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Age dependent increase in UVA-induced bleaching of the brown components of the human lens

With age, human lenses accumulate a yellow color. This yellow color is a risk-marker and possibly risk-factor to the formation of cataracts. The yellow color, that is accumulated, is at its highest concentration in the lens nucleus, which is also the major site of cataracts, which may be related to UVA light. Evidence suggested that the yellow color accumulates when lens proteins react with the oxidation products of ascorbic acid. In this process, the oxidation of ascorbic acid causes modification of lens proteins, which is called glycation. Glycation is the reaction of carbonyl compounds and lysine residues in proteins. Glycation results in yellow compounds, which can absorb UVA light. Those products are called Advanced Glycation End products (AGEs). Previous research has shown that *in vitro* UVA irradiation of lenses causes bleaching of their yellow color. This is due to reduction of the yellow AGEs in the absence of oxygen, along with a concomitant oxidation of ascorbic acid. Providing that UVA light makes up ninety-seven percent of our solar spectrum and that 1000 times more UVA light reaches the lens than UVB light, it is likely that UVA light is the major factor for photochemical transformations in human lens. The aim of present investigation is to study the effect of UVA-light irradiation on human lens proteins of different ages. Proteins from human lenses of four different age groups(0-20yrs, 20-40yrs, 40-60yrs, 60+yrs) were separated into Water Insoluble(WI) and Water Soluble(WS) fractions. Amount of protein, absorbance at 330 nm and fluorescence at excitation/emission = 350/450 nm were measured for all fractions. To measure the activity, or the effectiveness of these fractions to oxidize ascorbic acid in the absence of oxygen, samples containing 1 mg/ml of WI and WS proteins were irradiated by UVA light, at 17 C^{o}, for one hour. Spectra in the range 200-400 nm were taken every 15 minutes. Ascorbic acid oxidation was measured by the absorbance at 265 nm. Irradiation of lens proteins also caused the yellow compounds to bleach, which was followed by the absorbance at 330 nm. Dark controls were prepared and treated in the same way except that they were kept in the dark instead of being irradiated. A decrease in absorbance at 265nm was observed of the irradiated samples. In contrast, there was no significant change in the dark controls at 265nm. The data argues that oxidation of ascorbic acid was caused by UVA light and that a photochemical reaction occurred due to the irradiation of the samples. Similarly, a decrease in absorbance at 330 nm was observed in the irradiated samples. The dark controls remained unchanged, which indicates that the bleaching of the yellow compounds was caused by the irradiation of the samples. In older lenses a larger percent loss of absorbance at 330 nm was observed compared to younger lenses. This result correlates with the higher absorbance per milligram in older lenses. There is also an increase in moles of ascorbic acid oxidized with age. WI and WS proteins were examined to find out where the yellow compounds are located. Data from the experiments will be further analyzed to determine how age and solubility affects the activity in lenses. Current data shows that proteins in older lenses are more susceptible to UVA light than younger lenses. The increased oxidation of ascorbic acid in older lenses will further accelerate formation of glycation products, which are potential protein cross-linkers, and cause of cataract formation.