Factors in the prevalence of cataract in India — Analysis of the recent Indo-US study of age-related cataracts

D. Balasubramanian, K. Seetharam Bhat **and** ***Gullapalli N. Rao***

CATARACT is the leading cause of blindness all over the world. Prevention of cataract or delaying its progression would constitute a major achievement in human welfare. If the development of cataract could be delayed by 10 years, the number of cataract operations would decrease[1] by 45%. Of the world's 40 million blind people, cataract affects about 17 million and about a third of these are in India. The recognized causes for visual impairment and blindness in India are: cataract (55%), trachoma and eye infections (20%), malnutrition (20%), injuries (1.2%) and glaucoma (0.5%), with other causes accounting for the remainder[2]. Thus any plan aimed at combating blindness must necessarily deal with cataractogenesis. One approach towards this is to identify the aetiological factors governing cataract through epidemiological analysis of the disease.

Physiology and anatomy of the lens

Figure 1 illustrates the various parts of the human eye. The lens and cornea together constitute the major refractive components of the eye. The lens is a uniquely transparent, biconvex intraocular structure, whose shape can be altered by compressing or relaxing the ciliary muscles attached to it. It is enclosed in an elastic capsule suspended from the ciliary body by many fine zonular fibres which are attached radially at its equator. Bounded anteriorly by the iris and posteriorly by the vitreous gel, and devoid of any blood supply or innervation, it derives its nutrition from the aqueous humour and the vitreous gel[3,4].

The anatomical details of the lens are shown in Figure 2. The lateral border of the lens is the equator and is formed from the union of the flatter anterior surface and the more convex posterior surface. The zonules are attached to an area approximately 1 mm in diameter on each side of the equator. The equatorial diameter of the lens is 6.5 mm at birth, and increases to 9.0 mm by about age fifteen, and remains constant thereafter. The anterior–posterior diameter of the lens is 3.5 mm in the newborn and reaches 5.0 mm in the adult. The average radius of curvature is 10 mm for the anterior surface and 6 mm for the posterior surface. These measurements vary during accommodation, the adjustment of the focal length of the lens by changes in its curvature. The elastic lens capsule, made up of largely collagenous material, increases in thickness until approximately 35 years of age.

The lens epithelium is a single layer of cuboidal cells which extends laterally to the equator, where the cells begin to elongate. The pre-equatorial epithelial cells multiply by mitotic division. The young cells frequently migrate to the equator, where they rotate and become lens fibres. The cells lie on their basement membrane and the capsule and their lateral ends are joined by large interdigitations. The lens cells resulting from the equatorial epithelium

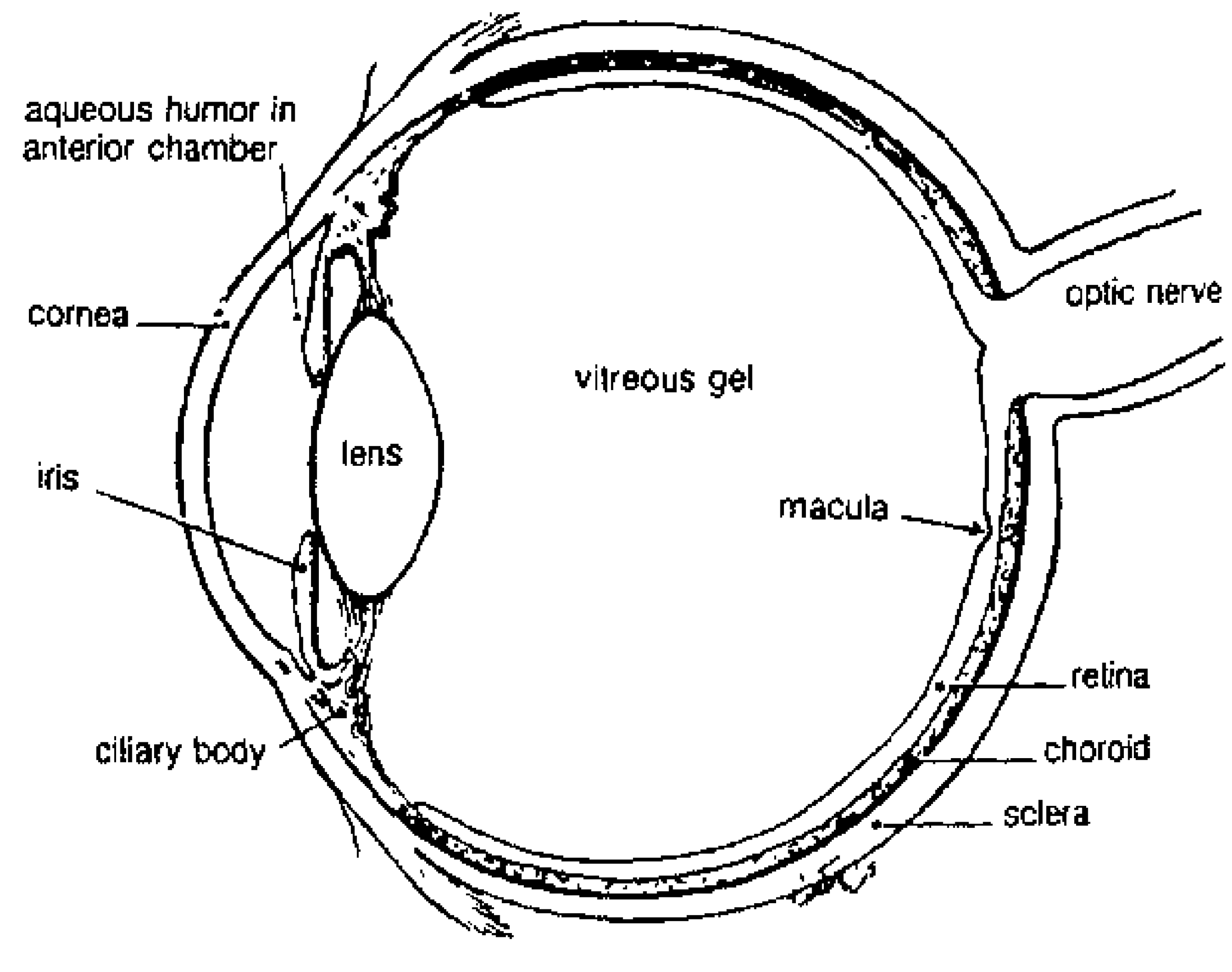

Figure 1. The human eye.

