Human-brain ferritin studied by muon spin rotation: a pilot study

Lucia Bossoni1©, Laure Grand Moursel2,3, Marjolein Bulk2,3,4, Brecht G Simon1, Andrew Webb3, Louise van der Weerd2,3, Martina Huber1, Pietro Carretta5©, Alessandro Lascialfari6 and Tjerk H Oosterkamp1

1 Huygens-Kamerlingh Onnes Laboratory, Leiden University, 2333 CA Leiden, Netherlands
2 Department of Human Genetics, Leiden University Medical Center, Leiden, Netherlands
3 Department of Radiology, Leiden University Medical Center, Leiden, Netherlands
4 Percuros BV, Leiden, Netherlands
5 Department of Physics, Pavia University, Pavia, Italy
6 Dipartimento di Fisica, Università Degli Studi di Milano, Milano, Italy

E-mail: bossoni@physics.leidenuniv.nl

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Abstract
Muon spin rotation is employed to investigate the spin dynamics of ferritin proteins isolated from the brain of an Alzheimer’s disease (AD) patient and of a healthy control, using a sample of horse-spleen ferritin as a reference. A model based on the Néel theory of superparamagnetism is developed in order to interpret the spin relaxation rate of the muons stopped by the core of the protein. Using this model, our preliminary observations show that ferritins from the healthy control are filled with a mineral compatible with ferrihydrite, while ferritins from the AD patient contain a crystalline phase with a larger magnetocrystalline anisotropy, possibly compatible with magnetite or maghemite.

Keywords: muon spin rotation, nanomagnetism, Alzheimer’s disease, ferritin

(Supplementary material for this article is available online)

1. Introduction
Ferritin is a protein that attracts much interest, not only because of its crucial role in iron storage and ferroxidase activity [1, 2], but also because of its magnetic properties. Ferritin is a nanoscopic hollow protein made of a shell (apoferritin) of molecular weight 450 kDa, containing a core of trivalent iron (Fe(III)) in the mineral form of ferrihydrite, a nano-crystal quite elusive to x-ray diffraction. Ferritin acquires Fe(II), catalyzes iron oxidation, and induces mineralization within its cavity [3]. The outer diameter of the shell is 12 nm, regardless of the iron loading, whereas the iron core diameter can vary between 2–3 nm and 7 nm [4], depending on the number of stored ions.

It is generally agreed that the ferritin core is antiferromagnetic (AFM) below a temperature in the range of 340–500 K [4–6]. However, some AFM sublattices do not cancel out completely due to the small particle size. This results in an excess of spin orientation, giving rise to a magnetic moment of 225–400 $\mu_B$ [4, 7, 8]. Because of its hexagonal crystal structure (space group $P6_3mc$), the particle possesses a unique easy axis of magnetization [9]. The ‘giant’ magnetic moment of the particle can rotate about the crystal easy axis, if it overcomes an energy barrier $E_a$, which depends on the volume of the particle $V$ and on the magnetocrystalline anisotropy constant, $K$ [10]. If, upon decreasing the temperature, the dynamic time constant of the moment crossing the barrier ($\tau_c$) is greater than the measuring time of the specific experimental technique ($\tau_m$), the magnetic moment is said to be blocked [11]. This formalism was introduced by Néel to describe the magnetism of nanoscopic single-domain particles with an internal magnetic order. The magnetic behaviour of an assembly of these ultra-fine particles was termed superparamagnetism (SPM). This blocking occurs at about 12 K, for
ferritin, when $\tau_m \sim 100$ s [12], in a DC magnetometry measurement. In addition to the AFM and SPM phases, it was also proposed that a Curie–Weiss-like behavior could be found at the core-shell interface, as a result of the reduced Weiss field [7, 10]. More recently, other models have been proposed, yet the exact spin structure of the nanoparticle and its magnetic properties are still a matter of debate [4, 7, 8, 13].

