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MELANIN GRANULE MODELS FOR PULSED LASER INDUCED RETINAL INJURY*

W. P. HANSEN AND S. FINE

Department of Biophysics and Biomedical Engineering
Northeastern University, Boston, Massachusetts

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Box #: 3
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Biologic Effects of Laser Radiation

INTRODUCTION

Experimental and theoretical studies of laser induced threshold retinal injury have been carried out by several groups (1,2,3,7). Models for threshold injury to the retina on exposure to laser or intense light sources where the duration of exposure exceeds approximately 1 millisecond, have been proposed (1,7). It is the purpose of this report to present models for retinal threshold injury on exposure to intense light pulses ranging from one second to 10^{-8} seconds in duration.

The retinal energy density required to produce minimal ophthalmoscopically visible lesions of the pigmented rabbit retina has been found to be 1.5×10^{-1} to 8.5×10^{-1} joules/cm^2 using conventionally pulsed ruby lasers (1,2). These investigators have reported threshold retinal energy densities of 4.5×10^{-3} to 7×10^{-2} joules/cm^2 using Q-switched ruby lasers. In studies by Ham et al. the retinal image diameter was 800 microns for both conventionally pulsed and Q-switched irradiation. In the studies by Kohtiao et al. the retinal image diameter was 250 microns. The histologic characteristics of the threshold lesions produced at Q-switched laser pulse durations (16 to 80nanosec) may differ from those produced by conventionally pulsed lasers (175 to 500 μsec). Microscopic examination of ophthalmoscopically visible threshold lesions produced by Q-switched ruby lasers often reveals marked disruption of the pigment epithelium (1,3). The ophthalmoscopically visible threshold lesions produced by conventionally pulsed ruby lasers appear as mild coagulations (retinal burns), and in the absence of laser "hot spots", are not usually accompanied by severe alteration of the architecture of the retinal pigment epithelium (1).

Ham and his associates have developed a model for threshold retinal injury which applies to both conventionally pulsed ruby lasers and pulsed white light sources at pulse durations ranging from 175 μ sec to several seconds (1,4). This model assumes that heat is generated uniformly within the irradiated volume of pigment epithelium and uniformly within the choroid.

Washington National Record Center
Office of the Army Surgeon General
Record Group 112
Accession #: 75-13
Box #: 3
File: 1304-14 Fine, Samuel MD 2436
Biologic Effects of Laser Radiation

Under these conditions, Ham et al., have calculated maximum temperatures at the retinal pigment epithelium that are below 100°C for threshold retinal injury (4). In these cases a retinal lesion is thought to be caused by thermal denaturation at a temperature at which steam is not produced (4).

The solid curve shown in Figure 1 on a semi-log plot is taken from Ham et al.(4). This curve is based on maximum retinal temperatures which have been calculated by using the model for uniform heat generation described above (4). Linear extrapolation of their curve to shorter pulse durations is shown by the dotted line. This extrapolation indicates that at a pulse duration of approximately 4μ sec, a temperature of 100°C would be required to produce a threshold lesion.

According to Figure 1, steam production must therefore be considered for pulse durations shorter than 4μ sec., if extrapolation of the curve is continued. Possible steam production in the retinal pigment epithelium is a complicated process which, for the purposes of this report, will be analyzed from the viewpoint of simple, classical models for nucleation of vapor bubbles in a liquid phase (5). In general, a liquid to vapor transformation begins with the appearance of very small vapor bubbles, or "nucleii", in the liquid which grow until the liquid is completely transformed to a vapor phase. If insoluble stable impurities are present in the liquid phase, nucleii tend to form at interfaces separating the liquid, vapor, and impurity regions. For a one component system, the liquid to vapor phase transformation occurs as a constant temperature process. In the case of water, where stable impurities are normally present, the maximum nucleation rate occurs at the normal boiling point (100°C at one atmosphere of pressure) (6). In the retinal pigment epithelium, which will be considered as salt water containing stable impurities, it is probable that nucleii will form at impurity sites such as melanin granules at the normal boiling temperature of salt water. If the intracellular hydrostatic pressure is assumed to be of the order of one atmosphere, nucleii will form in the retinal pigment epithelium when the temperature at the boundaries of the individual melanin granules or other "impurities" reaches about 100°C.