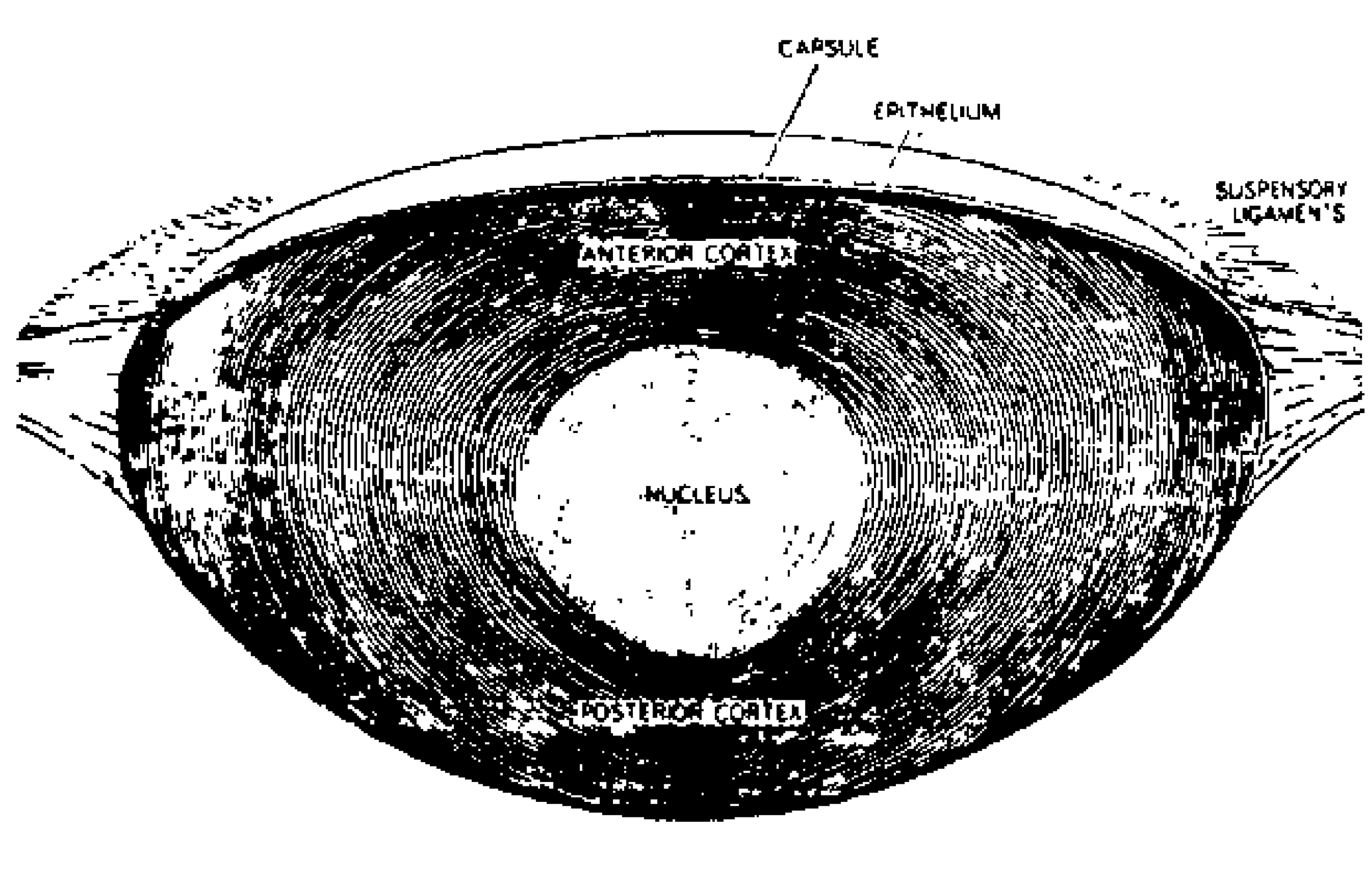

Figure 2. The human eye lens. The anterior faces the iris while the posterior abuts the vitreous gel.

D. Balasubramanian is in the Centre for Cellular and Molecular Biology, Uppal Road, Hyderabad 500 007; K. Seetharam Bhat in the National Institute of Nutrition, Jamai Osmania P.O., Hyderabad 500 007; and Gullapalli N. Rao in the L. V. Prasad Eye Institute, Road No. 2, Banjara Hills, Hyderabad 500 034. Correspondence on this paper should be addressed to D. Balasubramanian.

elongate to form fibre-like cells, which are hexagonal in shape in cross-section.

The two main functions of the lens are refraction and accommodation. In a relaxed state the combination of lens shape and the refractive media acts to converge the parallel light rays from a distant object to focus on the retina[3,5]. When near objects are viewed, the divergent rays striking the eye must be converged by a greater amount to retain the focus on the retina. The lens accommodates by increasing its convexity to supply the additional convergence power necessary. Accommodation is relaxed when the lens assumes a flattened shape for viewing distant objects.

The index of refraction of the lens (1.390) is greater than that of the aqueous or the vitreous humour (1.334). Therefore any increase in the convexity of its surface increases its convergence power and permits near objects to be focused on the retina. The total convergence power of the eye is approximately 60 dioptre (dioptre D is the reciprocal of focal length), of which 14 D is contributed by the lens. The amount of dioptric change that the lens can produce by altering its convexity is termed the amplitude of accommodation. This is greatest in children (approximately 14 D) and decreases linearly to about 1–2 D at about 55 years of age.

Any opacity in the lens, whether it is a small, localized one or one involving the entire lens, is called a cataract (see Figure 5, *a*). Trauma, inflammation, metabolic or nutritional defects, and radiation damage can all induce such a change. However, age-related change is the most common and is known as senile cataract. This type of cataract appears usually after an age of 30 to 40 years, is progressive, and ultimately results in severe visual impairment.

Age-related cataracts are classified either according to the location within the lens or according to the stage of development[6,7]. The major division is between the nuclear and cortical types of cataract, with subcapsular cataracts being a special subdivision of cortical cataracts. This classification is particularly relevant in early stages of cataract, when it is easier to observe the different zones of the lens. Mixed cataracts are more common in advanced cases. Each of these different types of cataracts can be further graded based on the degree of change, i.e. whether it is early, moderate or advanced.

The clinical symptoms of cataract are largely two-fold: (i) Visual impairment, which is gradually progressive, is the most common symptom of cataract. In nuclear cataract, there may be an initial shift to myopic refraction (which may lead to improvement in near vision, the so-called second sight), ultimately resulting in complete loss of vision. Of the subcapsular cataracts, the posterior subcapsular is the most common type. The initial symptoms are in the form of bright lights, and when the pupillary axis is involved, the effect can be devastating. Visual impairment progresses at a very rapid rate. (ii) Mononuclear diplopia or poor hue discrimination may be another common symptom of nuclear cataract. Typically, nuclear cataracts take longer before they interfere sufficiently with vision to warrant extraction of cataract.

The only effective treatment for cataract is surgery, namely cataract extraction. Significant advances have been made in the technique, making this one of the most successful and gratifying surgical expereinces. The traditional method has been to remove the lens *in toto* by the 'intracapsular technique'.

In recent years the technique of 'extracapsular cataract extraction' has become popular since it is considered safer, with fewer postoperative complications of any severe nature. In this technique, the entire posterior capsule along with the peripheral rim of the anterior capsule are left behind to form the 'capsular bag'. This also facilitates the implantation of a posterior chamber lens. This has become the most accepted method of visual rehabilitation following cataract surgery around the world and provides the most optimal visual result. The widespread application of this technique has limitations in India, mainly owing to economic considerations. While traditional methods of cataract extraction can be accomplished at a cost of about Rs. 200 to 300, the addition of intraocular lens implantation escalates the cost to Rs. 2000 to 3000. This is mainly due to the need to provide better infrastructure for performing surgery as well as the cost of the implant.

Molecular features of the lens

As mentioned above, the lens forms as a tissue from the lenticular epithelial cells which differentiate into the lens fibre cells. The earliest protein to be expressed is γ-crystallin, while α and β crystallins are expressed just when the epithelial cells are about to differentiate. The fibre cells become enucleated and all particulate organelles are expelled from the fibre cells, which consequently become metabolically sluggish, with little turnover. They simply deposit on one another, much as onion layers do, compactly and densely, to form the transparent, colourless, non-scattering tissue[8].

