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Imbalanced Biphasic Electrical Stimulation: Muscle Tissue Damage

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The effects of imbalanced biphasic stimulation were studied on cat skeletal muscle to determine if greater charge densities can be safely used than with balanced or monophasic stimulation. The results of the study indicate that imbalanced biphasic stimulation can be tolerated safely by tissue at or below a net dc current density of 35 $\mu A/mm^2$ and not safely tolerated at or above a net dc current of 50 $\mu A/mm^2$. Monophasic stimulation has been shown to be safe at or below net dc current levels of 10 $\mu A/mm^2$ and in these studies we found it was not safe at or above net dc current levels of 20 $\mu A/mm^2$. Stimuli were applied to muscles via coiled wire intramuscular electrodes using a regulated current source. Since the safe average current density was higher for imbalanced biphasic stimulation than for monophasic stimulation, this suggests that: (a) pH change is not the primary reaction causing tissue damage and (b) the damaging electrochemical process that takes place during a cathodic stimulation pulse can be reversed by an anodic pulse having substantially less charge than its companion cathodic pulse. We conclude that greater cathodic charge densities can be safely employed with imbalanced biphasic stimulation than with either monophasic stimulation or balanced charge biphasic stimulation.

Keywords—*Electrical stimulation, Tissue damage, Electrode.*

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INTRODUCTION

The electrical activation phenomenon of the nervous system provides an opportunity to restore missing or impaired body function to systems normally under neural control. Assistance devices based on this phenomenon are often referred to as neural prostheses. Because small electrodes are often needed to effect selective activation of small groups of nerves or it is necessary to depolarize axons at great distances from the electrode, it is desirable to be able to inject as much current as possible through these electrodes. The long-term viability of these neural prosthesis requires that neither the activated cells nor the activating electrode be damaged by the process of electrical stimulation. In a previous study (Mortimer *et al.* (9) it was shown that cathodic monophasic stimulation must be limited to 0.2 $\mu C/mm^2$/phase, 50 Hz to avoid excessive tissue damage and balanced charge biphasic stimulation must be limited to 0.4 $\mu C/mm^2$/phase, 50 Hz to avoid electrode corrosion. Since this study indicated that muscle tissue could tolerate a net dc current (net dc current is defined as (cathodic charge per phase − anodic charge per phase) × (stimulation frequency)), we hypothesized that *imbalanced* biphasic stimulation could be tolerated safely by both the electrode and the muscle at cathodic charge densities greater than that possible under balanced or monophasic charge conditions.

In the previous study (9) (Fig. 1) it was shown that muscle tissue could safely tolerate cathodic monophasic stimulation at a net dc current of 10 $\mu A/mm^2$ ((0.2 $\mu C/mm^2$/phase − 0.0 $\mu C/mm^2$/phase) × 50 Hz). At 50 $\mu A/mm^2$ (1.0 $\mu C/mm^2$/phase, 50 Hz), the next value tested, unacceptable muscle tissue damage occurred. The charge transfer during monophasic stimulation at this current density resulted in a large pH increase (*in vitro* tests), in the immediate vicinity of the electrode, that was presumed to reflect the hydrogen evolution reaction. These results were interpreted to indicate that a hydroxyl ion generation rate corresponding to 10 $\mu A/mm^2$ could be adequately buffered by normal body processes and that rates corresponding to 50 $\mu A/mm^2$ could not be adequately buffered.

An examination of the effect of balanced charge biphasic stimulation was also reported as part of the same study. The pH change under these conditions (*in vitro*) was minute when compared with equal current and charge densities during cathodic monophasic stimulation. At charge densities of 1.0 and 2.0 $\mu C/mm^2$ per phase, at which monophasic stimulation was judged unsafe, the tissue damage with balanced charge biphasic stimulation was not distinguishable from the tissue response to a passive implant and was judged to be safe. However, electrode corrosion was observed during biphasic stimulation at these charge densities making it unfit for chronic stimulation.

Combining the results of the monophasic study with those of the balanced charge biphasic study suggested that a charge imbalanced biphasic stimulation pulse could be tolerated by both the electrode and muscle tissue. Furthermore, a charge imbalanced biphasic stimulus waveform should permit greater charge densities for the cathodic phase of the stimulating pulse than could be safely tolerated with either the monophasic or balanced charge biphasic stimulation waveform. The parameters of the waveform were predicted using the following assumptions. First, that the imbalanced portion of the stimulus (which yields a net dc current) produces an electrochemical reaction that results in tissue damage at a rate corresponding to the net dc current.

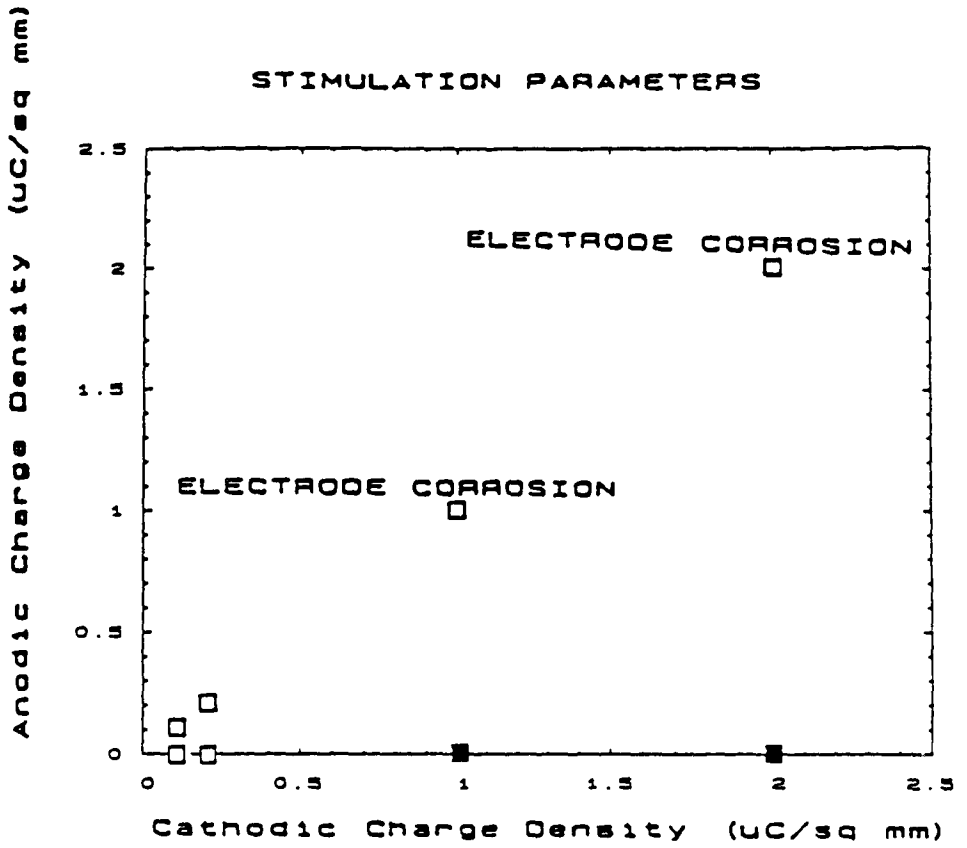


FIGURE 1. A plot of the stimulation parameters used in the tissue damage study of Mortimer et al. (9). Two types of stimulating waveforms—both regulated current—were studied: one rectangular monophasic and the other, balanced charge biphasic with an exponentially decaying current during the anodic portion of the wave. In all cases, the cathodic phase was rectangular and applied first (in time) to the intramuscular electrode. Pulse amplitudes of the rectangular phase were, in all cases, 20 mA; durations of 50, 100, 500, and 1000 μS were used. "Open" symbols represent results that showed no significant tissue damage. "Filled" symbols represent results that showed significant tissue damage. Symbols marked "electrode corrosion" indicate that corrosion was observed after removal from the test muscle.

