Albrecht v. Graefes Arch. klin. exp. Ophthal.
204, 67–72 (1977)

Graefes Archiv
für klinische und experimentelle
Ophthalmologie

Investigations on Trace Elements in Normal and Senile Cataractous Lenses

Activation Analysis of Copper, Zinc, Manganese, Cobalt, Rubidium, Scandium, and Nickel

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Summary. A total of 125 human lenses (97 senile cataractous and 28 clear) were analyzed by means of neutron activation analysis for trace element concentrations of copper, zinc, manganese, cobalt, rubidium, scandium, and nickel. It was found that the rubidium concentration remarkably decreased and the copper concentration moderately increased with the progression of cataract. The concentrations of manganese, zinc, and cobalt showed some fluctuations without a definite increase or decrease, and the scandium and nickel content was found to be very low.

Introduction

The inorganic ion content of clear and cataractous lenses has been the subject of several investigations, and especially the potassium, sodium, and calcium ions have been analyzed in detail (Van Heyningen, 1972; Maraini and Mangili, 1973; Maraini and Torcoli, 1974; Stankiewicz, 1974; Duncan and Bushell, 1975; Jedziniak et al., 1976; Rácz and Kellermayer, 1977). A few experiments have been concerned with the trace element contents of lenses, and changes in their concentrations have been found in cataractous lenses (Shlopak, 1962; Nath et al., 1969; Swanson and Truesdale, 1971; Murata et al., 1972; Murata and Taura, 1975). Unfortunately, the reported data are not unambiguous; e.g., as to the copper content, both remarkable increase (Shlopak, 1962; Nath et al., 1969) and decrease (Swanson and Truesdale, 1971) have been observed, while Murata et al. (1972) and Murata and Taura (1975) could not find any change in the copper level. Similarly, for zinc, a reported significant decrease (Shlopak, 1962; Swanson and Truesdale, 1971) is contradicted by reports of slightly increased values (Murata et al., 1972; Murata and Taura, 1975). There are few data on manganese, cobalt, and nickel, and none at all on rubidium and scandium. On the base of these contradictory data it seemed

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interesting to reinvestigate the trace element contents in normal and different types of senile cataractous lenses by means of activation analysis. The present work covered the following trace elements: manganese, copper, zinc, cobalt, rubidium, scandium, and nickel.

Material and Methods

A total of 125 lenses were analyzed, 97 of which belonged to patients with different types of senile cataract, the rest being clear. The ages of the patients varied from 40 to 90 years, except that one of the clear lenses belonged to a child of 3 years.

Senile cataractous lenses were classified before the operation by slit-lamp examination with maximum mydriasis. The samples were grouped as follows:
I. Nuclear cataract – the cortex remains clear and nuclear opacity appears yellow-brown
II. Cortical cataract – the nucleus remains clear
III. Nuclear plus cortical cataract
IV. Complete opacity.

Normal lenses were obtained from eyes enucleated because of tumors of the posterior pole and removed from cadaver eyes within in 4 h after death. The cataractous lenses with intact capsule were removed by cryoextraction and immediately analyzed. The single lenses were dried at 105° C to constant weight and always weighed before (wet weight) and after drying (dry weight).

Neutron activation analysis was conducted on each lens. The irradiations were performed in a vertical channel of the VVR-SZ-type reactor of the Central Research Institute for Physics, Budapest at a thermal neutron flux of approximately 3×10^{13} n/cm^2 s. The short-time irradiations were performed by the aid of a pneumatic tube system built into the core of the reactor. Because of the nuclear properties of the analyzed elements not all of them could be determined in a single lens, thus three lenses were needed for the determination of seven elements.

For the determination of manganese, a single lens was irradiated for 6 min, then destroyed in a mixture of sulfuric acid and concentrated nitric acid in the presence of inactive manganese carrier. A two-step chemical separation followed the destruction: first, manganese sulfide was precipitated from the hot alkaline solution. The precipitate was filtered, washed once with water saturated with hydrogen sulfide, then dissolved in 1 N hydrochloric acid. The hydrogen sulfide was boiled out from the solution and manganese ammonium phosphate monohydrate was precipitated from the solution neutral to methyl red. The gamma spectrum of this precipitate was measured.