There is thus not only a temporal but a spatial segregation of matter and events in the lens. The core or the interior nucleus of the lens largely comprises γ-crystallin, and since this part is the oldest part of the lens, this also contains the oldest protein population in the organism. γ-Crystallins are monomeric proteins of molecular weight around 19–21 kDa, and highly homologous among themselves. The crystal structure of γ_2 (now called γ_B) crystallin was elucidated in 1981 by the Birkbeck group[9]. It is a kidney-shaped molecule with a Greek-key folding motif, and the secondary structure is largely antiparallel pleated sheet conformation. α and β Crystallins are found more in the midregion and cortical regions of the lens. β-Crystallins are oligomeric proteins, with each protomer ranging in size from 21 to 34 kDa. The crystal structure of β_{B3a}

crystallin has been recently solved[10], and it is quite similar to that of γ_B, as the high sequence homology would have it. α-Crystallin is also homologous to β- and γ-crystallins, but is a very large aggregate, almost micellar, of monomers of molecular weight 20 kDa[11].

About 35% of the weight of the lens is made up of the crystallins and most of the remainder is water. The eye lens is thus the most concentrated gel of proteins in water known so far in nature, devoid of any long-range order and at once plastic and elastic. Being a slowly-turning-over tissue, the eye lens is an excellent reporter of cellular and molecular events during senescence and ageing. Chronic low-level assault and modifications on its components tend to accumulate over the years, ultimately causing covalent modifications and loss of solubility of the crystallins, leading to light scattering and precipitation, or lens opacity and cataract.

Such opacification of the lens can be reversible or irreversible. Reversible cataract occurs upon a drop in the temperature of the lens and is termed 'cold cataract'. It is now understood to be a phase separation of the proteins at low temperature, exemplifying the lower critical solution phenomenon[12], and occurs both at the lenticular and constituent molecular levels[13]. Upon increasing the temperature, the precipitated protein redissolves in the medium, and transparency is restored. Another case of reversible opacification of the lens is encountered in the early stages of the induction of cataract in experimental animals by a high-sugar diet. When rats are administered a 30% galactose diet, lens opacification starts within a week and becomes irreversible after three weeks on the diet. However, if normal diet is restored, even after 10–12 days after the start of the high-galactose diet, the opacity can be cleared. This reversible 'cataract' is essentially a situation of vacuole formation in the lens upon osmotic stress, and is cleared upon withdrawal of the stressful diet early enough.

Irreversible opacity usually occurs owing to covalent chemical changes in the crystallins, leading to loss of solubility. Intermolecular aggregation and cross-linking of proteins are seen in cataractous lenses[14]. Figure 3 shows the electrophoretic gel pattern of the proteins from a cataractous lens extracted at the L. V. Prasad Eye Institute. Indeed, even as a normal lens ages, the relative amounts of high-molecular-weight and water-insoluble proteins in it increase (Figure 4), and the extent of light scattering also increases while transmission decreases[15,16]. Cross-linking might involve intermolecular disulphide bridges, as well as non-disulphide bonds whose exact nature is yet to be clarified.

Besides having high-molecular-weight cross-links and insoluble proteins, a cataractous lens is often also coloured (Figure 5), or contains chromophores other than the phenylalanyl, tyrosyl and tryptophanyl residues of the protein chains[17]. These chromophores might occur either in the protein-bound or in the free form, and are thought to

Figure 3. Polyacrylamide gel electrophoresis of human cataractous lens crystallins in presence of sodium dodecylsulphate and β-mercaptoethanol. Notice the presence of high-molecular-weight proteins in the gel. In comparison, a young infant lens has proteins only in the 18–20 kDa region.

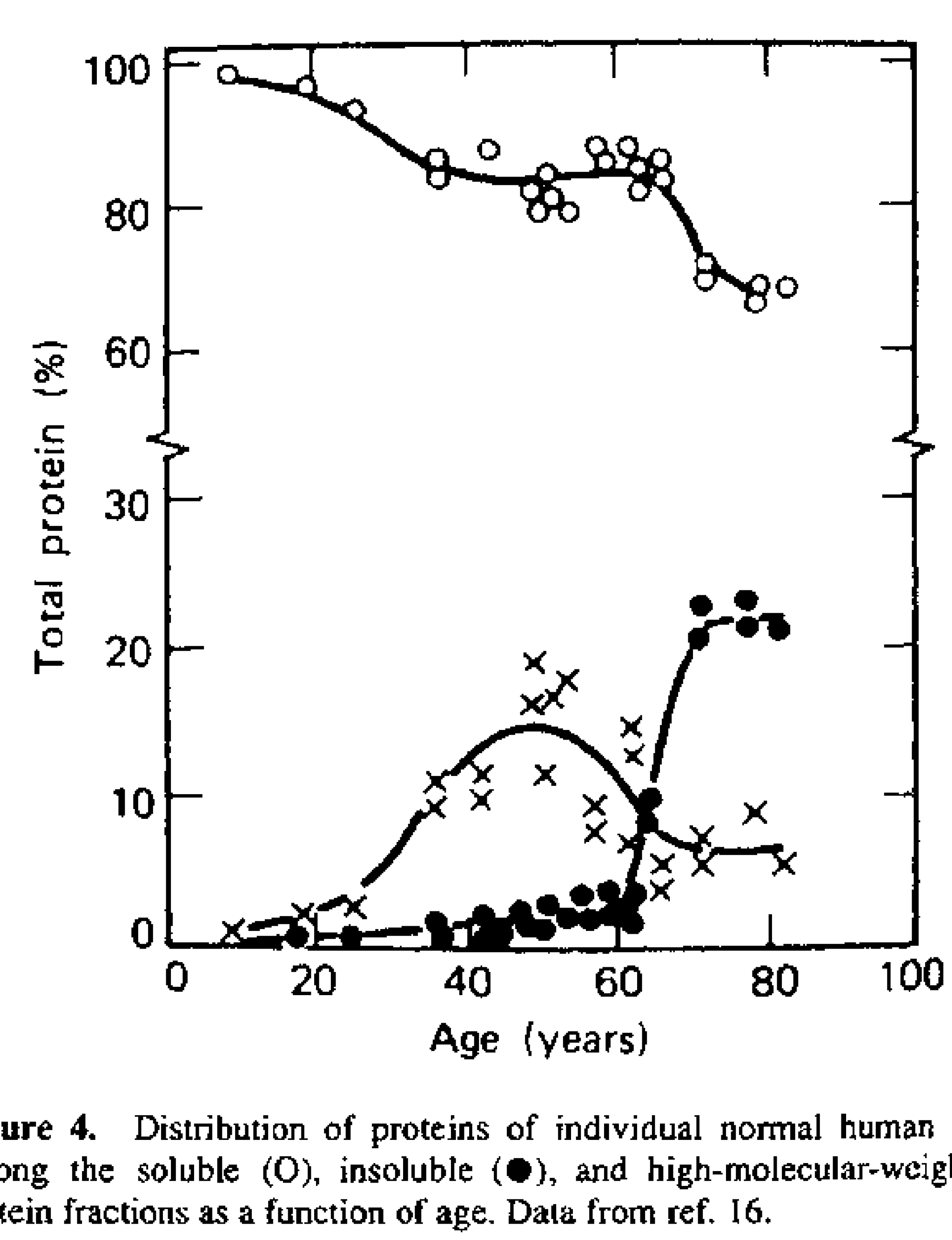

Figure 4. Distribution of proteins of individual normal human lenses among the soluble (O), insoluble (●), and high-molecular-weight (x) protein fractions as a function of age. Data from ref. 16.

accumulate in the lens with time as reaction products of endogenous molecules, photochemically or in the 'dark'. Some of them arise as oxidation products of amino-acid residues, e.g. kynurenines from tryptophan[18]. The lenses of squirrels, monkeys and humans contain hydroxykynurenine and related molecules, perhaps in the free form[19]. These also appear to be produced enzymatically

