Retinal Effects of Ruby Laser Photocoagulation

Microelectrode Recordings in the Rabbit

Richard D. Banyard, MD; Kimiharu S. Noyori, MD;
Charles J. Campbell, MD, New York; and Elwin Marg, PhD, Berkeley, Calif

Using tungsten microelectrodes, electrical potentials were recorded directly from the rabbit retina. These responses were not obliterated by isolated clinical photocoagulations of grade 1 to 2 intensity, using the ruby laser.

THE RUBY laser, since its introduction a decade ago, has been used successfully in the treatment of various pathologic ocular conditions. Coincident with these clinical applications has come research into its fundamental effects on normal ocular structures, especially the retina. Histological,[1,2] biochemical,[3] and thermal[4] alterations have been studied. To our knowledge there have been no reports as yet demonstrating the electrophysiological effects of ruby laser lesions at the site of injury.

Materials and Methods

Equipment (Fig 1).—Modified clear plastic contact lenses were fitted with hollow plastic cylinders. The diameter of the hole within the cylinder was made large enough to accept a standard 30-gauge needle easily. Several sized lenses were found necessary to conform to varying globe diameters between large and small animals.

Standard 30-gauge needles were cut to lengths of 18 mm, preserving the sharp beveled end.

Insulated tungsten microelectrodes, detailed previously by Marg[5] and Marg and Adams,[6] were made to measure approximately 10 cm in length. These electrodes tapered to a 1μ tip, were rugged, could be used repeatedly, and had the added advantage of being sterilizable by any method without affecting the tip.

A light weight plastic micromanipulator (E. Marg et al, unpublished data) was used to advance the electrode. This device had a detachable clamp for spreading the apparatus in coupling to the contact lens. It also had a drive wheel for fine up and down adjustments of the microelectrode.

Recordings were monitored visually and audibly with a dual, alternating current preamplifier (Grass P 9) a power pack with audiomonitor (Grass model 109/AM4-A), and an oscilloscope (Tektronix 502).

Permanent records were made with a polaroid attachment on the oscilloscope.

Photocoagulations were made throughout the A-O ruby laser photocoagulator without modification, described fully elsewhere.[7]

Procedure (Fig 2 and 3).—Gray chinchilla rabbits weighing between 3 and 5 kg were anesthetized by intravenous injection of pentobarbital sodium. Pupils were dilated maximally with solutions of cyclopentolate 2% and phenylephrine 10%.

The modified scleral lens was placed between the lids and over the cornea of the animal. A precut 30-gauge needle held at the blunt end with a jewelers pin vise was inserted through the hollow cylinder of the plastic lens and then used to penetrate the eye by a twisting motion through sclera and choroid into the vitreous cavity at a position corresponding to the ora serrata. The microelectrode was then inserted through the needle until it could be seen ophthalmoscopically emerging into the vitreous. The micromanipulator was spread and fastened onto the cylinder of the contact lens and around the electrode so that its drive wheels were engaged. Connection was then made from the monitoring system to the electrode. Record-

---

Submitted for publication May 26, 1970.

From the Department of Ophthalmology, Columbia-Presbyterian Medical Center, College of Physicians and Surgeons, New York.

Reprint requests to School of Optometry, University of California, Berkeley, Calif 94720 (Dr. Marg).

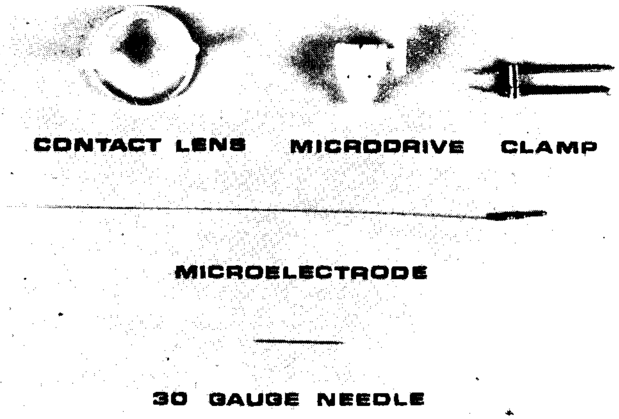
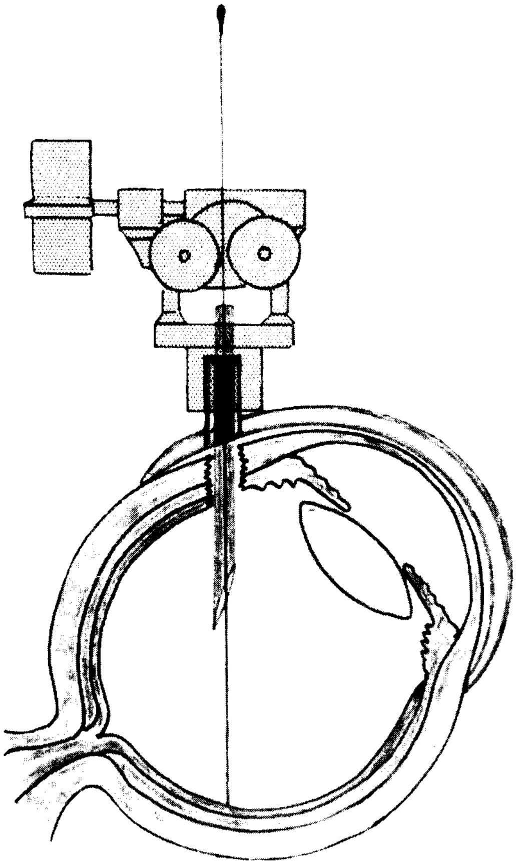


