IOP PUBLISHING PHYSICS IN MEDICINE AND BIOLOGY

Phys. Med. Biol. **53** (2008) 1701–1713 doi:10.1088/0031-9155/53/6/014

Time dependence of electrical bioimpedance on porcine liver and kidney under a 50 Hz ac current

J Spottorno[1], M Multigner[2,3], G Rivero[1], L Álvarez[4], J de la Venta[1] and M Santos[4]

[1] Instituto de Magnetismo Aplicado (ADIF-UCM-CSIC), PO Box 155, Las Rozas, Madrid 28230, Spain
[2] Departamento de Física de Materiales, UCM, Ciudad Universitaria, 28040 Madrid, Spain
[3] Centro Nacional de Investigaciones Metalúrgicas (CSIC), Avda, Gregorio del Amo 8, 28040 Madrid, Spain
[4] Centro de Investigación Biomédica en Red en Bioingeniería, Biomateriales y Nanomedicina (CIBER-BBN), Madrid, Spain
[5] Hospital Universitario Puerta de Hierro, San Martín de Porres 4, 28035 Madrid, Spain

Received 3 August 2007, in final form 27 January 2008
Published 29 February 2008
Online at stacks.iop.org/PMB/53/1701

Abstract

The purpose of this work is to study the changes of the bioimpedance from its '*in vivo*' value to the values measured in a few hours after the excision from the body. The evolution of electrical impedance with time after surgical extraction has been studied on two porcine organs: the liver and the kidney. Both *in vivo* and *ex vivo* measurements of electrical impedance, measuring its real and imaginary components, have been performed. The *in vivo* measurements have been carried out with the animal anaesthetized. The *ex vivo* measurements have been made more than 2 h after the extraction of the organ. The latter experiment has been carried out at two different stabilized temperatures: at normal body temperature and at the standard preservation temperature for transplant surgery. The measurements show a correlation between the biological evolution and the electrical bioimpedance of the organs, which increases from its *in vivo* value immediately after excision, multiplying its value by 2 in a few hours.

(Some figures in this article are in colour only in the electronic version)

1. Introduction

During the last 30 years there has been an increasing interest in the effects of interaction between the electromagnetic fields (EMF) and the human body and specially their eventual health hazard. Different studies refer to leukaemia in children under magnetic fields (Ahlborn *et al* 2000, Draper *et al* 2005).

Because of this social concern, different organizations, like the International Commission on Non-Ionizing Radiation Protection (ICNIRP 1998) or the American Conference of

Governmental Industrial Hygienists (ACGIH 2000), have created guidelines and standards for restricting human field exposure establishing the dose that the human can be exposed to (ICNIRP 1998).

The EMF dosimetry that studies these doses could be split into two levels (Repacholi and Greenebaum 1999):

- Macrodosimetry, related to exposure of the whole body or specific tissues and organs.
- Microdosimetry, related to exposure at the cellular level.

These restrictions impose limits on macrodosimetry levels for different frequencies, on the magnetic and electric fields as well as on the induced electric current density inside the body, which is the value the ICNIRP emphasizes. These currents cannot be measured precisely by non-invasive methods. This is the reason why in the past 20 years great efforts have been made to procure a numerical calculation method with the aid of magnetoresonance images and using the impedance method, the finite-difference time-domain (FDTD) calculations, the electromagnetic finite element (EMFE), etc (e.g. Dimbylow 2005, Gandhi and Kang 2001, Dawson and Stuchly 1998).

To obtain a realistic result it is necessary to have a precise knowledge of the electric properties of the different organs and tissues that are involved in the calculation. So the study of the electric properties of tissues has become an important task, in order to correctly calculate the induced electrical currents that appear in the human body because of field exposure. There are a lot of works on this issue from the 1950s that give values to the conductivity and permittivity of the tissues (Schwann 1957, 1963). The available experimental data cover a great range of frequencies, but the properties at low frequencies come mainly from non-living or at best freshly excised tissues (Foster and Schwan 1989, Gabriel *et al* 1996a, 1996b). Probably the most complete and divulgated database of electric parameters of biological tissues is that of Gabriel *et al* (1996a and 1996b). In this case animal tissues were used within 2 h after death and the human material was obtained 24–48 h after death except for the cases of human skin and tongue, which were made *in vivo*. In that work it can be appreciated that in the bibliography there is a remarkable dispersion in the values obtained by different authors on the conductivity of tissues and organs. One of the possible reasons for this dispersion could be the reasonably expected biological changes of the organs from the moment of their extraction, giving rise to different values of electrical conductivity depending on the time after the excision when the measurements were performed. As McLeod (1992) notes: 'tissues conductivity *in vivo* may appear significantly different that in non-living tissue'.