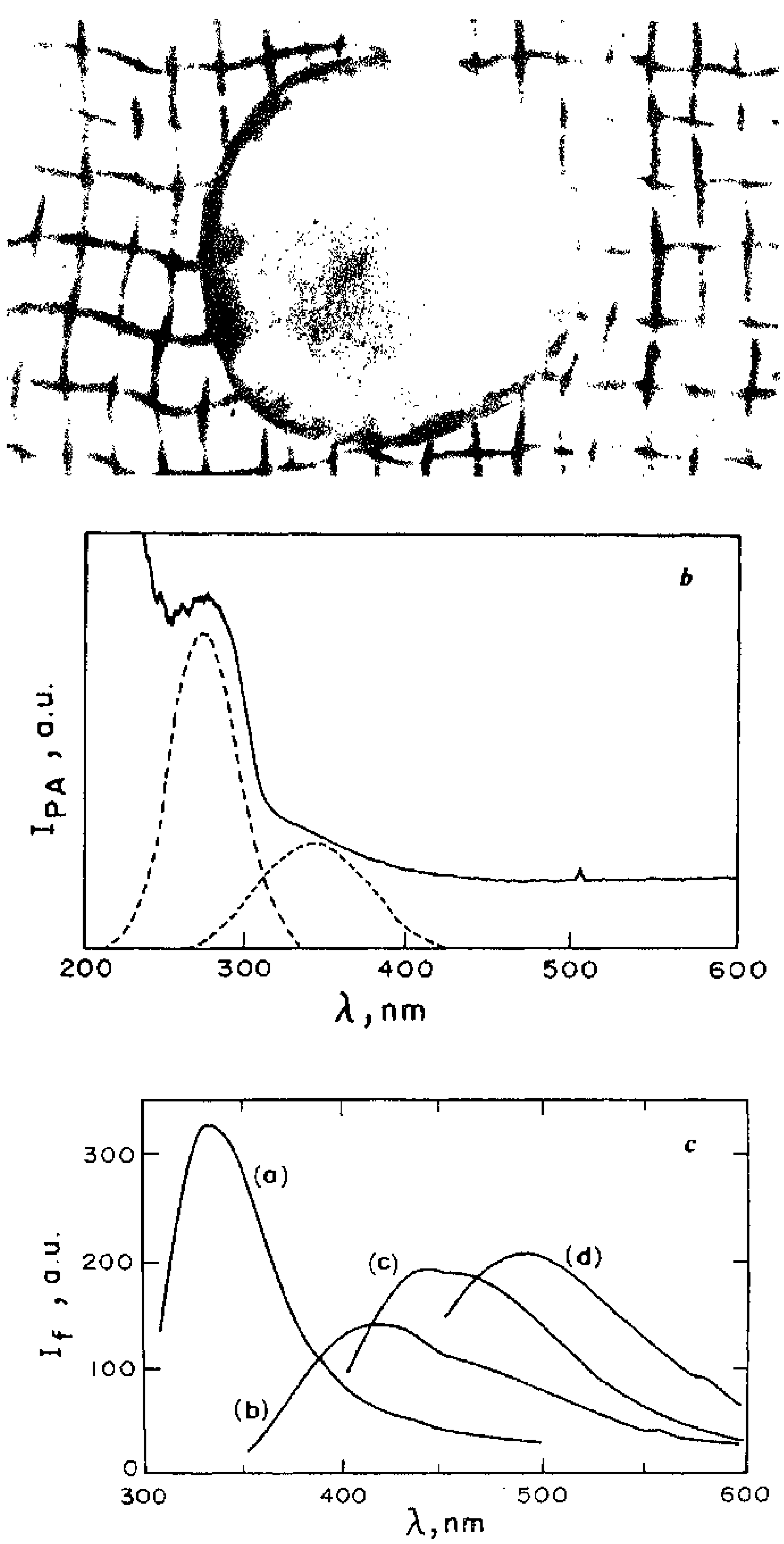

Figure 5. *a.* An enlarged photograph of a cataractous lens of a diabetic patient, extracted at the L. V. Prasad Eye Institute. The lens has been placed on a grid to facilitate inspection of transparency. Opacity is highest at the nucleus and least at the cortical exterior. *b.* Photoacoustic (PA) spectrum of a cataractous lens, showing the two major resolved bands at 280 and 360 nm. PA spectroscopy was used so as to avoid difficulties due to the solid state of the intact lens and the light scattering due to the sample. (Spectrum by Ch. Mohan Rao, CCMB) A young and normal lens displays only the 280 nm band due to the aromatic amino acids of the constituent crystallins. The 360 nm band accumulates with age and upon modification of the crystallins. *c.* Fluorescence spectrum of the cataractous lens shown in *a* shortly after extraction. Band (a) around 340 nm is largely due to the tryptophan residues of the lens crystallins, and is obtained upon exciting at 295 nm. Band (b) is obtained upon excitation at 320 nm, band (c) upon excitation at 360 nm, and band (d) upon excitation at 410 nm. Notice that the intensities of the (c) and (d) bands approach that of band (a) in this lens; a young and healthy lens does not display bands (c) and (d), and only a minor band (b). A Hitachi F-4000 spectrofluorimeter was used along with a solid-state sample holder, using the procedure given in ref. 53.

from tryptophan in the human lens[20]. Other chromophores arise as a result of glycation of the crystallins and subsequent Maillard (or 'browning') reaction[21]. Some of these chromophores act as photosensitizers[22]; they absorb light and transfer the excited-state energy to substrates such as the crystallins, leading to further coloured products and oxidative changes. The sensitization efficiencies of the kynurenine derivatives and of some coenzymes present in the eye have recently been evaluated by us[23], since electromagnetic radiation, particularly in the ultraviolet region, has been suggested to be a risk factor in cataractogenesis[24–27].

With age, the human lens accumulates as many as two dozen substances that absorb and/or fluoresce above 350 nm (termed non-tryptophan fluorescence), which might also explain the colours of yellow, brunescent and nigrescent cataracts. The presence of visibly coloured substances in the lens should also lead to a colour-filter effect, in which light of some wavelength regions are transmitted to greater extents than that of other regions. Usually, the region above 550 nm seems to be transmitted more than blue and green light. Figure 6 shows the age-dependent deterioration of light transmission and also the progressive colour filtering of the human lens. It is interesting to ask how these coloured compounds originate in a tissue that is metabolically inert. Several of these compounds relate to the oxidation products of tryptophan, and occur at least in part through direct or through sensitized photochemical mechanisms, as mentioned above[23–27].

There is a correlation between diabetes and high endogenous glucose levels and the prevalence of

