Impedance Properties of Metal Electrodes for Chronic Recording from Mammalian Nerves

RICHARD B. STEIN, DEAN CHARLES, TESSA GORDON, JOAQUIN-ANDRES HOFFER, AND JACK JHAMANDAS

Abstract—Over the last few years methods have been developed for recording chronically from mammalian nerves with implanted electrodes contained in silastic cuffs. The impedance of the electrodes and the changes that take place over time were studied. Platinum-iridium electrodes remain stable over long periods of time, whereas the impedance of silver wire electrodes increases and eventually the silver fractures. The impedance at low frequencies (e.g., 10 Hz) provides a measure of the capacitance of the metal electrodes, whereas the impedance at higher frequencies (1 kHz) mainly measures the resistance of the tissue filling the cuff. Increases in resistance due to growth of connective tissue around intact nerves have been studied as well as changes in resistance after cutting a nerve. Impedance measurements provide a useful way to follow the properties of electrodes and nerves in basic neuroscientific research and in future clinical applications of these chronic recording methods.

Introduction

METHODS have recently been developed [4, 7, 16] for recording from mammalian nerves for months or even years. These methods have considerable potential for use in basic neuroscientific research and also in clinical applications; for example, in the neural control of artificial limbs. For clinical use electrodes must be stable and bio-compatible for long periods of time. The stability of chronic stimulating electrodes has been thoroughly studied (see [6] for a recent review), but the properties of chronic recording electrodes have rarely been reported [14]. We have used routine impedance measurements as a way of following the electrical properties of chronic recording electrodes. The measured impedance depends on the properties of the tissue, as well as the electrodes, and the measurement of impedance proved a sensitive method of following changes in the nerves and connective tissue, as well as changes in the electrodes. The purpose of this report is to describe the impedance characteristics of chronic recording electrodes in terms of quantitative models, and to show how changes in impedance can be attributed to changes in the electrodes themselves or to changes in the nerves and connective tissue.

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Methods

The method used for recording chronically from peripheral nerves in cats and mice involves placing a silastic cuff containing three or more electrodes around an intact nerve (Fig. 1). The implantation is carried out under aseptic conditions. In cats the electrode leads form a flexible insulated cable, ending in a miniature socket within a very hard, smooth vitreous carbon button [15] which acts as skin interface. Further technical details can be found in previous publications [16, 17]. In mice the procedure is similar except that a smaller Teflon and Dacron percutaneous connector substitutes for the vitreous carbon button [8].

Impedances at 1000 Hz were measured for each lead with respect to an indifferent electrode outside the cuff, using a Hewlett-Packard Vector Impedance Meter (Model 4800A). In addition, the impedance for the configuration shown in Fig. 1 was measured for a range of frequencies from 10 to 10,000 Hz. This particular configuration was chosen because it had proved to be the most effective way of recording neural activity with least contamination from the much larger EMG signals generated in adjacent muscles [17]. This configuration records the second spatial difference between the voltages at the three electrodes, and is called a *tripolar* configuration [17].

In using the impedance meter it was not possible to control the current density of the signal, as this varied with impedance. However, a number of measurements were also checked with a simple voltage divider and signal generator. These measurements verified that the current densities used were small enough to remain within the linear range of the metal electrodes [11].

Results

Fig. 2A shows the magnitude and phase of tripolar configurations of 75 μm diameter silver and platinum-iridium electrodes in cuffs around the sciatic nerve of cats. The cuff diameter was 3.4 mm and the distance between the end electrodes was 3 cm. Fig. 2B shows the corresponding values for 25 μm Pt-Ir electrodes in smaller cuffs around the peroneal and tibial nerves of mice. The diameters were 0.3 and 0.4 mm and the distance between the end electrodes was 4.5 mm. The simplest circuit that can approximate the impedance of the electrode configuration is the *RC* circuit shown in the insert. The impedance of this circuit, given by the solid lines, deviates consistently from the measured impedance at both low and high frequencies; this deviation will be considered in more detail in the discussion.