Second, that the electrochemical reaction produces a reduction in the local hydrogen ion concentration that can be buffered to a limited extent by naturally available mechanisms. Third, that the electrochemical reaction and its effect on muscle tissue has the same damaging effect whether it is applied as a monophasic stimulus or as an imbalanced biphasic stimulus at the same net dc current. When these assumptions and the previous data were used, it was predicted that an imbalanced stimulus waveform that (a) does not subject the electrode to potentials that produce corrosion and (b) has a net dc component of 10 $\mu A/mm^2$ or less can be safely tolerated by the muscle tissue. Furthermore, a similar waveform but with a net dc component of 50 $\mu A/mm^2$ or more cannot be tolerated.

METHODS AND MATERIALS

Electrode Manufacture

Electrodes were formed from polyurethane insulated 316LVM stainless steel wire 46 μm in diameter (California Fine Wire Co.) and inserted into 22 gage needles (4). The overall diameter of the coil electrodes ranged from 200 to 250 μm in diameter. The length of the noninsulated portion of the coil (the stimulating tip) was 10 mm (the manufacturing tolerance of the tip length was $\pm 5\%$). The effective stimulating surface area (geometric) was approximately 10 mm^2. (The geometric stimulating area was determined by calculating the outside surface area of the stimulating tip modeled as a close wound helix.)

Electrode Implantation

Six adult cats weighing between 3.0 and 4.5 kg were used in these studies. The animals were anesthetized with intravenous administered sodium pentobarbital, intubated, and prepared for surgery by shaving and cleaning the implant sites with Povidone-Iodine, 10% solution. Test electrodes were implanted bilaterally, approximately parallel to the muscle fibers, under aseptic conditions, in the following muscles: **tibialis anterior, medial gastrocnemius, lateral gastrocnemius,** and **triceps brachii.** Implantation was achieved by insertion of a loaded hypodermic needle, at an acute angle in relation to the skin surface, into the center of the test muscle. The needle was then withdrawn leaving the electrode in place. Counterelectrodes were attached to the test animal by lightly wrapping stainless steel cable and saline-soaked pads to each of its paws. A schematic of the test setup is shown in Fig. 2. Subsequent to electrical stimulation, the animals were placed on a respirator and curare was administered intravenously to minimize animal motion due to motor nerve excitation at the higher stimulus levels.

Stimulation

Electrical stimulation was applied immediately after implantation to all of the coil electrodes continuously for 6 h. (A 6-h stimulation time was chosen because (a) it was a reasonable period to maintain the animal under anesthesia, (b) it was sufficient time to permit damage to the muscle tissue to develop (8) and (c) it permitted a direct comparison with previous work (8) that also used a 6-h stimulation period.) Low levels of muscle contractions, lasting less than 30 min, were observed near the implanted electrodes. The current through each lead, and the voltage, measured from each lead to the counter electrode, were monitored to ensure that lead-electrode continuity was maintained throughout the experiment. The stimulating waveform used was produced from a regulated current source. The waveform and the method of generating the waveform are illustrated in Fig. 3. In all cases, the stimulating frequency was 50 Hz and the stimulation waveform was rectangular. (The test frequency, 50 Hz, was chosen because it is an upper limit for most motor prosthesis applications.) When biphasic stimulation was applied to an electrode, the cathodic phase was always applied first. Electrodes were stimulated at various charge densities at pulse widths of 75 or 150 μs. A list of the stimulation parameters used is given in Table 1. A plot of these stimulation parameters (based on the anodic and cathodic charge densities of the stimulus pulse) is shown in Fig. 4. At the end of the stimulation period the animals were

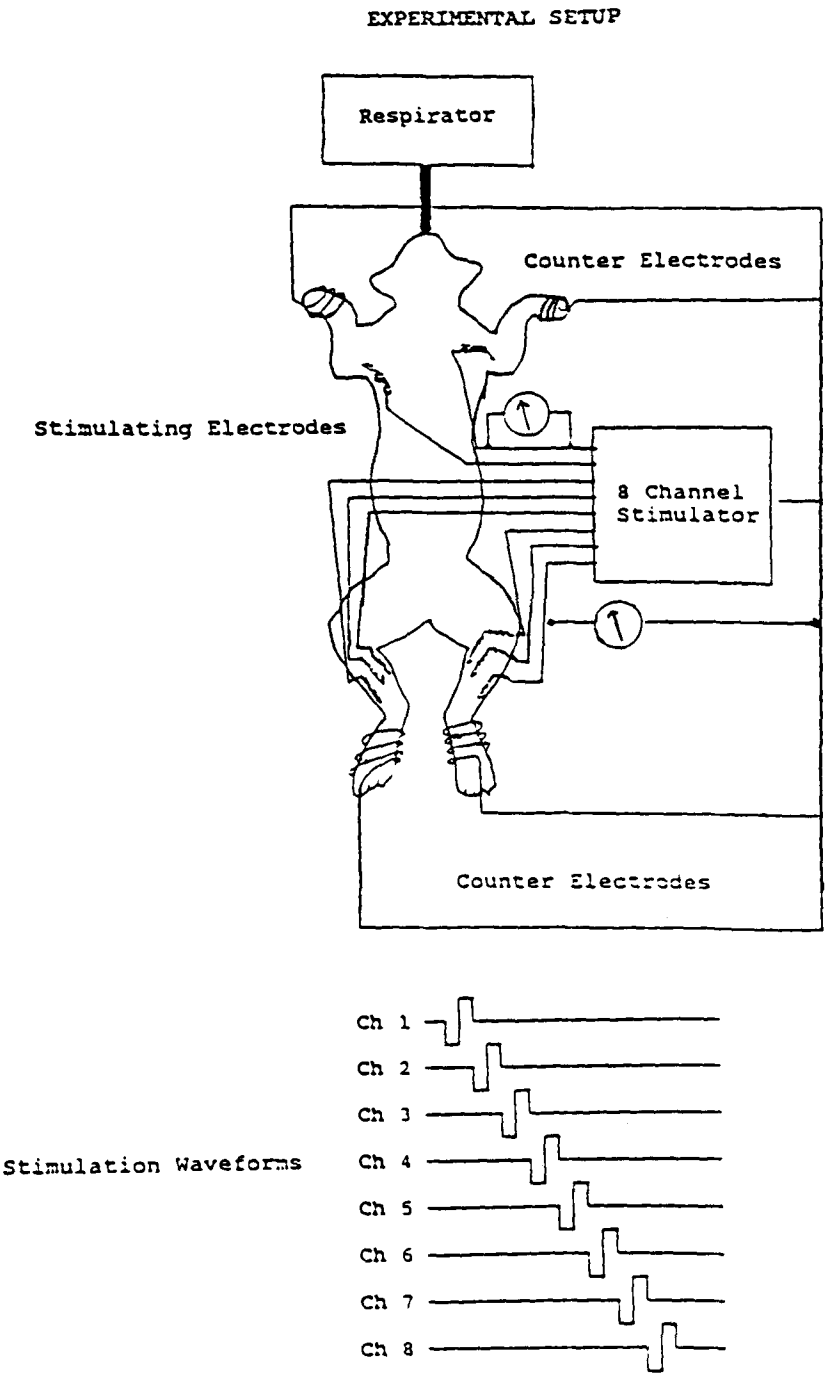


FIGURE 2. Schematic of the test setup. Eight electrodes were implanted in *tibialis anterior, medial gastrocnemius, lateral gastrocnemius,* and *triceps brachii* muscles. Counter electrodes were attached to each paw of the test animal. Each of the test electrodes were stimulated sequentially at a frequency of 50 Hz. Subsequent to stimulation, the animal was given curare and placed on a respirator.