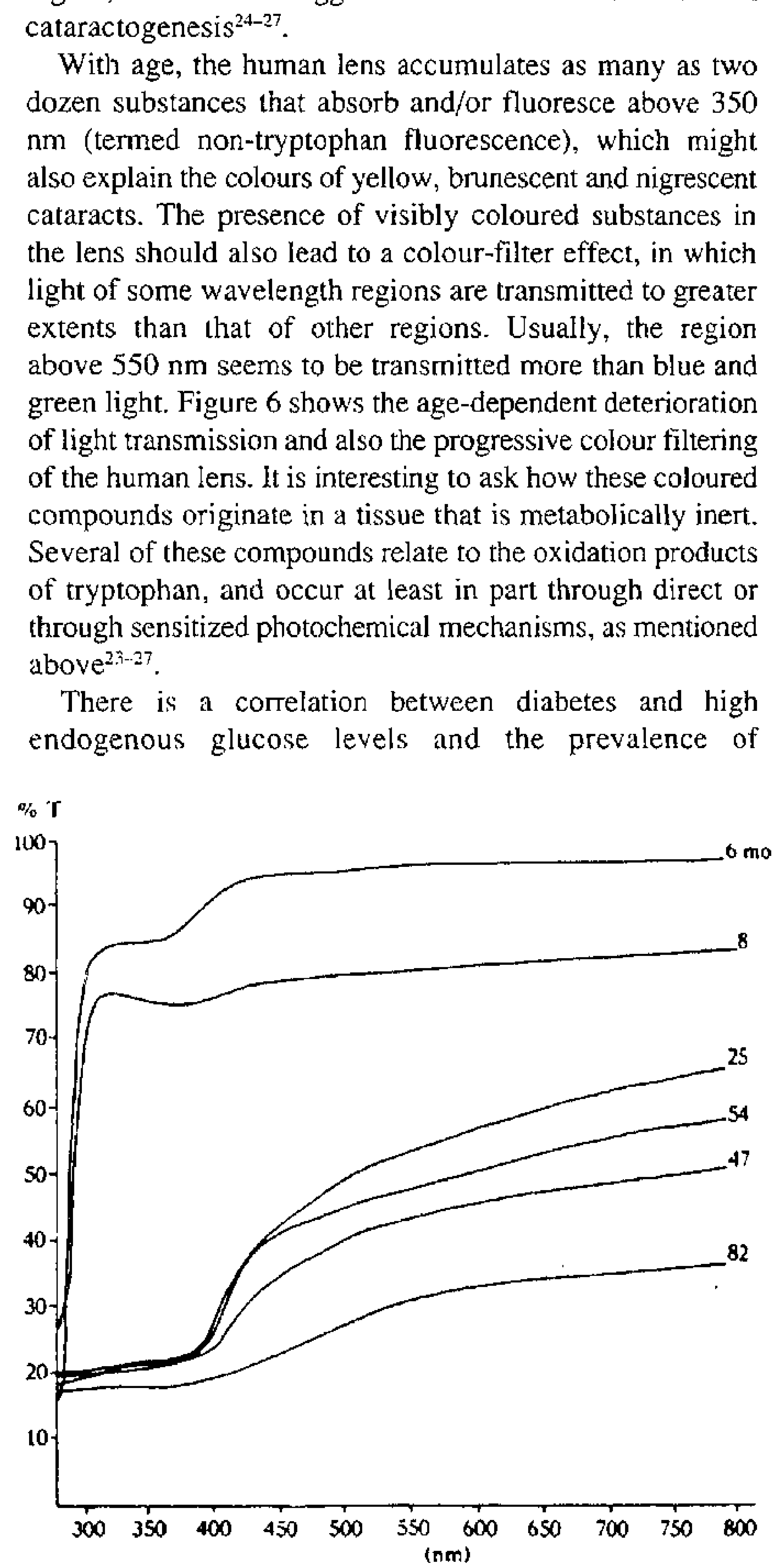

Figure 6. Transmission characteristics of normal human lenses of various ages. Note the drop in the transmission and also the colour filtering (positive slope in the curves) as a function of age.

cataract[28-30]. In this connection, analysis of the crystallins from aged or cataractous lenses has also revealed glycosylation of amino groups in the proteins. Glycosylated lysine has been identified in acid hydrolysates of crystallins from diabetic cataracts[31-33]. The accumulation of polyols in the lens would also offer a potential source of active oxygen species and radicals, which would lead to protein cross-linking and precipitation. Inhibition of the endogenous enzyme aldose reductase would lead to decreased production of polyols and thus offer cataract prophylaxis[34,35]. The search for safe and effective aldose reductase inhibitors (sorbinil, quercitrin, ICI 105552, AL 1576) and their use as anti-cataract drugs is an active area of investigation at present.

The Indo-US cataract survey

It is in this background that we wish to review the recent Indo-US epidemiological study of age-related cataract published last year[36]. This was a cooperative study spanning the period August 1984–December 1987, and covered 1441 cataract patients and 549 controls, all in the age group of 37–62 years. The patients were referred to the Rajendra Prasad Centre for Ophthalmic Sciences at New Delhi. This study is the latest in a series of papers on cataract epidemiology in the subcontinent[37-42] and the most extensive so far. The earlier ones had found that:

(i) Lenses removed at eye camps in Pakistan were darker brown in colour than those removed in the Oxford eye hospital in England[37]; this is similar to the observation[43] that Filipino lenses are darker brown than those from New York state (see Figure 5 for the colour of a cataractous lens from Hyderabad). This might very well reflect photo-oxidative processes in the lens that could be more prevalent in the tropical sunshine belt where cloud cover is on an average lower[26]. Of relevance here are the studies of B. N. Srivastava of the National Physical Laboratory, New Delhi, who found that the levels of UV-B (280–320 nm) radiation are higher in the South Asian latitudes, and observed that these levels also show a seasonal rhythm (about 0.9 units of normalized surface UV radiance at 20° latitude on 16th July, dropping to about 0.6 on 16th January; the ozone content being about 280 Dobson units at the same 20° latitude on 16th July, dropping to around 240 units on 16th January)[44].

(ii) The incidence of cataract or aphakia is strongly age-dependent, jumping from 13% of the population of Punjab and Haryana over 30 years of age to 31% of those over 60 years[38,39].

(iii) There might be a dietary correlation with cataract, e.g. high mineral intake caused by drinking up to 5 litres of the local water daily and heavy consumption of yogurt[39,40]. This could lead to the chronic accumulation of metal ions such as Ca^{2+} in the lens. Abnormal levels of Ca^{2+} lead to lenticular opacity through a variety of means, such as membrane permeability changes, precipitation of calcium oxalate and phosphate, binding to the crystallins and precipitation, transglutaminase-mediated crystallin cross-linking, and so on[45-47].

(iv) Malnutrition[48] and gastrointestinal infection resulting in diarrhoea leading to malnutrition, acidaemia, uraemia and dehydration[49] might correlate with cataract incidence. Figure 7 is a speculative scheme suggested by Harding, indicating possible relationships between diarrhoea and cataract. We highlight it here in order to draw attention to the role of cyanate, which (a) was seen both *in vitro* in rabbit lenses[50] and in human cataract lenses from Pakistan[51] and (b) could have been important in the Bhopal gas tragedy of 1984, in which methyl isocyanate caused damage to the eyes and other tissues of the affected population[52].

(v) The influence of genetic factors in cataract is a possibility.

The Indo-US study analysed the cataracts of the 1441 patients and classified them largely into four types, namely pure posterior subcapsular (P), pure cortical (C), pure nuclear (N) and mixed nuclear-cortical (M). To the extent that γ-crystallins are largely localized in the nuclear region while α-and β-crystallins occur in the cortex and midregion, this classification would allow us to hazard some guesses about the molecular features of the cataracts, a liberty that we take guardedly. The study is thorough in that it has attempted to look at as many as 55 physical, biochemical and behavioural parameters as possible correlates, and implicates at least six of them as very likely correlates of cataract. We comment on each of these below.