In the frame of a recent research project funded by Red Eléctrica de España which is the company responsible for the transmission network and for operation of the Spanish Electricity System, the authors of this paper have measured the electric properties of different organs and tissues at low frequencies around 50 Hz (the industrial frequency in Europe) in order to accurately calculate the induced currents for different situations that electrical company workers may be in. For these characterizations, *in vivo* measurements of the conductivity of porcine organs have been carried out and it has been found that the values were systematically higher than the ones in the literature. The most remarkable difference between our experiments and the ones in the literature was that in our work these experiments were performed *in vivo*. This fact led us to think that the organs could vary their properties soon after excision from the living body, which was confirmed in later experiments. Also there are works that show changes in the *in vitro* conductivity of the liver at frequencies over 10 kHz (Raicu *et al* 2000). This would mean that the conductivities measured in the hours after the excision would be lower than the *in vivo* one, and thus the numerical simulation of electric currents created by

EMF of industrial frequency calculated by numerical programs would give a value smaller than the real ones.

In order to check this assumption, measurements both *in vivo* and in a few hours after its extraction from the body have been performed in two porcine organs: the liver and the kidney.

2. Experimental procedures

As we comment above, the aim of this work was to measure the evolution of the electrical properties of the organs with the time past after the excision. The impedance of the two organs (porcine liver and kidney) has been measured. These organs were chosen for two reasons.

- They are accessible in an easy way for the *in vivo* measurements.
- From the available transplant organs, they present two different delay times for surgery: 12 h for the liver and more than 24 h for kidney. In other words, they present very different resistance to biological (and presumably electrical) degradation.

So, measurements of the impedance were registered on each organ in two conditions:

1. *in vivo*, with the organ in the alive and anaesthetized pig;
2. *ex vivo*, with the organ just excised and in the hours following its extirpation.

In both cases the impedance of the organs was obtained by means of two electrode measurements of the current–voltage relationship. Electrodes, impregnated with conductive gel such as the one used in electrocardiograms, in order to assure a proper electric contact, were placed at opposite sides of the organ studied and attached to organ surfaces using tubular elastic retention bandages.

The impedance measured this way depends on the shape and material of the electrodes, the distance between them and the average conductivity of the organ under study. We fixed the electrodes to the organ with a tubular elastic retention bandage so that when the organ is excised, the electrodes could be maintained in the same position relative to the organ. Therefore, the changes in the impedance of the organ should only depend on its electrical properties and not on the geometrical features of the electrodes.

An ac voltage difference of the frequencies studied was applied between the electrodes, so that an electric current passes through the organ under study. The voltage signal was created and measured by means of a Signal Recovery 7265 DSP 'lock-in' amplifier, at frequencies ranging from 20 to 120 Hz in order to avoid the influence of environmental electromagnetic noise. A second 'lock-in' amplifier was used to simultaneously measure the electrical current at every frequency and the phase shift from the voltage signal (figure 1). For each frequency, the pairs of I–V values were registered and the impedance was obtained from the minimum square adjust. The voltage introduced ranged from 50 μV to 1 mV depending on the electrodes and the organ measured, while the currents were of the order of μA.

Three different electrodes were used in order to disregard its influence in the results and all of them are made of noble metals to avoid its degradation over the process of measurement:

- Gold electrodes: circular plates with 4.5 cm in diameter.
- Platinum electrodes: square plates 2.5×2.5 cm^2.
- Silver electrodes: rectangular plates 0.6×0.5 cm^2.

Furthermore, the use of different electrodes combined with different frequencies helps to evaluate the influence of the capacitive effects in the measurements.

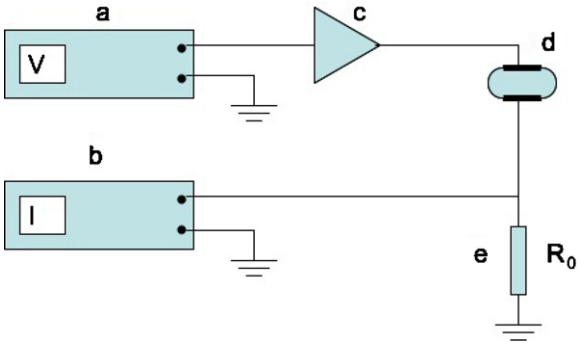

Figure 1. Experimental setup. (a) Lock-in to applied and measured ac voltage. (b) Lock-in to measured current. (c) Voltage follower. (d) Organ with electrodes. (e) Standard resistance.

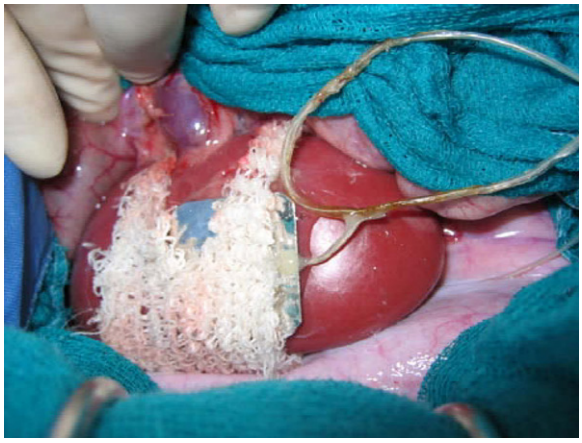

Figure 2. Fixation of the Pt electrodes on the kidney for the *in vivo* measurements.