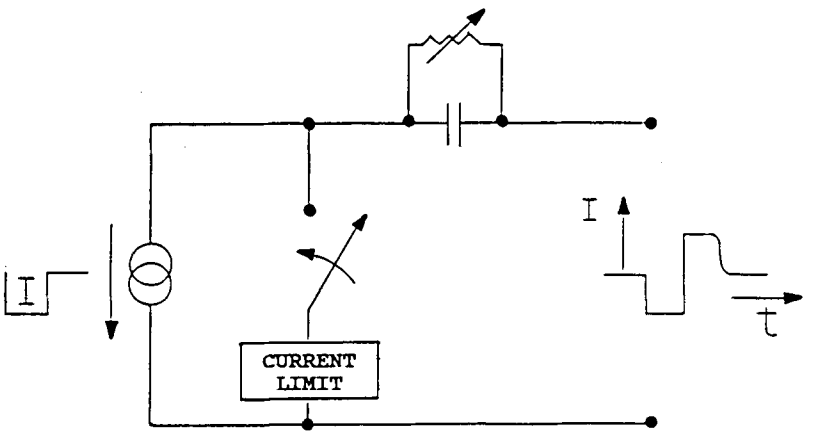


FIGURE 3. Schematic of the biphasic stimulator used in these experiments. The cathodic phase was generated by the regulated current pulse generator, passing current through the series combination of the load (tissue), and the capacitor in parallel with a resistor. The shunting switch was open during the cathodic phase but was closed at all other times. Some charge flowing during the cathodic phase was shunted in the parallel resistor and some charge flowed in the reverse direction during the interpulse interval. The current flow during the anodic phase was limited to produce a pseudorectangular wave. The net result was a rectangular biphasic imbalanced waveform. The degree of imbalance can be varied by varying the resistor values.

allowed to recover from the anesthesia and neuromuscular block. After recovery, no change in the behavior of any animal was noticed suggesting that the aftereffects of the tests did not cause the animal discomfort or gross changes in motor movement.

Tissue Analysis

Five to six days after stimulation the animal was anesthetized as previously described, the test muscles were excised, and the animal was killed with an overdose of sodium pentobarbital. The electrodes were carefully removed and examined with a light microscope for signs of corrosion or mechanical damage. Four serial blocks (2 to 3 mm thick) were cut from each muscle and frozen in liquid nitrogen. The muscle blocks that were taken are shown schematically in Fig. 5. Block I was taken 5 to 10 mm distal to the electrode tip. Blocks II and III were located in the region surrounding the active portion of the electrode. Block IV was cut from tissue surrounding the insulated region of the electrode as far as possible from the active region. Frozen sections, 16 μm thick, were cut and stained with hematoxylin and eosin. Areas of the muscle showing abnormally staining tissue, necrotic or degenerating fibers, cellular infiltrates, increased connective tissue, or regenerating fibers were defined as "damaged" and measured from a projected microscopic image ($\times$40) by tracing the image with a digital graphics analyzer.

RESULTS AND CALCULATIONS

Tissue specimens were evaluated by two criteria—quantitative measurement of the cross-sectional area damaged (damaged as defined in methods section) by stimulation and a qualitative examination of the type of damage observed. Tissue stimulated with monophasic stimulation (10 sites) produced a larger average damaged area than bi-

TABLE 1. Stimulation parameters used in this study. The cathodic current, anodic current, and pulsewidth were independently set for each test site. Stimulation frequency was 50 Hz. TA = tibialis anterior, LG = lateral gastrocnemius, MG = medial gastrocnemius, TB = triceps brachii.

			Stimulation Parameters		
Site No.	Animal No.	Muscle	Current Amplitude I_a/I_c (mAmp)	Pulse Width (μs)	Charge Density Q_a/Q_c ($\mu C/mm^2$)
1	1	TA	33/48	150	0.5/0.7
2	1	TA	13/27	150	0.2/0.4
3	1	LG	33/60	150	0.5/0.9
4	1	LG	13/40	150	0.2/0.6
5	1	TB	33/80	150	0.5/1.2
6	1	TB	13/60	150	0.2/0.9
7	2	TA	33/80	150	0.5/1.2
8	2	LG	33/48	150	0.5/0.7
9	2	LG	13/40	150	0.2/0.6
10	2	TB	13/27	150	0.2/0.4
11	2	TB	33/60	150	0.5/0.9
12	3	TA	0/27	150	0.0/0.4
13	3	TA	0/27	150	0.0/0.4
14	3	LG	0/68	150	0.0/1.0
15	3	TB	0/100	150	0.0/1.5
16	3	TB	0/120	150	0.0/1.8
17	4	TA	33/60	150	0.5/0.9
18	4	TA	13/40	150	0.2/0.6
19	4	LG	33/80	150	0.5/1.2
20	4	LG	13/60	150	0.2/0.9
21	4	MG	33/100	150	0.5/1.5
22	4	MG	13/100	150	0.2/1.5
23	4	TB	13/80	150	0.2/1.2
24	4	TB	13/80	150	0.2/1.2
25	5	TA	13/27	150	0.2/0.4
26	5	TA	27/53	75	0.2/0.4
27	5	LG	27/53	75	0.2/0.4
28	5	LG	33/48	150	0.5/0.7
29	5	MG	66/95	75	0.5/0.7
30	5	MG	27/80	75	0.2/0.6
31	5	TB	66/95	75	0.5/0.7
32	5	TB	27/80	75	0.2/0.6
33	6	TA	0/27	150	0.0/0.4
34	6	TA	0/68	150	0.0/1.0
35	6	LG	0/48	150	0.0/0.7
36	6	MG	0/48	150	0.0/0.7
37	6	TB	0/48	150	0.0/0.7
38	6	TB	13/60	150	0.2/0.9

phasic stimulation (28 sites) for the same level of net dc current. The area of damage increased as the net dc current increased for both monophasic and biphasic stimulation. Two distinct types of tissue damage were observed, a zone of degenerating and regenerating muscle fibers with scattered polymorphonuclear leukocytes,