Ferritin has also been extensively studied by neuroscientists, due to its central role in cellular iron homeostasis. Ferritin is at the center of many debates concerning iron toxicity in relation to neurodegeneration, and in particular to Alzheimer’s disease (AD). In the brain of AD patients, iron dys-regulation has been reported [14]. This may indicate a malfunction of the storage protein [15], from which iron can leak out and take part into toxic reactions such as the Fenton and Haber–Weiss reactions [1, 16–19]. Additionally, a recent longitudinal study showed that baseline cerebral spinal fluid (CSF) ferritin levels are negatively associated with cognitive performance over 7 years, and predicted conversion of mild cognitive impairment into AD [20]. Moreover, ferritin was found to be strongly associated with CSF apolipoprotein E levels and was elevated in presence of the Alzheimer’s risk allele, APOE-ε4. Microscopy studies of ferritin cores revealed the existence of ferritins with a poly-phasic structure: ferritins found in the brain of AD patients seem to contain a higher amount of cubic crystalline phases consistent with magnetite and wüstite [21, 22], whereas the ‘healthy type’-ferritins may be more abundant in the hexagonal ferricydride phase. In this scenario, pathological ferritin would be better described by magnetoferritin, an artificial complex made of apoferritin containing a magnetite or maghemite crystal [23]. However, these observations were not confirmed by nuclear magnetic resonance (NMR) [24]. Since magnetoferritin carries a magnetic moment larger than ferricydride, i.e. typically between 2000 $\mu_B$ and 9000 $\mu_B$ [25], one would expect a 200-fold (or larger) increase in the longitudinal and transverse relaxation rates of the water protons surrounding the protein. However, no significant enhancement of the relaxation rate was observed.

Additionally, Pan et al showed that in human-liver ferritin, an increasing percentage of octahedrally coordinated Fe(III) migrated to tetrahedral sites and was partially reduced to Fe(II), upon increasing the electron dose in electron microscopy experiments [26]. Although it is rather unlikely that such alterations would happen only in the ‘pathological’ ferritin [27], the poly-phasic composition of physiological versus pathological ferritins remains a debated issue.

In order to unravel these controversies, here we propose a muon spin rotation ($\mu$SR) experiment, as an alternative investigation technique.

$\mu$SR has been successfully employed in the past to study the magnetism of fine-particles systems. In particular, horse-spleen ferritin [28, 29] and similar artificial compounds [30] have shown a two-component relaxation of the muon asymmetry: while the fast-decaying asymmetry (exponential- or Kubo–Toyabe-like) is informative of the superparamagnetism taking place in the iron core, the slow exponential tail has been ascribed to the interaction of the muon spin with the protons of the organic shell. However, a study of ferritin purified from human tissue has not been undertaken yet.

In this manuscript, we present a $\mu$SR study of a ferritin sample isolated from the brain of an AD patient, and an age- and gender-matched healthy control (HC). As a third sample and reference, a lyophilized commercial horse-spleen ferritin sample (HoSF) was used. Firstly, we evaluate the feasibility of $\mu$SR on a human sample. Then we propose a model to interpret the spin dynamics of the ferritin iron core, associated with the SPM effect. Secondly, we draw some preliminary conclusions on the mineral composition of the protein core based on the magnetocrystalline anisotropy constant derived from our model, showing that all steps of the analysis are feasible. Finally, we discuss the limitations and the improvements that should be undertaken in future studies.

2. Sample preparation and characterization

One freshly frozen human-brain hemisphere from an AD case (Braak stage 6C, age 90 yrs, female) and one from a healthy age- and gender-matched control (age 88 yrs, female) were obtained from the Netherlands Brain Bank (NBB) of Amsterdam. Patient anonymity was strictly maintained and informed consent was obtained from all prospective donors by NBB in accordance with EU regulations. All tissue samples were handled in a coded fashion, according to Dutch national ethical guidelines (Code for Proper Secondary Use of Human Tissue, Dutch Federation of Medical Scientific Societies).