For the analysis of copper, a single lens was likewise used. It was irradiated for 1 h, then destroyed as for manganese. The destroyed sample was diluted in distilled water, and copper (I) thiocyanate was precipitated from a slightly acidic system upon the addition of sodium sulfide reducing agent and of solid potassium thiocyanate. If the sample was well destroyed, the precipitate was purified from ^{24}Na by a factor close to 10^5 and a radiochemically pure precipitate was obtained in a single step.

The long-lived isotopes were measured after 115 h irradiation and cooling for 10 days, without destruction of the sample.

The measurements were made with 45 cm^3 Nuclear Diodes Ge/Li semiconductor detector and ICA 70 4096-channel analyzer. For nondestructive gamma spectrometry (Rb, Zn, Ni, Co, Sc) the 2048-channel range of the analyzer was used, and for the measurement of Cu and Mn, the 1024-channel range.

Table 1. Trace element amounts in clear and different types of senile cataractous lenses

Clear lenses			Nuclear cataract			Cortical cataract		
	µg/g wet weight	µg/g dry weight	µg	µg/g wet weight	µg/g dry weight	µg	µg/g wet weight	µg/g dry weight
Cu 0.045 (7) ±0.0082	0.20 (7) ±0.045	0.58 (7) ±0.10	0.071 (4) ±0.033	0.32 (4) ±0.15	0.72 (3) ±0.29	0.051 (13) ±0.017	0.27 (13) ±0.092	0.73 (13) ±0.23
Mn 0.012 (5) ±0.0044	0.058 (5) ±0.023	0.19 (5) ±0.076	0.008 (3) ±0.0046	0.045 (3) ±0.023	0.13 (3) ±0.067	0.012 (12) ±0.0062	0.071 (12) ±0.034	0.19 (12) ±0.086
Zn 1.52 (8) ±0.29	8.2 (8) ±1.05	25.4 (8) ±9.8	1.62 (5) ±0.54	6.9 (5) ±1.2	19.3 (5) ±4.7	1.69 (10) ±0.23	9.1 (10) ±1.9	24.0 (10) ±5.4
Rb 0.41 (8) ±0.17	2.2 (8) ±0.98	6.8 (8) ±3.1	0.29 (6) ±0.12	1.5 (6) ±0.47	4.3 (6) ±1.7	0.32 (10) ±0.16	1.7 (10) ±0.77	4.6 (10) ±2.4
Co 0.00061 (8) ±0.00047	0.0029 (8) ±0.0021	0.0091 (8) ±0.0058	0.00071 (5) ±0.00020	0.0034 (5) ±0.0026	0.01 (5) ±0.0084	0.00047 (10) ±0.00045	0.0026 (10) ±0.0026	0.0071 (10) ±0.0074
Sc < 8.15^{-5}	–	–	< 8.10^{-5}	–	–	< 8.10^{-5}	–	–
Ni < 0.1	–	–	< 0.1	–	–	< 0.1	–	–

Nuclear plus coritcal cataract			Completely opaque lenses		
µg	µg/g wet weight	µg/g dry weight	µg	µg/g wet weight	µg/g dry weight
Cu 0.051 (13) ±0.026	0.25 (13) ±0.12	0.74 (13) ±0.28	0.046 (9) ±0.026	0.28 (9) ±0.17	0.88 (9) ±0.57
Mn 0.011 (3) ±0.0057	0.060 (3) ±0.028	0.19 (3) ±0.087	0.0087 (6) ±0.0042	0.054 (6) ±0.029	0.18 (6) ±0.10
Zn 1.42 (8) ±0.34	8.4 (8) ±3.13	25.6 (8) ±7.3	1.61 (8) ±0.34	8.2 (8) ±2.5	29.7 (8) ±7.95
Rb 0.60 (8) ±0.025	0.34 (8) ±0.12	1.09 (8) ±0.50	0.053 (9) ±0.044	0.29 (9) ±0.18	0.97 (9) ±0.52
Co 0.00032 (8) ±0.00024	0.0019 (8) ±0.0014	0.0057 (8) ±0.0037	0.00059 (8) ±0.00052	0.0024 (8) ±0.0013	0.0093 (8) ±0.0058
Sc < 8.10^{-5}	–	–	< 8.10^{-5}	–	–
Ni < 0.1	–	–	< 0.1	–	–

Values are expressed as mean ± SD.
The numbers of samples are shown in parentheses

The experimental procedure was checked by use of the Bowen's kale (international biological standard) sample (Ördögh, 1976). The reliability of the method was also confirmed by the data obtained on clear lenses of cadavers, which were expected to be practically equal.

