Hence, the results presented suggest that the sympathetic nervous system can modulate IL-1 production by the macrophages through the release of NGF, affecting both the immune cells and adrenergic neurons. Disturbances in these regulatory processes probably take place during carcinogenesis.

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EXPERIMENTAL BIOLOGY

Circadian Changes of the Hemocoagulation Indexes in Healthy Persons

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There have been few investigations of the circadian rhythm of hemocoagulation [2-5,8-11], and some of these studies were performed using obsolete techniques.

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In recent years refined methods of assaying hemocoagulation have been created, one of which is the anticoagulation test [13] in a modification [6] which assesses the kinetics of both the coagulation and anticoagulation processes. The hemolysate aggregation test [1] is suitable for studying platelet aggregation capacity.

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TABLE 1. Circadian Rhythm of Hemostasis Indexes in Healthy Persons Aged 18–57 Years (Confidence Interval)

Index	Mean	Amplitude	Acrophase
ACT:			
A, %	17.84 (17.36–18.32)	2.33 (1.67–3.00)	14.41 (12.58–15.35)
MA, %	89.92 (88.36–91.48)	6.65 (4.78–8.57)	14.75 (12.38–15.51)
T, min	9.86 (9.79–9.92)	0.24 (0.11–0.37)	2.93 (0.44–3.60)
TII	2.06 (2.04–2.07)	0.08 (0.06–0.11)	3.04 (1.40–4.30)
AT III, %	95.97 (94.33–97.60)	8.48 (6.95–9.99)	3.21 (2.03–4.24)
SFMC, +,–	0.18 (0.11–0.25)	0.31 (0.15–0.47)	15.72 (14.50–16.26)
Fibrinogen, g/liter	2.43 (2.39–2.47)	0.19 (0.17–0.22)	15.42 (15.03–15.45)
Thrombin time, sec	24.32 (24.12–24.52)	1.85 (1.57–2.13)	2.88 (1.40–3.59)
Prothrombin time, sec	24.21 (23.94–24.48)	1.68 (1.42–1.94)	2.79 (1.27–4.03)
Prothrombin index, %	78.74 (77.98–79.51)	5.50 (4.72–6.29)	14.75 (13.32–15.56)
Fibrinoligase activity, %	88.70 (87.42–89.97)	8.63 (6.80–10.47)	16.75 (15.57–17.29)
Fibrinolytic activity, min	135.18 (133.72–136.64)	10.20 (8.37–12.04)	15.65 (14.54–16.21)
HAT:			
10^3/ml, %	85.00 (82.88–87.11)	8.07 (5.71–10.43)	15.03 (13.50–15.58)
10^7/ml, %	96.09 (92.80–99.37)	11.44 (8.75–14.12)	15.22 (14.03–16.10)
TAI	1.13 (1.12–1.14)	0.03 (0.02–0.04)	16.06 (14.26–17.30)

This study deals with the circadian changes of the hemocoagulation indexes in essentially healthy persons with the use of modern methods.

MATERIALS AND METHODS

Fifteen healthy persons 18-57 years old (7 men and 8 women) were included in the investigation. They were mainly medical students and hospital co-workers. None of them had any disorders of the internal organs according to clinical, laboratory, X-ray, and functional examinations. For assessment of the functional state of the coagulation system the circadian dynamics of the following indexes was determined: 1) the anticoagulation test (ACT after [6,13]); 2) antithrombin III (AT III), using a table converting the thromboplastin and thrombin inactivation indexes (TII) to ACT in percent of the AT III content (after to B.F.Arkhipov, 1983); 3) fibrinogen "B" (+,-), soluble fibrin-monomer complexes (SFMC) (Cummine and Lyons, 1948); 4) serum fibrinogen, g/liter (R.A.Rutberg, 1961); 5) thrombin time, sec (Searman, 1957); 6) prothrombin time, sec (Quick, 1943); 7) the prothrombin index, % (Quick, 1943); 8) fibrinoligase activity, % (Sigg and Dukert, modified by V. P. Baluda et al., 1965); 9) serum euglobulin fibrinolytic activity (E. Kowalski, 1959); 10) the hemolysate aggregation test (HAT) [1]. An examination was performed six times a day (at 10:00, 14:00, 18:00, 22:00, 02:00, and 06:00 h). The results were subjected to statistical analysis using "Cosinor" software [14].

RESULTS

The data on the circadian changes of the hemocoagulation indexes are listed in Table 1.

The acrophase is expressed in the decimal system: 0.1 h is equal to 6 min.

A reliable circadian rhythm is evident in the fluctuations of all the hemocoagulation indexes.