Washington National Record Center
Office of the Army Surgeon General
Record Group 112
Accession #: 75-13
Box #: 3
File: 1304-14 Fine, Samuel MD 2436
Biologic Effects of Laser Radiation

Vos (7) has qualitatively considered liquid to vapor transformation temperatures in the retinal pigment epithelium on retinal irradiation with intense light sources. He has suggested that the forces of surface tension in the retinal pigment epithelium are greater than those for water and consequently the nucleation temperature in the pigment epithelium can be higher than 100°C. If nucleation in the pigment epithelium does indeed occur at temperatures above 100°C, then referring to Figure 1, two mechanisms for injury are possible for laser pulse durations shorter than four microseconds. The first possible mechanism is that denaturation injury will occur above 100°C, but at a lower temperature than that necessary for boiling. The second is that boiling may occur before denaturation temperatures are reached. There is, of course, the additional possibility that the denaturation injury temperature and the boiling temperature coincide.

While estimates of temperatures for threshold thermal injury can be obtained from the solid curve in Figure 1, sufficient data is not available for one to estimate the temperature at which boiling occurs in the pigment epithelium. However, for the purposes of this report, two limiting cases will be examined. In the first case, it will be assumed that a liquid to vapor phase transformation will occur at a temperature of 100°C at the boundary of melanin granules with a latent heat of vaporization, L = 540 cal/gm, as would be the case for stable granules suspended in water at one atmosphere of pressure. Based upon the above conditions, the dotted curve in Figure 1 will be a 100°C isotherm for pulse durations shorter than four microseconds. The second limiting case will assume that nucleation does not occur and that the thermal denaturation injury temperature is always less than the boiling temperature for all pulse durations. In this second case, the solid curve would be linearly extrapolated for shorter pulse durations. A break point would therefore not occur at 4 μ sec.

The first limiting case will be referred to as a "two-mechanism" model for the formation of threshold retinal lesions inasmuch as thermal denaturation injury is considered for non-Q-switched laser pulse durations while mechanical injury from steam production at 100°C (8) is predominant for Q-switched laser pulse durations. The second limiting case will be referred to as a "single-mechanism" model for retinal injury since thermal denaturation is assumed to be the only injury mechanism

Washington National Record Center
Office of the Army Surgeon General
Record Group 112
Accession #: 75-13
Box #: 3
File: 1304-14 Fine, Samuel MD 2436
Biologic Effects of Laser Radiation

at all laser pulse durations. The main portion of this report will be devoted to a mathematical development of the "two-mechanism" model for injury. Some attention will be directed toward a comparison of the two models.

Accurate temperature calculations for either the "single" or "two-mechanism" model cannot be carried out assuming uniform heat generation within the irradiated region of the pigment epithelium for pulse durations shorter than several microseconds. For these short pulses, consideration must be given to heat generation in, and heat dissipation from, individual melanin granules of the pigment epithelium (9). Hansen and Fine et al. have presented a "single-mechanism" model for threshold injury to the perigranular zone based on absorption of radiation within the melanin granules (9). Injury to this perigranular zone will also be utilized in this report to develop a "two-mechanism" model for the formation of threshold retinal lesions at both Q-switched and non-Q-switched laser pulse durations. This "two-mechanism" model may more accurately describe the mechanism for retinal injury at Q-switched laser pulse durations.

Mathematical Development of the Two-Mechanism Model

The following two mechanisms for injury are assumed in the development of the present model:

1. Local steam production at 100°C at the boundary of melanin granules is the dominant process in the formation of ophthalmoscopically visible, threshold retinal lesions when the laser pulse duration is shorter than 4 μ sec.

2. Thermal coagulation or thermal denaturation at the boundary of the melanin granule is the dominant process responsible for the formation of ophthalmoscopically visible threshold retinal lesions when the pulse duration is longer than 4 μsec. Steam production is not required for the formation of ophthalmoscopically visible, threshold retinal lesions at these longer pulse durations.

