mately, leads to the conclusion that the addition of PP not only lowers the affinity of PHb for oxygen and produces an increase in the quantity of oxygen given up by it practically to the characteristic values for blood, but also increases (although by a rather lesser degree) the efficiency of interaction between the artificial carrier and other ligands (H^+ and CO_2), which are themselves also regulators of gas transport processes *in vivo*.

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LIPID PEROXIDATION IN TISSUES OF NORMAL AND HUNGRY RATS OF DIFFERENT AGES

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According to the free-radical theory [7, 9], aging is associated with the action of free radicals on the genetic apparatus and on the biological membranes of the cell [8]. **Unsaturated acyl** residues of fatty acids of phospholipids, which take part in the process of peroxidation, are most sensitive to their action. Unsaturated fatty acids are formed during oxidative catabolism of lipids. In this connection, investigation of lipid peroxidation (LPO) is important during starvation, when the body switches from the carbohydrate type of metabolism to the lipid type [4].

The results of the study of LPO in the blood serum, adipose tissue, and liver tissue in rats of different ages, under normal conditions and after starvation for 48 h are described below.

EXPERIMENTAL METHOD

Experiments were carried out on young (aged 4-5 months) and old (aged 24-26 months) Wistar rats. The malonic dialdehyde (MDA) concentration was determined by the method in [6]. The background MDA level and its concentration during spontaneous, nonenzymic, and enzymic LPO *in vitro* (incubation for 30 min at 37°C with continuous shaking) were investigated and the mean rate of MDA formation was calculated. The composition of the reaction medium for spontaneous LPO was: 50 mM Tris-HCl, 160 mM KCl (pH 7.4), and 0.1 ml blood serum or 50 mg of finely minced test tissue (liver, epididymal adipose tissue). The final volume of medium was 2 ml. For nonenzymic (ascorbate-dependent) LPO 2.5 μM $FeSO_4 \cdot 7H_2O$ and 0.2 mM ascorbic acid also were added to the medium for enzymic (NADPH-dependent) LPO, 1 mM NADPH, 20 mM nicotinamide, 4 mM ADP, and 2.5 μM $FeSO_4 \cdot 7H_2O$ were added to the medium. The optical density of the solutions was measured on the SF-26 instrument at 532 nm.

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TABLE 1. MDA Concentration in Blood Serum (in nmoles/ml), in Adipose Tissue, and Liver (in nmoles/g tissue) of Normal and Hungry Rats of Different Ages (M ± m)

Pathway of MDA accumulation	Young rats		Old rats	
	intact	starved for 48 h	intact	starved for 48 h
Blood serum				
Init. bkgr. level	8.6±1.5 (4)	6.9±1.3 (6)	18.4±2.3* (6)	8.5±1.3 † (20)
Spontaneous	2.6±0.6 (5)	8.7±1.7 (6)	13.0±1.7 (6)	5.2±0.6 (5)
Nonenzymic	26.2±5.1 (7)	10.6±2.8 (8)	34.3±1.5 (18)	26.1±2 (9)
Enzymic	149±3.7 (11)	107±6.2 (6)	134±5.4 (7)	130±4.5 (20)
Adipose tissue				
Init. bkgr. level	740±30 (5)	900±40 (6)	80±80 (6)	620±60 (6)
Spontaneous	550±10 (5)	590±30 (6)	696±90 (6)	380±20 † (6)
Nonenzymic	7720±10 (5)	1920±60 (6)	1695±84* (6)	1048±65 † (6)
Enzymic	2630±180 (5)	5130±50* (5)	4930±70* (6)	3910±100 † (6)
Liver				
Init bkgr. level	2210±250 (5)	1690±60 (6)	2630±170 (6)	1220±100 † (6)
Spontaneous	5170±410 (5)	6010±250 (6)	7000±300* (6)	3750±150 † (6)
Nonenzymic	4660±320 (5)	4660±250 (6)	6810±260* (6)	3680±120 † (6)
Enzymic	4590±210	7170±150 †	5930±80*	5770±70 †

Legend. Here and in Table 2: *) $P < 0.05$ (young and old animals), †) $P < 0.05$ (normal and hungry rats); number of animals in parentheses.

The experimental results were subjected to statistical analysis by Student's t test.

