi Mi, Sanguinetti M, Menna E, Lelli M, Berti D, Cicchi S, Bugli F, Richichi B. Ball milled glyco-graphene oxide conjugates markedly disrupted Pseudomonas aeruginosa biofilms, ***Nanoscale***, 2022, *14*, 10190-10199, doi: 10.1039/D2NR02027K (IF 8.307)

19. Parigi G, Ravera E, Luchinat C. Paramagnetic effects in NMR for protein structures and ensembles: Studies of metalloproteins. ***Curr Opin Struct Biol.,*** 2022, *74*:102386. doi: 10.1016/j.sbi.2022.102386. (IF 7.786)

20. Luchinat E, Banci L. In-cell NMR: From target structure and dynamics to drug screening. ***Curr. Op. Struct. Biol.***, 2022, *74*:102374. doi: 10.1016/j.sbi.2022.102374. (IF 7.786)

21. Conti L, Ciambellotti S, Giacomazzo G E, Ghini V, Cosottini L, Puliti E, Severi M, Fratini E, Cencetti F, Bruni P, Valtancoli B, Giorgi C, Turano P, Ferritin nanocomposites for the selective delivery of photosensitizing ruthenium-polypyridyl compounds to cancer cells, ***Inorg. Chem. Front.***, 2022, *9*, 1070. doi:10.1039/D1QI01268A. (IF 7.779)

22. Bernacchioni C, Squecco R, Gamberi T, Ghini V, Schumacher F, Mannelli M, Garella R, Idrizaj E, Cencetti F, Puliti E, Bruni P, Turano P, Fiaschi T, Donati C. S1P Signalling Axis Is Necessary for Adiponectin-Directed Regulation of Electrophysiological Properties and Oxidative Metabolism in C2C12 Myotubes. ***Cells****,* 2022, *11*, 713. doi: 10.3390/cells11040713. (IF 7.666)

23. Di Cesare F., Tenori L., Meoni G., Gori AM, Marcucci R, Giusti B, Molino-Lova R, Macchi C, Pancani S, Luchinat C, Saccenti E. Lipid and metabolite correlation networks specific to clinical and biochemical covariate show differences associated with sexual dimorphism in a cohort of nonagenarians, ***GeroScience****,* 2022, *44*, 1109–1128, doi: 10.1007/s11357-021-00404-3 (IF 7.581)

24. Ghini V, Meoni G, Pelagatti L, Celli T, Veneziani F, Petrucci F, Vannucchi V, Bertini L, Luchinat C, Landini G, Turano P. Profiling metabolites and lipoproteins in COMETA, an Italian cohort of COVID-19 patients. ***PLoS Pathog***, 2022, *18*: e1010443, doi: 10.1371/journal.ppat.1010443. (IF 7.464)

25. Di Cesare F, Luchinat C., Tenori L., Saccenti E. Age- and Sex-Dependent Changes of Free Circulating Blood Metabolite and Lipid Abundances, Correlations, and Ratios, ***The Jour-***

nals of Gerontology*: *Series A, 2022, *77*, 5, 918–926, doi.org/10.1093/gerona/glab335 (IF 6.591)

26. Cantini F, Andreano E, Paciello I, Ghini V, Berti F, Rappuoli R, Banci L. 2D NMR Analysis as a Sensitive Tool for Evaluating the Higher-Order Structural Integrity of Monoclonal Antibody against COVID-19, ***Pharmaceutics***, 2022, *14*, 1981 doi.org/10.3390/pharmaceutics14101981, (IF 6.525)

27. Burgalassi S, Fragai M, Francesconi O, Cerofolini L, Monti D, Leone G, Lamponi S, Greco G, Magnani A, Nativi C. Functionalized Hyaluronic Acid for "In Situ" Matrix Metalloproteinase Inhibition: A Bioactive Material to Treat the Dry Eye Sydrome, ***ACS Macro Lett***., 2022, *11*, 1190-1194, doi.org/10.1021/acsmacrolett.2c00455 (IF 6.434).