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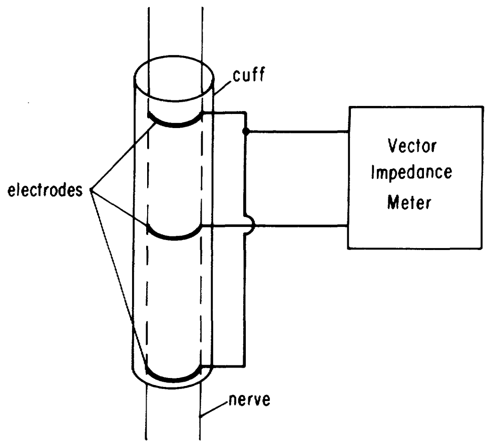

Fig. 1. Arrangement for measuring the impedance of electrodes in a silastic cuff implanted chronically around an intact nerve.

The capacitance C derives from the double layer that is formed by the electrode-fluid or electrode-tissue interface which acts as a capacitive barrier to current flow. This capacitor C is particularly important at low frequencies. The resistance R in the circuit is that of the tissue and fluid filling the cuff, and it is across this resistance that neural currents develop a voltage, which is represented as the input to the recording electrodes. The resistance will be particularly important at high frequencies; the effective resistivity of the tissue can be calculated, assuming that it is the *sole* contributing factor to the high frequency impedance. If the resistivity were known, the expected impedance could be calculated using a formula given by Mannard *et al.* [9] for impedance measurements between a central electrode and the two end electrodes in a cuff. Alternatively, knowing the resistance R, we can invert this formula to obtain the apparent resistivity ρ

$$\rho = \pi d^2 R/l \qquad (1)$$

where d is the diameter of the cuff and l is its length. If these dimensions are given in cm and the resistance in Ω, the resistivity will be given in Ω-cm.

Fig. 3 shows the impedance at a low frequency (10 Hz) and a high frequency (1000 Hz) of cuffs around various cat nerves as a function of time after implantation. Immediately after implantation the values were considerably higher than the control values measured in saline (S) before implantation. The apparent resistivity calculated from equation (1) indicates that the resistivity increased from about 80 Ω-cm in saline to slightly more than double this value in the body, probably because of the higher impedance of the tissue. The apparent resistivity increased further for several weeks after implantation, to values near 250 Ω-cm for both silver or platinum-iridium electrodes. This increase is attributed to the further replacement of fluid within the cuff by connective tissue.

Changes in the low-frequency impedance of the cuffs over time are shown in the upper part of Fig. 3. Low-frequency impedances differed between the two types of metals used and thus depended on electrode properties. The impedance of the silver electrodes tended to increase with time. Silver electrodes

