Non-invasive measurement of iron overload in the human body

Mauro Marinelli, Stefano Cuneo, Barbara Gianesin, Antonella Lavagetto, Martina Lamagna, Eraldo Oliveri, Giuliano Sobrero, Laura Terenzani, Gian Luca Forni

Abstract—We present a magnetic susceptometer to measure the iron overload in patients with pathology, like thalassemia, producing iron store in the body. The output signal is the difference between the induced voltages in two pickups that are located symmetrically with respect to an AC magnetic source. The overall dimensions allow the presence of the whole human torso between the magnetic source and one of the pickup. All the apparatus components work at room temperature; the symmetry, stability and the temperature control, at the milliKelvin level, of the complete apparatus gives the sensitivity, better than 10⁻⁷, necessary to detect the iron quantity of interest for these pathologies. The measurements of human beings recently started in the Center for Congenital Anemia of the "E.O. Ospedali Galliera" in Genoa Italy. The apparatus description, its calibration with healthy volunteers without iron overload, and the preliminary results with patients are presented.

Index Terms—Iron overload, Liver iron concentration, LIC, Susceptometer, Thalassemia.

I. INTRODUCTION

Accurate assessment of body-iron accumulation is essential for managing therapy of iron-chelating diseases characterized by iron overload such as thalassemia, hereditary hemochromatosis, and other forms of severe anemia. At present, the gold standard to determine liver-iron concentration (LIC) is liver needle biopsy. In this work, we present an alternative non-invasive technique to measure LIC based on a room-temperature susceptometer (the technology is subject matter of an international patent application). SQUID biosusceptometers [1]-[4], and MRI [5] are currently the only validated non-invasive methods for LIC measurements. However, SQUIDs are liquid helium-cooled superconducting devices, therefore costly and resource intensive. Furthermore, SQUIDs are only sensitive to a fraction of the liver volume because of their magnetic configuration [6]. MRI requires large magnets with dedicated software and hardware, trained operators, and its accuracy worsen at high iron concentration.

The susceptometer presented herein measures iron overload in the whole liver, as the entire human torso fits within its region of sensitivity. Since all of its components operate at room temperature, this susceptometer is more affordable than competing techniques and can reach a wider hospital base.

The signal picked up by the susceptometer has two sources: an overall magnetic background of the torso and an eventual contribution from liver iron excess. After measuring the magnetic signature of a patient, statistical analysis method is employed to estimate the background signal, given the patient anthropometric data. Liver-iron overload is then determined by subtraction of the estimated background from the total measured signal. The refinement of the methodology is in progress and, at present, the error in liver iron is about 1 g (SD), corresponding to a concentration of 0.5 mg/cm² for a 2 l liver volume.

Since February 2005, after the approval by the Ethics Committee, 40 patients and 68 healthy volunteers have been measured. A correlation study between iron overload and blood serum-ferritin concentration in the patient population attained a correlation coefficient R~0.68. Comparison with measurements of LIC via SQUID susceptometry on a subset of 30 patients participating in the present study (carried out by Dr. A. Piga at Ospedale S. Anna, Torino, Italy) yields a correlation coefficient R~0.77. Four patients (3 thalassemia major, 1 hereditary hemochromatosis) under intensive iron depletive therapy have been measured at least twice; our estimate of liver iron reduction is compatible with the clinical data (R~0.81). Comparison with LIC measured via biopsy is in progress. All comparisons were performed blind.

After the description of the apparatus, we report the phantom and the human being measurements, to show that the instrument intrinsic noise makes possible detection of the quantity of iron overload interesting for these illnesses.
II. INSTRUMENTATION

Paramagnetic materials have atoms arranged with permanent intrinsic magnetic moments. Susceptibilities arise from competition between the aligning effect of the applied field and thermal vibrations. The susceptibility $\chi$ of paramagnetic atoms [11] at the equilibrium temperature $T$ (Kelvin) is the ratio between their contribution to the body magnetization and the applied magnetic field; accounting for the iron atomic weight, the Curie law is

$$\chi_{Fe} = \frac{\mu_B N}{V} \frac{p^2 B^2 E_2}{3kT} = \left( 2.8 \cdot 10^{-9} \frac{K}{Kg/m^3} \right) \times \frac{(p^2 C)}{T}$$