28. Ghini V, Abuja PM, Polasek O, Kozera L, Laiho P, Anton G, Zins M, Klovins J, Metspalu A, Wichmann HE, Gieger C, Luchinat C, Zatloukal K, Turano P. Impact of the pre-examination phase on multicenter metabolomic studies, ***New Biotech.***, 2022, *68*, 37-47, doi: 10.1016/j.nbt. 2022.01.006. (IF 6.409)

29. Andreini C, Rosato A. Structural bioinformatics and deep learning of metalloproteins: recent advances and applications, ***Int.J Mol Sci.*, 2022**, *23* (14), 7684, (IF 6.208)

30. Hricovíni, M.; Owens, R.J.; Bak, A.; Kozik, V.; Musiał, W.; Pierattelli, R.; Májeková, M.; Rodríguez, Y.; Musioł, R.; Slodek, A.; Štarha, P.; Piętak, K.; Słota, D.; Florkiewicz, W.; Sobczak-Kupiec, A.; Jampílek, J. Chemistry towards Biology-Instruct: Snapshot**, *Int. J. Mol. Sci.***, 2022, *23*, 14815. doi.org/10.3390/ijms232314815 (IF 6.208)

31. Laveglia V, Giachetti A, Sala D, Andreini C, Rosato A. Learning to Identify Physiological and Adventitious Metal-Binding Sites in the Three-Dimensional Structures of Proteins by Following the Hints of a Deep Neural Network, ***J. Chem. Inf. Model.***, 2022, *62*, 12, 2951–2960 doi: 10.1021/acs.jcim.2c00522 (IF 6.162)

32. Ghini V, Maggi L, Mazzoni A, Spinicci M, Zammarchi L, Bartoloni A, Annunziato F, Turano P. Serum NMR Profiling Reveals Differential Alterations in the Lipoproteome Induced by Pfizer-BioNTech Vaccine in COVID-19 Recovered Subjects and Naïve Subjects, ***Front. Mol. Biosci.***, 2022**, *9,* 839809, doi: 10.3389/fmolb.2022, 9:839809. (IF 6.113)

33. Zinga, M.M., Abdel-Shafy, E., Melak, T., Vignoli, A., Piazza, S., Zerbini, L.F., Tenori, L. & Cacciatore, S. KODAMA exploratory analysis in metabolic phenotyping, ***Front. Mol. Biosci.***, 2022, *9,* doi: org/10.3389/fmolb.2022.1070394 (IF 6.113)

34. Ji Y, Wei L, Da A, Stark H, Hagedoorn PL, Ciofi-Baffoni S, Cowley SA, Louro RO, Todorovic S, Mroginski MA, Nicolet Y, Roessler MM, Le Brun NE, Piccioli M, James WS, Hagen WR, Ebrahimi KH. Radical-SAM dependent nucleotide dehydratase (SAND), rectification of the

names of an ancient iron-sulfur enzyme using NC-IUBMB recommendations. ***Front Mol Biosci.***, 2022, 9:1032220, doi: 10.3389/fmolb.2022.1032220 (IF 6.113)

35. Francesconi O, Donnici L, Fragai M, Pesce E, Bombaci M, Fasciani A, Manganaro L, Conti M, Grifantini R, De Francesco R, Nativi C, Roelens S. Synthetic carbohydrate-binding agents neutralize SARS-CoV-2 by inhibiting binding of the spike protein to ACE2. ***iScience***. 2022 *25*:104239. doi: 10.1016/j.isci.2022.104239. (IF 6.107).

36. Licciardi G, Rizzo D, Salobehaj M, Massai L, Geri A, Messori L, Ravera E, Fragai M, Parigi G. Large protein assemblies for high relaxivity contrast agents: the case of gadolinium-labeled asparaginase, ***Bioconj. Chem.***, 2022, *33*, 2411-2419, doi: org/10.1021/acs.bioconjchem.2c00506 (IF 6.069)

37. Invernici, M; Selvolini, G; Silva, J M; Marrazza, G; Ciofi-Baffoni, S; Piccioli, M; Interconversion between [2Fe-2S] and [4Fe-4S] cluster glutathione complexes, ***Chem. Commun.***, 2022, *58*, 3533-3536, doi.org/10.1039/D1CC03566E (IF 6.065)