Decreased cloud cover correlates with increased risk of cataract

The odds ratio for cataract formation, computed by using the polychotomous logistic regression (PLR) analytic

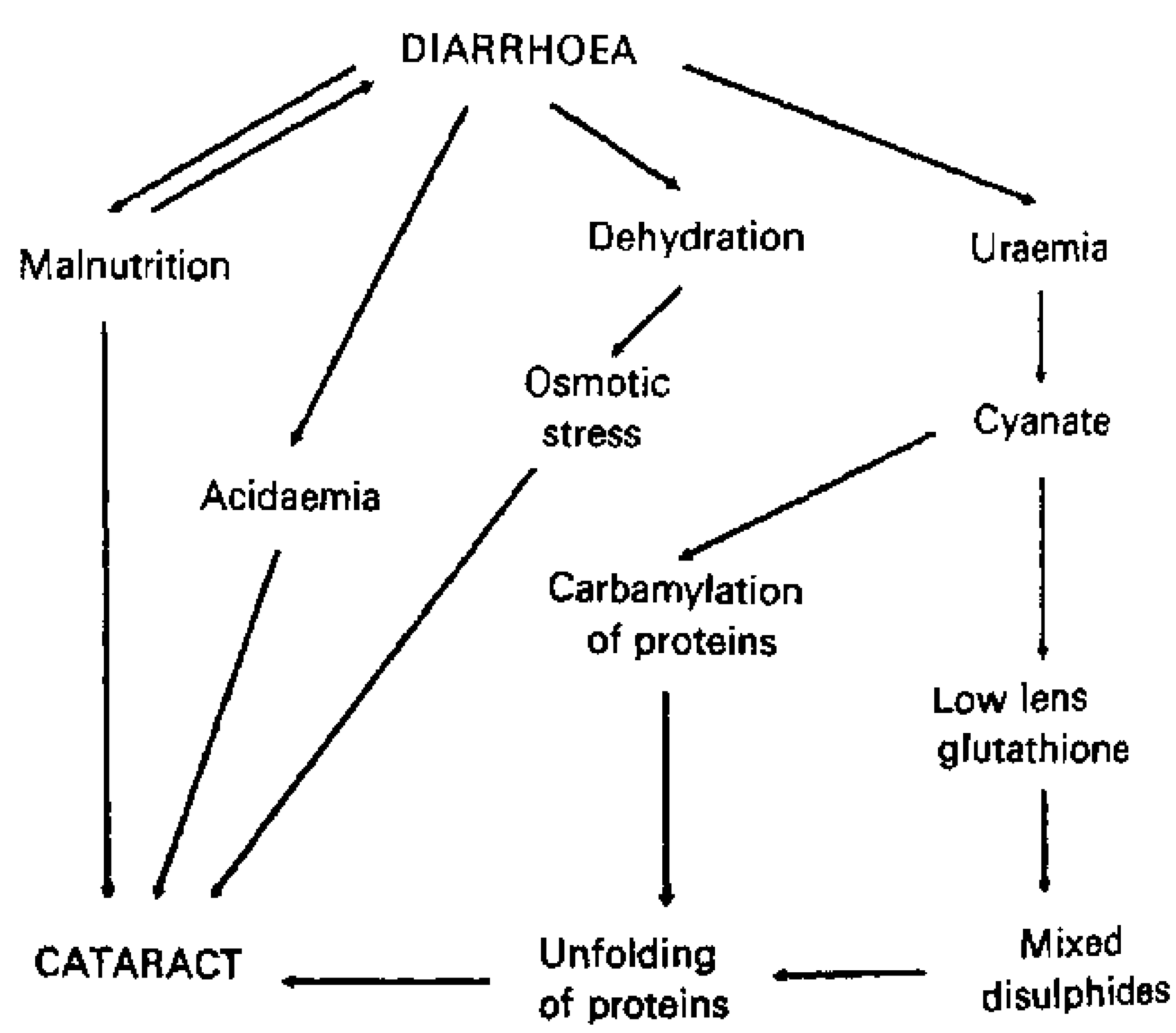

Figure 7. Harding's scheme indicating possible relationships between diarrhoea and cataract (refs. 49, 73).

method, is 0.78. Cloud cover below 4 okta appears to be involved, and cataracts of all the four types, namely P, C, N and M, appear likely (an okta is one-eighth of the sky with cloud cover).

We have commented on the possibility of light-induced covalent modifications that can be brought about both in the intact lens and in crystallins[53-55] (see above). While the average levels of UV-B radiation are higher in the Indian subcontinental latitude[44], we believe that most of this radiation is effectively filtered by the environment, and by the cornea, the aqueous humour and the lens capsule[26]; hence the photochemical changes *in vivo* are more likely due to the absorption of near-ultraviolet and visible radiation by endogenous chromophores in the lens and subsequent photosensitization of and damage to the crystallins[22, 53]. Some of these sensitizers would be the kynurenines, anthranilic acid[23] and glycated products[21]. That cataracts occur all through the lens (P, C, N, M) suggests equal vulnerability of all crystallins and/or accumulated events over time.

Diets low in selected nutrients increase the risk of cataract

The risk of cataract appears to be increased with lower levels of antioxidants (namely glutathione peroxidase, glucose-6-phosphate dehydrogenase, and vitamins C and E) and lower body mass index. The inclusion of blood biochemical factors and their assessment in each case allows the identification of systemic markers associated with each cataract type. Also, as mentioned above, oxidative mechanisms appear to play a role in cataractogenesis, and the lens has endogenous antioxidant molecules such as glutathione and its redox enzymes, superoxide dismutase, peroxidase and ascorbic acid to prevent or inhibit, either singly or (more likely) synergistically[56], oxidative damage to lens components. The Indo-US study took into account several nutritional factors — protein, thiamine, riboflavin, vitamins A, C and E, calcium and animal/vegetable sources in diet — appropriately weighted and found that:

(i) In general, the higher the nutritional level of an individual, the less risk he faces of cataract.

(ii) High nutrient levels protect against P, N and M cataracts; why the cortical (C) cataract is not equally protected is a puzzle here. Do the various crystallins differ in their susceptibility to cross-linking? Could this be a reflection of their individual molecular aggregation status (α-crystallin is the most aggregated)?

(iii) There is sufficiently good correlation between antioxidant levels, measured as AO3 (referring to the composite levels of glutathione peroxidase, vitamins C and E, and glucose-6-phosphate dehydrogenase), and protection against cataracts of type P and P+N.

(iv) There was no association between cataract occurrence and the status of thiamine, pyridoxine or vitamin E alone in the body.

(v) There was, however, association between cataract and low frequency of current use of protein.

Since many of these epidemiological findings are in accord with our nutritional studies, we comment in some detail on these.

Previous studies carried out by us at the National Institute of Nutrition (NIN) revealed a relationship between nutritional status and cataract[57, 58]. Such an association, beween cataract risk and low frequency of consumption of protein foods, was also reported earlier from other parts of India[40]. In undernourished cataract patients, i.e. those with decreased body mass index, the proportion of insoluble protein in the lens was significantly increased compared to that in well-nourished (with increased body mass index) patients while total protein remained unaltered, suggesting increased insolubilization of proteins in ocular tissue[57]. Further studies, involving protein fractionation, suggested that poor nutritional status in cataract patients accelerates the insolubilization process[58]. Our earlier work also suggested a protective role for some micronutrients, such as riboflavin[59, 60], zinc and copper[61], against cataractogenesis. No association was observed between cataract risk and vitamins like thiamine, pyridoxine and vitamin E in both studies[60, 62]. Both these points are confirmed in the Indo-US study.

A recent study[56] confirmed the observations regarding the B vitamin and cataract risk reported earlier by us[60]. In their study of 77 patients and 35 referents (controls) in the age group 40–70 years, the investigators found that higher concentrations of carotene and vitamin E were associated with reduced risk of cataract. However, unlike the Indo-US survey, Jacques *et al.*[56] did not find an association between cataract and antioxidant index including erythrocyte superoxide dismutase and glutathione peroxidase.