2.1. In vivo measurements

This study was performed in Landrace × Large White pigs weighing 19–22 kg. For the *in vivo* measurements animals were sedated with ketamine (20 mg kg^{-1} bw), diazepam (0.1 mg kg^{-1} bw) and atropine (0.02 mg kg^{-1} bw). Anaesthesia was induced with an i.v. bolus of propofol (2 mg kg^{-1} bw), midazolam (0.6 mg kg^{-1} bw) and fentanyl (5 μg kg^{-1} bw). After endotracheal intubation, anaesthesia was maintained with continuous i.v. infusion of propofol (9 mg kg^{-1} h^{-1}), midazolam (0.6 mg kg^{-1} h^{-1}), fentanyl (5 μg kg^{-1} h^{-1}) and pancuronium bromide (0.4 mg kg^{-1} h^{-1}). An Adult Star® ventilator (Infrasonics, Inc.) was used for mechanical ventilation. Electrocardiogram, heart rate and arterial pressure were monitored during the procedure.

Through a median laparotomy, the liver and both kidneys were exposed and electrodes were placed (figure 2).

Gold electrodes are wide enough to cover the kidney but (because of its large size) only one lobe of the liver was measured, with the electrodes at its centre. So, when the organ was excised we could easily cut the lobe without affecting the position of the electrodes.

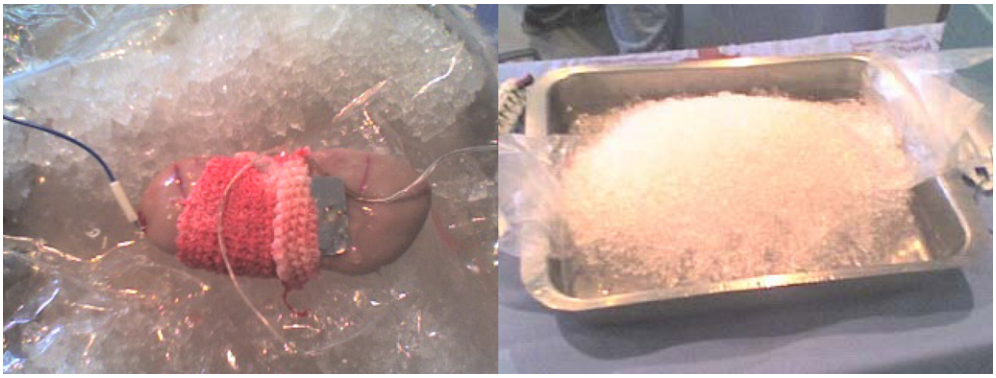

Figure 3. Ice bath for measurements in the kidney at 1 °C temperature.

2.2. *Ex vivo measurements*

Once the *in vivo* measurements were concluded, renal vessels were ligated jointly with 2-0 silk and divided to remove both kidneys. As we commented above, only a part of the left hepatic lobe was excised. In both cases the electrodes were maintained in their relative positions on each organ during this procedure, to exactly reproduce *ex vivo* the conditions established *in vivo*.

As is well known, the conductivity is also a function of the temperature. Two different temperatures have been chosen for the measurements.

- The normal body temperature (36 °C). This is the temperature that would normally be used when the *in vitro* measurements are performed (Gabriel *et al* 1996a, 1996b), because the conductivity values at this temperature are employed to calculate or simulate the effects on the human body by the EMF exposure.
  Each organ, when excised, was immersed into a thermostatic bath at 36 °C, enclosed into a sterilized plastic bag in order to avoid direct contact with the surrounding medium, but allowing the wiring of electrodes to get out.
  The *I–V* measurements were made continuously from the instant of excision until a few hours later. Tissular temperatures were monitorized with a thermistor probe (temperature monitor, Shiley Inc. Irvine, CA, USA) along the procedure.
- At the preservation organ's temperature (about 1 °C). This is the temperature at which the organs are usually kept in the standard conditions in order to minimize the tissue damage, not only for transplant surgery but also for further *in vitro* measurements. These experiments could be useful in order to see if the '*in vitro*' measurements are reliable under preservation conditions after a few hours have passed.

The designed procedure for kidneys was as follows: renal vessels were clipped and divided; kidneys were removed and immediately perfused, via arterial, one of them with cold (4 °C) autologous blood, and the other one with cold University of Wisconsin solution (Viaspan®). Finally, each organ was immersed in an ice bath (figure 3), enclosed in a sterilized plastic bag, in a similar way to the one employed for the 36 °C measurements.