Fig 1.—Components of microelectrode-microdrive-contact lens system.

ings were made while the microelectrode was advanced toward and into the retina. Using these methods, the electrode tip generally lay in the retina at a position midway between the disc and the equator.

Fig 2.—Schematic view of microelectrode system assembled on an eye (not to scale).



All experiments were conducted in a semidarkened room (dark with background lights from instruments). No attempts at dark adaptation of the animals were made. Spike potentials or unit activity from ganglion cells were elicited both spontaneously and with added light stimulation by moving a standard, battery-operated pocket flashlight in the animal's visual field (approximately four inches above the eye). Spikes were similarly obtained by arming the one-degree focusing spot in the laser viewing system on and off the tip of the electrode in the retina. It could thus be demonstrated that maximal responses occurred at the electrode with fall off as the light was moved away. The size of individual receptive fields was estimated in several pilot experiments using targets of varying sizes but was not incorporated in our laser studies. Barlow et al[8] have shown in rabbits that these units vary from 2° to 12° in diameter.

Preparations made in the foregoing manner were generally viable for some time and enabled us to work conveniently on the second phase of our experiments. This involved producing observable photocoagulations in the retina at the site of the recording electrode tip. Only the field stop setting on the laser corresponding to 2.5° burn size was employed. The laser was aimed through the contact lens and centered directly at the site of the microelectrode in the retina. Recordings were taken before, during, and after photocoagulation. Power

Fig 3.—Microelectrode system in place on a rabbit eye.

settings on the laser were always initially set below expected clinical threshold and increased slightly until a minimal observable lesion was produced. Burns produced by this method were in the range of grade 1 to grade 2, coinciding with previously published criteria.[1] Grade 1 lesions were frequently barely perceptable and were of smaller size than the field stop setting. The burn produced was gray with a faint central and peripheral accumulation of dark pigment. Grade 2 photocoagulations had more definite pigment disturbances and a faint gray corona. A small subretinal gas bubble was often present.

Lesions of greater than grade 2 intensity were also produced and the results monitored. In these treatments pigment changes were marked, they tended to be larger than the field stop setting, and a vitreous gas bubble or preretinal hemorrhage was found.

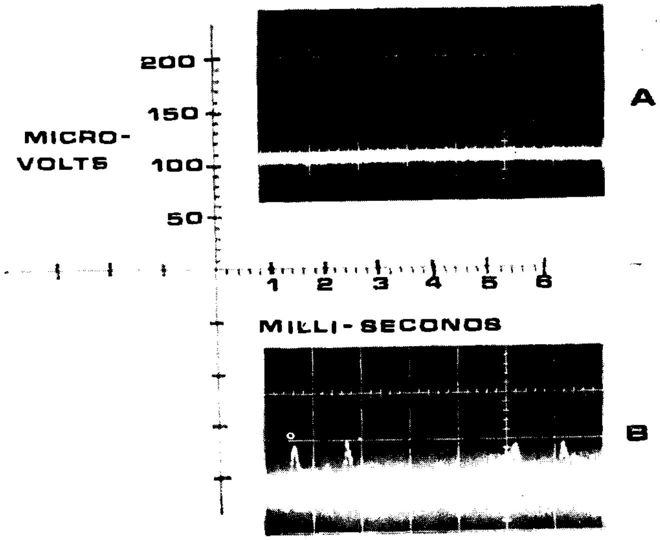


Fig 4.—Recording from rabbit retina. **A**, Baseline; **B**, spontaneous unit potentials prior to photocoagulation.

Fig 5.—Unit potentials obtained after photocoagulation of grade 1 to 2 intensity.

Results

Figure 4 shows recordings typical of those obtained both before and after microelectrode monitoring of unit receptive field activity (Fig 4, *A* is baseline). Characteristically, records of spontaneous responses consisted of waves of varying frequency with amplitudes of about 50μv. Light-induced responses had typically the same configuration but occurred in greater frequency.

Two types of responses were encountered following photocoagulations of grade 1 to 2 intensity.

