Regional Variation in Copper Content of Arterial Wall

Copper is unique in biological systems, as it forms an integral part of the electron-transfer oxidase which catalyze the reduction of molecular oxygen to water[1]. This element also promotes lipid autooxidation and thereby atherosclerosis[2]. Contrary to ITO[2], SCHROEDER et al.[3] reported a significant decline in the concentration of copper in aorta with increasing age and degree of atherosclerosis. Recently RAO[4] observed a significant variation in the magnesium content of different arteries. With these considerations the present work was undertaken.

Arterial samples removed at autopsy of 18 cases were analyzed. Each sample was roughly 1.5–2 cm in length. Aorta was taken about 2 cm above the origin of left renal artery. Pulmonary, renal, common carotid and illiac arteries of right side were also removed. Cross grading of atherosclerosis of all arteries was done by ITO's[2] modification of W.H.O. classification.

From the arterial samples, adventitia was removed and fat-free extracts of each of them were prepared by the methods described[5,6]. These were then weighed and ashing was carried out[4]. Dry ashing was preferred to the wet, as the latter has many disadvantages like contamination and lesser degree of accuracy[7]. Copper is a non-volatile element and hence dry ashing is a reliable procedure. Ash so obtained was dissolved in 3 ml of hydrochloric acid and copper content was determined[8]. Final results were calculated for copper content in 1000 g of fat-free dry weight of artery (FFDW) and the differences were statistically evaluated by Students' *t*-test (Table).

HARMEN[9] reported that the serum copper shows a linear rise with age and suggested that this change may increase the rate of lipid peroxidation in geriatric subjects. Serum copper level is also elevated in various vascular diseases[10,11]. Recently WAISMAN et al.[12] have shown that animals kept on a copper restricted diet survived with scarred vessels, but demonstrated a tendency towards premature atherosclerosis. Interestingly enough, in the present study copper content of all arteries decreased linearly with the increase in the degree of atherosclerosis. Moreover, copper content was highest in the pulmonary artery which had minimal degree of atherosclerosis, and was lowest in aorta and illiac arteries which exhibited severe grades of atherosclerosis.

The present study indicates that, though copper promotes atherosclesosis[2], its concentration in the arterial wall declines with the increase in the grade of atherosclerosis in various arteries. The mechanisms which either mobilize copper from arterial wall or inhibit its entrance into it are yet unknown.

Number of specimens of various arteries (shown in bracket) and mean average of copper-content of various arteries as related to degree of atherosclerosis of aorta. Copper-content in mg per 1000 g of FFDW of arteries

Grade of atherosclerosis	Aorta	Pulmonary	Illiac	Carotid	Renal
Grade I	$9.31\pm0.34^{c,d}$ (7)	$13.2\pm0.2^{a,f}$ (7)	9.56 ± 0.34 (6)	9.84 ± 0.6 (5)	9.56 ± 0.49 (6)
Grade II	8.9 ± 0.45 (6)	12.24 ± 0.6 (5)	9.0 ± 0.6 (5)	9.7 ± 0.7 (6)	9.35 ± 0.34 (6)
Grade III	7.4 ± 0.5 (5)	11.54 ± 1.54 (5)	9.6 ± 0.42 (5)	9.9 ± 0.5 (4)	9.75 ± 0.6 (4)
Average of 3 grades	$8.64\pm0.41^{c,e}$ (18)	12.43 ± 0.71 (17)	$9.35\pm0.45^{c,g}$ (16)	$9.52\pm0.67^{c,g}$ (15)	$9.39\pm0.49^{c,g}$ (16)

[a] $P<0.05$. [b] $P<0.005$. [c] $P<0.001$. [d] Between grade I and III of aorta. [e] Between aorta and pulmonary-artery. [f] Between grade I and III of pulmonary-artery. [g] Between pulmonary-artery and illiac, carotid or renal arteries.

Résumé. La teneur en cuivre dans l'aorte et les artères pulmonaire, rénale, illiaque et carotide a été déterminée. Une corrélation inverse entre le degré d'athérosclérose et celui du cuivre a été observée. La cause de ce phénomène reste à être établie.

J. D. KHANDEKAR, D. P. MUKERJI, G. D. NAIK and G. C. SEPAHA

M.G.M. Medical College and M.Y.H., Indore (India), 23 December 1971.

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Potentiation of Fever, Produced by Intravenous Leucocyte Pyrogen, Following the Injection of Paraffin Oil in the Cerebral Ventricles of the Unanesthetized Rabbit[1]

When leucocyte pyrogen[2] is injected into the brain of unanesthetized rabbits, the greatest increases in temperature, with the shortest latency, occur following injections into the anterior hypothalamic region[3,4]. There is recent evidence to suggest that leucocyte pyrogen, when given intravenously, enters the hypothalamus directly from the blood stream rather than via the cerebrospinal fluid (CSF)[5]. We now present evidence to suggest that fluid flow in the cerebral ventricular system could play a role in the termination of the febrile response.