38. Vignoli a, Fornaro A, Tenori L, Castelli G, Cecconi E, Olivotto I, Marchionni N, Alterini B, Luchinat C. Metabolomics Fingerprint Predicts Risk of Death in Dilated Cardiomyopathy and Heart Failure, ***Front. Cardiovasc. Med.***, 2022, *9*, 851905, doi: 10.3389/fcvm.2022.851905 (IF 5.848)

39. Carniato F, Ricci M Tei L, Garello F, Terreno E, Ravera E, Parigi G, Luchinat C, Botta M. High Relaxivity with No Coordinated Waters: A Seemingly Paradoxical Behavior of $[Gd(DOTP)]^{5-}$ Embedded in Nanogels. ***Inorg Chem***, 2022, *61*, 13, 5380-5387, doi: 10.1021/acs.inorgchem.2c00225 (IF 5.436)

40. Vignoli A, Tenori L, Morsiani C, Turano P, Capri M, Luchinat C. Serum or Plasma (and Which Plasma), That Is the Question. ***J. Proteome Res.***, 2022, *21*, 1061–1072, doi.org/10.1021/acs.jproteome.1c00935 (IF 5.370)

41. Di Cesare F., Tenori L., Luchinat C., Saccenti E., Association of Plasma Metabolites and Lipoproteins with Rh and ABO Blood Systems in Healthy Subjects, ***J. Proteome Res.***, 2022, *21*, 11, 2655–2663, doi: 10.1021/acs.jproteome.2c00375 (IF 5.370)

42. Licari C., Tenori L., Di Cesare F., Luchinat C., Giusti B., Kura A., De Cario R., Inzitari D., Piccardi B., Nesi M., Sarti C., Arba F., Palumbo V., Nencini P., Marcucci R., Gori A.M., Sticchi E., NMR-based metabolomics to predict early and late adverse outcomes in ischemic stroke treated with intravenous thrombolysis, ***J. Proteome Res.***, 2022, doi.org/10.1021/acs.jproteome.2c00333 (IF 5.370)

43. Torricella F, L Barbieri, V Bazzurro, A Diaspro, L Banci; Protein delivery to living cells by thermal stimulation for biophysical investigation. ***Sci. Rep.***, 2022, *12*, 1-10, doi: 10.1038/s41598-022-21103-9 (IF. 4.996)

44. Camponeschi F, Piccioli M, Banci L. The Intriguing mitoNEET: Functional and Spectroscopic Properties of a Unique [2Fe-2S] Cluster Coordination Geometry, ***Molecules***, 2022, *27*, 8218 doi.org/10.3390/molecules27238218 (IF 4.927)

45. Micaelli M, Dalle Vedove A, Cerofolini L, Vigna J, Sighel D, Zaccara S, Bonomo I, Poulentzas G, Rosatti EF, Cazzanelli G, Alunno L, Belli R, Peroni D, Dassi E, Murakami S, Jaffrey SJ, Fragai M, Mancini I, Lolli G, Quattrone A, Provenzani A. Small-Molecule Ebselen Binds to YTHDF Proteins Interfering with the Recognition of N6-Methyladenosine-Modified RNAs. ***ACS Pharmacol Transl Sci.***, 2022, *5*, 872-891, doi.org/10.1021/acsptsci.2c00008 (IF 4.88)

46. Massai, L.; Grifagni, D.; De Santis, A.; Geri, A.; Cantini, F.; Calderone, V.; Banci, L.; Messori, L. ***Biomolecules*** 2022, *12*, 1675; https://doi.org/10.3390/biom12111675. (IF 4.694)

47. Beniamino Y, Cenni V, Piccioli M, Ciurli S, Zambelli B. Characterization of the Ni(II) binding activity of the intrinsically disordered region of human NDRG1, a protein involved in cancer development. ***Biomolecules***, 2022, *12*,1272 doi:10.3390/biom12091272 (IF 4.694)

48. Schiavina M, Pontoriero L, Tagliaferro G, Pierattelli R, Felli IC. The Role of Disordered Regions in Orchestrating the Properties of Multidomain Proteins: The SARS-CoV-2 Nucleocapsid Protein and Its Interaction with Enoxaparin, ***Biomolecules***, 2022, *12*, 1302, doi: 10.3390/biom12091302. (IF 4.694)