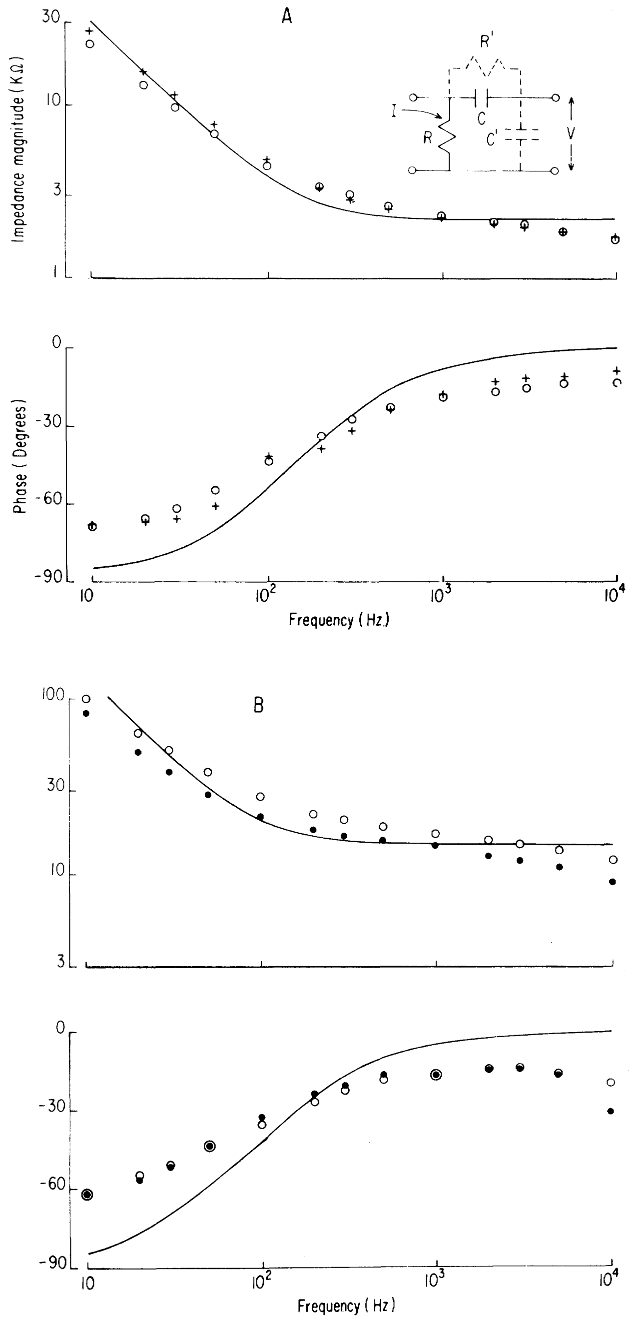

Fig. 2. A. Magnitude and phase of the impedance recorded from Ag (+) and Pt-Ir electrodes (o) several weeks after being implanted in two cats. The solid lines are predictions from the simple RC circuit shown in the inset with R = 2.2 kΩ and C = 0.51 μF. Note that the experimental points deviate somewhat from the predictions. The interpretation of these data, the RC circuit and the extra elements R' and C' are contained in the Discussion. B. Magnitude and phase of the impedance recorded from smaller Pt-Ir electrodes twenty weeks after being implanted in two mice. The corresponding solid lines are predictions with R = 15 kΩ and C = 0.15 μF.

became coated with silver chloride in physiological saline, and the increased impedance could be due to the formation of a thicker silver-silver chloride layer with time. A thicker layer would increase the impedance by decreasing the capacitance (and increasing the resistance). The low-frequency impedance

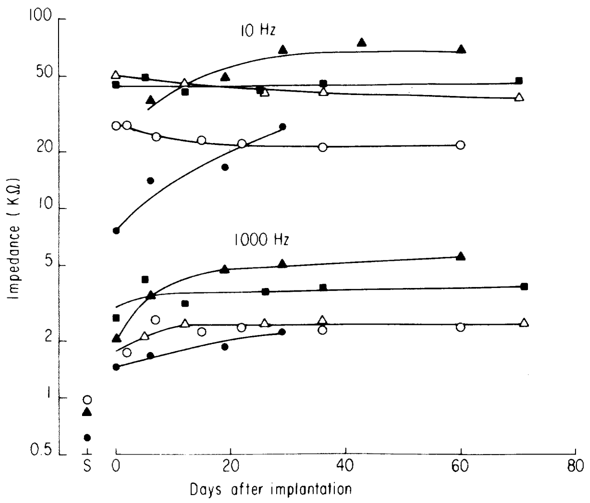

Fig. 3. Magnitude of the impedance for electrodes at a low frequency (10 Hz) and a higher frequency (1000 Hz) as a function of time after implanting the devices in cats. The impedance of some electrodes was measured prior to implantation at 1000 Hz in physiological saline (*S*). The impedances were measured between an electrode in the center of the cuff and the electrodes at the two ends of the cuff. The metals and nerves used were: ■ Ag, posterior tibial nerve; ● Ag, sciatic nerve; ▲ Ag, lateral gastrocnemius-soleus nerve; ○ and △Pt-Ir, sciatic nerves. The impedance of all leads tended to increase at high frequencies due to growth of connective tissue into the cuff. The behavior at low frequencies was different for the two metals. Further discussion in the text.