The first mechanism is compatible with the observations which show disruptive effects involving the pigment epithelium in which the scattering of melanin granules has been observed histologically (1,2). The particular extrapolation for pulse durations shorter than 4 μsec of the curve shown in Figure 1, is based on the

Washington National Record Center
Office of the Army Surgeon General
Record Group 112
Accession #: 75-13
Box #: 3
File: 1304-14 Fine, Samuel MD 2436
Biologic Effects of Laser Radiation

limiting case for steam production at $\sim 100\,^{\circ}C$ discussed above. The second mechanism is based on the success of the model for uniform heat generation in the irradiated region of the pigment epithelium developed by Ham et al. for long radiant pulse durations. At long pulse durations, the model for heat generation within an array of melanin granules in tissue reduces to the model for uniform heat generation within the irradiated tissue.

In order to calculate the incident retinal energy density required for injury at pulse durations ranging from 30nanosec to 10msec, the following assumptions are made pertaining to heat generation in, and heat dissipation from, melanin granules. The melanin granules are variable in size and shape, consisting principally of spheres and ovoids in the rabbit pigment epithelium. They will however, be considered to be spheres with diameters ranging from approximately 0.75 μ to 1.5 μ for purposes of calculation. These spheres are heated uniformly throughout their volume during a laser pulse. Both the granules and the surrounding medium have the thermal properties of water. This last assumption is only approximate, and is made for lack of exact data on melanin itself. The granule spacing is sufficiently wide so that the temperature at the boundary of one granule is independent of heat flow from neighboring granules. The surrounding medium is infinite in extent.

The temperature rise ΔT at a granule boundary then obeys the following relationship (10,11):

$$\Delta T\,(r = a, \tau) = \frac{E f(\tau)}{\tau} \tag{1a}$$

$$f(\tau) = \left\{ (\tau/\gamma) - \frac{4}{3\sqrt{\pi}}(\tau/\gamma)^{3/2} + \frac{2}{3\sqrt{\pi}} e^{-\gamma/\tau}\left[2(\tau/\gamma)^{3/2} - (\tau/\gamma)^{1/2}\right] + \left(\frac{2}{3} - \tau/\gamma\right) \operatorname{erfc}\sqrt{\gamma/\tau} \right\} \frac{3\sigma}{8\pi JK\,a} \tag{1b}$$

Washington National Record Center
Office of the Army Surgeon General
Record Group 112
Accession #: 75-13
Box #: 3
File: 1304-14 Fine, Samuel MD 2436
Biologic Effects of Laser Radiation

σ = radiation absorption cross section of granule (cm^2)

E = incident retinal energy density (joules/cm^2)

τ = laser pulse duration (sec)

K = thermal conductivity (cal/sec - cm^2)

erfc ( ) = complementary error function

r = distance from granule center (cm)

a = granule radius (cm)

γ = a^2/K

J = 4.18 joules/calorie

When the latent heat of vaporization (L = 540 cal/gm) is converted to an equivalent temperature, L/C_p, above 100 °C, one can write the condition for threshold steam production at a granule boundary for two different pulse durations as:

$$\frac{E_1 \; f(\tau_1)}{\tau_1} - \frac{L}{C_p} = \frac{E_2 \; f(\tau_2)}{\tau_2} - \frac{L}{C_p} \qquad (2)$$

where C_p is the specific heat at a constant pressure, and $\tau \leq 4$ μsec. The energy density, E, is the retinal energy density required for threshold injury. For ruby laser radiation, Ham et al. reported that the energy density required for threshold injury at a pulse duration of 30nanosec was 0.07 joules/cm^2 based on a calculated retinal image diameter of 800 microns (1). By letting E_1 = 0.07 j/cm^2 for τ_1 = 30nanoseconds in eq. 2, the portion of the curve labeled "A" in Figure 2 is generated for the range of granule radii shown.

Washington National Record Center
Office of the Army Surgeon General
Record Group 112
Accession #: 75-13
Box #: 3
File: 1304-14 Fine, Samuel MD 2436
Biologic Effects of Laser Radiation

It can be seen from Figures 1 and 2, that for $\tau = 4\,\mu$ sec a discontinuity will appear in the curve for E vs. τ if steam production is no longer necessary for injury. The magnitude of the discontinuity, ΔE, is given by L/C_p. If the lower energy at the discontinuity in Figure 2 is denoted by E_3, then the portion of the curve labeled "B" in Figure 2 can be generated from the equation:

$$\frac{E_3\, f(\tau_3)}{\tau_3} = \frac{E\, f(\tau)}{\tau}\; g(\tau) \qquad (3)$$

The factor, $g(\tau)$ is given by $(T(\tau) - 37\,^{\circ}C)/63\,^{\circ}C$ where $T(\tau)$, the temperature above $0\,^{\circ}C$ is obtained from the graph in Figure 1. It is assumed that the normal retinal temperature is $\sim 37\,^{\circ}C$ and $63\,^{\circ}C$ is the temperature rise necessary to reach $100\,^{\circ}C$.

