COMPARATIVE EFFECTS OF BACTERIAL AND LEUKOCYTIC PYROGENS ON CHANGES IN THE BLOOD 11-HYDROXYCORTICOSTEROID LEVEL

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An exogenous pyrogen (pyrogenal) and an endogenous (leukocytic) pyrogen were tested on adult rabbits (males and females). The fever induced by both substances was practically identical in height (the temperature was raised by 1.0 and 0.9°C, respectively). However, fever induced by pyrogenal was accompanied by raising of the blood 11-hydroxycorticosteroid level whereas in fever induced by leukocytic pyrogen no elevation of the blood 11-hydroxycorticosteroid level was observed.

Injection of endotoxins which are preparations of bacterial lipopolysaccharides (Pyrexal, Pyromen, etc.) into the blood stream causes elevation of the blood corticosteroid level [2, 3, 9, 11, 12]. Ozeretskovskii and Dzheksenbaev [6] showed in male guinea pigs that both pyrogenal (the Soviet analog of Pyrexal and other lipopolysaccharide preparations), belonging to the group of exogenous pyrogens, and serum pyrogen, belonging to the group of endogenous pyrogens, raise the blood 17-hydroxycorticosteriod level.

There is no direct information in the literature on the stressor action of leukocytic pyrogen (LP), belonging to the group of endogenous pyrogens (obtained from polymorphs and widely used currently in experimental work). To rectify this omission the present investigation was carried out.

EXPERIMENTAL METHOD

Altogether 70 adult rabbits of both sexes belonging to the Soviet Chinchilla breed, weighing 2.06-3.77 kg, were used. Rabbits of as far as possible equal weight were chosen for the different experimental groups.

In the experiments with pyrogenal (batch 267-1) 32 rabbits with a mean weight of 2.7 kg were used. The experiments were carried out to allow for tolerance to the repeated pyrogenic action of the pyrogenal. If pyrogenal was injected twice into the same animals, the interval between injections was 2.5 months. Pyrogenal was obtained from the N. F. Gamaleya Institute of Epidemiology and Microbiology, Academy of Medical Sciences of the USSR; doses were expressed in MPD units (minimal pyrogenic dose for rabbits). The animals received an intravenous injection of 0.5-1.0 MPD/kg body weight.

In the experiments with LP 36 rabbits with a mean weight of 2.4 kg were used. Although LP is known not to induce tolerance, the experiments to test its action were carried out not more often than once every two weeks in order to ensure that the animals received adequate rest between one experiment and the next. After several tests the recipient rabbits receiving LP (39 animals with a mean weight of 2.7 kg) became the donors and were used to obtain LP by the method of Bennett and Beeson [8] in the modification of Collins and Wood [10]. LP was obtained by incubation of a suspension of leukocytes (titer 10^8/ml) and was injected intravenously in a dose of 1-2 ml/kg.

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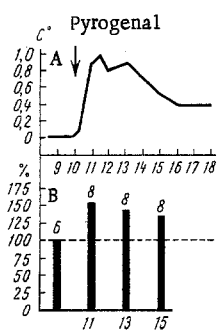

Fig. 1

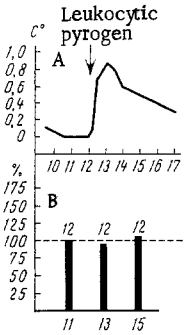

Fig. 2.

Fig. 1. Changes in body temperature (A) and 11-hydroxycorticosteroid level in plasma (B) of rabbits after injection of pyrogenal (marked by arrow). A) abscissa, time of day (in h); ordinate, changes in body temperature (in deg.). In B) ordinate, changes in 11-hydroxycorticosteroid level (in percent of initial level). Numbers below columns show hours, numbers above columns show number of cases.

Fig. 2. Changes in body temperature (A) and 11-hydroxycorticosteroid level in blood plasma (B) of rabbits after injection of leukocytic pyrogen (indicated by arrow). Legend as in Fig. 1.

The plasma 11-hydroxycorticosteroids were estimated by De Moor's method in the modification of Usvatova and Pankov [7]. Blood was taken from a vein on the rabbit's ear once only in a volume of 5 ml to prevent any possible stressor effect from previous blood sampling. In the experiments with pyrogenal the substance was injected at 10 A.M., while LP was injected at 12 noon, so that the peaks of the fever in both cases occurred at the same time.

Blood for estimation of the corticosteroid level in the experiments with pyrogenal was taken when the body temperature was rising (1 h after injection), when the temperature was stabilized at a high level (3 h after injection), and during its fall (5 h after injection). In the experiments with LP blood was taken for estimation of the corticosteroids during the rise (1 h) and fall of temperature (3 h after injection).

The numerical results were subjected to statistical analysis [1, 4, 5].