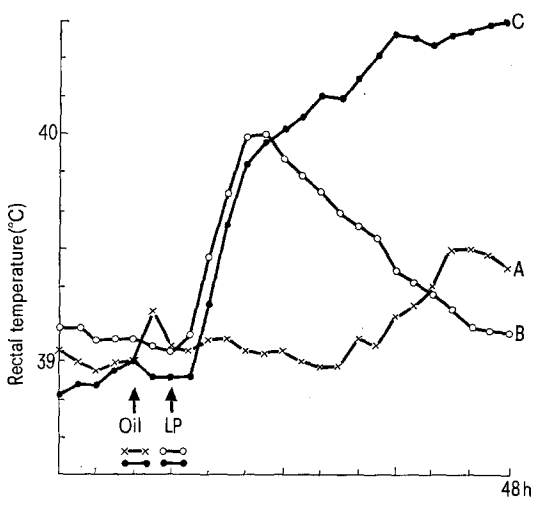
Materials and method. In each of 38 New Zealand white rabbits weighing between 2.6 and 3.6 kg, a Collison cannula, with no shaft, was screwed into the skull above the left lateral cerebral ventricle[6]. Following a recovery period of 1 week, rectal temperature was recorded and 0.15–0.3 ml sterile paraffin oil was injected into the lateral cerebral ventricle. A dose of leucocyte pyrogen[2], which had previously been shown to usually produce an increase in temperature of approximately 0.6 to 0.8 °C, was injected into an ear vein 15 to 45 min later. At the end of a series of experiments, paraffin oil mixed with *x*-ray-opaque oil was injected in volume of 0.15 to 0.3 ml into the lateral cerebral ventricle and radiographs were taken of the head in 2 planes to determine its spread through the ventricular system.

Results and discussion. Following the injection of paraffin oil into the lateral ventricle the time of onset of the temperature increase was 15.02 mean $\pm$ S.E. 1.13 min whereas that observed when leucocyte pyrogen was given without oil was 13.73 $\pm$ S.E. 0.91 min ($t = 0.88$; $df = 40$; $p > 0.1$). The mean and standard error of the maximum temperature increase within 2 h following an injection of leucocyte pyrogen alone were 0.74 $\pm$ 0.04 °C and those for leucocyte pyrogen following oil were 0.95 $\pm$ 0.06 °C ($t = 3.55$; $df = 40$; $p < 0.001$). This difference is equivalent to a very large increase in the dose of leucocyte pyrogen since the response is a logarithmic function of dose[2].

Comparisons of the duration of the febrile response showed that, of the 41 paired experiments, fever after oil and leucocyte pyrogen lasted longer in 32 experiments, and in 9 there was no change from that observed when leucocyte pyrogen was given alone. Of the 32 mentioned, 8 were recorded and remained febrile for either 1 or 2 days. Of the remaining animals, 6 died the following day although their temperature was not recorded. It is clear, therefore, that the application of standard statistical tests to this aspect of the results may not be appropriate but it is evident that there is a major difference in the duration of fever between the two conditions. In 15 control experiments in which oil was given alone there was essentially no change in the animals' temperature (mean + 0.08 $\pm$ S.E. 0.07 °C).

Typical temperature responses are illustrated in the Figure for 1 rabbit in 3 experiments each separated by a minimum of 48 h. Trace A (× — ×) is a record of the largest body temperature response observed following an injection of 0.15 ml sterile paraffin oil into the cerebral ventricle. Trace B (□ — □) shows the 1.0 °C increase in temperature following 0.5 ml leucocyte pyrogen, injected into an ear vein, with no oil in the ventricles. If 0.5 ml leucocyte pyrogen was given i.v. 20 min after an injection of oil into the cerebral ventricle, the temperature increased by 1.5 °C as shown by trace C (● — ●), and remained elevated for the next 2 h. X-ray examination of the brains of the rabbits, following injections of *x*-ray opaque oil mixed with paraffin oil, showed that the oil had filled the ventricular spaces including the ventral portion of the third ventricle.

Since there was no difference in time of onset of the febrile response to leucocyte pyrogen between the 'oil' and 'control conditions', it would appear that the oil was not interfering with the entry of leucocyte pyrogen into the hypothalamic tissue. This supports the view that leucocyte pyrogen injected i.v. reaches the hypothalamus via the blood stream. With oil in the cerebral ventricles, the temperature response to the leucocyte pyrogen was of greater amplitude and of longer duration than it was under the control conditions. Thus the oil may have decreased the egress, via the ependymal wall, of the leucocyte pyrogen, its metabolites, or substances which it caused to be released from the hypothalamic tissue, into the CSF. Alternatively, if calcium ions or other substances are involved in the lowering of body temperature[6], then the oil may have interferred with the movement of these substances between the CSF and the hypothalamic tissue.



Temperature record obtained from one unanesthetized rabbit in 3 experiments, each separated by a minimum of 48 hours. In the experiment represented by: trace A (x–x), 0.15 ml sterile paraffin oil was injected into a lateral cerebral ventricle at the time indicated by the arrow; trace B (□—□), 0.5 ml leucocyte pyrogen was injected i.v. at the arrow; and trace C (●—●), 0.15 ml sterile paraffin oil was injected into the lateral ventricle at a time indicated by the first arrow followed by an i.v. injection of 0.5 ml leucocyte pyrogen at the time indicated by the second arrow.

Zusammenfassung. Potenzierung und Verlängerung der Wirkung des i.v. injizierten (Kaninchen) Leukozyten-Pyrogens konnte durch die sterile intraventrikuläre (laterale Ventrikel) Injektion von Paraffinöl ausgelöst werden, dies im Vergleich mit der Wirkung des Pyrogens bei mit Paraffinöl nicht behandelten Tieren. Diese Ergebnisse bringen eine Grundlage für ein besseres Verständnis des pyrogenen Wirkungsmechanismus von Leukozyten-Pyrogen.

K. E. COOPER and W. L. VEALE

Division of Medical Physiology,
Faculty of Medicine,
University of Calgary,
Calgary 44 (Alberta, Canada),
6 December 1971.

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