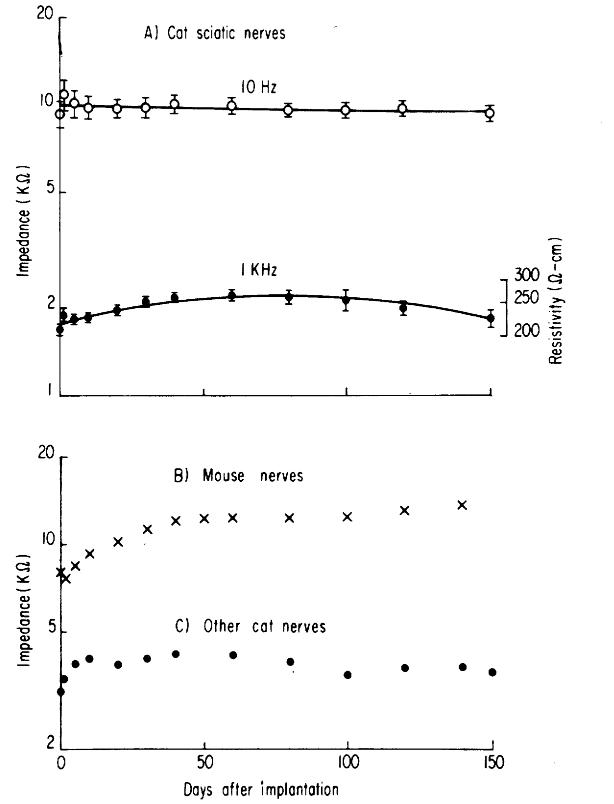

Fig. 4. A. Changes in impedance of eight platinum-iridium electrode units implanted on sciatic nerves of cats. The two sets of symbols give the means and standard errors of the mean at 10 Hz and at 1 kHz measured at varying times after implantation. Changes in impedance at 1 kHz of B) three electrodes units implanted on nerves in mice and C) ten other electrode units implanted on various branches of sciatic nerves in cats. The extent of the symbols in A) represents the standard errors of the mean, and the curves have been fitted so as to minimize the mean square deviations. Because of the smaller numbers of mice in B) and the variety of cat nerves in C), no statistical analysis or curve fitting was performed.

of the Pt-Ir electrodes tended to decline slightly over a period of months.

After about two months the silver wires became brittle and disintegrated. In contrast, the platinum-iridium wires proved to be much more stable. They have never broken except when insufficient slack was allowed in the cable so that considerable stress was exerted on the leads during normal animal movements. Thus, silver electrodes were abandoned for chronic use, and subsequent devices were made with platinum-iridium electrodes, despite their higher cost.

The electrical properties of Pt-Ir electrodes were followed over longer periods of time to assess their stability for chronic use. Measurements of impedance were made on eight additional cuffs, each implanted around cat sciatic nerves for over five months. Where measurements were not available for a given cat on a given day, values were interpolated between the two nearest days on which measurements were available. Fig. 4A shows means and standard errors for these measurements of impedance at 10 Hz and 1 kHz. Since all sciatic cuffs were a standard length and diameter, the ordinate on the right shows the apparent resistivity of the tissue calculated from equation (1). This more complete series of measurements showed that the impedance at 1 kHz increased approximately 30% in the first two months after implantation, but also began to decline again after two months. The data at 1 kHz could be fitted with the parabola shown in Fig. 4A, which accounted for two-thirds of the variance of the points about the mean value. The late decline was less clearly seen in ten cuffs on smaller cat nerves and was not observed in three mouse nerves which have been followed over this period of time. Possible reasons for these discrepancies will be discussed later.