49. Pontoriero L, Schiavina M, Korn SM, Schlundt A, Pierattelli R, Felli IC. MR Reveals Specific Tracts within the Intrinsically Disordered Regions of the SARS-CoV-2 Nucleocapsid Protein Involved in RNA Encountering. ***Biomolecules***, 2022, *12*, 929. doi: 10.3390/biom12070929 (IF 4.694)

50. Camponeschi F, Ciofi-Baffoni S, Calderone V, Banci L. Molecular Basis of Rare Diseases Associated to the Maturation of Mitochondrial [4Fe-4S]-Containing Proteins, ***Biomolecules***, 2022**,** *12*(7), 1009-1037, doi.org/10.3390/biom12071009, (IF 4.694)

51. Ciani C, Pérez-Ràfols A, Bonomo I, Micaelli M, Esposito A, Zucal C, Belli R, D'Agostino V G, Bianconi I, Calderone V, Cerofolini L, Massidda O, Whalen M B, Fragai M, Provenzani A. Identification and Characterization of an RRM-Containing, RNA Binding Protein in *Acinetobacter baumannii*. ***Biomolecules***, 2022, *12*, 922, doi: 10.3390/biom12070922 (IF 4.694)

52. Vignoli A, Tenori L, Luchinat C. An omics approach to study trace metals in sera of hemodialysis patients treated with erythropoiesis stimulating agents. ***Metallomics***, 2022, *19*;14(5), doi: 10.1093/mtomcs/mfac028 (IF 4.636)

53. Andreini C, Arnesano F, Rosato R. The zinc proteome of SARS-CoV-2. ***Metallomics***, 2022**,** *14*, mfac047. doi: 10.1093/mtomcs/mfac047 (IF 4.636)

54. Ghini V, Magherini F, Massai L, Messori L, Turano P. Comparative NMR metabolomics of the responses of A2780 human ovarian cancer cells to clinically established Pt-based drugs***. Dalton Trans.***, 2022, *51,* 12512-12523, doi: 0.1039/d2dt02068h (IF 4.569)

55. Licciardi G, Rizzo D, Ravera E, Fragai M, Parigi G, Luchinat C. Not only manganese, but fruit component effects dictate the efficiency of fruit juice as an oral magnetic resonance imaging contrast agent. ***NMR Biomed.***, 2022, 35, e4623. doi: 10.1002/nbm.4623. (IF 4.478)

56. Trindade IB, Coelho A, Cantini F, Piccioli M, Louro RO. NMR of paramagnetic metalloproteins in solution: Ubi venire, quo vadis? ***J Inorg Biochem.***, 2022**,** *234*, 111871. doi: 10.1016/j.jinorgbio.2022.111871. (IF 4.336)

57. Grifagni, D.; Silva, J.M.; Cantini, F.; Piccioli, M.; Banci, L.; ***J Inorg Biochem.***, 2022**,** doi: doi.org/10.1016/j.jinorgbio.2022.112089. (IF 4.336)

58. Bazayeva, M, V Laveglia, C Andreini, Rosato A. Metal-induced structural variability of mononuclear metal-binding sites from a database perspective. ***J Inorg Biochem***, 2022, *238*, 112025, doi: 10.1016/j.jinorgbio.2022.112025 (IF 4.336)

59. Lang L., Ravera E., Parigi G., Luchinat C., Neese F. Theoretical analysis of the long-distance limit of NMR chemical shieldings. ***J. Chem.Phys***, 2022, *156*, 154115, doi: 10.1063/5.0088162. (IF 4.304)

60. Berruyer P, Bertarello A, Björgvinsdóttir S, Lelli M, Emsley L, ^{1}H Detected Relayed Dynamic Nuclear Polarization, ***J. Phys. Chem. C*** 2022, 126, 7564–7570, doi: 10.1021/acs.jpcc.2c01077 (IF 4.304)

