Effect of catecholamines in normal and hypothyroid mice

| Treatment | Kidney weight (mg) | Total DNA (μg) | DNA specific activity [a] | Difference (%) |
|---|---|---|---|---|
| a) Control (10) | 398.6 ± 15.2 | 1419 ± 125 | 9710 ± 611 | |
| $^{131}$I (12) | 348.7 ± 9.6 | 1164 ± 98 | 7340 ± 721 | |
| b) Control (12) (6 mg isoproterenol) | 410.8 ± 18.3 | 1390 ± 142 | 23.060 ± 2.540 | 137 [c] |
| $^{131}$I (14) (6 mg isoproterenol) | 391 ± 12.5 | 1200 ± 153 | 12.510 ± 2.431 | 70 [d] |
| $^{131}$I (14) (6 mg isoproterenol + $T_3$) [b] | 429 ± 18.9 | 1681 ± 203 | 17.710 ± 2.101 | 141 [d] |

[a] dpm/mg DNA ± S.E. [b] $T_3$ injected for 1 week. [c] Compared to Control experiment a). [d] Compared to $^{131}$I experiment b). Number of animals in parenthesis.

tion of adenyl cyclase and a resultant decrease in cAMP in salivary glands of hypothyroid rats[10]. Therefore, in the present experiments, the effect of isoproterenol stimulated DNA synthesis in hypothyroid mice is consistent with the hypothesis that catecholamine receptors are intermediated by thyroid hormone increasing their sensitivity and that cAMP has a direct effect on isoproterenol-stimulated cell kidney proliferation.

*Resumen.* En el presente trabajo se estudia el efecto del hipotiroidismo sobre la sintesis de DNA y proliferación celular renal estimuladas por isoproterenol. Se observa que la hormona tiroidea actúa como intermediario de las catecolaminas en el proceso de multiplicación celular. Se sugiere que la hormona tiroidea modula el efecto de catecolaminas aumentando la sensibilidad en el receptor. El aumento de la concentración de cAMP podría estar involucrado en este proceso.

O. L. Catanzaro and Adriana Marzi[11]

*Departamento de Ciencias Biologicas, Catedra de Fisiologia, Facultad de Farmacia y Bioquimica, Junin 956, Buenos Aires (Argentina), 9 April 1974.*

[10] O. L. Catanzaro, M. Flawia, A. E. Dominguez, B. E. Fernandez and N. A. Vidal, IRCS (Biochem; Dent; Endocr; Physiol) 2, 1203 (1974).

[11] This work was supported by a grant from the Consejo Nacional de Investigaciones Cientificas y Tecnicas, Rep. Argentina.

## Intra-Retinal Absorption of Argon Laser Irradiation in Human and Monkey Retinae

The argon laser is now used extensively in the treatment of a variety of retinal conditions. The small spot size, together with the output reliability of this instrument has allowed treatment of retinal disease adjacent to the macula[1-3]. It has been a consistent observation in this hospital, that argon induced lesions placed within one disc diameter (5°) of the macula show a different damage topography to those placed in more peripheral retina. Using retinal biomicroscopy and fundus photography, two retinal damage planes may be clearly discriminated. Such observations have not been reported for macular lesions produced by other longer wave length lasers[4-6].

We have exposed the maculae of 3 rhesus monkey eyes, and 1 human eye (prior to enucleation for a malignant melanoma of the anterior uvea without posterior retinal detachment) to a series of irradiations from an argon laser. Exposures were placed in groups of 4 within 1°, 2°, 3° and 10° of visual angle from the fovea. In each group lesions were produced by power levels of 50, 100, 200 and 300 mW. A single foveal exposure of 100 mW was given in each eye. All exposures had a 50 μm spot size and a pulse duration of 0.05 sec. All above parameters were as recorded on manufacturers instrumentation. The system used was a Coherent Radiation 800 and exposures were delivered via the integral Zeiss slit lamp system in conjunction with a Goldmann fundus contact lens.

All eyes were removed within a few hours of exposure, and were processed for both light and electron microscopy[7]. Serial sections were cut of each lesion.

Histological preparations confirmed the ophthalmoscopic observations, and showed 2 discreet damage planes, one situated at the pigment epithelium and involving the overlying receptor cells, and a second site in the inner retinal layers (Figures 1 and 2). The degree of damage to the inner retinal layers was inversely related to the distance of the lesion from the fovea. The depth of the plane of this damage also varied with distance from the fovea. At the fovea the damage was situated in the fibre layer of Henle (Figure 3), whilst at distances of 2 and 3 degrees of visual angle, damage was found in the inner nuclear and inner plexiform layers respectively.

