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ACCUMULATION OF GASEOUS LIPID PEROXIDATION PRODUCTS IN THE EXPIRED AIR IN CHILDREN DURING HYPERBARIC OXYGENATION

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KEY WORDS: hyperbaric oxygenation; lipids; pentane.

Hyperbaric oxygenation (HBO) has recently found successful application in clinical practice [2, 3, 5]. However, the widespread use of this method is prevented by the possibility of development of toxic effects of oxygen, which are based on damage to membranous structures of the cells of different tissues [1, 4, 6]. The key mechanism of this harmful action of HBO is activation of endogenous lipid peroxidation (LPO) [6]. Consequently, a problem of practical importance is the development and use of quantitative methods of estimating concentrations of LPO products in the human body. The most promising method in this respect is that of recording gaseous products of endogeneous LPO in the expired air, for their content, as experiments on animals have shown [8], adequately reflects the intensity of LPO in the body. The method consists essentially of recording volatile hydrocarbons (methane, ethylene, ethane, propane, pentane, etc.) that are formed in vivo during LPO as a result of successive decomposition of lipid hydroperoxides [7, 10]. An important advantage of this method compared with all other methods of analysis of LPO products is that it requires no intervention on the subject or procedures of isolation of cellular or lipid fractions, which are inevitably associated with the appearance of artefacts.

The aim of the investigation described below was to study the possibility, in principle, of using a noninvasive method of recording gaseous products of endogenous LPO in man during HBO.

EXPERIMENTAL METHOD

Concentrations of gaseous LPO products in the expired air were determined immediately before HBO and also at different times (10 min, 1 and 2 h) after sessions of HBO conducted on seven children aged from 5 to 14 years with tumors in various situatons. HBO was carried out in a type OKA-MT single-person therapeutic pressure chamber in pure oxygen at 1 atm. The saturation time was 60 min. Quantitative analysis of hydrocarbons in the expired air was based on the assumption that the chief substrates of LPO are ω-6-polyunsaturated fatty-acid residues of phospholipids and, correspondingly, that the principal gaseous product of LPO quantitatively is pentane [9]. Accordingly, to ensure maximal sensitivity of the method the pentane concentration in the expired air was determined. The expired air was collected in polyethylene bags with a capacity of 2 liters, previously ventilated with pure nitrogen. Or-

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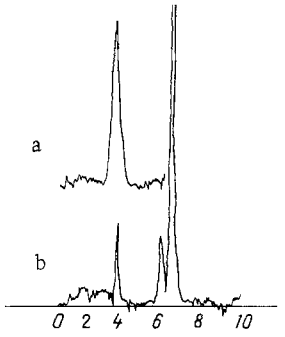

Fig. 1

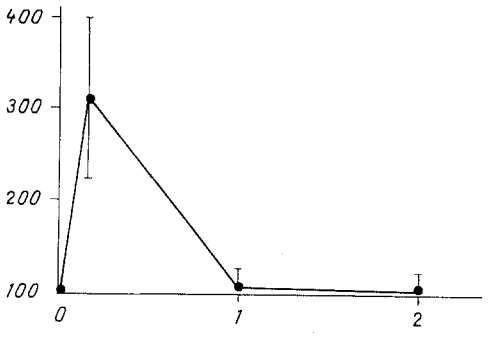

Fig. 2

Fig. 1. Typical chromatogram of expired air of a person before HBO (a) and 10 min after a session of HBO (b). Abscissa, elution time of peaks from separating column (in min).