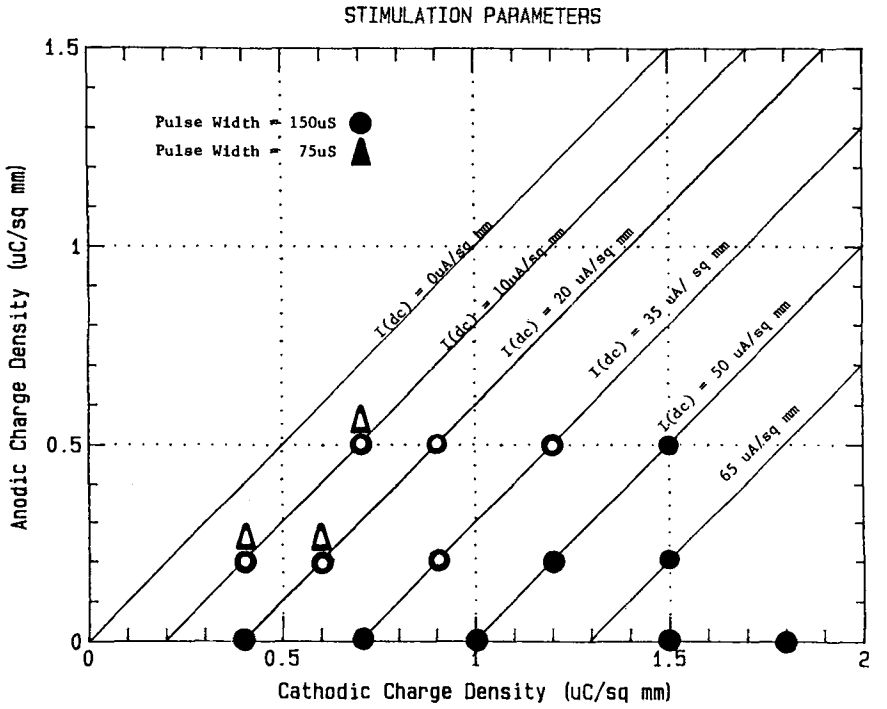


FIGURE 4. A plot of stimulation parameters used in this study. The ordinate and abscissa scale are the charge density per phase during the anodic and cathodic portion of the stimulus wave. The lines plotted at 45° represent different values of net dc current that are defined as: $I_{dc} = PW(I_c - I_a)/IPI$, where: I_{dc} = net dc current, PW = pulse width, I_c = cathodic current amplitude, I_a = anodic current amplutide, IPI = interpulse interval. Circles represent tests run at a pulse width of 150 μS. Triangles represent tests run with a pulse width of 75 μS. "Open" symbols represent test results that were judged to be no different than control. "Filled" symbols represent results that showed tissue damage that was significantly greater than control values.

and a zone of coagulation necrosis. The second type of damage was always observed in muscle stimulated with monophasic stimulation and occasionally in muscle stimulated with biphasic stimulation at the highest levels of net dc current.

Damaged Area Measurements

A quantitative list of the damaged areas measured from each tissue sample is presented in Table 2. The reduction in cross-section of the tissue samples during freezing was estimated to be a few percent and was ignored in these measurements. The measurement error of the projecting microscope–digitizer system was estimated to be within ±10% for 95% of the measured values. (This error was determined by performing multiple measurements on a group of samples and then calculating the variation between measurements.) Multivariate analyses using ordinary linear least squares regression (2) of the test data were performed to examine the relationship between the independent variables and the dependent variable, tissue damage area. The type of current waveform (monophasic vs. biphasic) and the amount of net dc cur-

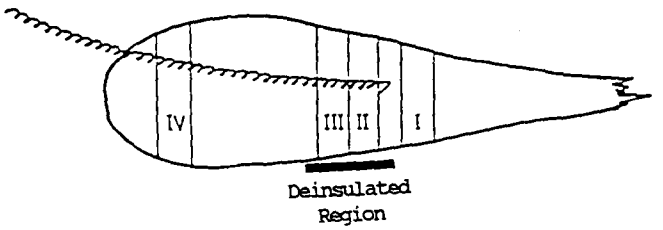


FIGURE 5. Schematic representation of test muscle showing the relationship between the electrode and tissue samples removed. Blocks II and III were in the regions surrounding the deinsulated tip of the electrode. Block IV was in the region surrounding the insulated section of the electrode (after Figure 2 in ref. 9).

rent (defined as (cathodic charge per phase − anodic charge per phase) × stimulation frequency) were determined to be the parameters that correlate best with the degree of tissue damage, presented in Table 3 and plotted in Fig. 6. Biphasic and monophasic stimulation data, at each level of net dc current, were compared with control data (Table 4) for statistical differences using Scheffe's method (5). The results of these tests showed that (a) imbalanced biphasic stimulation at average current densities of 35 $\mu A/mm^2$ or less causes no more damage to muscle tissue than a passive implant and (b) monophasic stimulation at average current densities of 20 $\mu A/mm^2$ and greater causes damage to muscle that is greater than that caused by a passive implant.

Qualitative Evaluation

Light microscope observation of the muscle sections from the insulated (control) region of the electrode (block IV) (Fig. 7) showed a small zone of polymorphonuclear leukocytes, macrophages, and degenerating and regenerating muscle fibers present around the electrode tract. The area occupied by abnormally staining tissue in these sections was taken as the baseline for damage (control) and measured to be 1.07 $mm^2 \pm 0.49$.

Muscle sections taken from the levels of the deinsulated tip (blocks II and III) showed damaged areas that varied with current intensity and waveform. Muscles stimulated with imbalanced biphasic current revealed a distinct pattern. Low net dc currents (10 to 35 $\mu A/mm^2$) resulted in a tissue reaction that was similar to that of the insulated portion of the electrode. As the level of net dc current was increased the area of damage increased. Tissue stimulated at the highest net dc current (50 to 65 $\mu A/mm^2$) (Fig. 8) occasionally showed a small zone of coagulation necrosis. Outside the zone of coagulation necrosis, there was a zone containing muscle cells showing degeneration and regeneration. The size of this zone increased with greater current amplitudes.

Muscles subjected to monophasic stimulation (from 20 $\mu A/mm^2$ to 90 $\mu A/mm^2$ net dc current) (Figs. 9 and 10) always showed a larger area of coagulation necrosis when compared to biphasic stimulation. Within this area, all of the muscle fibers were completely necrotic and there was no evidence of regeneration or scar formation. The zone of necrosis tended to increase with increasing net dc current and was surrounded by an area containing fibers undergoing degeneration and regeneration, similar to that seen in the tissue stimulated with biphasic stimulation.

TABLE 2. Damaged areas measured from each tissue sample. A dashed line indicates a sample was not available for measurement. (At some electrode sites, control samples were not available because the coil lead exited the muscle distal to section IV. Losses of tissue samples in blocks II and III were due to processing problems.)