61. Niero G, Meoni G, Tenori L, Luchinat C, Visentin G, Callegaro S, Visentin E, Cassandro M, De Marchi M, Penasa M. Grazing affects metabolic pattern of individual cow milk. ***J. Dairy Sci.***, 2022, *105*(12), 9702-9712, doi: 10.3168/jds.2022-22072. (IF 4.225)

62. Bobbo T, Meoni G, Niero G, Tenori L, Luchinat C, Cassandro M, Penasa M. Nuclear magnetic resonance spectroscopy to investigate the association between milk metabolites and udder quarter health status in dairy cows. ***J. Dairy Sci.***, 2022; *105*:535-548. doi: 10.3168/jds.2021-20906. (IF 4.225)

63. Lang SH, Camponeschi F, Joya E, Borjas-Mendoza P, Tekin M, Thorson W. Multiple Mitochondrial Dysfunction Syndrome Type 3: A Likely Pathogenic Homozygous Variant Affecting a Patient of Cuban Descent and Literature, Review. ***Genes****., 2022, 13*, 2044 doi 10.3390/genes13112044. (IF 4.141).

64. Gallorini R, Ciuffi B, Real Fernández F, Carozzini C, Ravera E, Papini AM, Rosi L. Subcritical Hydrothermal Liquefaction as a Pretreatment for Enzymatic Degradation of

Polyurethane. ***ACS Omega***, 2022 *42*, 37757–37763, doi: 10.1021/acsomega.2c04734 (IF 4.132)

65. Camponeschi F, Banci L. Metal trafficking in the cell: Combining atomic resolution with cellular dimension. ***FEBS Lett.,*** 2022 *597*(1):122-133, doi: 10.1002/1873-3468.14524 (IF 4.124)

66. Del Duca S, Puglia AM, Calderone V, Bazzicalupo M, Fani R. Effect of Non-Lethal Selection on Spontaneous Revertants of Frameshift Mutations: The *Escherichia coli hisF* Case. ***Microorganisms***., 2022,10, 692 doi: 10.3390/microorganisms10040692. (IF 3.864)

67. Ravera E, Gigli L, Fiorucci L, Luchinat C, Parigi G. The evolution of paramagnetic NMR as a tool in structural biology***, Phys. Chem. Chem. Phys.*** 2022**,** *24*, 17397-17416, doi: 10.1039/D2CP01838A. (IF 3.676)

68. Kaushik M, Lingua H, Stevanato G, Elokova M, Lelli M, Lesage A, Ouari O, Trehalose matrices for high temperature dynamic nuclear polarization enhanced solid state NMR, ***Phys. Chem. Chem. Phys.***, 2022, 24, 12167-12175, doi.: 10.1039/D2CP00970F (IF 3.676)

69. Dimitri G.M, Meoni G, Tenori L, Luchinat C, Pietro L. PROPAG-AGEING Consortium. NMR Spectroscopy Combined with Machine Learning Approaches for Age Prediction in Healthy and Parkinson's Disease Cohorts through Metabolomic Fingerprints. ***Applied Sciences***, 2022, *2*(18), 8954; doi: 10.3390/app12188954. (IF 2.838)

70. Vignoli A, Meoni G, Tenori L. Applications and Challenges for Metabolomics via Nuclear Magnetic Resonance Spectroscopy. ***Applied Sciences***, 2022, *12* (9), 4655, doi: 10.3390/app12094655. (IF 2.838)

71. Cerofolini L, Parigi G, Ravera E, Fragai M, Luchinat C. Solid-state NMR methods for the characterization of bioconjugations and protein-material interactions. ***Solid State Nucl Magn Reson***., 2022, 122, 101828. doi: 10.1016/j.ssnmr.2022.101828. (IF 2.812)

72. Martellini, T ; Sposato, L; Pucci, S; Meoni, G; Marinelli, C; Tenori, L; Luchinat, C; Giorgi, R; Sarti, C; Cincinelli, A. Influence of in-amphorae vinification on the molecular profile of Sangiovese and Cabernet Franc. ***Flavour and Fragrances Journal***, 2022, *37*, 219-233; doi: 10.1002/ffj.3697. (IF 2.5)

73. Ravera,E. C. Luchinat. Using simple algebraic concepts to understand chemical composition problems, ***Int. J. Math. Ed. in Sci. and Tech.***, 2022, *53,* 842.857. doi: 10.1080 0020739X.2020.1785569 (IF 1.33).