In order to know the role that could play in the results obtained the interval of time needed to achieve low temperatures, *I–V* measurements were also made during the cooling time.

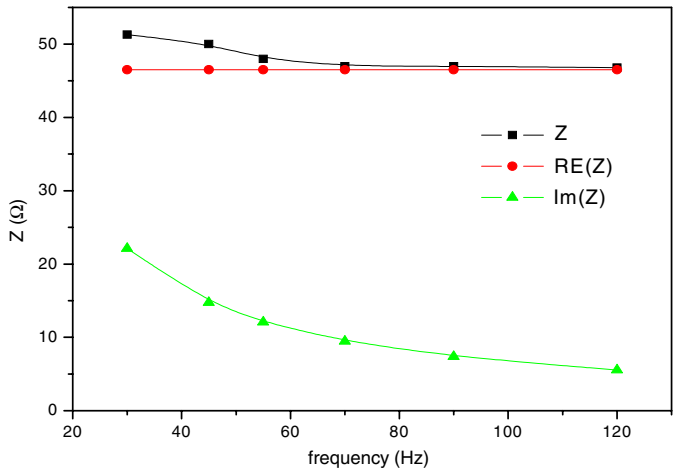

Figure 4. Impedance of the kidney and its real and imaginary components.

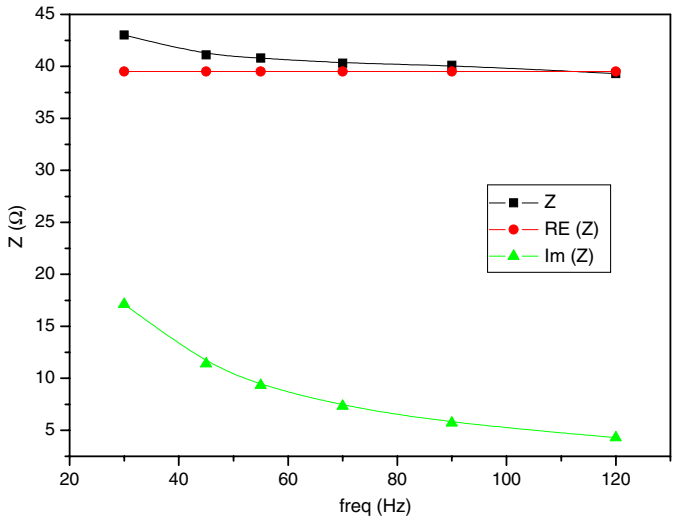

Figure 5. Impedance of the liver and its real and imaginary components.

3. Experimental results and discussion

3.1. In vivo results

The results obtained for the impedance of the organs from the *in vivo* measurements show the expected dependence on frequency of ac signal, due to the capacity effect of electrodes (Bard and Faulkner 2001). Figures 4 and 5 show the modulus, real and imaginary part of impedances of liver and kidney with gold electrodes from the experimental data. The real component is the one that has a zero phase shift with the voltage. This is the resistant part, which is inversely proportional to the conductivity. The imaginary component has a phase shift of 90° with the voltage and is due to the polarization effect on the electrodes. So the real component depends

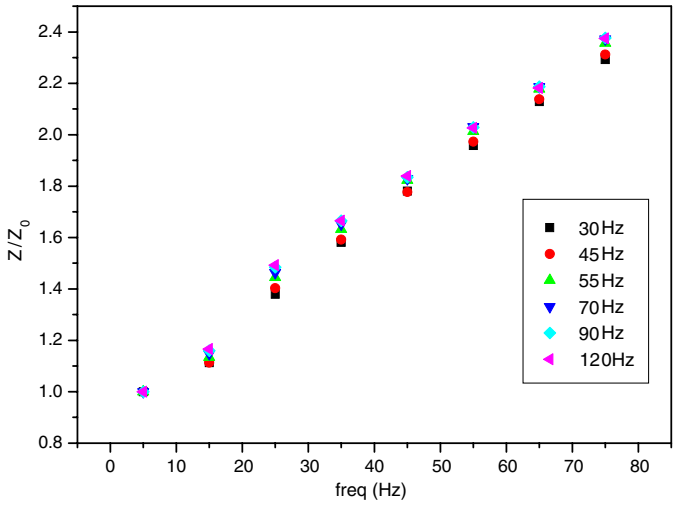

Figure 6. Impedance of the kidney normalized to the *in vivo* value at body temperature (Z_0).

on the conductivity while the imaginary one depends on the polarization of the electrodes. It can be observed that the influence of the capacity of the electrodes in the total value of the modulus of the impedance increases by about 5% over the value of the real part.

Similar results have been obtained with Ag and Pt electrodes, with the expected variation in the absolute value of the impedance (as well as in the real and imaginary parts) due to the different electrodes areas.