Throughout the months of recording, the impedance of all electrodes at low frequency (10 Hz) was remarkably steady, indicating excellent stability of the Pt-Ir wires in the biological environment. The impedance of the electrodes in these cats were all considerably lower than in the previous series shown in Fig. 3 because stranded wire was now used (Medwire Corp.). This increased the surface area of the electrodes markedly and improved their strength.

Having established that Pt-Ir electrodes remained stable for months on normal nerves, we were interested in their properties on severed nerves, such as would be found in applying these techniques to the control of artificial limbs in amputees. In these experiments we used 5 cm long cuffs on cut posterior tibial nerves of cats (Fig. 5) and sealed the distal ends of the cuffs to prevent axonal regeneration. Electrodes were spaced every cm along the cuff and the impedance of each lead (e.g., lead *A* in Fig. 5) could be measured with respect to a large, indifferent electrode (*G*) on the outside of the cuff. The *difference* in impedance between two adjacent leads at 1 Hz gave a measure of the resistance of the tissue within the cuff, and was measured in kΩ/cm. The pattern of impedance changes as a function of time after cutting the nerve is shown in Fig. 5 for the first (1), second (2), and other (3, 4, and 5) segments of the nerve at increasing distances from the cut end, which was near electrode *A*.

The most marked increase occurred in the first cm within a

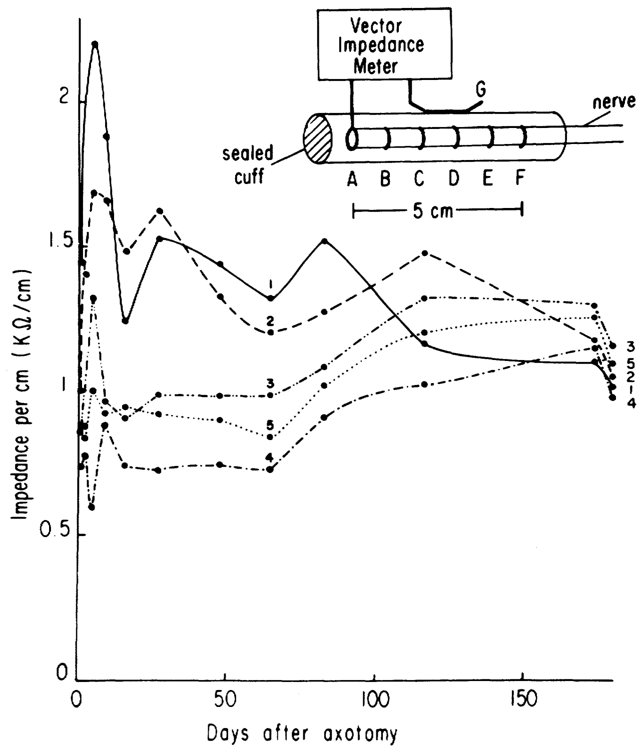

Fig. 5. Impedance changes over time at varying distances from the sealed end of a tibial nerve cuff. The impedances of each electrode (*A–F*) were measured with respect to a large, indifferent electrode (*G*) outside the cuff, shown in the inset. The difference in impedance between adjacent electrodes measures the impedance of nerve segments 1–5 cm from the sealed end. Initial changes are most pronounced closest to the sealed end (Curve 1). However, the later rise in impedances, due to connective tissue ingrowth, is more evident towards the end of the cuff (Curves 3–5). Further details of the degenerative and regenerative processes underlying the changes are discussed in the text.

couple of days after cutting the nerve, and was presumably due to the well-known swelling of a severed nerve as a result of the blockage of axoplasmic transport near the ligature. This increase in impedance was less marked in the second and third cm and was not obvious further from the cut end. In the two weeks after this initial increase, there was a decrease in impedance which was also marked in the first cm. This decrease is consistent with the finding from histological studies that a cut nerve degenerates back to the first node of Ranvier [12]. Within about three weeks there was a smaller and less distinct increase which may be associated with attempts by the nerve axons to grow back to their end organs [18]. Since the cuff was sealed and the attempted regeneration was unsuccessful, the impedance declined slowly as the fibers gradually decreased in diameter, a process which may continue for years [3]. The changes in impedance at the electrodes further from the cut end were much less dramatic. The major change was a slow increase in impedance which could be due to an ingrowth of connective tissue, as with intact nerves.

