Anterior chamber measurements on CO\textsubscript{2} laser corneal irradiation

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At threshold irradiation (~ 0.1 w./cm\textsuperscript{2}) of the rabbit cornea with a carbon dioxide (CO\textsubscript{2}) laser, there was no detectable rise in intraocular pressure and only a slight (1 to 2° C.) rise in aqueous temperature. On suprathreshold CO\textsubscript{2} corneal irradiation, the intraocular pressure rose to a plateau during irradiation and then gradually fell. There was a concomitant rise in temperature in the anterior chamber caused by heat conduction from the site of corneal irradiation. Further rise in the temperature of the corneal stroma to the critical temperature range for heat shrinkage of collagen causes the cornea to thicken. This thickening of the cornea helps to bring heated aqueous under the irradiated site more closely in apposition to the lens and iris, thus further increasing the temperature of the anterior lens. Lens indentation was observed in fixed eyes only following pulses of sufficient power and duration to raise the temperature near the surface of the anterior lens to a range of at least 60 to 70° C. Increase in aqueous protein occurred only with irradiation over regions of the iris diaphragm.

Key words: carbon dioxide laser, cornea, radiation effects, intraocular pressure, heat, aqueous humor, anterior chamber, corneal thickness, injuries of lens, proteins, pressure.
Measurements on CO₂ laser corneal irradiation

is about 0.1 watt per square centimeter (w./cm.²).

It has previously been reported that not only can corneal opacifications and occasional calcification (i.e., band keratopathy) be produced at suprathreshold levels of irradiation, but also that at these higher power densities lens alterations can result without corneal perforation. This study was carried out primarily to determine some physical aspects of the latter mechanism. Intraocular pressure and temperature alterations during and following CO₂ irradiation of the cornea were therefore measured. In addition, protein levels in the anterior chamber aqueous following irradiation of the cornea at various locations were also determined.

Materials and methods

A 20 watt CO₂ laser was used for this study. An adjustable pulse length was obtained by two electronically controlled shutters placed within the laser cavity between the NaCl Brewster angle windows and the end mirrors. At power levels between 0.1 and 0.35 w./cm.², a beam selector was used to provide a cross-sectional irradiation area of ~0.86 cm.². Higher power densities were obtained by focusing with an Irtran II lens or a gold-coated focusing cone. Output levels to 250 mw. were measured with a calibrated Eppley thermopile and at power levels to 10 w., with a Coherent Radiation Laboratory Model 201 detector. Over 200 albino rabbits were used in this study.* Each was systemically anesthetized with Innovar Vet (Sublimaze [fentanyl] 0.4 mg. per milliliter and with Inapsine [droperidol] 20 mg. per milliliter, McNeill) topically with Ophthetic drops (Proparacaine HC1, Allergan). Pupils were dilated when necessary with 2 per cent atropine sulfate or 1 per cent Mydriacyl (Bis-Tropicamide-Alcon). For intraocular pressure measurements, a 22 gauge hypodermic needle was sealed into the anterior chamber with Eastman 910 adhesive. The needle was connected through a small section of plastic tubing by a glass system to a Statham P23AA pressure transducer. The system was filled with heparinized normal saline. Intraocular pressure was adjusted to 20 mm. Hg prior to irradiation. Calibration was by means of a water manometer.

The temperature rise was measured by a Baldwin Lima Hamilton TC-RC-IT-200 chromel constantan thermocouple similarly sealed into the anterior chamber.

The pulse durations of the irradiation as well as the outputs from the pressure transducer and thermocouple were recorded on a Gilson model M8PM polygraph.

The aqueous humor was sampled, with a Hamilton 50 microliter syringe, from normal eyes and at intervals following irradiation from eyes that were not used for pressure and temperature determinations (i.e., from intact eyes). Care was taken during sampling to avoid touching the iris. Samples were diluted with 4 ml. of normal saline, and the protein level was determined by the UV method of Waddell in a Beckman DU spectrophotometer.* Bovine serum albumin standards were run with each determination.

Results

No pressure rise was detected in the anterior chamber on 10 minute irradiation of the entire cornea at 0.1 w./cm.². The temperature in the anterior chamber rose 4° C. after 3 minutes, plateauing at this level on irradiation at this power density. As the power density was increased, the aqueous temperature increased. At 0.15 w./cm.² the temperature increased, plateauing at approximately 5° C. and increased no more after 3 minutes of irradiation. At 0.35 w./cm.² there was a gradual increase of pressure, plateauing at 40 mm. Hg in 18 minutes with a temperature rise of about 12° C., which remained constant after 6 minutes. At higher power levels of irradiation, the maximum pressure elevation reached in the anterior chamber with unperforated corneas depended, within limits, upon the duration of the pulse. The effects of CO₂ corneal irradiation on intraocular pressure and temperature are shown in the sample recordings in Fig. 1.

In Fig. 1, A the entire cornea was irradiated at 6 w. for 1.5 seconds with a beam focused through the cone at the center of the anterior corneal surface. The pressure in the anterior chamber rose steadily from 20 to 79 mm. Hg at the end of the pulse. Following cessation of the pulse, the pressure fell to 40 mm. Hg in 10 seconds, then fell gradually to 30 mm.

*In conducting the research described in this report, the investigators adhered to the "Guide for Laboratory Animal Facilities and Care," as promulgated by the Committee on the Guide for Laboratory Animal Facilities and Care of the Institute of Laboratory Animal Resources, National Academy of Sciences-National Research Council.
Hg during the next 30 minutes. In the living eye, respiratory and arterial pulses were always readily observed. For temperature measurements, the thermocouple was placed in close approximation to the center of the anterior surface of the lens (i.e., in front of the pupil) along the axis of corneal irradiation. The temperature rose to 73° C. at the end of the pulse and returned to base-line levels (~ 35° C.) in several minutes. Peripheral displacement of the thermocouple resulted in a lower delayed temperature rise.