3.2. Ex vivo results

For each type of electrodes, the results obtained *ex vivo* show a dependence on the frequency and the time t passed from excision: $Z = Z(\omega, t)$.

As we commented above, the evolution of impedance in the two organs has been registered at two temperatures given below.

3.2.1. Normal body temperature (36 °C). The impedance evolution for the liver at 36 °C is shown in figure 6. In this figure it can be observed that for each frequency the impedance increases almost linearly with time after the excision, and the onset of the increase coincides with the instant of the excision from the organ. Figure 7 shows the evolution of the corresponding real and imaginary components at 50 Hz. (The real component of the impedance behaves similarly in all the frequencies studied.) All the impedances are normalized to the value of the *in vivo* impedance Z_0.

The results for the kidney, under the same experimental conditions, are shown in figures 8 and 9.

It is worth noting that the real part of bioimpedance of the liver doubles its value in around 1 h, and it depends only on the electrical properties of the organs, because the real part of bioimpedance does not depend on the polarization of the electrodes, and these have been fixed in such a way that there do not exist any changes in the position of the electrodes, or in the shape of the organ, so that the cell factor should be constant. Also, we believe that the effect of the anaesthesia is negligible both because of its very low concentration and because its

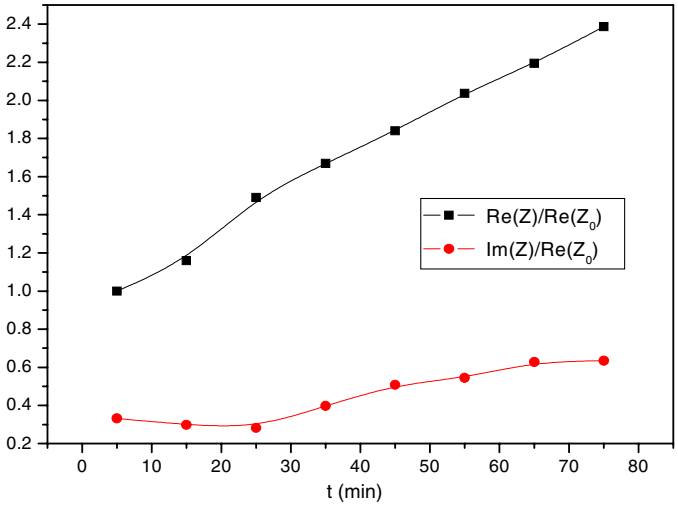

Figure 7. Real and imaginary components of the impedance of the liver at 50 Hz at body temperature, normalized to the *in vivo* value of the real component.

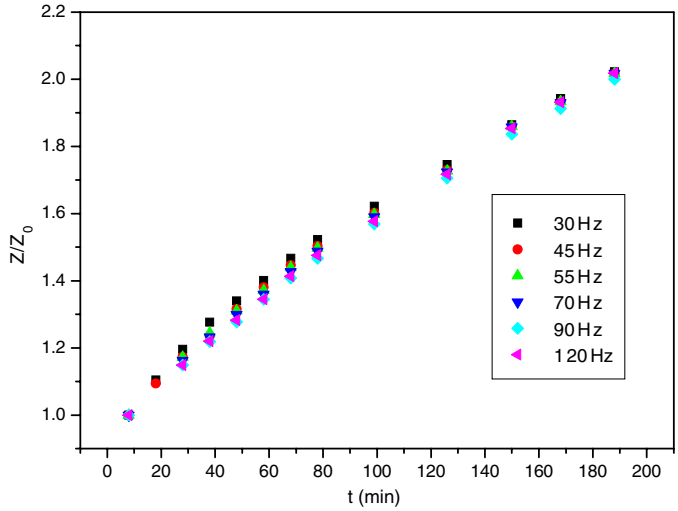

Figure 8. Impedance of the kidney normalized to the *in vivo* value at body temperature.

concentration is constant after the excision, so that it should not give rise to any change in the bioimpedance.

Also the average conductivity varies with temperature, but it should not be forgotten that the liver was kept at a constant temperature so that any changes in the bioimpedance should only be attributed to the time after the excision of the liver.

In the case of the kidney we can see a linear increase of the bioimpedance from the beginning of the measurements, doubling its resistance in about 3 h. This changing is slower than the liver one, which is coherent with the observed fact that the kidney degenerates slower than the liver after its excision from the body.