Before the tissue was processed for protein isolation, a section of the temporal region was selected from both the AD and the control individual for an MRI study (see supplementary information). Ferritin used in the $\mu$SR experiment was isolated from 2/3 of the hemispheres of the two individuals by following a modified version of the method reported by Cham et al [31]. The original protocol requires only a few steps and it is practical when dealing with a large amount of brain material. Moreover, this method retains most of the iron inside the protein, that is our main concern in this experiment. One part of the brain was homogenized on ice with three parts of phosphate-buffered saline solution, with a tissue homogenizer (Omni B style TH Motor 220 V, LA Biosystems). Subsequently, the nuclei and unbroken cells were removed by low-speed centrifugation: 2000 g for 15 min, at 4 °C. The supernatant was retained and put on ice again. This solution was further diluted with pure methanol 99.8% (Baker, product num.: 8045.2500) in order to reach a final concentration of 40% v v⁻¹. This new solution was heated for 10 min at 75 °C, and afterward put on ice again, and finally centrifuged at 2000 g for 15 min, at 4 °C. The ferritin-containing supernatant (final volume: 1.5–2 l) was retained in order to be further purified, desalted and concentrated with an Amicon Ultra-15 Centrifugal Filter Unit with an Ultracel-100 membrane (UFC910008) with a molecular cutoff of 100 kDa. Aliquots of the supernatants were assayed for ferritin by SDS gel electrophoresis, western blot, and transmission electron microscopy. The remaining solution was freeze-dried for the magnetic characterization. In order to prevent contamination with
magnetic material, no metal tools or containers were used in the dissection and handling of the tissue. Ceramic and plastic materials were used instead.

SDS-gel electrophoresis was done in parallel to western blot analysis: the results show the presence of both ferritin sub-bands in the human samples (figure 1).

One part of the gel was stained to assess purity and protein concentration, while the other part was used for western blot to determine the presence and molecular weight of ferritin. Protein solutions were loaded on the lanes of a 4–20% Criterion TGX™ pre-cast gel purchased from Bio-Rad. As a reference, precision plus protein™ dual Xtra Prestained Protein Standards (product number: 1610377) and ferritin from equine spleen purchased from Sigma-Aldrich (product number: F4503) were employed. Horse-spleen ferritin was loaded as a reference sample in three different concentrations. The gel was run using a standard Laemmli sample buffer and Tris/glycine/SDS running buffer. The gel was stained with PageBlue™ protein staining solution (ThermoFisher Scientific, product number: 24620) and imaged with the Li–Cor Odyssey™ imaging system. The AD ferritin concentration, while the other part was used for western blot. In these experiments, the static spin susceptibility is measured at 50 G after cooling the sample in two different ways. In the zero-field-cooled (ZFC) case, the sample is cooled from room temperature, down to 2 K without applying a magnetic field. Then the field is increased to 50 G and the static susceptibility is measured. In contrast, in the field-cooled (FC) case, the sample is cooled with the field of 50 G applied continuously. The static spin susceptibility (χ) of the human ferritin samples is shown in figure 3. The ZFC χ shows the typical peak of a superparamagnet after subtracting a Curie–Weiss trend from the data [12, 33] (inset figure 3). The AD ferritin-χ shows a broad peak, centered around 8.8 ± 0.9 K, indicating a blocking temperature (T_b) smaller than that observed in the HoSF sample (12 ± 1 K, χ data are shown elsewhere [34]). On the other hand, the HC ferritin shows a peak in χ, at the same temperature as the one of the HoSF sample. These values of T_b suggest that the average core size of the AD sample is smaller than the one of the healthy control and horse-spleen samples, in agreement with the TEM characterization. Please note that the FC curve does not show this peak, as 50 G is enough to block the superparamagnet at low temperatures.
temperatures. Furthermore, the presence of a peak in the ZFC curve in the range of ∼9–12 K rules out the possibility that a substantial part of our sample contains magnetite particles of tens of nm [34–37].