74. Silva J M, Grifagni D, Cantini F, Piccioli M. 1H, ^{13}C and ^{15}N assignment of the human mitochondrial paramagnetic iron-sulfur protein CISD3. ***Biomol NMR Assign***, 2022, *17*, 17–22 doi: 10.1007/s12104-022-10113-3 (IF 0.731)

BOOK chapters

Saccenti E, Tenori L. Chapter 17 - Metabolomics as a tool for precision medicine, in ***Metabolomics Perspectives: From Theory to Practical Application,*** Editor: Jacopo Troisi, Academic Press, 2022, Pages 605-624, https://doi.org/10.1016/B978-0-323-85062-9.00017-9.

Emwas A-H, Szczepski K, Poulson BG, McKay R, Tenori L, Saccenti E, Lachowicz J, Jaremko M. Chapter 5 - Nuclear magnetic resonance in metabolomics, in ***Metabolomics Perspectives: From Theory to Practical Application,*** Editor: Jacopo Troisi, Academic Press, 2022, Pages 149-218, https://doi.org/10.1016/B978-0-323-85062-9.00005-2.

Meetings and Events Organised by CERM

Seminars Held at CERM

Prof. Jeffrey C. Hoch
The Gregory P. Mullen NMR Structural Biology Facility U Conn Health "The Enduring and Emerging Value of BMRB" November 15, 2022 at 18:00 CERM Conference room

Prof. Lilian Quintanar
Departamento de Química Centro de Investigación y de Estudios Avanzados Ciudad de México "Copper-protein interactions in degenerative diseases: From the brain to the human lens" September 27, 2022 at 12:00 CERM Conference room

Dr. Maria-Grazia Concilio
Weizmann Institute of Science, IL. "J-driven Dynamics Nuclear Polarization: A proposal to enhance NMR sensitivity in solution state" July 28, 2022 at 12.00 CERM Conference room

Prof. Alejandro J. Vila
Instituto de Biologia Molecular y Celular de Rosario CONICET - Universidad Nacional de Rosario ARGENTINA "The adaptive success of New Delhi Metallo-beta-lactamase depends on the in-cell kinetic protein stability" May 12, 2022 at 12.30 CERM Conference room

Prof. Łukasz Jaremko
King Abdullah University of Science and Technology (KAUST) Kingdom of Saudi Arabia "Understanding biological motions - linking the structure with function of biomolecules" Monday May 2, 2022 at 12.30 CERM Conference room

Meetings and Conferences

Timb3 hands-on workshop
"NMR of paramagnetic proteins and applications" 7th - 8th April 2022 (In presence and Virtual)

Chianti Workshop
"Opening New Doors for Magnetic Resonance" Principina Terra (Grosseto), Italy 19-24 June, 2022

Instruct-ing Structural Biologists Towards Integration 18-21 July 2022 An Instruct Training Course (Virtual)

Group Meetings

Friday, December 2nd, 2022 at 1:00 pm **Veronica Ghini** "COVID-19: a complex disease with a unique metabolic signature" CERM Conference room

Friday, November 18th, 2022 at 1:00 pm **Alessia Vignoli** "Risk stratification in patients treated with oral anticoagulants: an NMR-based metabolomic and lipoproteomic study" CERM Conference room

Friday, November 11th, 2022 at 1:00 pm **Marco Schiavina** "Elucidating SARS-CoV-2 nucleocapsid protein NTD structure with a novel tool" CERM Conference room

Friday, October 28th, 2022 at 1:00 pm **Gaia Meoni** "NMR-based metabolomics to estimate chemical and sensorial profile of olive oil" CERM Conference room

Friday, October 21st, 2022 at 1:00 pm **Vincenzo Laveglia** "Protein structure-based prediction of metal binding sites" CERM Conference room

Friday, October 7th, 2022 at 2:00 pm **Maria Salobehaj** "Niosomes. Where origins of life and drug delivery meet" online and CERM Conference room

Friday, September 30th, 2022 at 2:00 pm **Valentina Vitali** "Enlarging the scenario of site direct 19F protein labelling on tyrosine residues" CERM Conference room