EXPERIMENTAL RESULTS AND DISCUSSION

Preliminary experiments showed no statistically significant differences in the blood corticosteroid level of the intact male and female rabbits. No statistically significant variations in the corticosteroid level were observed when blood was taken at 11 A. M. or 1 and 3 P.M. On the day of the experiment with pyrogens, blood was therefore taken from the intact rabbits to determine the normal 11-hydroxycorticosteroid level once only, at 11 A.M. The febrile rise of body temperature was observed 1 h after injection of pyrogenal, and after 1.5 h it reached its maximum, on the average 1°C above its initial level. After 4 h it began to drop, and after 7-8 h the temperature was approximately at its initial level.

The corticosteroid concentration under normal conditions varied between 5 and 18 μg%. During fever induced by pyrogenal (during the period of elevation of the body temperature, stabilization of the pyrexia, and its subsequent fall) the corticosteroid concentration also varied, but between 7 and 30 μg%. During all these fluctuations the corticosteroid level 1 h after injection of the pyrogenal had reached 156% of the normal level (100%), falling to 146% after 3 h and 135% after 5 h. A clear tendency for the corticosteroid level to fall as the fever subsided was observed after 5 h (Fig. 1).

After injection of LP the febrile rise of body temperature appeared within 30 min, and after 1 h it reached its maximum, on the average 0.9°C above its initial level. The temperature began to fall after 1.5 h, and was close to its initial level after 5 h. The corticosteroid concentration varied normally between 7.5 and 25.0 μg%. During fever induced by LP (in the period of rise and fall of the temperature) the corticosteroid concentration varied between 7.5 and 26.0 μg%. No significant differences in the corticosteroid level could thus be found under normal conditions and in the febrile state induced by injection of LP (Fig. 2).

Both under normal conditions and after injection of pyrogenal and LP, a tendency was observed for the corticosteroid level to be higher in males than in females.

Preliminary experiments showed that the mean height of the febrile response was almost the same after injection of pyrogenal (1.0°C) and after injection of LP (0.9°C). Meanwhile the 11-hydroxycorticosteroid level was definitely increased after injection of pyrogenal, in accordance with data in the literature, whereas after injection of LP the 11-hydroxycorticosteroid level was virtually unchanged and indistinguishable from normal.

The discrepancy between the results of these experiments and those described by Ozeretskovskii and Dzheksenbaev [6], who found an increase in the corticosteroid level after administration of both exogenous and endogenous pyrogens, is difficult to explain at present. It can only be noted that these workers carried

out their experiments on guinea pigs and injected an endogenous blood pyrogen, whereas in the present experiments leukocytic pyrogen was injected into rabbits.

The results described above show qualitative differences between the action of pyrogenal (exogenous) and leukocytic (endogenous) pyrogen on the pituitary–adrenocortical system in rabbits. The difference is expressed by the fact that pyrogenal induces not only a febrile response of the body temperature, but also a simultaneous and significant increase in the blood 11-hydroxycorticosteroid level, whereas leukocytic pyrogen, while causing a febrile elevation of the body temperature, does not change the 11-hydroxycorticosteroid level at the same time.

LITERATURE CITED

1. V. S. Genes, Tables of Significant Differences Between Groups of Observations for Qualitative Indices [in Russian], Moscow (1964).
2. O. M. Dzheksenbaev and N. A. Ozeretskovskii, Byull. Éksperim. Biol. i Med., No. 5, 31 (1964).
3. V. D. Lindenbraten, Probl. Éndokrinol., No. 3, 74 (1967).
4. E. I. Lyublina, A. Ya. Broitman, A. A. Golubev, et al., Recommendations for Statistical Analysis of Results of Experimental Toxicological Investigations [in Russian], Moscow (1965), p. 46.
5. N. A. Nechaev and V. I. Suslikov, in: Advances in Biometrics [in Russian], Moscow (1970), p. 112.
6. N. A. Ozeretskovskii and O. M. Dzheksenbaev, in: Pyrogen [in Russian], Moscow (1965), p. 45.
7. P. Ya. Usvatova and Yu. A. Pankov, in: Modern Methods of Determination of Steroid Hormones in Biological Fluids [in Russian], Moscow (1965), p. 38.
8. Y. L. Bennett and P. B. Beeson, J. Exp. Med., 98, 477 (1953).
9. E. Bliss, C. Migson, K. Eik-Nes, A. Sandberg, et al., Metabolism, 3, 493 (1954).
10. M. D. Collins and W. B. Wood, J. Exp. Med., 110, 1005 (1959).
11. J. S. Jenkins, Investigation of the Hypothalamic-Pituitary Adrenal Function, Cambridge (1968), p. 205.
12. B. Wexler, A. Dolgin, and E. Tracyzski, Endocrinology, 61, 488 (1957).