Discussion

The results of this study show that platinum-iridium but not silver electrodes remain stable for months when implanted around mammalian nerves. Signals recorded from nerves also remain stable, as shown elsewhere [17], and provide a reliable method for studying the activity of mammalian nerves during normal behavior and the changes that take place when nerves are cut, blocked, or subjected to other experimental procedures. In fact, measurements of impedance from cut nerves proved a remarkably sensitive way of monitoring the changes that took place, for comparison with the changes in neuronal activity or histological appearance.

Most of the changes in impedance could be readily attributed to changes in nerve diameter and growth of connective tissue. However, the reason for the decline in cat sciatic nerve impedance after two to three months of implantation was more puzzling. One possibility is that some nerve fibers degenerated and the high impedance of myelin was replaced by tissue of lower impedance. There is evidence for a selective loss of some nerve fibers [7, 9] due to the constraints of a cuff. Clinically in nerve repair operations, Ducker and Hayes [5] recommend use of cuffs 40% larger than the peripheral nerve being repaired. The sciatic nerves generally filled the cuffs quite fully, whereas the cuffs used on smaller nerves in the cat and mouse were more in line with Ducker and Hayes' [5] recommendation. Interestingly, the late decline in impedance was less obvious or absent in these other nerves (Fig. 4B, C). In any case, the small degree of degeneration, which might be indicated by the slight long-term decline in impedance, does not hinder the use of these cuffs in basic research and should not prevent their application to clinical problems.

A further goal of this study was to describe the impedance of the electrodes in terms of a simple circuit diagram. The circuit diagram of Fig. 2 assumes that the impedance can be considered as a resistance and a capacitance. The capacitance C may be due to the double layer which forms at the electrode-fluid or electrode-tissue interface. This double layer has been extensively studied by physical chemists (see, for example, Bockris and Reddy [2]). If the electrodes were non-polarizable (e.g., if the silver wires were chlorided to provide a good silver-silver chloride surface), then the resistance R', shown by the dashed lines in Fig. 2, should be considered. It would limit the impedance at low frequencies to some finite value. An unchlorided silver wire or a platinum-iridium wire is a polarizable electrode which does not pass dc currents (i.e., its impedance approaches ∞ near 0 Hz). Over the frequency range of interest (10 to 10,000 Hz) we rarely saw any evidence of the resistance R' shown by the dotted line and it will be ignored.

The voltage detected by such an electrode will therefore be capacitively coupled to the recording equipment. There will also be some stray capacity C' between the leads and ground. This stray capacity will produce a decrease in impedance and a loss of signal amplitude at sufficiently high frequencies. There was usually some indication of a downward trend in the impedance near 10 kHz attributable to this stray capacity, but its effect was small with large, low-impedance cuffs in the cat (Fig. 2A). The effects of stray capacity were more noticeable in the small cuffs implanted on nerves in the mice (Fig. 2B) and a decline in phase at high frequency can also be observed as a result of stray capacity at high frequency.