We have only examined 1 human eye, but for a given exposure within a 1 degree field of the fovea, greater retinal disturbance was seen in the human eye than in the monkey eyes. (Figures 3a and b).

Both ruby[6] and helium neon irradiations of the macula show a conventional damage distribution centred on the pigment epithelium[7]. Though damage to the inner retina

[1] J. D. M. Gass, Trans. Am. Acad. Ophthal. Otolaryng. *75*, 580 (1971).
[2] H. Schatz and A. Patz, Arch. Ophthal. *90*, 183 (1973).
[3] A. C. Bird, Br. J. Ophthal., in press (1974).
[4] W. T. Ham, W. J. Geeraets, H. A. Mueller, R. C. Williams, A. M. Clarke and S. F. Cleary, Arch. Ophthal. *84*, 797 (1970).
[5] P. W. Lappin and P. S. Coogan, Arch. Ophthal. *84*, 350 (1970).
[6] C. J. Blair and D. M. Gass, Arch. Ophthal. *88*, 167 (1972).
[7] J. Marshall, Invest. Ophthal. *9*, 97 (1970).

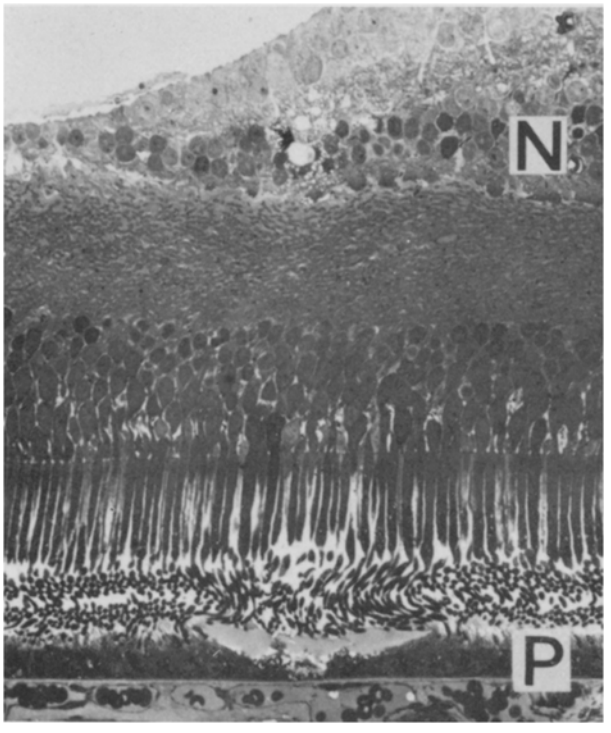


Fig. 1. Light micrograph of monkey retina showing a 50 mW Argon laser lesion within a 1° field of the fovea. Isolated damage sites can be seen in both the pigment epithelium (P) and the inner nuclear layer (N). ×600.

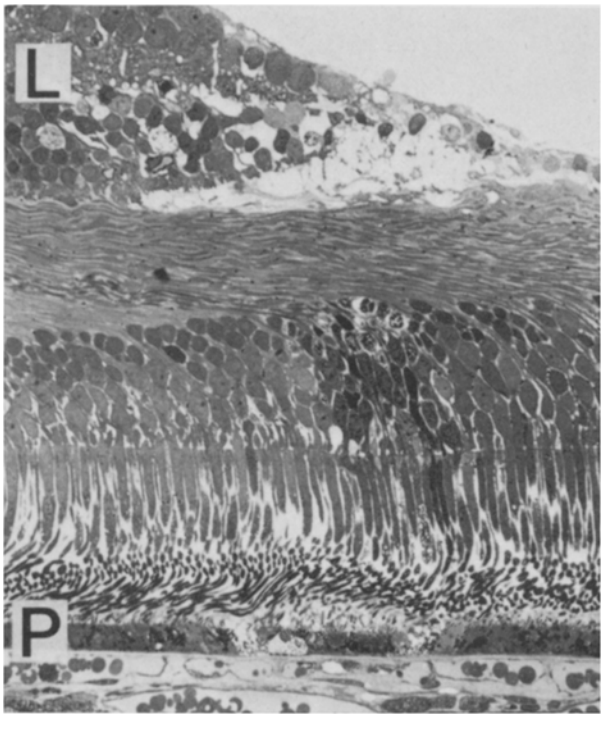


Fig. 2. Light micrograph of monkey retina showing 100 mW Argon laser lesion within a 1° field of the fovea. Note the increase in damage to the inner nuclear (N) and inner plexiform layers (L). ×600.