Site No.	Test Results			
		Damaged Area (mm^2)		
	Control	Area 2	Area 3	Average (2 + 3)
1	–	0.3	0.4	0.4
2	0.6	0.8	1.2	1.0
3	–	0.8	1.2	1.0
4	–	0.7	1.0	0.8
5	1.0	0.3	1.2	0.7
6	–	–	0.7	0.7
7	–	4.1	2.1	3.1
8	–	2.7	–	2.7
9	–	1.6	1.7	1.7
10	–	1.2	–	1.2
11	–	1.6	–	1.6
12	–	5.1	–	5.1
13	–	4.0	6.5	5.3
14	–	15.8	–	15.8
15	1.5	20.0	–	20.0
16	–	25.0	28.9	26.9
17	1.5	2.5	1.3	1.9
18	–	–	0.8	0.8
19	1.4	0.8	1.8	1.3
20	1.0	0.6	3.3	2.0
21	–	6.8	2.4	4.6
22	1.2	3.6	–	3.6
23	–	2.2	2.2	2.2
24	–	1.2	3.4	2.3
25	0.7	0.7	0.7	0.7
26	0.7	0.7	0.9	0.8
27	–	0.4	1.2	0.8
28	0.9	2.0	2.3	2.1
29	0.9	1.5	2.8	2.1
30	0.9	0.9	1.3	1.1
31	0.9	1.3	–	1.3
32	0.6	0.9	1.3	1.1
33	1.1	1.5	2.0	1.7
34	2.5	17.0	6.8	11.9
35	–	5.7	7.6	6.6
36	–	–	3.6	3.6
37	1.5	3.1	–	3.1
38	0.4	1.1	1.0	1.0

Blood vessels and axons, located in the area of coagulation necrosis, usually showed severe damage. Occasionally, undamaged vessels and axons could be seen. Blood vessels and axons located on the edge of the lesion area show slight damage or appeared undamaged. No damage attributed to the electrical stimulation could be

TABLE 3. Averaged damaged areas based on net dc current and type of waveform (area given in units of mm^2).

Net dc Current	Test Sites	*N*	Average	Standard Deviation
Biphasic Stimulation				
10	1, 2, 8, 10, 25 26, 27, 28, 29, 31	10	1.19	0.59
20	3, 4, 9, 11, 17 18, 30, 32	8	1.40	0.65
35	5, 6, 7, 19, 20, 38	6	1.77	0.94
50	21, 23, 24	3	3.03	1.36
65	22	1	3.60	
Monophasic Stimulation				
20	12, 13, 33	3	4.03	2.02
35	35, 36, 37	3	4.43	1.89
50	14, 34	2	13.85	
75	15	1	20.00	
90	16	1	26.90	
Control				
0	2, 5, 15, 17, 19 20, 22, 25, 26, 28 29, 30, 31, 32, 33 34, 37, 38	18	1.07	0.49

seen in tissue samples taken in block I or far from the electrode (outside the area immediately adjacent to the electrode tract) in tissue samples from blocks II, III, or IV. No corrosion products were observed in the tissue specimens.

Examination of Test Electrodes

No signs of corrosion could be seen on electrodes examined with microscopy after removal from test muscles.

DISCUSSION

Safe Stimulation Parameters

The results of these experiments show that charge imbalanced biphasic stimulation can be safely tolerated by muscle tissue at and below imbalanced levels that produce a net cathodic current of 35 $\mu A/mm^2$. This level of stimulation was judged safe because (a) no statistically significant difference could be shown in the area of tissue damage when comparing the sample population mean to the control and (b) no significant histological changes could be seen in the sample tissues. The important consequence of this result is that the charge density during the cathodic pulse (the excitatory phase of the stimulus) can be increased. At the present time, balanced biphasic waveforms are used for intramuscular stimulation. Because the magnitude of

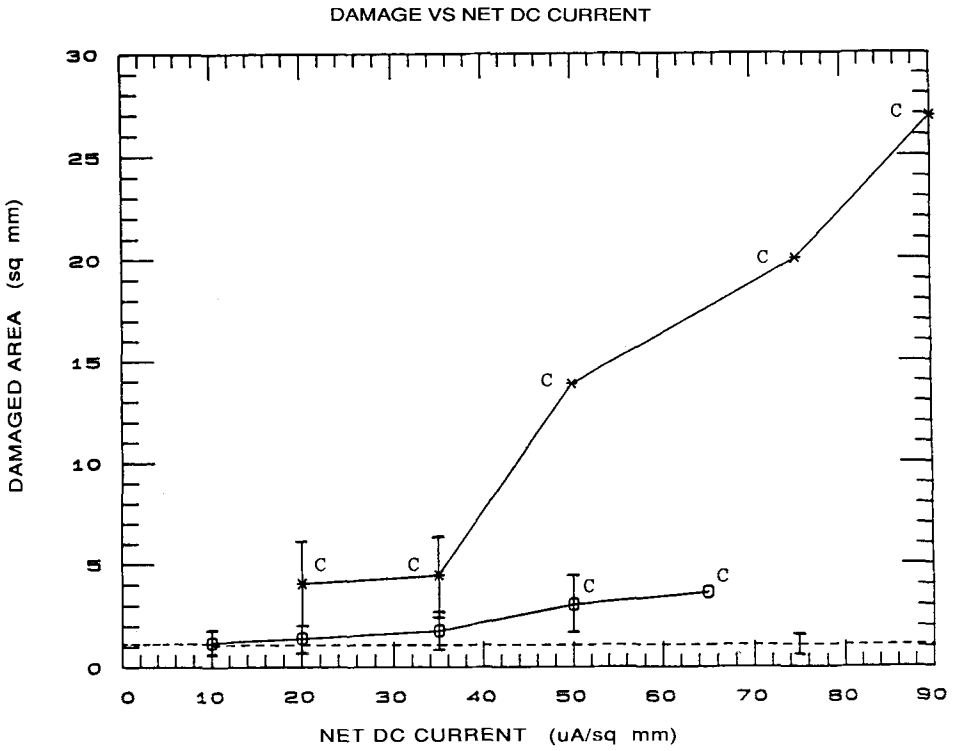


FIGURE 6. A comparison was made between the amount of damage to tissue near wire electrodes in the insulated region of the electrode (control) and the active region where the bare metal is exposed to muscle tissue. In the figure, the dashed line represents control data; the circles, biphasic stimulation; and the asterisks, monophasic stimulation. The letter "C" indicates coagulation necrosis was seen.

the anodic portion of the stimulus is limited by corrosion considerations to no more than 0.4 $\mu C/mm^2$, the cathodic charge density (the excitatory phase of the stimulus) is also no more than 0.4 $\mu C/mm^2$ (20 mA, 200 μs, 10 mm^2). These new findings indicate that the excitatory phase of the pulse can be as high as 1.2 $\mu C/mm^2$ (with an anodic phase of 0.5 $\mu C/mm^2$) without tissue or electrode damage with imbalanced biphasic stimulation. This means that the charge density in the excitation portion of the waveform may be up to three times greater than what has previously been recommended. The important consequence of this is (a) a single electrode can be used to activate a larger volume of muscle or (b) the surface area of the electrode could be reduced by a factor of 3 and still activate the same region of tissue as activated with balanced charge stimulation. A reduction of the electrode surface area and size may make the electrode tip less likely to break and provide a more localized stimulus to the tissue.

The predictions that a net dc current of 10 $\mu A/mm^2$ can be safely tolerated and 50 $\mu A/mm^2$ would cause tissue damage were verified. However, a conflict arises between the assumptions and the results obtained from the experiments. It was assumed that the electrochemical reaction producing tissue damage was a change in the hydro-

TABLE 4. Statistical comparison (using Scheffe's method (4) for multiple comparisons) of stimulation data to control. For biphasic stimulation, no difference can be seen in tissue damage area when comparing the sample population mean and control for net dc currents from 10 to 35 μA/mm^2. At a net dc current of 50 μA/mm^2, damage to stimulated tissue was greater than control. For monophasic stimulation, the amount of tissue damage area was greater than control for net dc currents of 20 and 35 μA/mm^2.