Finally, the presence of iron in all three powder samples was confirmed by laser-ablation inductively coupled mass spectrometry.

3. Results

Ferritin powders were loaded onto a kapton holder, with a thin (25 μm) kapton window to allow the muons to reach the sample. Experiments were carried out in longitudinal geometry, both in zero-field and longitudinal field mode. The experiments were performed at the GPS beamline of the Paul
Scherrer Institute, where a continuous wave (CW) muon beam is available. CW muons allow a high time resolution, which in turn enables strongly inhomogeneous internal fields to be probed. A forward and a backward detector were used at all times. The time-dependent asymmetry $A(t)$ function was calculated as:

$$A(t) = \frac{F(t) - \alpha B(t)}{F(t) + \alpha B(t)}$$

(1)

where $F(t)$ and $B(t)$ are the positron counts in the forward and backward detector respectively, while $\alpha$ is an instrumental parameter which compensates for the difference in efficiency between the two detectors. Our estimated $\alpha$ ranged between 0.787 and 0.859, and it was calibrated in a transverse field of 50 G, at high temperature (170 K). Data were bunched with the ‘constant error’ binning option of the Wimda software [38], in which the bin length exponentially increases with time from the initial value so that the counts per bin and the resulting error remain fixed (see supplementary information (stacks.iop.org/JPhysCM/29/415801/mmedia)). Data were fitted between 0 and 6 $\mu$s. Different fitting functions were tested, such as the Uemura function [39], various combinations of Lorentzian and Gaussian Kubo–Toyabe [40]. The dynamical Kubo–Toyabe and Keren functions [41] were also tested, but they could not capture all the features of the data, as the empirical model discussed in this manuscript. Muon spin rotation raw data were processed with Wimda, and fitted with WinDa, Matlab 2016a and OriginLab(R) Pro 2016.

### 3.1. Horse–spleen ferritin zero-field asymmetry

Before carrying out the $\mu$SR study on the human-brain ferritin, a sample of freeze-dried horse-spleen ferritin was investigated. Horse-spleen ferritin was chosen because it is similar to human ferritin and therefore a good reference. Zero-field $\mu$SR on HoSF shows a change in the muon asymmetry decay $A(t)$ as a function of the temperature (figure 4). We observed an initial asymmetry $(A_0)$ of $\sim 15\%$, which equals 2/3 of the full asymmetry, as it will be discussed later.

In the zero-field measurement data, two regimes were identified. At temperatures higher than 20 K, the asymmetry is well described by the sum of two contributions, namely an exponential term, decaying in the first 300–500 ns, and a slower stretched-exponential component, evolving from a Gaussian at high temperature, to a single exponential at low temperature. The decay of $A(t)$ was modeled by assuming that a fraction of muons probes the local magnetic field of the protein shell ($f_{\text{shell}}$), and the rest probes the core ($f_{\text{core}}$):

$$A(t) = A_0 (f_{\text{core}} e^{-\lambda_{\text{1}} t} + f_{\text{shell}} e^{-\lambda t^\beta}) + B.$$ 

(2)

We observed a fraction ratio of $f_{\text{core}} / f_{\text{shell}} \sim \frac{1}{4}$. A similar value was found by Cristofolini et al, who suggested that this fraction may originate from the core/shell mass ratio [28].

In equation (2), $\lambda_1$ represents the spin-lattice relaxation rate of the ‘core muons’, analogous to the NMR $1/T_1$ [43, 44]. As it will be shown, the fast decaying component is weakly dependent on the application of a longitudinal magnetic field. Finally, $\lambda$ is the spin relaxation rate of the muons probing the protein shell, and the stretched exponent in equation (2) can be associated with either a distribution of muon sites or with...
an anisotropic hyperfine coupling, leading to a distribution of relaxation rates.