has been described following a single neodynium exposure[8], observations by other workers[7,9] and figures for retinal transmission[10] would suggest that this damage arose from factors other than intra retinal absorption. The isolation of the damage planes observed in our figures suggests that argon laser light is being absorbed both within the neural retina and at the pigment epithelium. Differential thermal tolerances of inner retinal components to a heat front eminating from the pigment epithelium is not a tenable theory, as the depth of the damage plane varies with the site in the fundus in such a way, that with increase in retinal thickness the damage appears further from the pigment epithelium.

If it is accepted that the site of primary damage to a biological system is coincident or adjacent to the site of absorption of the incident radiation, then from our figures we must postulate the existence of a pigmented system in the neural retina which is localized around the macula, and which absorbs more radiation at 488 nm than at 633 or 694.3 nm. The macular pigment has these characteristics[11] and at the fovea would absorb approximately 70% of radiation at 488 nm. The concentration of this pigment decreases with distance from the fovea[11]. The Table shows the calculated relative absorption of argon laser irradiation by macular pigment[11], and the pigment epithelium[10] for representative exposures in the present study. It should, however, be stated that macular pigmentation varies both between individuals[12], races[13] and species.

The degree of retinal damage is a complex function of beam tissue interaction[14], but at the power levels used in the present study it may be related to the degree of tissue heating. Such heating is dependant on the absorbed energy density which is a function of the thickness of the absorbing layer. For this reason, it would be useful to identify the cell population which contains the macular pigment. At the fovea the retina is formed exclusively from the components of receptor cells and glia. Figure 3 shows damage centred on the inner aspect of the receptor cells, but further from the fovea damage occurs in the inner nuclear (Figures 1 and 2) and plexiform layers. From this distribution it would seem that either pigment is present in each of these retinal layers, or that the pigment is contained within a particular cell type which occurs in each of these cell layers, namely the Muller fibres.

A major specialization of the foveal cones is the development of long inner receptor fibres forming the fibre layer of Henle. If this layer contained a dichroic pigment it would be ideally suited to produce the entoptic phenomenon of Haidinger's Brushes[15]. Diffusely distributed pigmentation in other neuronal layers as well as the fibre layer of Henle would not significantly interfere with the production of this entoptic phenomenon, however, it could not be produced by pigmentation solely within the transversely oriented Muller's fibres. Whatever the pigment distribution, damage will occur in any layer when sufficient energy has been absorbed to produce tissue break down.

[8] M. L. WOLBARSHT, K. E. FLIGSTEN and J. R. HAYES, Science *150*, 1453 (1965).
[9] W. J. GEERAETS, Acta opthal. *45*, 846 (1967).
[10] W. J. GEERAETS and B. S. BERRY, Am. J. Ophthal. *66*, 15 (1968).
[11] K. M. RUDDOCK, Vision Res. *3*, 417 (1963).
[12] R. A. BONE and J. M. B. SPARROCK, Vision Res. *11*, 1057 (1971).
[13] I. G. H. ISHAK, J. opt. Soc. Am. *42*, 529 (1952).
[14] J. MARSHALL and J. MELLERIO, Br. med. Bull. *26*, 156 (1970).
[15] G. S. BRINDLEY, *Physiology of the Retina and Visual Pathway* (Arnold, London 1970), p. 141.

Differential absorption of Argon laser radiation (488 nm) in human retinal tissue at various distances from the centre of the fovea

| Exposure (incident power mW) | Calculated absorbed power mW | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | Neural retina (Macular pigment) | | | | Pigment epithelium (Melanin) | | | |
| | 1° | 2° | 3° | 10° | 1° | 2° | 3° | 10° |
| 50 | 35 | 18 | 4 | < 1 | 11 | 22 | 32 | 35 |
| 100 | 70 | 35 | 8 | < 1 | 21 | 46 | 64 | 70 |
| 200 | 141 | 70 | 17 | < 1 | 41 | 91 | 128 | 141 |
| 300 | 211 | 106 | 25 | < 1 | 62 | 136 | 193 | 211 |

The primary damage site described in the present paper could account for the recently reported secondary degenerative changes in the inner retinal layers of monkeys 1 year after being exposed to an argon laser[16], as all these irradiations were within 3 disc diameters of the fovea.