(1)

where $N/V$ is the number of iron atoms per unit volume, $C$ (kg/m$^3$) is their concentration, $p$ is the effective Bohr magneton number, $\mu_B = 9.27 \cdot 10^{-24} J/T$. The magnetic moment of the ferric (Fe$^{3+}$) ions is 5.9 $\mu_B$ and literature reports values ranging between 3.7 $\mu_B$ and 4.2 $\mu_B$ for ferritin iron and between 3.3 $\mu_B$ and 4.7 $\mu_B$ for hemosiderin iron [12]-[14]. For $p = 4.2$ and $T = 310$ K, the ratio between the iron susceptibility and the concentration is $1.6 \cdot 10^{-6}$ m$^3$/kg. The iron concentration in normal human liver is a few hundred micrograms per gram of wet tissue ($g_{wet}$) and more than 10 mg/g for overload states [15], [16]. The body susceptibility is determined by two contributions arising from magnetic susceptibilities of paramagnetic iron and of diamagnetic tissues. Normal body tissues have a magnetic susceptibility very close to the one of water ($\chi_w = -9.0 \cdot 10^{-6}$).

The present susceptometer is composed of two pickup coils (only one is shown in Fig. 1) that are symmetrically located in respect to the AC magnetic source (Fig. 1 and 4). A lock-in amplifier, locked with the magnet oscillating current, measures the difference between the induced voltages in the two pickups. A concentration of 620 $\mu g/cm^3$ of iron atoms, with an effective magnetic moment of 4 $\mu_B$ and at 37 C, gives a positive contribution to the susceptibility $\chi_{Fe} = +0.9 \cdot 10^{-6}$, which is 1/10 of the absolute value of the water susceptibility. So to measure the contribution of the iron, at concentrations of interest, the sensitivity must be better than one part over ten million. The stability of the apparatus temperature (room temperature) within a few milliKelvin is a necessary condition to get this sensitivity.

A fiber-reinforced resin structure holds the magnets and the pickups of the present susceptometer (Fig. 2). A thermal shield wraps this structure without touching it (Fig. 3). The magnets temperature is kept stable within
a few milliKelvin by temperature controlled water, flowing through their hollow copper conductors.

A sample, with susceptibility $\chi(\tau)$ and volume $V$ (m$^3$), placed between the magnet and one of the pickups (Fig. 1), generates the signal $\phi$ (volt) [17]

$$\phi = \int g(\tau)\chi(\tau)\,d\tau$$  \hspace{1cm} (2)

the weight function $g(\tau)$ (V/m$^3$) depends on the frequency and amplitude of the magnet current and on the magnet and pickup dimensions. Two concentric magnets and two sets of concentric pickups generate four weight functions. The diameters of the inner magnet (IM) and pickup (IP) (Fig. 4) have been chosen making a trade-off between reducing the spatial spread of the weight function $g$ and allowing a significant contribution to the signal by the inner part of the body. Both the IM and the outer magnet (OM) are made of 128 turns of hollow copper conductor; the mean IM diameter is 140 mm and the OM one is 400 mm. The IM current is 38 A rms at 234 Hz and the OM one is 19 A rms at 195 Hz. Layers of printed circuit boards build the pickups. Each IP is made of 740 turns and has a mean diameter of 161 mm. Similarly each OP has 380 turns and its mean diameter is 401 mm. The distance between the middle planes of the upper and lower pickups is 880 mm. The aperture of the thermal shield, available for the human body and the stretcher, is 265 mm wide. The magnetic field in this entire region is lower than $1.9 \times 10^{-2}$ T [18].