The theoretical values of E as a function of τ, that are generated in the manner described above, agree reasonably well with the experimental data points shown for part "B" of Figure 2. Order of magnitude calculations indicate that the slightly high values for E along part "B" are probably a result of neglecting temperature contributions from neighboring granules.

One of the interesting aspects of this model is the prediction of a threshold injury energy density that is lower at 4 μsec than has been measured at 30 nanoseconds (see Fig. 2). Therefore, it may not be possible to infer safe conditions for lasers operating in the pulse duration range of approximately 10^{-6} to 5×10^{-5} sec, by using a straight line interpolation of the experimental data points shown in Fig. 2. Further experimental studies, particularly in the microsecond range, are required for verification of the model presented above.

Washington National Record Center
Office of the Army Surgeon General
Record Group 112
Accession #: 75-13
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Biologic Effects of Laser Radiation

Discussion

A "two-mechanism" model for threshold, retinal, laser lesions based on steam production at short pulse durations but not requiring steam production for longer pulse durations has been presented. Melanin granules are considered as the primary energy absorption site in this model. Although the melanin granules in the rabbit pigment epithelium are not only spherical, but also ovoidal and ellipsoidal in shape (12), for purposes of calculation, spherical, homogeneous granules are assumed.

It should be noted that although a 100°C isotherm is assumed for injury in the region in which steam is produced, that is for pulse durations shorter than 4 μsec, other extrapolations are possible. Until further experimental data regarding temperature and pressure is obtained, there is no evident reason for the choice of other extrapolations.

Hansen and Fine et al. have previously reported on a melanin granule model for threshold retinal injury without steam production at short pulse durations — a "single-mechanism" model (9). This model was based on the assumption that injury occurs in the perigranular region, at the same temperature, for all pulse durations. The curves sketched in Figure 3 are based on this model. In developing these specific curves, the data point by Ham at 200 μsec (Fig. 3) was used as a starting point. These curves were then obtained from equations 2 and 3 with the term referring to the latent heat of vaporization deleted. The variation in the temperature required for perigranular injury which is based on the factor $g(\tau)$ in equation 3 has negligible effect in the shape of the curves in Figure 3.

Figure 3, based on the "single-mechanism", melanin granule model shows that as the pulse duration is decreased, the energy density required for a threshold lesion is decreased. The curves do not, however, indicate that a minimum threshold energy density for injury will occur at about 4 μsec.

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Accession #: 75-13
Box #: 3
File: 1304-14 Fine, Samuel MD 2436
Biologic Effects of Laser Radiation

When τ = 30 nanoseconds in Figure 3, it can be seen that the threshold retinal energy density is a strong function of the granule radius. A variation of a factor of five in the granule radius results in a variation of a factor of thirty in the threshold energy density when the granule boundary temperature is held constant. Under experimental irradiation conditions where a certain retinal energy density and pulse duration is chosen, a similar variation will be manifested in a statistical distribution of granule boundary temperatures as a function of granule radii. Therefore, as the laser pulse duration is shortened to the Q-switched range, microscopic, threshold retinal injury may depend strongly upon the distribution of melanin granule size rather than upon grossly observable "degree of fundus pigmentation".

It can be seen from Figure 3 that there is some variation in the results reported by various investigators. Kohtiao and co-workers have obtained threshold lesions in rabbits for retinal energy densities at 4.5×10^{-3} j/cm^2 at 80nanoseconds in 50% of the cases (2). It is difficult, however, to compare this value with the data from Ham (1) and Bergquist (3) because the criteria for injury, retinal image diameter, pigment granule size distribution, and statistical meaning of "threshold injury" may not be the same in each case.

Both the "two-mechanism" model where steam production is considered, and the "single-mechanism" model for thermal injury in the neighborhood of melanin granules, can be applied to laser studies on reversible depigmentation in rodent skin (13). These models may also be of significance in elucidating the mechanism for energy transformation on the interaction of laser radiation with pigmented tumor systems. The heat conduction analysis and mechanisms for injury presented in this report are applicable to laser studies on single cells.