The use of "Cosinor analysis" made it possible to obtain more accurate time limits of hyper- and hypocoagulation in the control group. During the course of 24 h the coagulation activity (second minute of incubation) varied from 17.36 to 18.32% and was on the average 17.84%. The amplitude limits were 1.67-3.00%, average 2.33%, with an acrophase at 14:24 h and confidence interval from 12:36 h to 15:18 h. The maximal coagulation activity was 89.92% with variations from 88.36 to 91.48%. The amplitude varied from 4.78 to 8.51%, average 6.65%. Maximal coagulation activity was observed from 13:24 h to 15:30 h

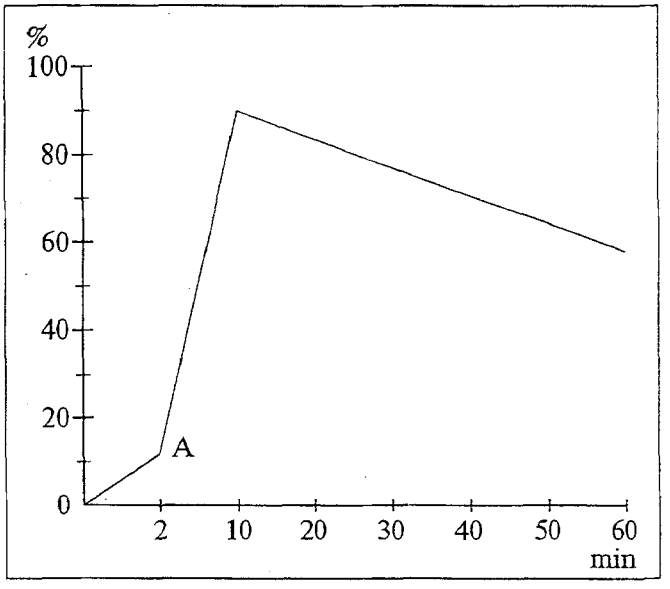

Fig. 1. Autocoagulogram (means) of healthy persons.

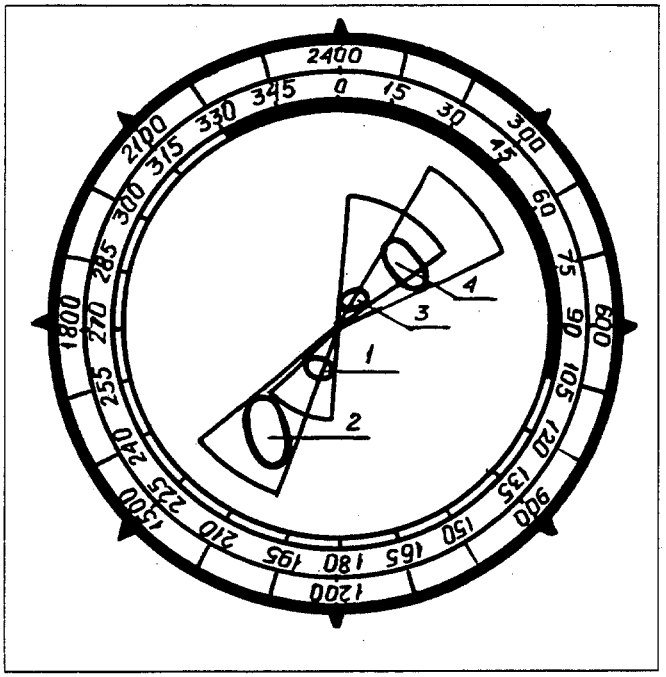

Fig. 2. Circadian ACT indexes of healthy persons 1) A; 2) MA; 3) T; 4) TII

with the acrophase at 14:42 h. The time it took for maximal coagulation activity to be reached was 9.86 min, with variations from 9.79 to 9.92 min. The amplitude of maximal activity varied from 0.11 to 0.37 min, average 0.24 min, with the acrophase at 02:54 h. The thromboplastin inactivation index was 2.06 with variations from 2.04 to 2.07 and amplitude 0.08 (0.06-0.11). A rise of the TII level was observed from 01:24 h to 04:18 h, the acrophase occurring at 03:06 h.

The obtained results can be depicted graphically by plotting the hemolysate incubation time on the abscissa, and the activity indexes on the ordinate. The ascending part of the autocoagulogram curve reflects the increase and the maximal activity of thromboplastin and thrombin in the blood samples under standardized contact and phospholipid (due to the erythrocyte hemolysis) activation of coagulation. The descending part of the curve reflects the rate and intensity of thrombin inactivation as a result of its sorption on fibrin (AT I), the effect of slow-acting (progressive) AT III and IV, and the fibrinolysis products (AT IV) (Fig. 1).

Thus, according to ACT, healthy people exhibit a two-phase circadian rhythm with a tendency toward hypercoagulation in the afternoon. This is confirmed by the stepped-up coagulation activity in the second minute (A), the increase of the maximal circadian activity (MA), and the shorter time taken to reach the maximal coagulation activity (T). It can be noted that there is a tendency toward an increase of the serum progressive AT activity that is revealed as a less steep slope of the descending part of the autocoagulogram curve.