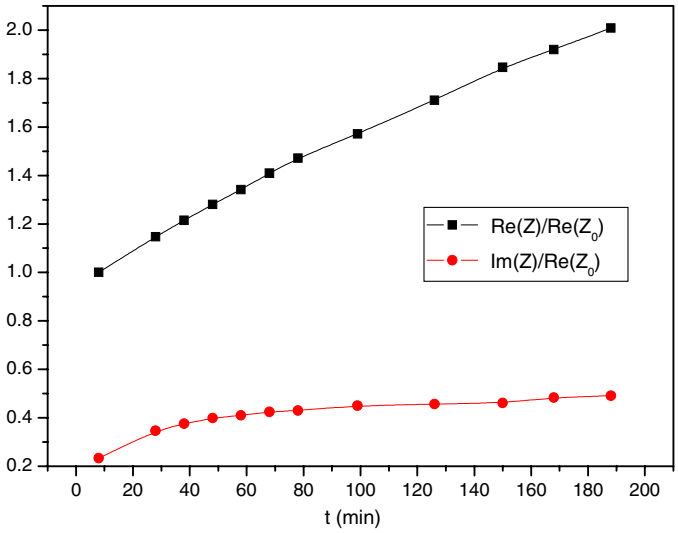

Figure 9. Real and imaginary components of the impedance of the kidney at 50 Hz at body temperature, normalized to the *in vivo* value of the real component.

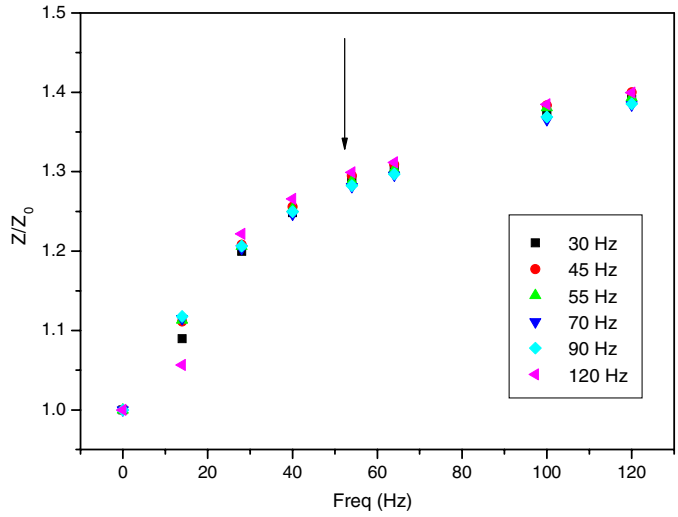

Figure 10. Impedance of the liver under preservation conditions normalized to its *in vivo* value. The arrow shows the point when the temperature is stabilized at 1 °C.

In both cases, it should be noted that the contribution of the imaginary component, mainly due to the capacity of the electrodes, is negligible.

Even though the absolute values of the impedance are different depending on the area of the used electrodes, the evolution of the normalized value to the *in vivo* one is almost identical.

3.2.2. Standard preservation temperature (1 °C). The evolution of the impedance as well as the real and imaginary components for liver and kidney is shown in figures 10–13.

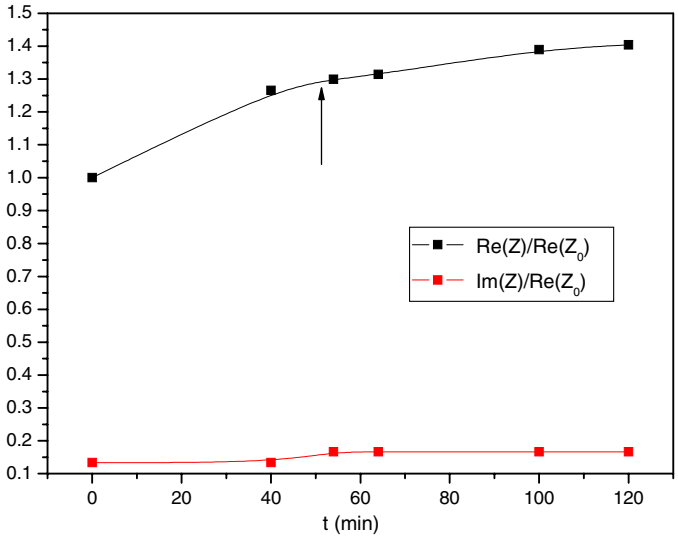

Figure 11. Real and imaginary components of the impedance of the liver at 50 Hz under preservation conditions normalized to the *in vivo* value of the real component. The arrow shows the point when the temperature is stabilized at around 1 °C.

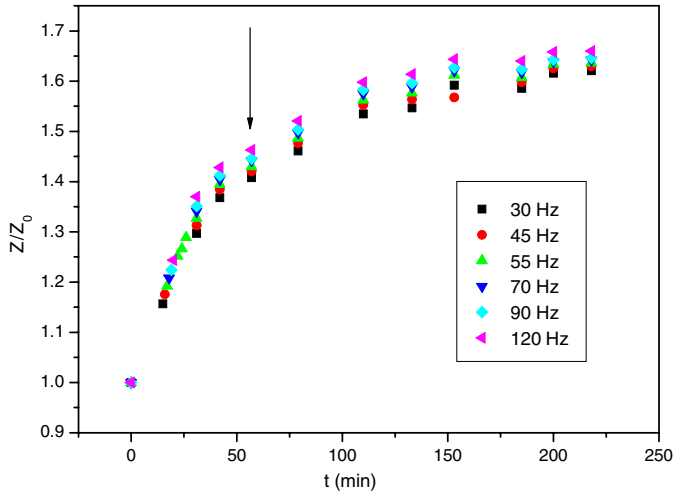

Figure 12. Impedance of the kidney under preservation conditions normalized to its *in vivo* value. The arrow shows the point when the temperature is stabilized at 1 °C.

