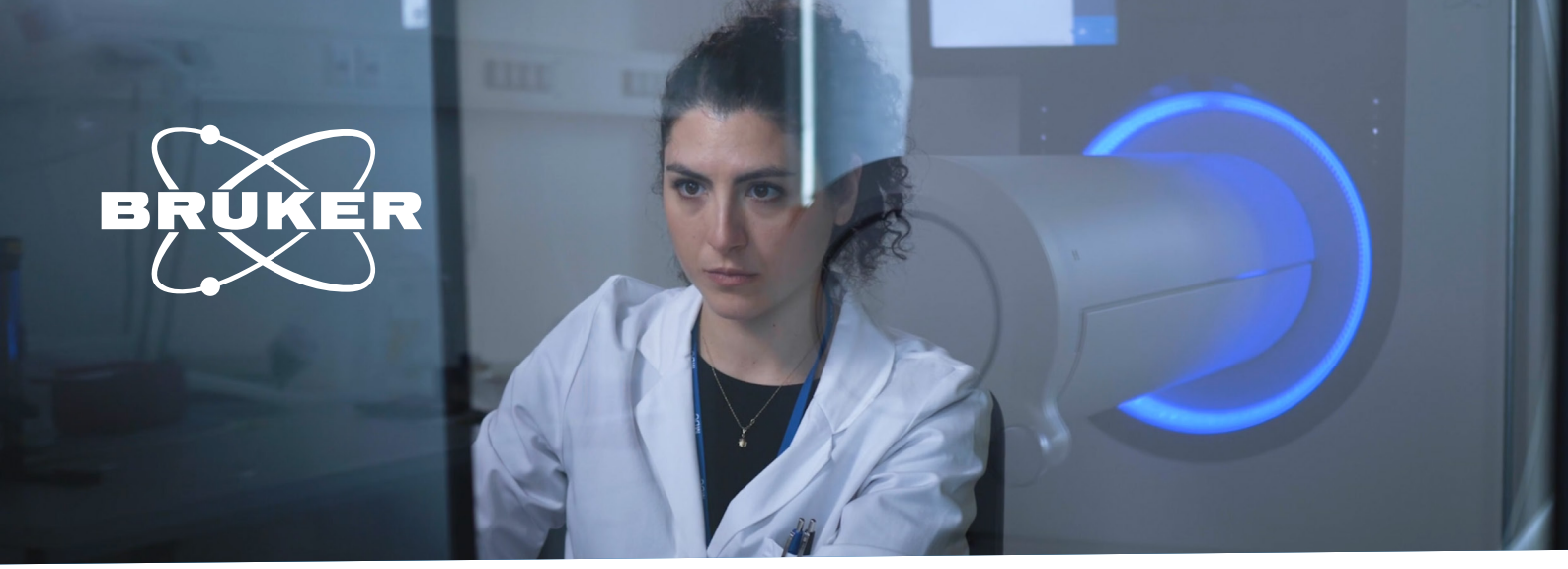


MRI

Promoting best practices and standardization in preclinical animal research

Innovation with Integrity

As one of the world's leading biomedical research institutions, the Max Delbrück Center (MDC) for Molecular Medicine in the Helmholtz Association in Berlin, Germany, is working towards a better understanding of the molecular basis of health and disease in an ongoing effort to bring these discoveries to patients for better prevention, diagnostics, and treatment. With the establishment of its Preclinical Research Center (PRC) in 2020, the MDC proudly supports the best possible conditions for animal research by working in accordance with the principles of humane experimental techniques, known as the 3Rs – reduce, refine, and replace. The center's focus on imaging technology in preclinical studies is one way the PRC works to reduce the number of animals needed for research, while also minimizing their stress from testing procedures.