EXPERIMENTAL RESULTS

The experiments showed that the MDA concentration in the blood serum was higher in old rats than in young (Table 1). Incubation for 30 min without addition of LPO activators lowered the serum MDA level. Due to incubation the MDA was evidently metabolized, and this process took place more rapidly than its formation. Addition of Fe^{++}, a catalyst of the reaction, and substances reducing Fe^{+++} by nonenzymic (ascorbate) and enzymic (nicotinamide, ADP, NADPH) pathways, to the medium increased the MDA concentration in the blood serum of both young and old rats; in young rats MDA accumulated more rapidly (Table 2).

The rise in the serum concentration of MDA with age accompanied by a fall in the rate of its accumulation may be the result of depression of functional activity of the systems protecting the body against LPO products and, in particular, against those inactivating hydroperoxides. This last process involves the use of thiol compounds [9], the content of which in the body diminishes during aging [3, 5].

The MDA concentration did not differ significantly in the adipose tissue of young and old rats. Incubation of adipose tissue without activators, just as in the blood serum, led to a fall in the MDA concentration in animals of both age groups. Activation of LPO by nonenzymic and enzymic methods increased MDA formation; in the young rats the ascorbate-dependent pathway was more effective, in the old rats the NADPH-dependent pathways.

In the liver tissue, spontaneous, nonenzymic, and enzymic pathways of LPO led to the accumulation of MDA, almost twice as rapidly in the old rats. This could indicate that the liver tissue contains LPO activators, the concentration of which rises during aging, or that activity of the antioxidant system falls with age [1]. There were no age differences, incidentally, in the background level of MDA in the liver.

The divergence of the results which was observed was evidently not accidental. Age changes, it must be noted, were heterogeneous and asynchronous, even within the same organ or tissue. Differences in the intensity of LPO, reflecting to some degree the character of aging, point to the existence of tissue specificity in the process utilized. Starvation for 48 h lowered the MDA level a little in the blood serum and liver and adipose tissues of the

TABLE 2. Rate of Accumulation of MDA in Blood Serum (in nmoles/ml/min) and in Adipose Tissue and Liver (in nmoles/g tissue/min) in Rats of Different Ages (M ± m)

Pathway of MDA accumulation	Young rats		Old rats	
	intact	starved for 48 h	intact	starved for 48 h
Blood serum				
Nonenzymic	0,7±0,1 (7)	0,27±0,05 † (8)	0,52±0,19 (18)	0,63±0,07 (19)
Enzymic	4,7±0,3 (11)	3,15±0,26 † (16)	3,8±16* (19)	3,8±0,16 (19)
Adipose tissue				
Nonenzymic	232,4±13,2 (5)	40,1±1,1 † (6)	43,78±5,18* (6)	14,3±2,5 † (6)
Enzymic	60,7±17,1 (5)	148,5±3,0 † (5)	135,3±3,46 (6)	109,9±4,3 † (6)
Liver				
Nonenzymic	81,5±10,4 (5)	98,4±4,4 (6)	139,1±12,5* (6)	82,3±3,9 † (6)
Enzymic	79,3±9,1 (5)	181,6±5,2 † (6)	127,8±8,2* (6)	135,0±4,7 (6)

old rats. Changes in the young rats were not significant. The intensity of enzymic and nonenzymic MDA formation was lowered in the blood serum of the young hungry rats, but in the old it was unchanged. During spontaneous LPO the process of metabolization of MDA in the blood serum was delayed a little under the influence of starvation. Whatever the case, it took place more slowly in the hungry rats, especially the young ones. After starvation the rate of spontaneous and nonenzymic LPO decreased in the adipose tissue of the young rats and the enzymic pathway of oxidation was intensified. In old rats, starvation led to inhibition of all LPO pathways in the adipose tissue. Starvation led to acceleration of all the LPO pathways studied in the liver of the young animals. In the old animals considerable inhibition of the spontaneous and nonenzymic pathways was observed but the enzymic pathway was unchanged.

Consequently, during aging the MDA concentration was unchanged in the adipose and liver tissues but raised in the blood serum. A contributory factor may have been exhaustion of the defensive systems of the body responsible for supplying antioxidants of different types, and inactivating potential LPO initiators (hydroperoxides). Under the influence of starvation for 48 h the MDA concentration in the blood serum and liver and adipose tissues fell in the old rats and was unchanged in the young. Since the energy requirements of the body on the second day of starvation are satisfied mainly through mobilization of fat from the fat depots, the intensity of lipolysis largely determines the character of metabolic processes in the body. **Mobilization of fat** from fat depots during starvation has been shown to be less **intensive in old** animals than young [2]. This could be responsible for the fall in the intensity of LPO in the old rats. The possibility likewise cannot be ruled out that inhibition of LPO processes under the influence of starvation in old rats may be connected with activation of the antioxidant system.

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