A worrying feature of our observations is the sensitivity of the human macula. To relate the results of the monkey exposures to those in the human, one must consider the respective retinal flux densities. Under our experimental conditions any differences in flux densities can only be related to different transmission properties of the ocular media in the 2 species. Average figures from 28 human and 14 monkey eyes[10] show transmission properties to be identical at the argon wavelength, however the transmission of light through the lens decreases with increase in age[17]. Our human subject was 49, thus the total energy reaching the retina may well have been lower in the human exposures than those in the monkey. We appreciate that this is data from a single individual, and that our exposure energies are well above threshold levels, however, we feel this result should be considered when examining codes of practice for laser workers. Obviously we would like more data from the maculae of both primates and human retina, and from energy values nearer threshold before a detailed analysis of the dangers of neural retinal absorption can be defined. Most laser safety codes are based on experimental data obtained from monkey irradiations, and there is a factor of 10 between the recommended maximum permissible exposure and the level at which ophthalmoscopic damage occurs in these animals. In all codes throughout the visible region of the spectrum the absorption of laser light is assumed to take place in the melanin of the pigment epithelium, and absorbed energy densities have, therefore, been calculated solely on this basis. The pigmentation in the rhesus monkey eye is about twice as dense as that of a caucasian, thus for most laser irradiations, the monkey eye requires less energetic exposures to produce threshold damage. Hence when these threshold exposure energies determined empirically in monkey are applied to man there is a small additional safety factor. If the neural retinal damage we have shown in Figure 3 still occurs at threshold exposure levels, then for argon irradiation of the fovea this small additional safety factor would be removed, and may even be reversed.

These observations are also important to the therapeutic use of the argon laser. Inner retinal damage caused by argon laser photocoagulation in the parafoveal region may cause poor visual results due to foveal denervation.

We feel that the threshold for argon laser damage to the macula should be further investigated, and that some attempts should be made to determine the functional differences inherent in lesions produced in different retinal layers.

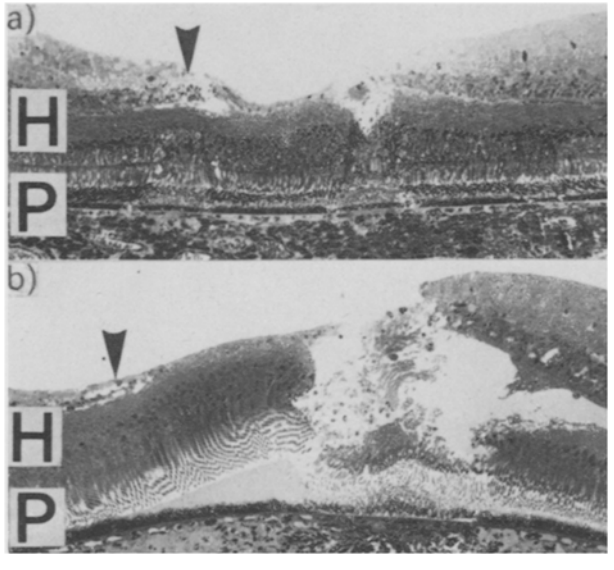


Fig. 3. Light micrographs of (a) monkey and (b) human retinae showing 100 mW Argon laser lesions at the fovea. Damage can be seen in both the pigment epithelium (P) and in the fibre layer of Henle (H). The detachment of the retina, and separation of the retinal layers is not a preparative artefact but arises due to the accumulation of exudates. More damage is seen in the human exposure than in the monkey. In both cases inner retinal damage induced by a second exposure can be seen arrowed. ×150.

*Zusammenfassung.* Die histopathologische Untersuchung von Argon Lasers Fotokoagulation der Macula bei Menschen und Affen konnte zwei Angriffsebenen an der Netzhaut zeigen: 1. Pigmentepithel und 2. innere retinale Schichten.

J. Marshall, A.M. Hamilton and A.C. Bird[18]

*Department of Visual Science and Department of Clinical Ophthalmology, Institute of Ophthalmology and Moorfields Eye Hospital, Judd Street, London WC1H 9 QS (England),*
*6 May 1974.*

[16] G. D. Frisch, P. D. Shawaluk and D. O. Adams, Nature, Lond. *248*, 433 (1974).

[17] R. A. Weale, Ciba Foundation Symp. *19*, 5 (1973).

[18] We would like to thank Mr. P. L. Ansell for technical assistance. This project was supported by the Medical Research Council and the Wellcome Trust.