From the onset, the PRC was outfitted with equipment and technology that was not previously available at the MDC, such as a two-photon microscope and special equipment for neuroscience behavioral experiments. Many of the tests are also used routinely on hospitalized patients, including ultrasound and clinical chemistry, the latter of which involves the analysis of urine samples, serum or blood plasma. A liquid cryogen-free-filling 3 Tesla Bruker BioSpec Maxwell magnetic resonance imaging (MRI) instrument was purchased specifically for the facility, making it possible to examine animals without any radiation exposure.

"The most requested service in our facility is undoubtedly structural neuroimaging, and cardiac imaging is the second," Dr. Giovanna Diletta Ielacqua explains. "We are fortunate to have a comprehensive research center equipped not only with MRI but also with computed tomography (CT), traditional ultrasound, functional ultrasound, and various other lab tests, providing a comprehensive approach to research needs. This versatility allows us to choose the most suitable modality for a given biological question."

In partnership with Bruker, the MDC PRC team is using its deep knowledge of advanced imaging technology to develop standardized practices that will improve animal studies in preclinical research.

Preclinical research on small animals

Non-invasive small animal imaging remains a critical resource in biomedical and pharmaceutical research. Investigators use imaging techniques to accurately assess developmental, behavioral, cardiovascular, and metabolic characteristics in rodent disease models over long periods of time and to sensitively screen for phenotypic variations. Researchers can also use imaging modalities to track changes in the same animal, and a single analysis can reveal detailed information about molecules and functions.

MRI is a powerful, non-invasive imaging technique, particularly suitable for imaging of soft tissues. It allows for acquisition of anatomical, as well as functional information in diverse research areas. The technique has gained popularity in the preclinical imaging community for its suitability in a wide range of applications.

The PRC team's work includes preclinical applications for both structural and functional measurements in different fields such as neuroimaging, oncology, cardiology, cardiovascular, and metabolic research via measurements in different organs, as well as in joints, bones, and whole-body imaging.[1] The facility's platform provides validated approaches for investigating physiological and morphological characteristics in mice and rats, as well as screening for phenotypic variations. As a result, the PRC provides researchers with an important resource for studying animal disease models.

"MRI, an exceptionally versatile imaging technique, can be applied across various domains and research areas," Dr. Ielacqua says. "This encompasses neuroimaging projects, such as the examination of brain structure, function, and perfusion. Additionally, it extends to addressing various heart malformations and assessing cardiac function. Our services go beyond data acquisition; we provide fully analyzed data. We collaborate closely with research groups to design studies and identify the best MRI sequences for optimal results."

The PRC team also uses multimodal imaging to provide deeper insights into complex biological phenomena by combining MRI with other techniques, such as CT and ultrasound. Multimodal imaging enhances the ability to gather detailed and diverse information, improve the accuracy of findings, and uncover new knowledge across various fields of study.

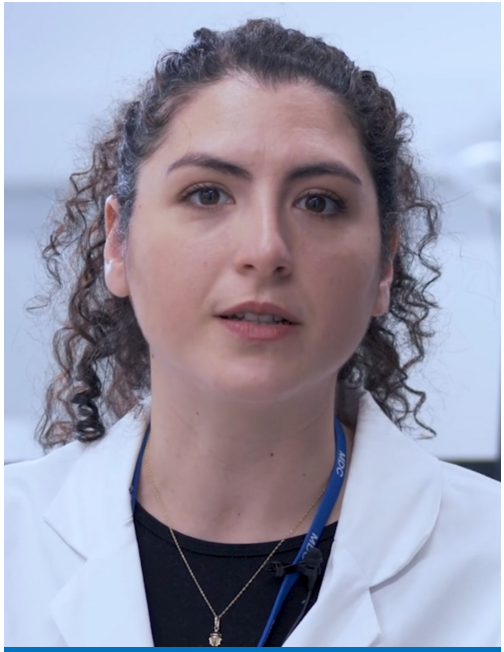
"Having this range of techniques under the same roof is advantageous," Dr. Ielacqua explains. "For instance, we might combine MRI with CT for bone studies. I also have conducted cardiac studies by combining MRI with conventional ultrasound, with MRI providing exceptional spatial resolution for anatomical assessments. We're not just an MRI lab. We are a comprehensive research center."