Fig. 2. Pentane concentration in expired air of persons subjected to HBO. Abscissa, time after HBO session (in h); ordinate, accumulation of pentane (in % of initial level).

ganic substances contained in the samples were concentrated on a 40 × 40 mm column packed with "Tenax-Gc" 60-80 mesh (The Netherlands) – a sorbent which selectively traps organic compounds and allows inorganic compounds to pass through [11]. Air from the bag was passed through the column for 10 min at the rate of 30-40 ml/min. The absorbing column was then connected to the system of a gas chromatograph. The carrier gas, after passing through the absorbing column, which was intermittently heated to 300°C in 15 sec, entered a V-shaped capillary tube of the chromatograph injector, cooled with liquid nitrogen. During the next 3 min the capillary tube was heated for 60 sec to 300°C. Under these conditions compounds, including pentane, adsorbed in the capillary tube were vaporized and passed with the flow of carrier gas into the separating column of the chromatograph. A Perkin-Elmer F22 gas chromatograph (Sweden) with flame-ionization detector was used. Glass columns measuring 3 mm × 2 m were packed with F-1 Activated Alumina (Perkin-Elmer, Sweden). Helium (high purity grade, USSR) was used as the carrier gas. The rate of flow of helium was 10 ml/min, and of air 400 ml/min. The temperature of the column was 190°C, of the injector 210°C, and of the detector 220°C. Peaks were integrated on an M-1 Computing Integrator (Perkin-Elmer, Sweden). The elution time of pentane was 2-3 min. The error of the method did not exceed 5%.

EXPERIMENTAL RESULTS

A typical chromatogram of the expired air before and 10 min after HBO is shown in Fig. 1. The first peak on the left of both chromatograms corresponds to pentane. The average pentane concentration (in the control) before HBO was 0.08 ± 0.01 nmole/liter air. HBO caused activation of LPO, which was recorded as a two to threefold increase in the pentane concentration in the expired air (Fig. 2). This increase was observed immediately after the HBO session and was no longer present 1 h after hyperbaric oxygenation.

After HBO under the chosen conditions, activation of LPO thus had the character of a sudden short-term process. This differs essentially from the time course of accumulation of gaseous LPO products in man during stress caused by intensive intellectual activity with insufficient time available. It was shown, for instance, that the pentane concentration in the expired air increases by more than 1.3 times immediately after stress, reaches a maximum (by 1.7 times) on the 2nd day, and returns to its initial level only after 5 days [6].

To conclude, the method of recording the endogenous LPO level as reflected in the pentane concentration in the expired air is sufficiently sensitive to detect changes caused by HBO. The high specificity and sensitivity, and also the relative simplicity of the method make a further study of the possibility of its use with a view to selecting the optimal HBO program for clinical use, allowing for individual differences, worthwhile.

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EFFECT OF PREGNANCY ON STATE OF THE HEMOSTASIS SYSTEM IN INTACT AND THYMECTOMIZED RATS

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Hypercoagulation develops in women during pregnancy and fibrinolysis is inhibited [8, 9, 11]. Immunologic conflict between mother and fetus also frequently develops during pregnancy, and this is bound to aggravate disturbances in the hemostasis system [14].

For the reasons given above, the study of the role of the thymus, the central organ of cellular immunity, in the mechanism of development of coagulopathy during pregnancy is a matter of considerable interest, and the investigation described below was devoted to it.

EXPERIMENTAL METHOD

Experiments were carried out on 48 female rats. The thymus was removed from 24 of them at the age of 2 months. Mating of both thymectomized and intact females took place at the age of 4 months. All investigations were carried out in the last stage of pregnancy according to the following scheme: blood was taken simultaneously from nonpregnant intact (control 1) and thymectomized (control 2) rats and from pregnant intact (experiment 1) and thymectomized (experiment 2) rats.

The following parameters of the hemostasis system were determined: the blood clotting time and plasma recalcification time, the kaolin and cephalin times, prothrombin and thrombin times, total antithrombin activity, fibrinogen concentration, euglobulin fibrinolysis, and the ethanol test. The total platelet, leukocyte, and erythrocyte counts also were determined and the aggregometer tracing recorded. All the methods listed above are described in a recent textbook on investigation of the hemostasis system [1].

The numerical results were subjected to statistical analysis.

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