It can be seen that the impedances and corresponding real components also increase with time, but at a slower rate than at 36 °C, as could be expected. In the measurement procedure, as mentioned in the experimental procedures, the organs were immersed in an ice bath immediately after the excision, reaching the stable measurement temperature (1 °C) after about 50 min for both organs. As can be shown in figures 10–13, during the cooling of the organs the evolution of the impedance and the resistance is very sharp. However,

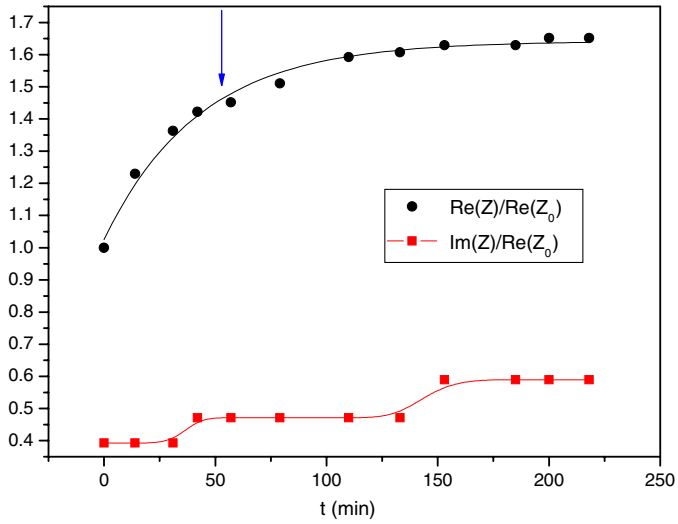

Figure 13. Real and imaginary components of the impedance of the kidney at 50 Hz under preservation conditions normalized to the *in vivo* value of the real component. The arrow shows the point when the temperature is stabilized at around 1 °C.

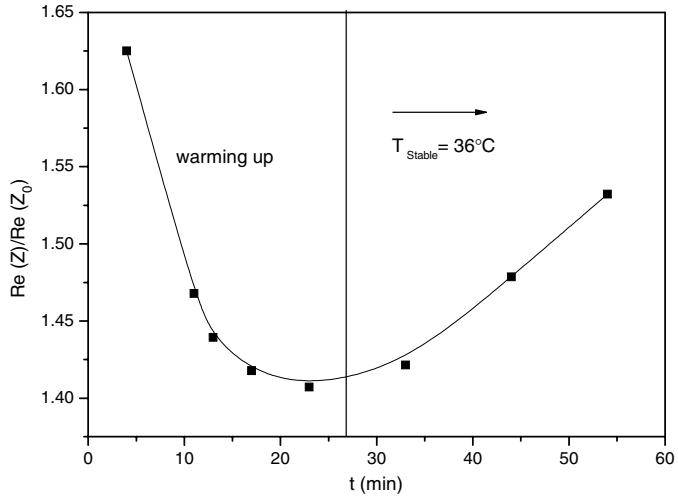

Figure 14. Variation of the real component of the impedance of the kidney when it is heated up to 36 °C after being kept under preservation conditions for 3 h.

this change could not only be attributed to the temporal evolution of those magnitudes but also to the well-known dependence of the ionic conductivity with temperature. When the stable temperature is reached the evolution is slower but relevant, 15% in 60 min and 20% in 160 min, for the liver and the kidney, respectively.

In order to check the changes suffered by the organ after the cool preservation during 3 h, the organ was warmed again to 36 °C, registering continuously the impedance values. As can be observed in figure 14, the warming process gives rise to a decrease of the impedance value

due to the increase of the ionic conductivity with temperature. However, the reached value at 36 °C is 40% higher than the *in vivo* one.

3.3. Discussion

We have seen that after the excision from the animal the bioimpedance measured on the kidney and the liver increases just after the excision in an appreciable way. The organs' impedance and conductivity behaviour, observed after surgical excision, could reflect deleterious changes originated by the interruption of the blood flow: in ischaemia, anoxic injury starts with a decrease in mitochondrial energy production; in parallel, cellular ion homeostasis becomes impaired resulting in increased cytosolic calcium and sodium concentrations, which may activate lytic enzymes, cause osmotic swelling, disruption of plasma membrane and, ultimately, cell death (De Groot and Rauen 2007).