When irradiation was carried out at approximately the same power levels for 0.75 seconds (Fig. 1, B), the pressure and temperature elevations were correspondingly less—to 66 mm. Hg and 49° C.

Changes in pressure and temperature were similar on irradiation of eyes in dead animals (Fig. 1, C), except for loss of the respiratory and vascular pulses. One
AQUEOUS HUMOR PROTEIN LEVEL (mg./100 cc.)
FOLLOWING CO₂ LASER IRRADIATION

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Fig. 2. Protein elevation in anterior aqueous chamber following CO₂ laser irradiation of the cornea. The letters A to D are keyed to the sites of irradiation.

minute following the peak pressure, the per cent increase of peak pressure in live eyes was significantly higher than in eyes of dead animals (29.6 per cent ± 2.2 S.E.M., compared with 18.9 per cent ± 2.1 S.E.M.).

In all eyes in which the corneas were perforated, the pressure dropped rapidly to zero.

When eyes were subsequently fixed in 3 per cent glutaraldehyde and opened, indentations of the lens surface were found only following laser pulses that raised the temperature near the lens surface to 60 to 70°C.

Indentations were never found in lenses from eyes that had been similarly fixed but not irradiated, or from eyes irradiated for shorter periods of time (i.e., with pulses of power and time insufficient to raise the underlying aqueous humor to a temperature greater than 60°C.). When whole fixed lenses were stained by immersion in 1 per cent acid orcein, the entire lens tissue beneath the capsule was colored maroon, except for that immediately beneath the deformed area.

A comparison of aqueous protein levels in normal and irradiated eyes is shown in Fig. 2. The irradiated eyes were exposed through the focusing cone at 6 watts for 1.5 seconds. Each eye was sampled only once. The protein levels obtained in the control eyes (60 mg. per 100 c.c.) were similar to that reported by others (50 mg. per 100 c.c.). The height and duration of protein elevation depended on the irradiation site (A to D). It was greatest 24 hours after irradiation of the cornea over the midperiphery of the iris, reaching a level of 636 mg. per 100 c.c.

Discussion

The temperature elevations obtained at the lens surface in this study were not due to possible direct irradiation of the thermocouple. If the thermocouple, with a time constant of 8 msec. (260 msec. in hot air, 8 msec. in boiling water), were directly irradiated, the temperature curves
would be peaked rather than flat over a short period of time, as they are in the sample curves in Fig. 1. This plateau is compatible with heat conduction to this region. In addition, there was a time delay between irradiation of the cornea and the onset of pressure and temperature rise (Fig. 1). On direct irradiation of the thermocouple, the rise in temperature would start immediately. The onset of increase of pressure in the aqueous preceded slightly the onset of temperature rise at the lens surface. This could be due to the time necessary for heat conduction to the thermocouple and also possibly to the corneal flattening. As the thermocouple was placed more peripherally, there was an increase in the time necessary for heat conduction to the thermocouple. This effect has been previously reported. 14 Other studies indicate that the absorption coefficient for biologic tissue at 10.6 μ is in excess of 150 cm⁻¹. At the power-density levels used, even if the thermocouple tip were only 2 mm. posterior to the irradiation site, the direct irradiation of the thermocouple should therefore have been far less than 1 μw/cm². Consequently, the temperature measured by the thermocouple accurately reflected the temperature at that site. These considerations should be contrasted with the use of thermocouples in measurement of elevation in the temperature of tissue upon irradiation with a ruby laser, with which inaccurate readings may often be obtained because of direct irradiation of the thermocouple. 16, 17

The indentation of the lens that was observed subsequent to corneal irradiation was probably due to a combination of temperature at the lens surface in excess of 60°C, together with a rise of pressure in the anterior chamber. The pressure increase observed in these experiments is apparently caused by concomitant heating of the aqueous and temporary alteration (flattening) of the anterior chamber. This lens indentation, occurring in nonpenetrated eyes, may be dependent on collagen shrinkage in the corneal stroma with or without corneal swelling 1. 5 resulting in cornea-lens approximation which may be augmented by possible collagen shrinkage (i.e., microfilaments) in the anterior lens capsule.

In studies discussed by Gustavson, 12 mammalian collagen in contact with water at 60 to 70°C, contracts sharply to about one third to one fourth of its initial length. Under tension these fibers return to their original length. Consequently, above 60°C, there may have been some shrinkage of the collagen in the cornea. The alteration in stress patterns in the cornea caused by collagen shrinkage would result in an increase in corneal radius. Gustavson notes that denatured collagen absorbs more water than native collagen. The thickening of portions of the irradiated corneas reported previously is due to foci of denaturation of the collagen in the cornea, which, together with the addition of water, appears to explain the swelling.

Preliminary experiments using hot water applications to isolated fresh whole lenses, with and without their restraining capsules, indicates that the lens alterations are produced deep to the capsule (i.e., to the superficial cortical cells).

As expected, similar elevations in pressure and temperature, together with subsequent lens deformation upon fixation, were observed when a heated glass rod was applied to the corneal surface without penetration into the eye.

The aqueous protein level increased up to 10 times normal following CO₂ irradiation of the cornea over the midperiphery of the iris. Similar irradiation of the center of the well-dilated eye did not result in observable protein elevation. These findings indicated a production of a form of traumatic (i.e., thermal) iritis.

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REFERENCES