Friday, September 23rd, 2022 at 6:00 pm **Lorenzo Bracaglia** "A step by step approach for the characterisation of IDPs at ultra-high field NMR" CERM Conference room

Friday, September 16th, 2022 at 2:00 pm **Lucrezia Cosottini** "Ferritin-based anticancers" CERM Conference room

Friday, June 17th, 2022 at 2:00 pm **Valentina Monaci** "Optimizing vaccine design for prevention of neonatal sepsis" CERM Conference room

Friday, May 27th, 2022 at 2:00 pm **Lan Pham** "^{19}F in-cell NMR Endogenous incorporation of ^{19}F-amino acid into target proteins in human cells" CERM Conference room

Friday, May 20th, 2022 at 2:00 pm **Giulia Licciardi** "How deep can we investigate olive oil relaxation?" CERM Conference room

MEETINGS & EVENTS

Friday, May 13th, 2022 at 2:00 pm **Milana Bazayeva** "Metal sites from a database perspective" CERM Conference room

Friday, May 6th, 2022 at 2:00 pm **Naomi Anna Consoli** "Characterization of innovative materials for hydrogen storage by Solid State NMR" CERM Conference room

Friday, April 29nd, 2022 at 2:00 pm **Francesco Bruno** "Journey to the center of the noise" CERM Conference room

Friday, April 22nd, 2022 at 2:00 pm **Luis Padilla** "Biophysical Characterization of Siglecs and Siglec-Sialoglycan Complexes" CERM Conference room and on line

Friday, April 1st, 2022 at 2:00 pm **Giulia Roxana Gheorghita** "Expression and biophysical characterization of Siglec proteins" online

Friday, March 25th, 2022 at 2:00 pm **Francesca Sacco** "Development of an innovative analytical platform, NMR- and structural MS- based, for advanced product knowledge" online

Friday, March 18th, 2022 at 2:00 pm **Lorenzo Niccoli** "Efficient Dynamic Nuclear Polarization up to 230 K with Hybrid BDPA-Nitroxide Radicals at High Magnetic Fields" online

Friday, March 11th, 2022 at 2:00 pm **Deborah Grifagni** "A multidisciplinary approach for investigating the CISD3 protein: from spectroscopic to cellular studies" online

Friday, March 4th, 2022 at 2:00 pm **Letizia Fiorucci** "Paramagnets in high field magnets" online

Friday, February 25th, 2022 at 2:00 pm **Beatrice Bargagna** "Chaperones in the CIA machinery involved in the assembly of [4Fe-4S] clusters" online

Friday, February 18th, 2022 at 2:00 pm **Anna Perez Y Rafol** "Expression of single and multidomain RRMs for the characterization of protein-RNA interactions" online

Friday, February 11th, 2022 at 2:00 pm **Francesco Torricella** "Novel living systems protein delivery for EPR based experiments" online

Friday, February 4th, 2022 at 2:00 pm **Francesca Di Cesare** "NMR-based metabolomics to investigate the association between AB0/Rh blood group systems and plasma metabolomic and lipoproteomic profiles" online

Friday, January 28th, 2022 at 2:00 pm **Francesco Milanesi** "Synthesis of a new glycomimetic as ligand of human galectin-3 and galectin-8" online

FUNDING INSTITUTIONS

Acknowledgements

University of Florence

Tuscany Regional Government

European Commission

Italian Ministry of University and Research

Fondazione Cassa di Risparmio di Firenze

Italian National Research Council

Italian National Institute of Health

FUNDING INSTITUTIONS

Cariplo Foundation

National Institutes of Health

Italian Association for Cancer Research

National Recovery and Resilience Plan (NRRP)

Italian Ministry of Agricolture

Italian Ministry of Health

CONTACT INFORMATION

Contact Information

CERM

Via Luigi Sacconi 6

50019 Sesto Fiorentino (FI)

Italy

www.cerm.unifi.it

Phone: + 39 055 4574270

Fax: + 39 055 4574923

E-mail: cerm@cerm.unifi.it

@cerm_cirmmp

https://www.linkedin.com/company/cerm-cirmmp/