Standardization and best practices in preclinical imaging

The PRC team prides itself on following best practices, starting with the approval of an animal experiment only if there are no alternative methods to achieve the desired scientific objectives.[2] Even then, scientists may only use a minimum number of animals, and experiments must be humanely managed. Technological advances have made this possible, for example, the facility's BioSpec Maxwell 3 Tesla MRI is capable of temporal resolution that can produce detailed images of the beating heart. Mice are scanned under anesthesia, and a few minutes after the procedure is over, they are freely released onto a warmed platform.

Additionally, the PRC team strongly believes that standardization enhances the credibility, reliability, and efficiency of scientific research by providing a common framework for conducting experiments and collecting data. At times, even in the same institution, it can be challenging to compare results because they rely on various intricate factors like anesthesia, how animals are managed, physiological conditions, data collection, and analysis methods. That sparked Dr. Ielacqua's interest early in her career.

"I began to notice subtle differences such as mouse preparation techniques, statistical analysis, and the methods for reporting findings between different labs but also within the same one," she says. "These disparities extended to how we documented our research in publications. I started to realize the broader implications when I engaged with other research groups during conferences and lab visits.



Dr. Giovanna Diletta Ielacqua, DVM
As a senior scientist in the MDC's PRC, Dr. Ielacqua uses MRI technology to perform measurements in different animal organs or provide whole-body imaging services. Originally trained as a veterinarian, Dr. Ielacqua completed a PhD at ETH Zurich and then a postdoc at Stanford University. She joined the PRC, moving to Germany in March 2020 right before the nation's first lockdown. It took months until she finally had a chance to introduce herself in person to colleagues. While COVID-19 made the transition challenging, Dr. Ielacqua has found the mission of MDC and her experience with imaging technology to be an excellent fit.

"Over the course of a decade, I've been deeply immersed in MRI research," she said. "The prospect of taking on a challenging role in this innovative center appealed to me greatly. It felt like the perfect opportunity because it was a brand-new facility where I could apply my extensive expertise in this technique. This work will have implications far beyond our laboratory, improving scientific methodology that will impact both small animal studies and medical research."

It became increasingly evident that the lack of standardized protocols was hindering scientific progress. This realization raised significant concerns about how we could compare studies, even when conducted on the same animal models and with identical instruments."

While these factors can significantly affect experiment outcomes, Dr. Ielacqua believes that they are manageable and, to some extent, preventable. Having consistent protocols and established standard procedures would prevent redundant studies and, as a result, enhance research.

"This lack of transparency contributes to the difficulty of reproducing research findings, which is a critical component of the scientific method," she explains. "I came to recognize that true scientific advancement required a shift toward more open collaboration, data sharing, and standardized protocols to ensure that we were comparing apples to apples. This shift was essential to avoid redundant work and to foster genuine progress in the field."

Acting upon her concerns, Dr. Ielacqua has been working with the European Society for Molecular Imaging (ESMI) for several years to promote standardized imaging protocols for both animal welfare and fundamental research in pharmaceutical and biomedical fields. Currently serving as the co-chair of the Standardization in Small Animal Imaging group, she is leading efforts to create standardized imaging procedures that enable consistency of results across various laboratories or offer reference methods for creating comparable imaging protocols.

The ESMI study group created a social media survey in 2021 to gather knowledge and evoke discussion in the preclinical imaging community. The group then published a community-led consensus paper on best practices when collecting, analyzing, and publishing preclinical imaging data. Data collected in this project showed a need to promote and disseminate already available tools to standardize preclinical imaging practices.[3]

"As more people began to realize the benefits, enthusiasm grew," Dr. Ielacqua says. "Participants gained access to analysis tools and codes, while data sharing occurred anonymously to maintain confidentiality. The overarching aim was knowledge sharing, without any sense of exclusivity."