Although changes in glutathione reductase activity (as well as its activation coefficient as a measure of riboflavin status) were reported to reflect changes in erythrocyte as well as lens riboflavin levels in laboratory animals[63-65], the nature of such a relationship with respect to other nutrients in human subjects is not available at present. In the absence of a precise knowledge of such a relationship, it becomes difficult to interpret these results to explain their role in cataractogenesis. A recent study[66] showed higher glutathione reductase activity in the lens epithelium of cataract patients taking thyroxine or riboflavin supplements than in that of non-consumers of these agents, and suggests a positive correlation between riboflavin status and glutathione reductase activity. Thyroxine is known to have a role in the conversion of riboflavin to flavin dinucleotide, the cofactor of glutathione reductase. However, our recent study on brunescent cataracts revealed no significant difference in the activity of the enzyme, in either the whole lens[67] or its different regions, between yellow and brown lenses (Bhat *et al.*, unpublished results). Riboflavin status, as measured by activation coefficient ratio, was also found to be normal in both groups of cataractous lenses. Analysis of lens and erythrocytes from the same subjects with cataract might throw further light on the relationship between riboflavin status and cataract. It appears possible that in cataract the oxidant–antioxidant equilibrium is shifted towards oxidant stress and there might be an increased demand for vitamins, trace metals and enzymes concerned with meeting such oxidant stress. It is hoped that restoration of antioxidants by dietary manipulation may delay the onset of oxidative stress-induced cataract in humans.

There is an association between increased blood pressure and nuclear (N) and mixed (M) cataracts

This is in keeping with earlier studies that reported such an association[68, 69], though one of them[68] found higher systolic blood pressure in persons with P-type cataracts than in persons without lens opacities. The present survey gives the following odds ratios for hypertension cases in Delhi: 1.44 for N- and M-type cataracts, 1.16 for C, and 1.07 for P. The propensity in this case for nuclear and mixed cataracts is a new finding and needs further clarification. (Here again, the intriguing molecular question would be whether the nuclear γ-crystallin is more vulnerable in the present circumstances than, say, α-crystallin.) There is some evidence, however, that brunescent cataracts (nuclear cataracts with a high degree of scattering plus dark pigmentation) increased sharply depending on the geographic latitude of the eye camps[70].

Taking one or more aspirin tablets a month appears to protect against posterior subcapsular cataracts

This is a controversial issue not only in cataract research but in medical research in general. Cotlier and Sharma[29, 71, 72] had proposed aspirin as an anticataract agent. Harding's group[73] has suggested that aspirin prevents the carbamylation of soluble lens proteins and prevents cyanate-induced phase separation opacities *in vitro* (see Figure 7). Sen and Chakrabarti[74] studied the conformational stability of the crystallins upon acetylation with aspirin, and found the thermodynamic stability of α_L and γ-crystallins to be reduced. α_L-Crystallin aggregated at lower temperature upon incubation with aspirin. It is not clear how these results may tie in with the Delhi finding of a lower odds ratio for P-type cataracts. Other population-based studies have found no evidence of aspirin prophylaxis against cataract[75–77]. The Oxford-based randomized six-year trial of prophylactic daily aspirin in over 3000 British male doctors revealed no statistically significant difference from control subjects in disease prevention or protection, for either myocardial infarction and stroke or incidence of cataract[78]. Further study on this problem is warranted, particularly in the light of Harding's suggestion that, in diarrhoea and dehydration, or in renal failure situations, the high levels of urea would lead to cyanate accumulation and consequent carbamylation of crystallins, and also in the light of the very high risk of cataract due to diarrhoea and dehydration in the Indian context[42].

Burning cheap cooking fuels such as cowdung or wood leads to an increased risk of cataract of the C, N and M types

It is not easy to rationalize or explain this finding right away. One might suspect that the smoke and fumes, and the higher levels of particulates from burning these fuels — all of which are absent or negligible when cooking gas is used as the fuel — are involved. But then, as the study group itself pointed out, this finding is to be viewed in the larger context of the socioeconomic status of the individuals, since the study also finds *that there is a strong association between lower educational achievement and cataract.* Socioeconomic variables related to educational achievement are a likely explanation of this finding. In this Indian context, and particularly in the urban areas of Delhi where the study was made, lower educational levels usually also mean lower economic brackets, lower quality of living, lower levels of nutrition and diet, poorer sanitation, and higher rates of infection and illness.

Smoking as a behavioural variable significantly associated with more cataract types in univariable analysis

Smoking was selected along with dietary protein, aspirin use, marital status, education and cooking fuel as a six-variable input for the PLR model analysis. Smoking was significantly associated with more cataract types in univariable analysis. So were aspirin use, marital status, education and fuel. Subsequently, single PLR model analysis left aspirin, education, fuel and dietary protein as significantly associated with one or more cataract types, and were retained for continued analysis. However, that smoking is associated with cataract is a tantalizing finding, since it has recently been shown[79] that quitting smoking can reduce the risk of prematurely developing cataract by half. It is believed that substances in inhaled cigarette smoke are carried internally to the eye and may contribute to the damage. It would also be of interest to study whether a similar pathway might operate in the case of smoke inhaled from cheap cooking fuels.

1. Kupfer, C., *Eye*, 1984, **104**, 1.
2. Siva Reddy, P., Keynote address at the National Workshop on Current Priorities in Eye Research, Chandigarh, India, October 1989.
3. Olson, D., *Clinical Opthalmology* (ed. Duane, T. D.), Harper and Row, Philadelphia, 1986, Chapter 71, p.1.
4. Worg, W. L., in *Biomedical Foundations of Opthalmology* (eds. Duane, T. D. and Jaeger, E.I.), Harper and Row, Philadelphia 1986, Chapter 15, p. 1.
5. Moses, R. A., *Adler's Physiology of the Eye*, C. V. Mosby Co, St. Louis, 1981, p. 304.
6. Phelps, C. D., in *Clinical Opthalmology* (ed. Duane, T. D.), Harper and Row, Philadelphia, 1986, Chapter 72, p. 1.