Some deviations from the frequency characteristics of the model circuit were consistently observed. Maximum phase shifts of about 70° are seen in Fig. 2, rather than the 90° expected with a capacitor. Similarly, the slope of the gain curve was consistently less than one (i.e., the impedance decreases as a fractional power of frequency [11, 13], rather than linearly). The data can be fitted better by assuming that the capacitor in Fig. 2 is of fractional order, i.e.,

$$C = (j\omega)^{-n}$$

where $j = \sqrt{-1}$, n is a fraction between 0.5 and 1 and ω is a frequency in radians/s. However, there is no compelling theoretical reason for assuming a capacitor of fractional order, and the better fit may simply be due to adding an extra parameter. In any case, since the tissue impedance hardly varies with frequency, the electrode impedance becomes dominant at low frequencies and changes in the low-frequency impedance with time (Figs. 3 and 4) reflect changes in electrode properties.

Finally, the phase at high frequencies does not approach zero as expected, and the magnitude of the impedance is not constant at high frequencies. In addition to effects of the stray capacity mentioned above, some of the deviations may be due to the diffusion layer outside the electrode. This layer produces the so-called Warburg impedance [11] which decreases in magnitude with frequency according to a square root relation (a slope of $-\frac{1}{2}$ on a log-log plot such as Fig. 2) and has a constant phase lag of 45°. However, the dependence of impedance on frequency was never this steep at high frequencies.

In summary, electrode properties may add somewhat to the impedance at high frequencies and stray capacity may detract from it, but the major contribution to the impedance at high frequencies is due to the fluid and tissue filling the cuff. This conclusion is strengthened by comparing the effective resistivity calculated from equation (1) with measurements of the resistivity of myelinated nerves which give values near 200 Ω-cm [20]. Reasonable values of resistivity are obtained for large nerves in the cat and for smaller nerves in the cat and mouse. Thus, by measuring the changes in the values of R and C with impedance measurements at 1 kHz and 10 Hz, it is possible to follow changes in tissues and electrode properties, respectively.

Acknowledgment

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References

[1] A. R. Allan, "Regeneration electrode impedance analysis and recording amplification method," *M.Sc. Thesis*, Department of Electrical Engineering, University of Alberta, 1976.

[2] J. Bockris and A. Reddy, *Modern Electrochemistry*. Plenum Press: New York, 1970.

[3] B. G. Cragg and P. K. Thomas, "Changes in conduction velocity and fibre size proximal to peripheral nerve lesions," *J. Physiol. Lond.* vol. 157, pp. 315-327, 1961.

[4] C. J. DeLuca and L. D. Gilmore, "Voluntary nerve signals from severed mammalian nerves: long-term recordings," *Science* vol. 191, pp. 193-195, 1976.

[5] T. B. Ducker and G. T. Hayes, "Experimental improvements in the use of silastic cuffs for peripheral nerve repair," *J. Neurosurg.* vol. 28, pp. 582-587, 1968.

[6] A. M. Dymond, "Characteristics of the metal-tissue interface of stimulation electrodes," *IEEE Trans. Bio. Med. Engng. BME* vol. 23, pp. 274-280, 1976.

[7] J. A. Hoffer and W. B. Marks, "Long-term peripheral nerve activity during behaviour in the rabbit," *Adv. Behav. Biol.* vol. 18, pp. 767-768, 1976.

[8] J. A. Hoffer, T. E. Milner, and R. B. Stein, "Long-term peripheral nerve and muscle recordings in intact mice," *Neurosci. Abstr.* vol. 2, No. 1040, 1976.

[9] A. Mannard, R. B. Stein, and D. Charles, "Regeneration electrode units: implants for recording from single peripheral nerve fibers in freely moving animals," *Science (N. Y.)* vol. 183, pp. 547-549, 1974.

[10] V. Pollak, "Computation of the impedance characteristic of metal electrodes for biological investigations," *Med. Biol. Engng.* vol, 12, pp. 460-464, 1974a.

[11] V. Pollak, "Impedance measurements on metal needle electrodes," *Med. Biol. Engng.* vol. 12, pp. 606-612, 1974b.

[12] S. W. Ranson, "Retrograde degeneration in the spinal nerves," *J. Comp. Neurol.* vol. 16, pp. 265-293, 1906.