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File: 1304-14 Fine, Samuel MD 2436
Biologic Effects of Laser Radiation

FIGURE CAPTIONS

Fig. 1. Calculated and extrapolated values of retinal temperature required for threshold injury.

Fig. 2 Experimental and theoretical values for the retinal energy density required for threshold injury in rabbits as a function of radiant pulse duration, τ . These curves assume two mechanisms for injury: steam production for $\tau < 4\,\mu$ sec., and thermal denaturation for $\tau > 4\,\mu$ sec. The parameter, a, is the granule radius.

Fig. 3 Experimental and theoretical values for threshold injury in rabbits as a function of radiant pulse duration, τ . These curves assume a single mechanism for injury, i.e., thermal denaturation, for all pulse durations. The parameter, a, is the granule radius.

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Biologic Effects of Laser Radiation

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Biologic Effects of Laser Radiation

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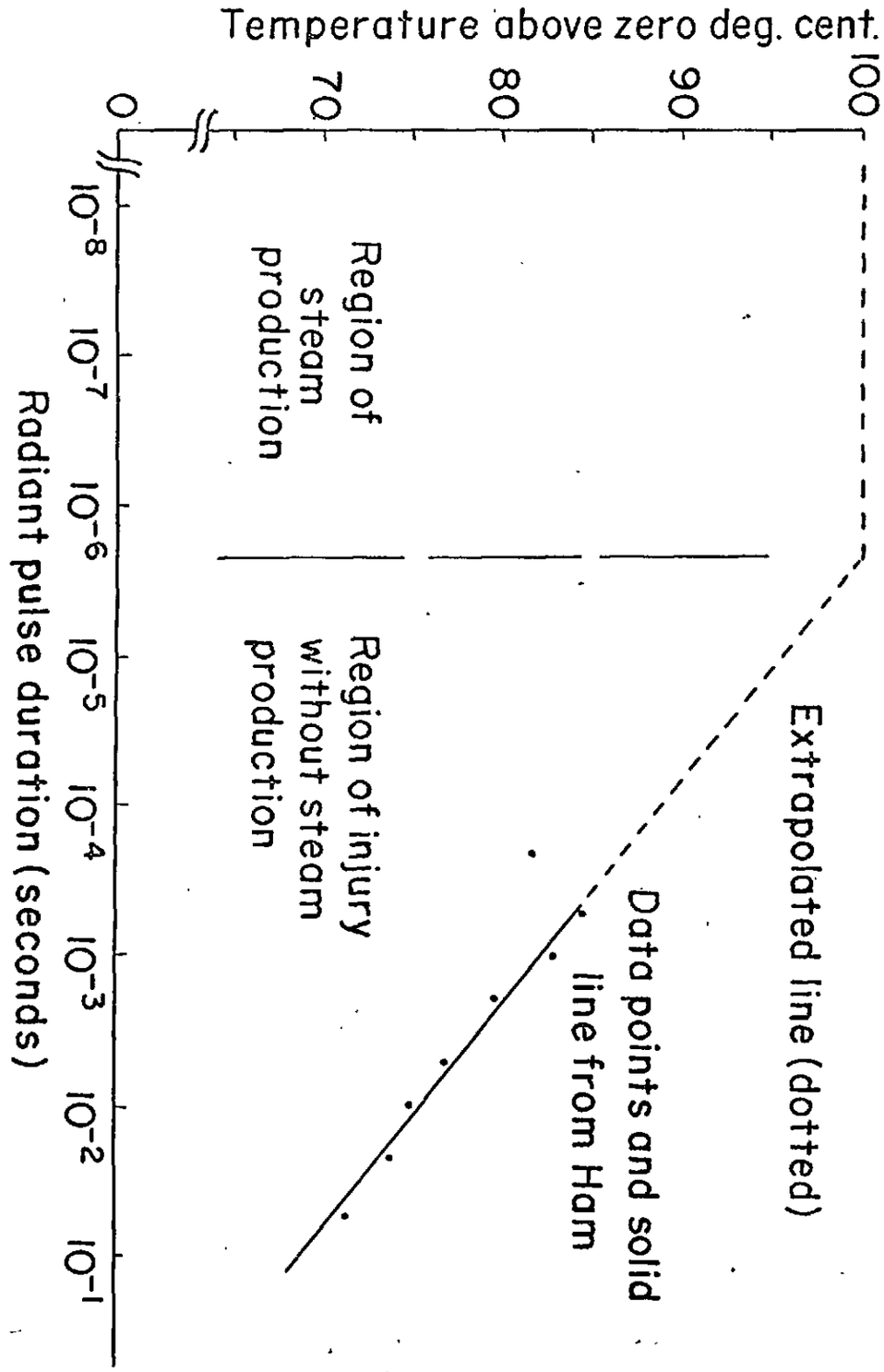