Aqueous solutions of hexahydrate ferric chloride ($\text{FeCl}_3 \cdot 6\text{H}_2\text{O}$) have been used to calibrate the instrument. The contribution to the solution susceptibility of the diamagnetic Cl$^-$ ions is negligible (the Fe$^{3+}$ molar susceptibility at room temperature is $+1.9 \times 10^{-3}$ m$^3$/mole and the one of Cl$^-$ is $-3.04 \times 10^{-10}$ m$^3$/mole). Equation (1) has been verified in function of the ferric (Fe$^{3+}$) ions concentration and the solution temperature. The magnetic signal of the eddy currents, induced in the sample by the oscillating magnetic field, lags the magnetization signal by 1/4 of period. Considering the inductance of the eddy currents loops within the sample and resistivity of the solutions, it is possible to show that, at the magnetic field frequencies, the delay of the eddy currents relative to the induced electric field is completely negligible. This is true also for the eddy currents induced in the human body. The phase-locked detection allows measuring both the magnetization and the eddy current signals.

The susceptometer works in a normal hospital room, without using any magnetic shield. In order to account for the changes of the environment magnetic properties (arrival or departure of a car from the nearby parking lot, shifting of a metallic furniture in the next rooms...), we average a few differences between the signals with the sample (and the stretcher) in and out of the apparatus sensitivity region. The stretcher (Fig. 5), made by a light honeycomb structure of electrical insulating fiber, is manually translated on rails. With its center on the axis of the magnetic field, the signal produced by the IM-IP configuration is $+0.68 \pm 0.07$ µV and by the IM-OP configuration is $+1.34 \pm 0.18$ µV. The apparatus reproducibility is checked measuring the signal of the stretcher with a plastic box (rectangular base 16 cm x 21 cm and containing 6.0 l of deionized water) positioned on its center, the signal of the IM-IP configuration is $-11.2 \pm 0.1$ µV and the signal of the IM-OP configuration is $-16.8 \pm 0.2$ µV.

The horizontal x-axis (Fig. 5) is linked with the body and crosses the liver center of mass; the body center defines its

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Fig. 4. The weight function $g$ of the inner magnet and inner pickup (IM-IP) magnetic configuration. The weight function peak is 1500 V/m$^3$ for 38 A rms at 234 Hz.

Fig. 5. The susceptometer and the stretcher. The horizontal x-axis is linked with the body, crosses the liver center of mass and its origin falls on the body center.
origin. Percussion makes the delimitation of the hepatic area. To scan the liver region of the body (or the phantom), the body longitudinal position is such that the axis of the magnetic field travels along the x-axis, when the stretcher moves horizontally. The body center is located on the symmetrical longitudinal axis of the stretcher. Simply shifting the body along the stretcher allows measuring the magnetic signal of other body parts, for instance the head. All the measures presented in this work have been done with the person lying on his back.

The instrument sensitivity has also been checked measuring a phantom. It is made up of 35 plastic, 2.5 cm thick, slices. About 60 small holes, normally stuffed with a plastic cylinder, are evenly distributed on each slice. We removed the plastic cylinders from the holes placed in the phantom liver region and poured paramagnetic powder inside them. The powder magnetic susceptibility was formerly measured. The phantom longitudinal position on the stretcher allows scanning the liver region. The magnetization signals, by the IM-IP configuration, of the phantom with no added powder and with two quantities of paramagnetic powder having respectively the magnetic moment of 15g of Fe$^{3+}$ and 3g of Fe$^{3+}$ are in Fig. 6. Because of ~100 nV error the minimum quantity of detectable iron inside the entire liver region of the phantom is ~130 mg of Fe$^{3+}$ or ~270 mg of iron with an effective magnetic moment of 4 $\mu_B$.

![Fig. 6. Magnetization signals of the phantom with and without the paramagnetic powder distributed in its liver region. (a) Plain phantom. (b) Phantom with paramagnetic powder in its liver region equivalent to 3 g of Fe$^{3+}$. (c) Phantom with paramagnetic powder in its liver region equivalent to 15 g of Fe$^{3+}$.](image)

III. RESULTS

Fig. 7 shows the magnetization and eddy current signals of a patient with liver iron overload and of a healthy volunteer having similar anthropometric characteristics in function of the position of the magnetic field axis relative to the center of the torso (x axis in Fig. 5). Fig. 8 shows the signals of the same patient before and after 4 months of iron depletive therapy. The skewness of the patient's magnetization curve (Fig. 7c) is similar to the one of the phantom with the paramagnetic powder added to the liver region (negative x values). Knowing the magnetization signal of the plain phantom (Fig. 6a) and the one with the added paramagnetic powder (for instance Fig. 6b), we obtain the contribution of the powder simply by making the difference between these two signals. Similarly we obtain the contribution of the patient's iron overload making the difference between the actual magnetization signal of the patient and the estimate of the signal of his body, supposed depleted by the iron overload.