Below 20K, the asymmetry shows an increase at times longer than $\sim 4$ μs (figure 4), indicating the onset of a static magnetic phase. We note that, in contrast to the work of Cristofolini et al on HoSF [28], here the Gaussian decay is not reached at any temperature below 20 K. The best fit to the data was obtained by phenomenologically adding a slow oscillating term, with a small amplitude ($f_{sm} \sim 6\%$), a frequency $\omega$, and a phase $\phi$:

$$A(t) = A_0(f_{core} e^{-\lambda t} + f_{shell} e^{-\lambda_1 t} + f_{sm} \cos(\omega t + \phi)) + B, (3)$$

where the baseline was deduced from the high-temperature regime, and $f_{sm}$ represents the fraction of ‘shell muons’ partially recovering the asymmetry due to slowing down of iron spin fluctuations in correspondence with the blocking transition.

3.2. Human-brain ferritin zero-field asymmetry

The asymmetry decay of the human-brain ferritin is remarkably similar to that of HoSF, as far as the high-temperature limit is concerned (figure 5), therefore we fitted the data with the high-temperature model of equation (2). However, by a more careful inspection of the data, some differences can be observed between the human and the horse data sets. The most remarkable is that the human ferritin asymmetry does not show the same clear increase at long times and at low temperature, as the one seen in figure 4. This may be due to the lower ferritin concentration in the human sample. Secondly, the stretched exponent does not decrease below 1.4 in both human samples, even when $\beta$ is left free to vary, thus showing that the mono-exponential limit is never reached (figure 6). Finally, the initial asymmetry of the human data is smaller than in the HoSF data (figure 5), as it will be discussed in the next section.

Figure 6 summarizes the best-fit parameters for the relaxation rates ($\lambda, \lambda_1$), stretched exponent ($\beta$), and the initial asymmetry ($A_0$) as a function of the temperature, for the three samples. We recall that the symmetry decay of human ferritins was described by a model where the ratio of the core-shell muon fraction was fixed. The baseline was left free to vary, but it did not show significant variations with the temperature. The stretched exponent was fixed below the temperature of the peak observed in $\lambda_1$, in order to avoid parameter covariance ($\beta = 1.4$ for AD ferritin and $\beta = 1.5$ for the HC).

3.3. Longitudinal-field $\mu$SR measurements on human-brain and horse-spleen ferritin

Longitudinal-field (LF) $\mu$SR on the horse and human ferritin shows two effects in the muon asymmetry. Firstly, the application of a weak field enhances the initial asymmetry $A_0$, as also observed by other authors in a HoSF sample [28, 29]. Secondly, the stretched exponential relaxation is partially quenched by the longitudinal field (figure 7). The loss of $A_0$ was earlier ascribed to muonium formation (a radioactive isotope of hydrogen) after the $\mu^+$ captures an electron, as often occurs in organic matter and in polymers [45, 46]. In this case, the polarization can be fully recovered by the application of a longitudinal magnetic field, large enough to decouple the hyperfine interaction between the electron and the muon. Given the large amount of organic material in our sample and the partial recovery of $A_0$ with the application of an LF, it is reasonable to assume that the signal loss at $t = 0$ is mostly due to...
to muonium formation. Moreover, since the human sample is about seven times more dilute than the HoSF, one may ascribe the larger loss of initial asymmetry to a larger fraction of other proteins in the human sample (figure 5).