7. Lentz, M. H., *Clinical Opthalmology* (ed. Duane, T. D.), Harper and Row, Philadelphia, 1986, Chapter 73, p. 1.
8. Maisel, H., Harding, C. V., Alcala, J. A., Kusjak, J. and Bradley, R., in *Molecular and Cellular Biology of the Eye Lens* (ed. Bloemendal, H.), John Wiley, New York, 1981, p. 49.
9. Blundell, T. *et. al.*, *Nature*, 1981, **289**, 771.
10. Slingsby, C., Driessen, H. P. C., Mahadevan, D., Bax, B. and Blundell, T., *Exp. Eye Res.*, 1988, **46**, 375.
11. Siezen, R. J. and Berger, H., *Eur. J. Biochem.*, 1978, **91**, 397.
12. Tanaka, T., Ishimoto, C. and Chylack, L. T., Jr, *Science*, 1977, **197**, 1010.
13. Siezen, R. J., Fisch, M. R., Slingsby, C. and Benedeck, G. B., *Proc. Natl. Acad. Sci. USA*, 1985, **82**, 1701.
14. Harding, J. J., in *Molecular and Cellular Biology of the Eye Lens* (ed. Bloemendal, H.), Wiley-Interscience, New York, 1981, p. 327.
15. Hoenders, H. J. and Bloemendal, H., in *Molecular and Cellular Biology of the Eye Lens* (ed. Bloemendal, H.), Wiley-Interscience, New York, 1981, p. 279.
16. Zigman, S., Groff, J., Yulo, T. and Griess, G., *Exp. Eye Res.*, 1976, **23**, 555.
17. Bessems, G. J. H., Keizer, E., Wollensak, J. and Hoenders, H. J., *Invest. Ophthalmol. Vis. Sci.*, 1987, **28**, 1157.
18. Pirie, A., *Biochem. J.*, 1971, **125**, 203.
19. Zigman, S. and Paxhia, T., *Exp. Eye Res.*, 1988, 47, 819.
20. von Heyningen, R., in *The Human Lens—in relation to Cataract*, Ciba Foundation Symposia, (eds. Elliot, K. and Fitzsimmons, D.W.), Elsevier, Amsterdam, 1973, vol. 19, p. 151.
21. Hayase, F., Nagaraj, R. H., Miyata, S., Njoroge, G. and Monnier, V. M., *J. Biol. Chem.*, 1989, **263**, 3758.
22. Dayhaw-Barker, P. C., *Photochem. Photobiol.*, 1987, **46**, 1051.
23. Murali Krishna, C., Riesz, P., Zigler, J. S. and Balasubramanian, D., manuscript under preparation.
24. Zigman, S., Schultz, J. and Yulo, T., *Exp. Eye Res.*, 1973, **15**, 201.
25. Hollows, F. and Moran, D., *Lancet*, 1981, **2**, 1249.
26. Sliney, D., *Invest. Ophthalmol. Vis. Sci.*, 1986, **27**, 781.
27. Taylor, H. R., West, S. K. and Rosenthal, F. S., *N. Engl. J. Med.*, 1988, **319**, 1429.
28. Kuck, J. F. R., Jr, *Biochemistry of the Eye*, Academic Press, London, 1970, p. 319.
29. Cotlier, E. and Sharma, Y. R., *Can. J. Ophthalmol.*, 1981, **16**, 113.
30. Harding, J. J., *Adv. Protein Chem.*, 1985, **37**, 247.
31. Harding, J. J., in *Ageing of the Lens* (eds. Regnault, F., Hackwin, O. and Courtois, Y.), Elsevier, Amsterdam, 1980, p. 71.
32. Monnier, V. M., Stevens, V. J. and Cerami, A., *J. Exp. Med.*, 1979, **150**, 1098.
33. Pande, A., Garner, W. H. and Spector, A., *Biochem. Biophys. Res. Commun.*, 1979, **89**, 1260.
34. Kinoshita, J. H., *Invest. Ophthalmol. Vis. Sci.*, 1974, **13**, 713.
35. Crabbe, M. J. C., Wolff, S. P. and Lloyd, P., in *The Lens: Transparency and Cataract.* (ed. Duncan G.), EURAGE, Rijswijk, The Netherlands, 1986, p. 119.
36. Mohan, M., *et al.*, *Arch. Ophthalmol.*, 1989, **107**, 670.
37. Jamieson, S. W., *Postgrad. Med. J.*, 1970, **46**, 557.
38. Franken, S. and Mehta, K. R., A Survey to Determine the Ophthalmic Morbidity in the Population Living in the Dry Belt of Southern Punjab and Haryana, Christian Medical College, Ludhiana, India, 1968.
39. Chatterjee, A., in *The Human Lens — in relation to Cataract*, Ciba Foundation Symposia, Elsevier, Amsterdam, 1973, vol. 19, p. 265.
40. Chatterjee, A., Milton, R. C. and Thyle, S., *Br. J. Ophthalmol.*, 1982, **66**, 35.
41. Brilliant, L. B., *et al.*, *Am. J. Epidemiol.*, 1983, **118**, 250.
42. Minassian, D. C., Mehra, V. and Jones, B. R., *Lancet*, 1984, **7**, 751.
43. Zigman, S., Datiles, M. and Torczynski, *Invest. Ophthalmol. Vis. Sci.*, 1979, **18**, 462.
44. Mitra, A. P., personal communication.
45. Hightower, K. R., in *The Lens: Transparency and Cataract* (Ed. Duncan, G.), EURAGE, Rijswijk, The Netherlands, 1986, p. 87.
46. Lorand, L., Conrad, S. M. and Velasco, P. T., *Biochemistry*, 1985, **24**, 1525.
47. Sharma, Y., *et al.*, *J. Biol. Chem.*, 1989, **264**, 12794.
48. Singh, D., in *Proceedings of the Seventh Asia-Pacific Academy of Ophthalmology Congress*, 1980.
49. Harding, J. J. and Rixon, K. C., *Metabol. Pediat. Ophthalmol.*, 1981, **5**, 161.
50. Kinoshita, J. H. and Merola, L. O., *Invest. Ophthalmol.*, 1973, **12**, 544.
51. Harding, J. J. and Rixon, K. C., *Exp. Eye Res.*, 1980, **31**, 567.
52. Project Advisory Committee Report, Bhopal Gas Disaster Research Centre, Indian Council of Medical Research, 1989.
53. Mohan Rao, Ch., Balasubramanian, D. and Chakrabarti, B., *Photochem. Photobiol.*, 1987, **46**, 511.
54. Chakrabarti, B. and Mandal, K., *Proc. SPIE: Lasers in Medicine*, 1986, **712**, 154.
55. Taylor, H. R. *et al.*, *N. Engl. J. Med.*, 1988, **319**, 1429.
56. Jacques, P. F., Hartz, S. C., Chylock, L. T., Jr, McGandy, R. B. and Sadowski, J.A., *Am. J. Clin. Nutr.*, 1988, **48**, 152.
57. Bhat, K. S., *Br. J. Nutr.*, 1982, **47**, 483.
58. Bhat, K. S., *Exp. Eye Res.*, 1983, **37**, 267.
59. Bhat, K. S. and Gopalan, C., *Nutr. Metabol.*, 974, 17, 1.
60. Bhat, K. S., *Nutr. Rep. Int.*, 1987, **36**, 685.
61. Bhat, K. S., *Nutr. Rep. Int.*, 1988, **37**, 157.
62. Bhat, K. S. and Nadiger, H., *Indian J. Clin. Biochem.*, 1986, **1**, 124.
63. Sauberlich, H. E., Dowdy, R. P. and Skala, J. H., *CRC Crit. Rev. Lab. Sci.*, 1973, **4**, 215.
64. Bamji, M. S., in *Vitamins in Human Biology and Medicine* (ed. Briggs, M. H.), CRC Press, Boca Raton, Florida, USA, 1981, p.1.
65. Bhat, K. S. and Rao, P. V., *Nutr. Res.*, 1987, 7, 1203.
66. Horwitz, J., Dovrat, A., Straatsma, B. R., Revilla, P. J. and Lightfoot, D. O., *Curr. Eye Res.*, 1987, **6**, 1249.
67. Bhat, K.S., John, A. and Reddy, V. N., *Proc. Intl. Soc. Eye Res.*, 1985, **5**, 130.
68. Hiller, R., Sperduto, R. D. and Ederer, F., *Am. J. Epidemiol.*, 1986, **124**, 916.
69. Kahn. H. A., *et al.*, *Am. J. Epidemiol.*, 1977, **106**, 33.
70. Zigman, S., Datiles, M. and Torczynski, E., *Invest Ophthalmol. Vis. Sci.*, 1979, **18**, 462.
71. Cotlier, E., *Lancet*, 1981, **1**, 338.
72. Cotlier, E., Sharma, Y. R., Niven, T. and Brescin, M., *Am. J. Med.*, 1983, **74**, 83.
73. Crampton, M., Rickson, K. C. and Harding, J. J., *Exp. Eye Res.*, 1985, **40**, 297.
74. Sen, A. C. and Chakrabarti, B., *Invest Ophthalmol. Vis. Sci.*, 1989, **30**, 3305.
75. Seigel, D., Sperduto, R. D. and Ferris, F. L., *Ophthalmology*, 1982, **89**, 47A.
76. West, S. K., Munoz, B. E., Newland, H. S., Emmet, E. A. and Taylor, H. R., *Arch Ophthalmol.*, 1987, **105**, 1229.
77. Klein, B. E., Klein, R. and Moss, S. E., *Diabetes Care*, 1987, **10**, 495.
78. Peto, *et al.*, *Br. Med. J.*, 1988, **296**, 313.
79. West, S. K., Munoz, B. E., Emmet, E. A. and Taylor, H. R., *Arch. Ophthalmol.*, 1989, **107**, 1166.