Dr. Ielacqua attributes part of the success of the initiative to a growing interest in collaboration, sparked by the scientific community's challenges during the COVID-19 pandemic, which mirrored her own experiences.

"The COVID-19 situation prompted a desire for collaboration, a trend we observed in the small animal MRI community," she explains. "Researchers were actively engaged online and receptive to collaborative studies. This marked the beginning, and since then, several MRI studies have followed a similar pattern of collaborative efforts."

Dr. Ielacqua's long association with Bruker also helped as the group worked towards the development of best practices and standardization in preclinical research.

"Bruker was involved in one of the early sessions about standardization," she says. "We recognized that while we could standardize many aspects, 84% of the surveyed users had Bruker scanners. So, collaborating with Bruker proved beneficial for everyone. More recently, we initiated a project with the Aswendt Group at University of Cologne where we developed a tool that could be integrated within Bruker ParaVision 360 MRI software. It takes a list of data sets and provides essential metrics like signal-to-noise (SNR) ratio. These parameters can help determine which data sets are suitable for analysis, making the process more efficient and standardized."

Bruker's preclinical instrumentation

The Bruker BioSpec Maxwell 3 Tesla has served as the cornerstone for much of the PRC's work, and Bruker and the PRC established a partnership early on. This relationship also resulted in ways for the PRC to share feedback on the Bruker technology used in its laboratory.

"When the PRC purchased the Bruker BioSpec Maxwell 3 Tesla, it was a relatively new technology, and Bruker established a formal collaboration with the PRC to gain insight into user experience," Dr. Ielacqua explains.

The BioSpec Maxwell 3 Tesla offers multiple advantages for the PRC team. The instrument's magnet technology requires no liquid helium filling, thereby extending the range of multi-purpose, preclinical MRI and MRS instruments. Its 170 millimeter bore allows imaging of not only mice and rats, but also other small rodents and the self-shielded instrument does not require a Faraday cage. The instrument is also equipped with a motorized animal handling system, including touchscreen operation for a fast and straightforward workflow to enable time-efficient and accurate animal positioning.

"One noteworthy feature of the Bruker instrument is the automated transport system for positioning animals," Dr. Ielacqua says. "Specifically, this feature is beneficial for placing mice within the scanner precisely, saving time, and minimizing disruptions to research activities. Moreover, the Bruker Maxwell magnet technology eliminates the need for liquid helium filling, providing long-term cost savings. The availability of diverse coils further enables us to focus on specific areas of the body or perform whole-body scans as needed."

Future plans

For the near future, the PRC team plans to continue engaging in discussions with other preclinical research scientists about MRI standardization. Dr. Ielacqua sees potential in improving standardization for related imaging technologies as well, such as the growing use of functional MRI (fMRI).

"Nowadays, there are more studies involving conscious animals, especially in fMRI, with no need to anaesthetize the animal," she explains. "From a technological standpoint, it would be beneficial to have larger bores where different techniques can run in parallel, such as with Bruker's multi-modal PET-MRI instruments."

Dr. Ielacqua also plans to continue the facility's on-going collaboration with Bruker by providing feedback on all aspects of her instrumentation, such as the Bruker MRI CryoProbe. *"This coil can potentially increase the signal to noise ratio (SNR) by a factor of two to five, as well as enhance the scan resolution,"* she says. *"That's crucial for fMRI as these sequences are demanding and very fast. By increasing the SNR, we can improve the accuracy of reading cerebral activity."*

Last, but certainly not least, Dr. Ielacqua sees the potential of machine learning in preclinical research data analysis. She's very interested in contributing to its successful implementation, particularly best practices regarding data quality, model interpretability, and ethical considerations. *"Several research groups at the MDC have expressed interest in analyzing data using machine learning algorithms,"* she explains. "*This trend is already taking hold and will undoubtedly grow as the technology develops. I'm looking forward to seeing how it will shape our work in the future."*

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