Fig. 1

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Biologic Effects of Laser Radiation

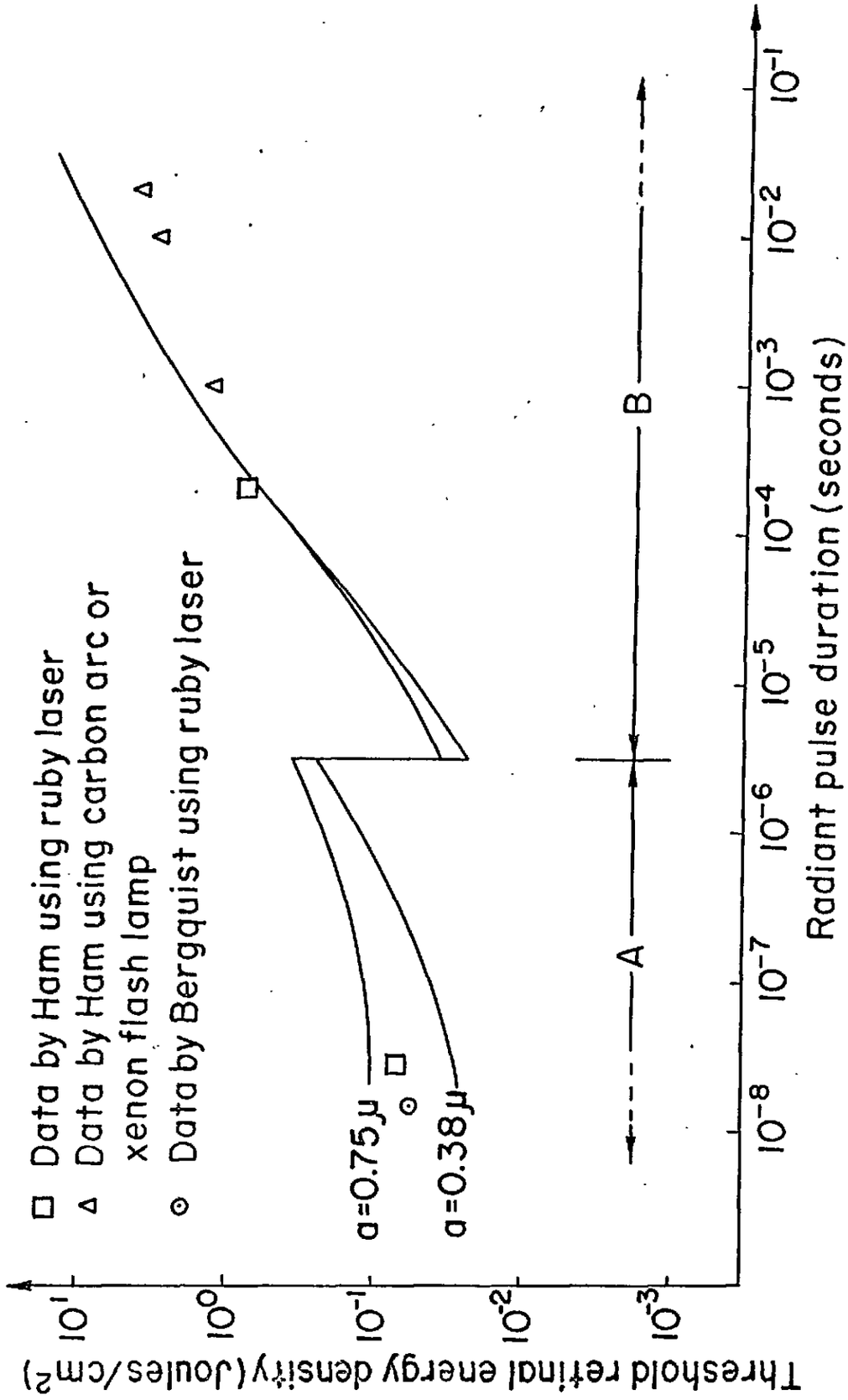

Fig. 2