The magnetization and eddy current signals, a few anthropometric data (height, weight, torso cross section, BMI...), the body fat percentage measure by bioelectric impedance (the fat has the highest electrical impedance) have been measured for each of the 68 controls. Both the eddy current and magnetization signals of a person without iron overload have a similar dependence on the body size. We obtained a model to calculate the ratio between these two signals (they are both proportional to the magnetic field) from the statistical analysis of the correlations between the ratios of the two signals of 55 controls (training set), and...
their anthropometric data. The expected magnetization signal is subsequently obtained multiplying the calculated ratio with the measured eddy current signal. The results have been checked with the remaining 13 controls (test set). The standard deviation of the distributions of the differences between the measured magnetization signals and those calculated by this procedure for the test set, the training set and the total controls is lower than 330 nV, which is more than three times the intrinsic noise of the instrument. The improvement of this procedure is in progress; recently we also started applying the neural-network simulation. Note that the eddy current signal does not depend on the iron overload. In all the patient measurements we never noticed the iron overload skewness on the eddy current signal (for example Fig. 7d). Reference [19] reports that the concentration of iron in the tissues as free aqua ions is not significant. The calculation of the liver iron overload is presently done making the differences between the values, of the measured and expected magnetization signals, in the positions near the center of mass of the liver and using the calibration that has been done adding paramagnetic powder to the phantom liver region (Fig. 6). The next step will be evaluating the iron contribution to the magnetization signal making the volume integral on the real shape of the patient liver measured by an ultrasonic probe.

Fig. 9 shows the signals of a patient before and after the spleenectomy. The spleen iron overload is the reason for the skewness of the magnetization signal (Fig. 9a) for positive values on the x-axis. The magnetization signal of the same patient after the spleenectomy is symmetrical (Fig. 9c). Note that the eddy current signals are symmetrical and almost the
same before and after the operation. The expected magnetization signal of this patient, supposed depleted of the iron overload, has been calculated by the model using the eddy current signal (Fig. 9b) and the other patient's data measured before the splenectomy.

A hemochromatosis patient (Fig. 10) was measured before and after 12 phlebotomies carried out during a period of three months. The total removed iron is 2.7 g (~225 mg of iron depletion per phlebotomy). The iron reduction in his liver was estimated to be 2.5 ± 1.5 g, this is the difference between the iron overloads 10 ± 1 g (March 7) and 7.5 ± 1 g (May 27). These two overloads were obtained, making the differences between the two measured signals and the related expected signals of the patient, assumed without overload. The same procedure applies to the other three thalassemia-major patients, under iron intensive depleting therapy, that have been measured more than once. Fig. 11 reports these four measures of the iron reduction compared with the reductions expected from the applied therapies. The correlation of the iron overloads of all 40 patients measured with their blood serum-ferritin is in Fig. 12. The measurements of LIC via SQUID susceptometry, on a subset of 30 patients, were compared (Fig. 13) with the LIC calculated for each patient dividing the measured iron overload by its liver volume (calculated assuming the average value of 25 ml per each kg of body weight) and adding to this concentration the average LIC (~0.5 mg/cm³) of normal human.

IV. CONCLUSION

The presented susceptometer has the sensitivity to measure the iron overload in patients with pathology producing iron store in the body. All its components work at room temperature. The whole human torso fits within the sensitivity region, so the iron present in the entire liver contributes to the signal. The signal picked up has two sources: an overall magnetic background of the torso and an eventual contribution from liver iron excess. After measuring the magnetic signature of a patient, statistical analysis method is employed to estimate the background signal, given the patient anthropometric data. Liver-iron overload is then determined by subtraction of the estimated background from the total measured signal. The refinement of the methodology is in progress and, at present, the error in liver iron is about 1 g, corresponding to a concentration of 0.5 mg/cm³ for a 2 l liver volume.

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