[13] D. A. Robinson, "The electrical properties of metal microelectrodes," *Proc. IEEE* vol. 56, pp. 1065-1061, 1968.

[14] M. Salcman and M. J. Bak, "Design, fabrication and behavior of chronic recording intracortical microelectrodes," *IEEE Trans. Biomed. Engng. BME* vol. 20, pp. 253-260, 1973.

[15] C. L. Stanitski and V. Mooney, "Osseus attachment to vitreous carbons," *J. Biomed. Materials Res.* vol. 7, *Biomed. Mater. Symp.* vol. 4, pp. 97-108, 1973.

[16] R. B. Stein, D. Charles, L. Davis, J. Jhamandas, A. Mannard, and T. R. Nichols, "Principles underlying new methods for chronic neural recording," *Can. J. Neurol. Sci.* vol. 2, pp. 235-244, 1975.

[17] R. B. Stein, T. R. Nichols, J. Jhamandas, L. Davis, and D. Charles, "Stable long-term recordings from cat peripheral nerves," *Brain Res.*, vol. 128, pp. 21-38, 1977.

[18] R. B. Stein, J. Jhamandas, and T. R. Nichols, "Long-term recording of electrical activity of mammalian nerves following axotomy," *Soc. Neurosci. Abstr.* vol. 1, No. 795, 1976.

[19] R. E. Strain, Jr., and W. E. Olson, "Selective damage of large diameter peripheral nerve fibers by compression: an application of Laplace's Law," *Exp. Neurol.* vol. 47, pp. 68-80, 1975.

[20] I. Tasaki, "A new measurement of action currents developed by single nodes of Ranvier," *J. Neurophysiol.* vol. 27, pp. 1199-1206, 1964.

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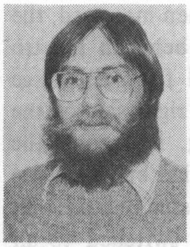

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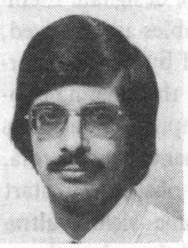

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Decompression Studies Using Ultrasonic Imaging of Bubbles

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Abstract—Single bubbles ranging down in size to under 1 μm (less than capillary size) can be noticed, localized, and measured in ultrasonic images of intact subjects using 7.5 MHz ultrasound for which the wavelength is 200 μm. Subjects included humans, fish, and guinea pigs. A combined brightness modulation and deflection display was most effective. Bubble reality during decompression and association with symptoms has been demonstrated, as have asymptomatic bubbles, a tendency for bubble formation in fat, recompression bubble showers, and decompression without diving tables. In guinea pigs there were age and male-female differences in susceptibility. Adjacent tissue inert gas pressure, supersaturation, and time constant can be measured by adjusting ambient pressure until bubbles cease to grow. Present data generally favor a supersaturation rather than a phase equilibration model for bends onset. An increase in allowable supersaturation was observed when decompression was to altitude rather than to sea level. Goldfish were seen to survive bubbling that would kill the mammals studied, and some simultaneous observations by light and sound were made in transparent fish.

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Introduction

THE formation of small gas bubbles has long been hypothesized to be involved in decompression sickness or the bends. Indeed, some effects were hypothesized as due to the formation of preclinical or "silent" bubbles that had no effect but could later grow to cause problems. One would not expect to observe such small bubbles in normal x-ray images (though manipulation of a joint in ordinary circumstances is known occasionally to produce a large noticeable bubble). Small bubbles interact strongly with sound waves and thus one would expect to detect them with an ultrasonic system. The original suggestion and observation with a pulsed "B scan" imaging system [1] reported increased tissue opacity in a decompressed rat observed at 15 MHz.

The original equipment [2] was modified by removal of the compound scan to reduce the number of confusing background echos. Subsequent observations were made [3, 4, 5, 6] during the process of decompression, either by taking the equipment into a large pressure chamber or by imaging through an acoustic window into a chamber for small animals.

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