Net dc Current	df[a]	F	Accept H_1 ($p = 0.05$)
Comparing Biphasic Data to Control			
10	26	0.33	No
20	24	2.05	No
35	22	5.71	No
50	19	24.12	Yes
Comparing Monophasic Stimulation to Control			
20	19	34.97	Yes
35	19	49.13	Yes

Null hypothesis H_0: $U_s \leq U_c$
Alternate hypothesis H_1: $U_s > U_c$
where U_s = stimulated sample population
U_c = control sample population
df = $N_s + N_c - 2$

$$F = \frac{(X_s - X_c)^2}{(S_w^2/N_s) + (S_w^2/N_c)}$$

S_w = within group variance

Net dc Current	Confidence Interval
Confidence Intervals for Sample Population Means	
Control	$C(0.87 \leq U_s \leq 1.27) = 0.95$
Biphasic Stimulation	
10	$C(0.77 \leq U_s \leq 1.61) = 0.95$
20	$C(0.86 \leq U_s \leq 1.94) = 0.95$
35	$C(1.02 \leq U_s \leq 2.56) = 0.95$
50	$C(-0.35 \leq U_s \leq 6.41) = 0.95$
Monophasic Stimulation	
20	$C(0.62 \leq U_s \geq 7.44) = 0.95$
35	$C(1.24 \leq U_s \leq 7.61) = 0.95$

[a]df = degree of freedom.

gen ion concentration and that the net change in the local hydrogen ion concentration would be the same for monophasic and biphasic stimuli of equal net dc current densities. This may not be correct because monophasic cathodic stimulation at 20 and 35 μA/mm^2 resulted in muscle tissue damage that was judged to be significant while imbalanced biphasic stimulation at the same net cathodic current densities did not result in tissue damage. At the 50 μA/mm^2 net dc current amount, both the monophasic and biphasic stimulation showed significant damage. However, the area of tissue damaged by monophasic stimulation was much greater (Figs. 4 and 6). Furthermore, the nature of histological change was radically different with coagulation necrosis

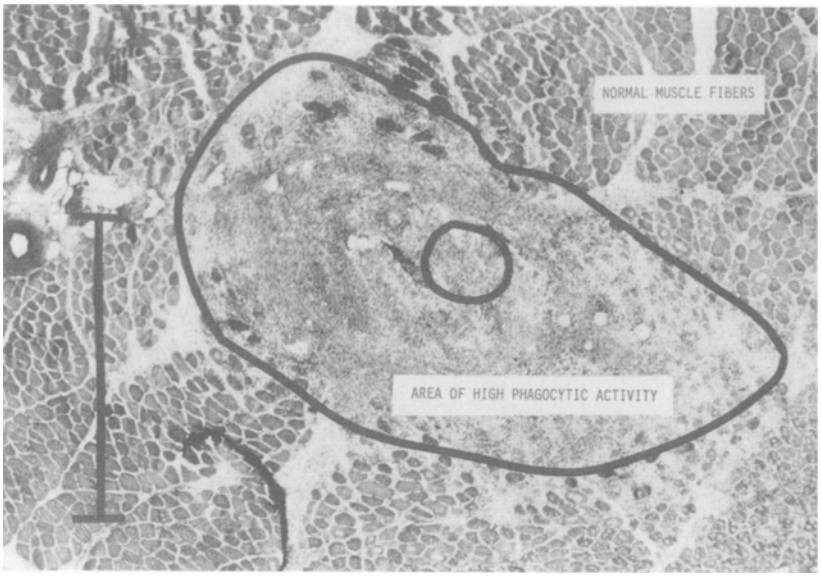


FIGURE 7. Photomicrograph from implant study (site #30). Tissue taken from block IV (control), in the insulated region of electrode. Section stained with hematoxylin and eosin. The small enclosed area shows the approximate size of the stimulating electrode. The large enclosed area (0.9 mm^2) is the tissue that has been identified as injured. The bar shown in the photograph is 1 mm long.

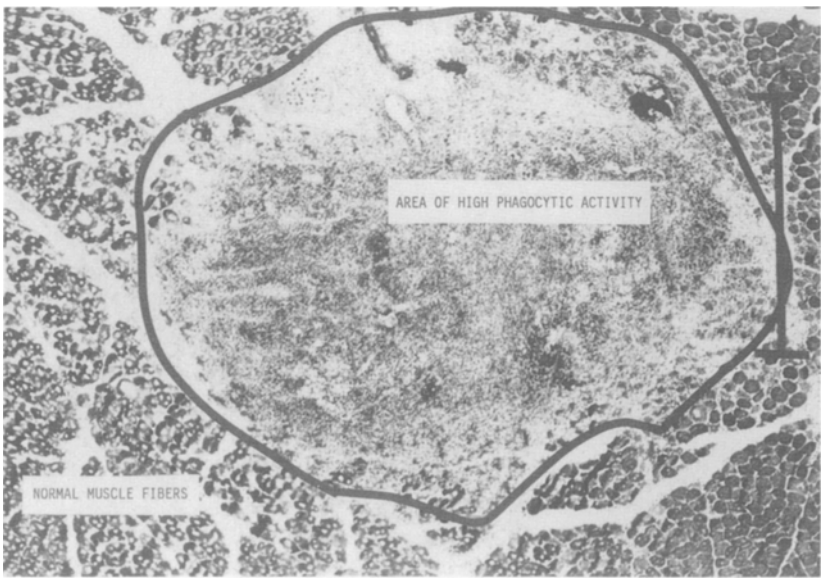


FIGURE 8. Photomicrograph from implant study (site #24). Tissue taken from block III (deinsulated) region of electrode. Stimulation parameters are cathodic current 80 mA, anodic current 13 mA, frequency 50 Hz, and a pulse width of 150 μS resulting in a net dc current of 50 $\mu A/mm^2$. Section stained with hematoxylin and eosin. The large outlined area is the tissue which has been identified as injured (3.4 mm^2). The bar shown in the photograph is 1 mm long.

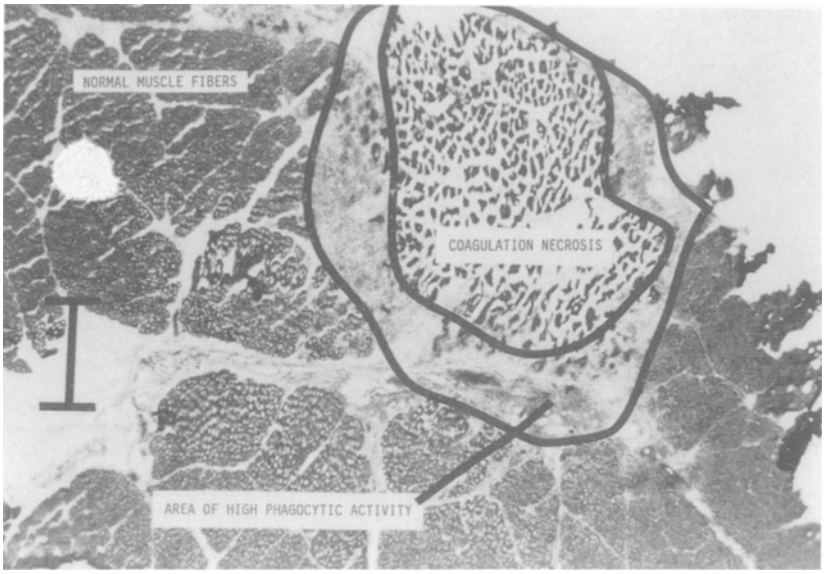


FIGURE 9. Photomicrograph from implant study (site #13). Tissue taken from block II (deinsulated) region of electrode. Stimulation parameters are a cathodic current of 27 mA, frequency of 50 Hz, and a pulse width of 150 μS resulting in a net dc current of 20 μA/mm^2. Section stained with hematoxylin and eosin. The large outlined area is the tissue that has been identified as injured (4.0 mm^2). The bar shown in the photograph is 1 mm long.