The quenching of the stretched exponential relaxation by small fields is in agreement with the hypothesis that this decay originates from the muons implanting in the protein shell, and probing weak dipolar fields of the proton nuclei. Moreover, the fast exponential term is not affected by the LF, thus suggesting that in this case, the local field is either static and strong (i.e. a few Tesla) or dynamic, as expected for the iron core. Indeed, another possible source of initial asymmetry drop might be the fast relaxation of muons stopping inside the magnetic core of the particle. However, this second contribution is unlikely to be recovered by a longitudinal field of a few tens or hundreds of Gauss.
4. Discussion

Among the different magnetic resonance techniques, muon spin rotation benefits from an almost 100% spin-polarized muon beam, leading to a signal which is not limited by the Boltzmann polarization. Since the spin probe (the muon itself) is implanted into the sample, the technique is neither limited by the sensitivity nor by the natural abundance of the internal probe, as it is in the case of NMR. Additionally, the signal intensity is not dependent on the strength of the external magnetic field. In μSR, zero-field experiments are possible and the muon current is so small (∼pA) that no or minor sample damage is expected.

Moreover, muon spin rotation offers several advantages to the study of nanoscopic magnetic systems: it directly probes the stray field produced by the metallic nanoparticle, and it captures its spin dynamics in a broad dynamic range (between $10^{-12}$ s and $10^{-5}$ s). On the other hand, obtaining structural information from relaxation rates still remains a challenge. In the next section, we propose a model to interpret the spin-lattice relaxation rate of the muon fraction stopping in the iron core of the protein ($\lambda_1$).

There are different channels for the spin relaxation of muons implanting in ferritin: (i) spin excitations of the AFM ordered lattice, which would appear at low temperatures, in the form of a power law [47, 48], (ii) Néel relaxation of the particle giant moment, activated by thermal energy, and (iii) muonium relaxation driven by the hyperfine coupling with electron spins undergoing a magnetic transition. While the first mechanism is not observed here, distinguish between the last two mechanisms is beyond the scope of this study. However, the data can be nicely fitted in the first ns to a single fast exponential, that we ascribe here to a magnetic transition. We recall that only a relaxation driven by the hyperfine coupling with electron spins is in the case of NMR. Additionally, the signal intensity is not dependent on the strength of the external magnetic field. In

$$\lambda_1(T, V) \propto \gamma^2(h_0^2) \int (S_i(0)S_i(t))e^{-\omega_0^2t^2}dt$$

$$= \gamma^2(h_0^2) \frac{\tau_0}{\tau_0^2 + \omega_0^2} + 1$$

(6)

where $\langle h_0^2 \rangle$ is the static $rms$ value of the hyperfine field, $\omega_0 = \gamma B_0$, $\gamma = 8.516 \times 10^4$ rad s$^{-1}$ G$^{-1}$ is the muon gyromagnetic ratio, and $B_0$ is the local field probed by the muons implanting in the core [53]. In zero-field, the core-muons will probe the dipolar field of the diluted iron spins. A local field of 8 G was used in the simulations. A similar field was found by Cristofolini et al. [28], and it is consistent with a point dipolar field produced by a single iron spin at 1.8 nm distance. Moreover, since the ferritins’ diameters are log-normally distributed, as observed by TEM, equation (6) should be integrated over the volume distribution:

$$\lambda_1(T) \propto \int V_0 \lambda_1(T, V) \exp\left(-\frac{(\ln(V) - \langle \ln(V) \rangle)^2}{2\sigma^2}\right) \Delta V \sqrt{2\pi} dV$$

(7)

where $\Delta$ and $\langle \ln(V) \rangle$ are respectively the standard deviation and the mean of the log-normal distribution. In order to interpret $\lambda_1$, we choose $\tau_0$ and $K$ from literature values for ferrhydr (see figure 8 and its caption).