Table 2. Data obtained on clear pairs of lenses

Patient	Sample Age	Sex	No	g wet weight	g dry weight	Cu μg	Mn μg	Zn μg	Rb μg	Co μg	Sc μg	Ni μg
K.J.	62	♀	o.d.	0.2155	0.0772	0.0566						
			o.s.	0.2150	0.0801	0.0464						
D.D.	62	♀	o.d.	0.2322	0.0743	0.035						
			o.s.	0.2197	0.0761	0.035						
L.J.	70	♀	o.d.	0.2007	0.0569		0.0074					
			o.s.	0.1987	0.0560		0.0066					
Dr.Sz.N.	78	♀	o.d.	0.2060	0.0740			1.76	0.63	0.0015	8.15^{-5}	0.1
			o.s.	0.2320	0.0764			1.81	0.58	0.0011	8.10^{-5}	0.1
S.A.	49	♂	o.d.	0.2240	0.0686			1.87	0.29	0.00055	8.10^{-5}	0.1
			o.s.	0.2239	0.0694			1.57	0.24	0.00061	8.10^{-5}	0.1
B.G.	50	♀	o.d.	0.1871	0.0686			1.31	0.32	0.00029	8.10^{-5}	0.1
			o.s.	0.1865	0.0674			1.05	0.25	0.00020	8.10^{-5}	0.1

o.d. = *oculus dexter*; o.s. = *oculus sinister*

Results

Table 1 shows the values obtained for the analyzed elements, as averaged over the number of samples given in parentheses. (The Bowen's kale analysis showed the following standard errors of the determinations: 6% for zinc, 8% for manganese, 5% for rubidium.) Each element shows fluctuations in either the positive or the negative sense, attributable in no way to the error of the analytical method in the case of the most important trace elements (Cu, Zn, Mn). Only the lower limits could be established for scandium and nickel. The analytical error of the cobalt determination was quite large, nevertheless the data provide sufficient information about the trace element contents of the lenses.

Clear pairs of lenses, taken from both eyes of cadavers were also analyzed. These measurements, which showed the reliability of the method, provided further useful data on clear lenses. The results are listed in Table 2. The amounts of trace elements are given here as found in single lenses. The values show a good agreement, proving the reliability of the method, and let us recall Table 1, where the trace elements of clear lenses obtained either from cadaver or by enucleation agree well, within the experimental error.

Discussion

In the present experiments we analyzed 97 senile cataractous and 28 clear human lenses by means of neutron activation analysis for several trace elements. The scandium and nickel content was found to be very low. The concentrations of manganese, zinc, and cobalt showed some fluctuations, but a definite increase or decrease could not be established. It was found that the rubidium concentration decreased remarkably and the copper concentration moderately increased with the progression of cataract.

At present we do not have enough information on the role of trace elements in human lenses in normal pathologic condition; even the available data concerning the changes in the trace element concentration of clear and cataractous lenses may be considered ambiguous. As to the copper, increase as well as decrease and no change have been found in senile cataractous lenses, as was mentioned above (Shlopak, 1962; Nath et al., 1969; Swanson and Truesdale, 1971; Murata et al., 1972; Murata and Taura, 1975; Kanai et al., 1974). At the present stage of our experiments no attempt can be made to explain the observed increase in the copper concentration converted to wet and dry weight of lenses.

Though the rubidium content of clear and cataractous lenses is lower by orders of magnitude, compared with their potassium content, simultaneous reference to the two ions is assumed to be reasonable. The rubidium concentration decreases remarkably with the progression of cataract and is reduced by more than a factor of 7 in group IV, as compared to the rubidium content of the clear lens. Similarly, in the case of complete opacity the potassium content of the lens decreased from the normal value by a factor of about 7, as found by our flame Photometric measurements (Rácz and Kellermayer, 1977). The two ions seem to leave the increasingly opaque lens at almost the same rate. This fact is a further confirmation of the observation that rubidium and potassium show similar behavior in biologic system (Solomon, 1952; Love and Burch, 1953).

In biochemistry the trace elements are usually discussed as key components of active enzymes. Our data cannot confirm or contradict this interpretation and, for the time being, no one has been able to establish whether the changes in the concentration of trace elements in cataractous lenses are prior or concomitant events.

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Received March 28, 1977