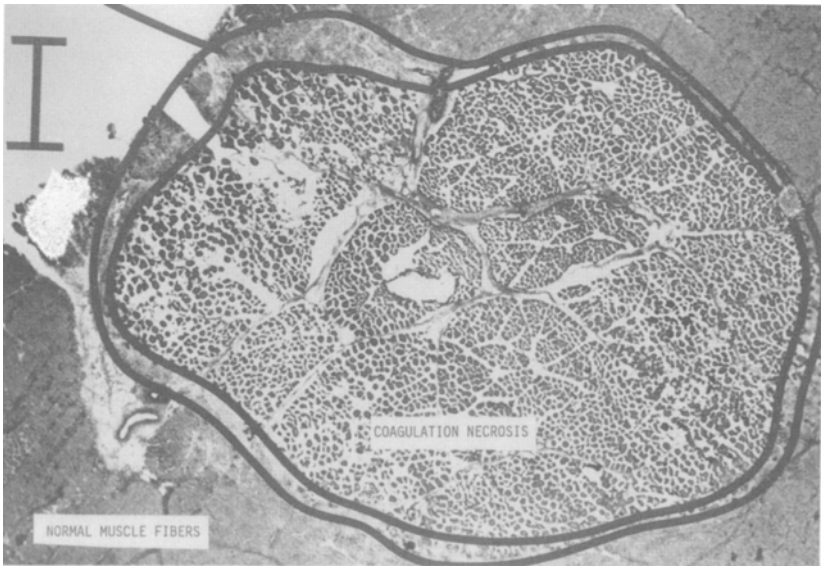


FIGURE 10. Photomicrograph from implant study (site #16). Tissue taken from block II (deinsulated) region of electrode. Stimulation parameters were a cathodic current of 120 mA, frequency of 50 Hz, and a pulse width of 150 μS resulting in a net dc current of 90 μA/mm^2. Section stained with hematoxylin and eosin. The large outlined area is the tissue which has been identified as injured (25.0 mm^2). The bar shown in the photograph is 1 mm long.

forming the most prominent effect of the monophasic stimulation. This type of necrosis occurs rarely in muscle and is produced usually by heat, deprivation of oxygen or bacterial toxins. These results imply that there is something different about the reactions produced by monophasic and imbalanced biphasic stimulation waveforms when operated at the same net dc cathodic current levels. The possible mechanisms of producing the damage under the described experimental conditions are discussed as follows.

Causes of Observed Tissue Damage

Mechanisms that may cause damage during electrical stimulation include heating, loss of blood flow to the area of stimulation, direct field effects, and electrochemical reactions that take place at the electrode–tissue interface.

Heating. Theoretical calculations of the temperature rise produced in body tissue by a spherical electrode have been published by Ruggeri, Beck, and Ballestrasse (1). They conclude that if the electrode radius is greater than 10 μm and the current density is less than 10 A/cm^2, then the peak temperature rise in the surrounding tissue will be less than 1°C. This model can be applied to coil electrodes. In this study, the intramuscular coil electrodes have a surface area of 10 mm^2 and stimulation current densities of not more than 1.2 A/cm^2. This should eliminate heating as a damage mechanism. The test results support this conclusion. Power dissipation at the electrode is proportional to the square of the current amplitude. Because more tissue damage was seen at sites stimulated with lower current amplitudes and monophasic stimulation than higher current amplitudes and biphasic stimulation, we conclude the damage was not caused by heat.

Loss of Blood Flow. The geometry of the vascular system within a musele can be characterized as a few main arteries entering the muscle body at various points, branching of these vessels to small arterioles and capillaries, then converging again to venule and main veins that exit the muscle. These blood vessels often run perpendicular to the muscle fibers as they spread out through the muscle. Thus, a branch may supply blood to groups of muscle fibers that are far apart in relation to the long axis of the muscle. If blood flow were halted in that branch, and the loss of blood flow caused tissue damage, then cross sections of the muscle should show damage in multiple areas. The lesions observed in this study were all continuous and confined to the area adjacent to the electrode track. Also, undamaged blood vessels have been observed in lesion areas where the muscle fibers are completely necrotic. From these observations, we cannot conclude that the tissue damage that we have seen was caused by loss of blood flow.

Direct Field Effects. Jones *et al.* (6,7) have investigated shock-induced electrical breakdown of cardiac membrane using monophasic and biphasic waveforms. Their working hypothesis is, using monophasic waveforms, "that during the shock, transient microlesions are formed in the cell membrane so that its structure breaks down and it no longer acts as a barrier to ions going back and forth across it. . . ." If there is excessive ion exchange, calcium overload within the cell may result and such severe injury can lead to cell death by inhibiting mitochondrial function or energy production, or by causing severe contracture. The reverse pulse of the biphasic waveform helps to "seal" the cell by reorienting the molecules more quickly instead of waiting

for the normal membrane functions to reseal it. The likelihood of this mechanism affecting skeletal muscle at the current amplitudes normally used for electrical stimulation is unknown.

Electrochemical Reactions. Electrochemical reactions on the surface of the stimulating electrode are the most likely cause of tissue damage. Reactions that may cause damage include production of metallic dissolution products, formation of oxidized organic and inorganic species, local pH shifts that can cause irreversible changes in tissue proteins, and gas evolution.

We conclude that corrosion did not contribute to tissue damage because (a) no corrosion was seen (using a scanning electron microscope) on specimens stimulated *in vitro* at current amplitudes corresponding to those used *in vivo*, (b) no signs of corrosion could be seen on electrodes examined after removal from the test muscles, (c) no corrosion products could be seen in the tissue specimens, and (d) sites of greatest tissue damage were stimulated with a waveform (cathodic monophasic stimulation) that will prevent corrosion through cathodic protection (8).

The generation of hydrogen or oxygen, which are potentially dangerous because (a) the pH shifts that can accompany the reactions and (b) the mechanical damage caused by gas bubbles, may contribute to tissue damage but are not considered the most important mechanism. The most likely electrochemical reactions (Table 5) (10) that can take place during the cathodic and anodic phases of stimulation will usually produce H^+ cations and OH^- anions in direct proportion to the number of electrons involved in the charge-carrying process. This suggests that the degree of hydrogen ion consumption (or pH change) during stimulation will be approximately proportional to the net charge passed (or average net dc current). The test results show that tissue damage gradually increases as net dc current increases (Fig. 6). However, the increase in damage was more dramatic when the type of waveform was changed from biphasic to monophasic at the same level of net dc current. From this we conclude that the pH change in the muscle tissue may be a contributing factor but is not the most important mechanism causing tissue damage. Furthermore, the increase in tissue damage that occurs as the level of net dc current is increased may be due to increases in other reactions that are discussed in the following paragraphs.