This hypothesis would be consistent with the differences observed between liver and kidney, and between the different temperatures at which the measures were performed: susceptibility to ischaemia times varies for different organs and tissues, and cold storage at 0°–4 °C greatly decreases the rate at which anoxic cell injury proceeds, but there are no decisive differences in the mechanisms of production of this damage.

So we think that the change in the impedance must be attributed to changes in the electric properties of the organ due to its degradation after its excision from the body. Other reasons for this change have been disregarded by the measurement method and by the usage of different electrodes. This degradation of the organ brings an increase in its bioimpedance. As could be expected, the increment is slower when the temperature is near 0 °C and is faster for the liver than for the kidney.

So, in order to measure the average conductivity of a tissue, it is very important to know exactly both the time elapsed from the time the organ was removed and temperatures to which it has been maintained, since these are determining factors for the production of cellular necrosis and tissular disruption. The values of the average conductivity for different organs affect in a dramatic way the calculated values of the induced currents in the human body by EMF exposure, so it is necessary that they be measured in the most precise way.

4. Conclusion

The measurements point to a correlation between the biological evolution and the electrical bioimpedance of the organs, which can reduce its *in vivo* value by 2 in a few hours. This effect is low but not negligible under the standard preservation conditions for transplant surgery.

On the other hand, these changes observed in the impedance values should be attributed to the corresponding changes in the bioimpedance of the organ because, under the conditions of experiment, the change of the imaginary component of the impedance (which is the most affected by the effects of the polarization on the electrodes) is almost negligible.

Therefore, it seems necessary to determine the bioimpedance and the average conductivity values of the organs and tissues *in vivo* or immediately after extraction, in order to calculate in a reliable way the induced currents in the human body.

Acknowledgments

The authors would like to gratefully acknowledge the helpful advices and conversations about this work with Dr Juan Bernar, Dr José María García Páez and ProfessorAntonio Hernando. Financial support from Red Eléctrica de España is acknowledged.

References

ACGIH 2000 TLVs and BEIs: threshold limit values for chemical substances and physical agents and biological exposure indices *American Conf. Governmental Industrial Hygienists (Cincinatti O H)*

Ahlborn A *et al* 2000 A pooled analysis of magnetic fields and childhood leukaemia *Br. J. Cancer* **83** 698–8

Bard A J and Faulkner L R 2001 *Electrochemical Methods: Fundamental and Applications* New York Wiley) pp 1–42

Dawson T W and Stuchly M A 1998 High-resolution organ dosimetry for human exposure to low frequency magnetic fields *IEEE Trans. Magn.* **34** 708–17

De Groot H and Rauen U 2007 Ischemia-reperfusion injury: processes in pathogenetic networks: a review *Tansplant. Proc*. **39** 481–4

Dimbylow P 2005 Development of the female voxel phantom, NAOMI, and its application to calculations of induced current densities and electric fields from applied low frequency magnetic and electric fields *Phys. Med. Biol.* **50** 1047–70

Draper G, Vincent T, Kroll M E and Swanson J 2005 Childhood cancer in relation to distance from how voltage power lines in England and Wales: a case-control study *BJM* **330** 1–5

Foster K R and Schwan H P 1989 Dielectric properties of tissues and biological materials: a critical review *Crit. Rev. Biomed. Eng.* **17** 25–104

Gabriel C, Gabriel S and Corthout E 1996a The dielectric properties of biological tissues: I. Literature survey *Phys. Med. Biol.* **41** 2231–49

Gabriel S, Lau R W and Gabriel C 1996b The dielectric properties of biological tissues: II. Measurements in the frequency range 10 Hz to 20 GHz *Phys. Med. Biol.* **41** 2251–69

Gandhi O P and Kang G 2001 Calculation of induced current densities for humans by magnetic fields from electronic article surveillance devices *Phys. Med. Bio.* **47** 2759–71

ICPNIRP 1998 Guidelines for limiting exposure to time-varying electric, magnetic and electromagnetic fields (up to 300 GHz) *Health Phys.* **74** 494–522

McLeod K J 1992 Microelectrode measurements of low frequency electric field effects in cells and tissues *Bioelectromagn. Suppl.* **1** 161–78

Raicu V, Saibara T and Irimajiri A 2000 Multifrequency method for dielectric monitoring of cold-preserved organs *Phys. Med. Biol.* **45** 1397–407

Repacholi M H and Greenebaum B 1999 Interaction of static and extremely low frequency electric and magnetic fields with living systems: health effects and research needs *Bioelectromagnetics* **20** 133–60

Schwann H P 1957 Electrical properties of tissues and cell suspensions *Adv. Phys. Med. Biol.* **5** 147–209

Schwann H P 1963 *Determination of biological impedances Physical Techniques in Biological Research* vol VI B, ed W L Nastuk (New York: Academic) p 323