AT III plays the leading role in the maintenance of the blood's fluidity and in the prevention of thromboembolism and other types of intravascular clotting.

More than 80% of all primary anticoagulation activity of the blood is due to AT III. This protein is the main serum heparin cofactor. The level of AT III was determined using the table converting TII to ACT (Arkhipov, 1983).

The circadian AT III level varied from 94.33 to 97.60%, the mean being 95.97%. A tendency toward an increase was revealed from 02:06 h to 04:12 h. The acrophase was at 03:12 h.

According to the data of Machabeli [7], there is no fibrinogen "B" (SFMC) in the blood of healthy persons. Indeed, in the morning hours, when the majority of clinical examinations are usually performed, SFMC is not found in healthy individuals. However, at noon and later in the day the SFMC level rises significantly [8].

The fibrinogen level was on the average 2.43 g/liter varying from 2.89 to 2.47 g/liter, amplitude 0.19 g/liter with a confidence interval from 0.17 to 0.22 g/liter. An increased level of fibrinogen was observed from 01:06 h to 15:24 h, the acrophase being at 01:24 h.

The fibrinoligase activity (FA) was on the average 88.7% (confidence interval 87.4-89.9%), amplitude 8.6% (6.8-10.4%). A pronounced FA increase was observed between 15:36 h and 17:18 h, the acrophase being at 16:42 h.

Platelet function was assayed by HAT after Barkagan and Arkhipov (1980).

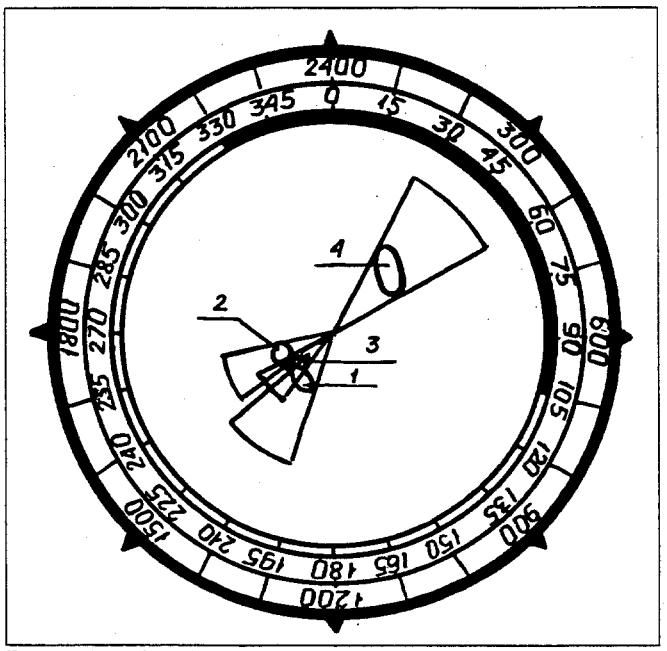

Fig. 3. Circadian hemocoagulation indexes of healthy persons. 1) prothrombin index; 2) fibrinoligase activity; 3) fibrinolytic activity; 4) AT III

The circadian platelet aggregation for exposure to a subthreshold hemolysate concentration (10^3/ml%) varied from 82.8% to 87.1%, the average daily level being 85%. There was a tendency toward an increase of platelet aggregation from 13:30 h to 15:36 h with the greatest value at 15:06 h. The circadian variation of platelet aggregation for exposure to the maximal hemolysate concentration (10^7/ml%) was 92.8-99.3%, the average daily level being 96.1%. The greatest value was observed from 14:06 h to 16:06 h, the acrophase being at 15:12 h.

The platelet aggregation index varied from 1.12 to 1.14 conventional units, the average daily level being 1.13, and the amplitude was 0.03, varing from 0.02 to 0.04. There was an increase of the platelet aggregation index from 14:18 h to 17:18 h, the acrosphase being at 16:06 h.

Synchronously with the fluctuations of the described indexes there was a depression of the blood fibrinolytic activity (AF) in the period from 14:30 h to 16:12 h, with the AF peak at 15:36 h.

Thus, essentially healthy persons exhibit two-phase circadian changes of the hemocoagulation indexes. There is an increase of blood coagulation in the period from 12:36 h to 17:18 h according to ACT, AT III, FG, PI, SFMC, FA, AF, platelet aggregation for exposure to subthreshold and maximal erythrocyte hemolysate concentrations, and the platelet aggregation index assays. The variations of TT and PT and the tendency toward a prolonging of T and TII in ACT point to a decrease of blood clotting from 0:24 h to 04:18 h (Figs. 2, 3).

It may be assumed that the two-phase circadian changes of the hemocoagulation indexes in healthy persons are related to the circadian rhythm of the metabolic activity and neuroendocrine and humoral regulation of the metabolism. In recent years new data have appeared confirming the leading role of the pineal gland and retina and of the circadian rhythm of melatonin production in the triggering of biological rhythms [12].

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