Reactions at the Electrode Surface. The flow of current between the metal electrode and the ionic medium occurs in many different processes. When a cathodic first biphasic stimulation waveform is used for stimulation, the reactions that occur during the cathodic half of the pulse are double-layer charging, oxide reduction, O_2 reduction, H_2 evolution, and H absorption process. On the anodic half of the pulse the reactions that occur are a reoxidation of the absorbed H, double-layer charging, oxide formation, metal corrosion, and O_2 evolution. The rates at which each of these reactions occur and the total time for their completion may vary considerably. This may result in many of these reactions occurring simultaneously. Brummer et al. (3) state that "Reversible charge injection is possible either by a capacitive process (such as double-layer charging) or by faradaic reactions involving species that remain bound to the electrode surface." Experimental evidence suggests that if the potential is reversed quickly enough, some of the reactions that are otherwise considered irreversible (and would be damaging to muscle tissue) can be reversed by "recapturing" the reaction products before they migrate away from the electrode surface. Those electrochemical processes that are not reversed, and account for the net charge imbalance,

TABLE 5. Possible charge transfer processes at metal electrodes during neurostimulation. Most reactions (2C, 3C, 5C, 6C, 7C, 2A, 3A, 4A, 7A, and 8A) produce H^+ and OH^- in direct proportion to the amount of electrons consumed or produced. Reaction 5A (metal corrosion) is unlikely for the charge densities used in these experiments. Reactions 1A and 1C are reversed during the end of each stimulation phase. The likelihood of reactions 4C, 6A, and 9A taking place is unknown. The E_0 values listed are steady-state electrode potentials; because of kinetic factors, the measured electrode potential at which a reaction occurs during stimulation may differ from the steady-state value (from Ref. 10).

Reaction		E_0 (pH = 7.0) (mV vs. SHE)
Cathodic Processes (negative pulse)		
1C	Double-layer charging	–
2C	$2MO + H_2O + 2e^- = M_2O + 2OH^-$	[a]
3C	$M_xO_y + yH_2O + 2ye^- = xM + 2yOH^-$	[a]
4C	$O_2 + e^- = O_2\cdot^-$	−0.29
5C	$O_2 + 2H_2O + 2e^- = H_2O_2 + 2OH^-$	0.27
6C	$O_2 + 2H_2O + 4e^- = 4OH^-$	0.82
7C	$2H_2O + 2e^- = H_2 + 2OH^-$	−0.41
Anodic Processes (positive pulse)		
1A	Double-layer charging	–
2A	$H_2 + 2OH^- = 2H_2O + 2e^-$	−0.41
3A	$M + yH_2O = M_xO_y + 2yH^+ + 2ye^-$	[a]
4A	$M_2O + 2OH^- = 2MO + H_2O + 2e^-$	[a]
5A	$M = M^{n+} + ne^-$	[a]
6A	$O_2\cdot^- = O_2 + e^-$	−0.28
7A	$H_2O_2 + 2OH^- = O_2 + 2H_2O + 2e^-$	0.27
8A	$2H_2O = O_2 + 4H^+ + 4e^-$	0.82
9A	$2Cl^- = Cl_2 + 2e^-$	1.4

[a]Values depend on the specific metal and oxide or hydroxide species involved.

are not as damaging to muscle tissue. The exact sequence of the electrochemical reactions that cause the tissue damage we have observed is not known. However, oxidation of inorganic and organic species appears to be a likely candidate. Reactions that may be responsible for the tissue damage are, the formation of hydrogen peroxide,

$$O_2 + H_2O + 2e^- \Rightarrow O_2H^- + OH^-$$

or

$$2OH^- + O_2 \Rightarrow 2O_2H^-$$

and

$$O_2H^- + H^+ \Rightarrow H_2O_2$$

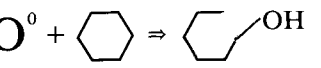
which is very toxic and can react easily with organic species,

$$H_2O_2 + \text{organic} \Rightarrow H_2O + \text{oxidized organic}$$

and the formation of nascent oxygen,

$$2O_2H^- \Rightarrow 2OH^- + 2O^0$$

which may also react with organic species.



Further studies must be carried out to understand the reactions so that insight may be gained into the mechanisms of muscle tissue damage resulting from electrical stimulation.

An important consequence of the results of this study is the possibility of reducing the area or size of the stimulating tip of the electrode. At present, the electrodes used for intramuscular stimulation typically support cathodic charge densities (the excitatory phase of the stimulus) of no more than 0.2 $\mu C/mm^2$ (20 mA, 100 μs, 10 mm^2). The results of this study indicate that the excitatory phase of the pulse can be as great as 1.2 $\mu C/mm^2$ without tissue damage or electrode damage with imbalanced biphasic electrical stimulation. This means that the electrode surface area may be reduced by a factor of 6. A reduction of the electrode area by a factor of 6 may make the tip less likely to break and provide a more localized stimulus to the tissue. It must be noted that the above studies have not considered the effects of a net anodic charge imbalance on the indifferent electrode and that any applications using imbalanced biphasic stimulation must take this into consideration.

REFERENCES

1. Beck, T.R.; Ruggeri, R.T.; Ballestrasse, C.L. Calculations of temperature rise produced in body tissue by a spherical electrode. Ann. Biomed. Eng. 13:177–194; 1985.
2. Box, G.E.P.; Hunter, W.G.; Hunter, J.S. Statistics for experimenters—an introduction to design, data analysis, and model building. New York: Wiley, 1978.
3. Brummer, S.B.; Robblee, L.S.; Hambrecht, F.T. Criteria for selecting electrodes for electrical stimulation: Theoretical and practical considerations. Ann. N.Y. Acad. Sci. 405:159–171; 1983.
4. Caldwell, C.W.; Reswick, J.B. A percutaneous wire electrode for chronic research use. IEEE Trans. Biomed. Eng. 22:429–432; 1975.
5. Ferguson, G.A. Statistical analysis in psychology & education. New York: McGraw-Hill, 1976:p. 295.
6. Jones, L.J.; Proskauer, C.C.; Paull, W.K.; Lepeschkin, E.; Jones, R.E. Ultrastructural injury to chick myocardial cells in vitro following "electric countershock." Circ. Res. 46:387–394; 1980.
7. Jones, J.L.; Jones, R.E. Decreased defibrillator-induced dysfunction with biphasic rectangular waveforms. Am. J. Phys. H792–H796; 1984.
8. Mchardy, ; Geller, J.D.; Brummer, S.B. An approach to corrosion control during electrical stimulation. Ann. Biomed. Eng. 5:155–149; 1977.
9. Mortimer, J.T.; Kaufmann, D.; Roessmann, U. Intramuscular electrical stimulation: tissue damage. Ann. Biomed. Eng. 8:235–244; 1980.
10. Contract Proposal #NIH-NINCDS-87-01, Electrodes for neuromuscular stimulation. Applied Neural Control Lab., Case Western Reserve University, July 1, 1987.