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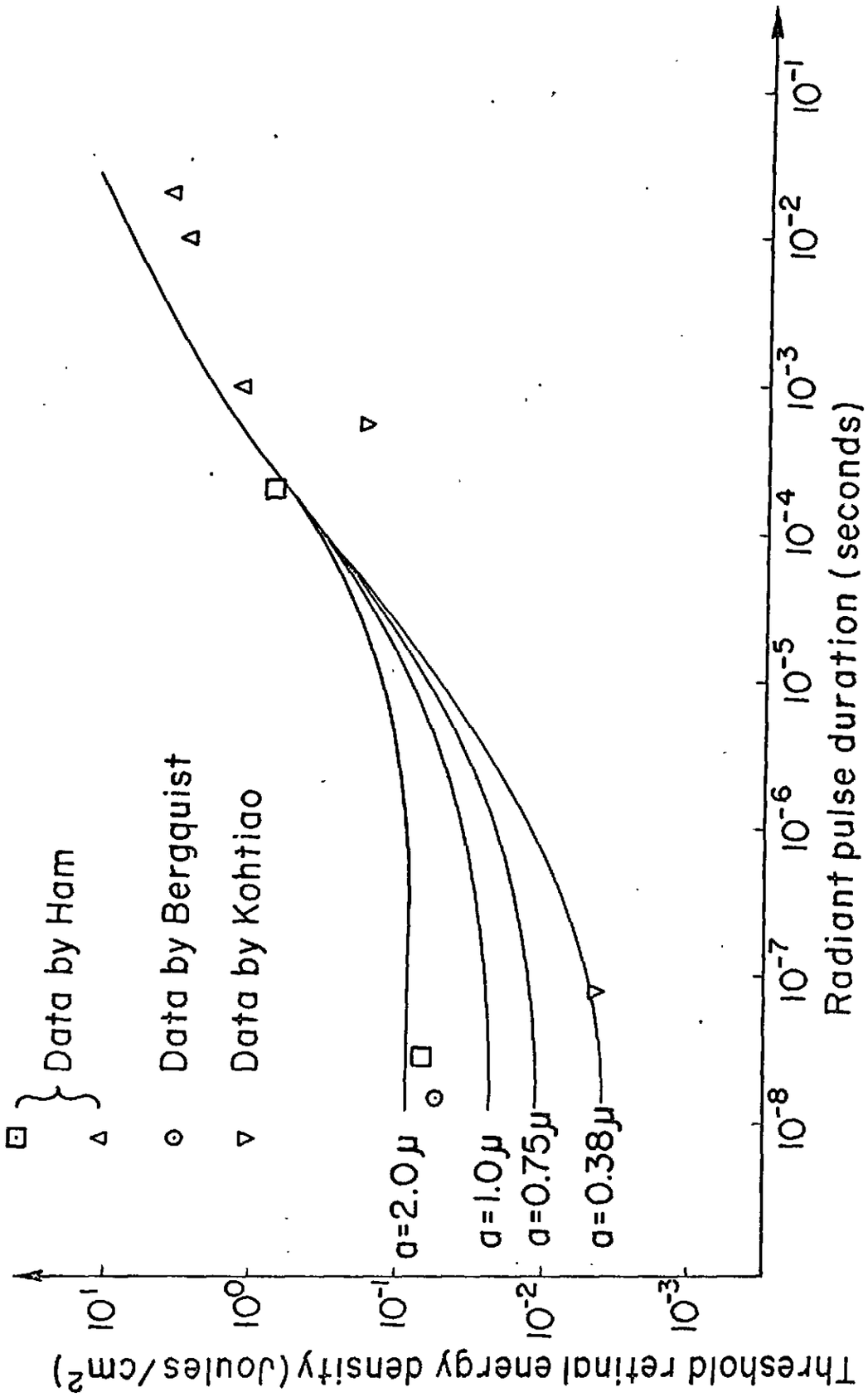

Fig. 3

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Biologic Effects of Laser Radiation