The simulation in figure 8 (a) shows that particles with a larger mean volume display a peak in $\lambda_1$ at higher temperature, and that particles with a broader particle-size distribution display a broader peak in $\lambda_1$ (as observed for HC). Figure 8 (b) shows a positive correlation between the value of $K$ and the temperature of the peak in $\lambda_1$. Here, instead of choosing a single volume for $K$, we present a range of values, as reported in literature. Indeed, $K$ depends on several factors such as chemical composition, kind of magnetic ion, particle shape, amount of frustrated spins at the particle surface, distribution of nucleation sites inside the core, and level of crystallinity. Plausible values for $K$, in the case of the ferrhydrate mineral, range from 0.7 to 2.5 K nm$^{-3}$ [55–57], while for magnetoferritin, and magnetite nanoparticles they can be found between 2.7 and 9.7 K nm$^{-3}$ or higher [23, 58]. However, intermediate values of 2.97 K nm$^{-3}$ have also been measured for HoSF [54]. In figure 8 (b) the chosen $K$ values are limited to 7.7 K nm$^{-3}$, for illustration purposes.

A comparison between the experimental and simulated $\lambda_1$ is shown in figure 9, for the three investigated samples. All the samples show a broad peak in $\lambda_1$, in the same temperature range observed in other works on HoSF and its synthetic analogues [28, 30]. In the case of HoSF, the acceptable values for $K$ fall between 2.5 and 2.7 K nm$^{-3}$ (figure 9 (a)), which is in line with the literature findings for HoSF, as discussed above. We note that $K$ may be overestimated, if the actual mean particle size was larger than considered here. The HC sample shows a broader peak centered in a temperature region compatible with $K \sim 2$ K nm$^{-3}$, which suggests the presence of a single ferrhydrate phase (figure 9 (c)), in agreement with the expectations for ferrihydrate and possibly characterized by a
lower degree of crystallinity with respect to the HoSF. Finally, the AD sample shows a narrower peak in $\lambda_1$, which results from a narrower particle-size distribution (figure 9 (b)), and a larger anisotropy constant of $K \sim 3.7$ K nm$^{-3}$, which is 45% higher than what can be expected only from a size effect [58].

This constant falls in the experimentally observed range of magnetoanisotropy for magnetoferritin nanoparticles.

It is worth stressing that this is the first attempt to use muon spin rotation to determine the mineralization form of the ferritin-iron core in human-brain ferritin, as a complementary tool to electron microscopy. Therefore, in order to improve the current data, and confirm our preliminary results, the human ferritin concentration and purity should be improved. A possibility would be to add a further purification step, such as affinity chromatography, at the end of our protocol. As many of these steps can be further improved, future experiments are certainly feasible and will be sensitive enough to determine differences with greater accuracy.

5. Conclusions

We isolated ferritin from the brain of an AD patient and a healthy age- and gender-matched individual. The protein solution was characterized by biochemical and physical techniques assessing the protein concentration and verifying the superparamagnetic properties, and the presence of iron in the sample. The characterization shows that the isolation protocol needs further improvements in order to significantly increase the concentration and purity of the ferritin solution.

Zero-field $\mu$SR was then used to probe the spin dynamics of the iron core of human-brain ferritin, as a function of the
temperature. Our pilot experiment showed asymmetry spectra that significantly resembled those of commercial and highly pure horse-spleen ferritin, which was used as a reference in this study. A broad peak in the spin-lattice relaxation rate of the muons stopping in the core allowed us to identify the blocking temperature of ferritin. We proposed a model to interpret the spin-lattice relaxation rate, based on the Néel theory of superparamagnetism, and on the experimental size distribution of the obtained ferritin particles. The comparison between simulation and experimental data gave us an indication of the magnetocrystalline anisotropy constant of the ferritin iron core. Our analysis suggests that ferritin isolated from the Alzheimer patient contains an iron mineralization form with a larger K constant, in the range observed for magnetoferritin, when the size distribution obtained from electron microscopy is taken into account. However, to draw further conclusions about the composition of ferritin in relation with AD, ferritin isolated from different AD patients should be investigated. This pilot study shows that μSR is a promising approach to the study of iron-loaded human-brain proteins.

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ORCID iDs

Lucia Bossoni  https://orcid.org/0000-0003-3156-4696
Pietro Carretta  https://orcid.org/0000-0002-0605-7310

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