1. In the majority of cases, solitary grade 1 or 2 lesions produced no detectable change in the unit responses. Both spontaneous and induced spikes appeared to be of the same amplitude, frequency, and duration as before treatment (Fig 5).

2. In several experiments our recorded responses fell silent following grade 1 to 2 photocoagulations and did not resume activity. This phenomenon was encountered when the lesions produced were more clearly grade 2 than grade 1 in intensity. We found, however, in each instance that by moving the microelectrode slightly up or down within the area of the burn that spikes were again elicited. Spontaneous and induced responses were recorded for up to an hour after photocoagulation. In several experiments the electrode was moved up and away from the original recording site and then repositioned in a different sector of the grade 1 to 2 burn. In each instance we could demonstrate preservation of unit activity irrespective of whether the electrode was central or in a peripheral portion of the lesion.

Following photocoagulations of greater intensity than grade 2, no unit spikes were either spontaneously produced or could be induced by light within the area of treatment. This observation held true 100% of the time despite efforts in searching for a viable unit (Fig 4, *A*).

Comment

Much interest has been generated recently about the retinal effects of lasers. Clinical studies with the pulsed ruby device have demonstrated its usefulness in the management of various pathological conditions. For example, coagulations which subtend an angle of 2.5° have been used in the management of retinal breaks, peripheral degenerations,[9] and diabetic retinopathy.[10]

Our understanding of the effects of the pulsed ruby laser which make it a safe and formidable clinical tool has come from basic research along several lines. As a result of these studies observable changes produced in the retina by laser photocoagulation have been separated into alterations produced by thermal and mechanical effects.

Thermocouple measurements[4] of ruby laser burns in the rabbit indicate that the temperature change in the plane of the retina is narrowly delimited and suggest that widespread destructive thermal injury does not occur.

Microscopic studies[1-3] have shown that controlled low energy coagulations cause changes limited to the external retinal segments with preservation of the nerve fiber and ganglion cell layers. Burns of greater intensity show marked changes in all layers.

Using radioautographic techniques, Marshall[3] suggests that thermal changes induced by ruby lasers are generally confined to the outer retinal layers. By comparison, he has demonstrated mechanical alterations in the inner segments dependent on beam size and energy output. Photocoagulations of greater than 300μ and relatively low intensity were found to create little mechanical effect.

Our results tend to support those earlier observations of limited changes in the retina by demonstrating intact electrical responses within photocoagulations of therapeutic size and intensity. Laser treatments greater than grade 2 which have been shown to cause widespread histological damage[1] obliterated unit activity in our experiments.

Since with our techniques it was impossible to reposition the microelectrode back to an original treatment site once the apparatus had been withdrawn from the eye, we do not know if these results hold true over long periods.

The characteristics of electrical potentials obtained with microelectrodes implanted in retinas have been outlined previously.[9,11] Individual variations in receptive field size and type were not specifically incorporated in our studies for the sake of uniformity and simplicity. Our recordings were graded purely on an all or none basis with regard to the effect of photocoagulation. Presumably with refined recording techniques it might be possible to demonstrate subtle changes in unit potentials with increasing intensities of laser burns.

References

1. Noyori KS, Campbell DJ, Rittler C, et al: The characteristics of experimental laser coagulations of the retina. *Arch Ophthal* **72**:254-263, 1964.
2. Zweng CH, Little HL, Peabody RR: *Laser Photocoagulation and Retinal Angiography.* St. Louis, CV Mosby Co, 1969.
3. Marshall J: Thermal and mechanical mechanisms in laser damage to the retina. *Invest Ophthal* **9**:97-115, 1970.
4. Campbell DJ, Noyori KS, Rittler C, et al: Intraocular temperature changes produced by laser coagulation. *Acta Ophthal* **76**:22-31, 1963.
5. Marg E: A rugged, reliable and sterilizable microelectrode for recording single units from the brain. *Nature* **202**:601-603, 1964.
6. Marg E, Adams JE: Indwelling multiple microelectrodes in the brain. *Electroenceph Clin Neurophysiol* **23**:277-280, 1967.
7. Campbell CJ, Rittler C, Koester CJ: Photocoagulation of the retina, in *International Ophthalmology Clinics.* Boston, Little Brown & Co, vol 6, 1966, p 2.
8. Barlow HB, Hill RM, Levick WR: Retinal ganglion cells responding selectively to direction and speed of image motion in the rabbit. *J Physiol* **173**:377-407, 1964.
9. Campbell CM, Koester CJ, Curtice V, et al: Clinical studies in laser photocoagulation. *Arch Ophthal* **74**:57-65, 1965.
10. Beetham WP, Aiello LM, Balodimos MC, et al: Ruby laser photocoagulation of early diabetic neovascular retinopathy. *Arch Ophthal* **83**:261-272, 1970.
11. Kuffler SW: Discharge patterns and functional organization of mammalian retina. *J Neurophysiol* **16**:37-68, 1953.