Age-Related Changes in Activity of Enzymes Catalyzing Oxidation-Reduction of Endogenous Aldehydes in the Liver of Rats during Immobilization Stress

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Activities of aldehyde dehydrogenase and aldehyde reductase in the liver 1.5-, 12-, and 24-month-old rats were measured after 30-min immobilization. Changes in activities of aldehyde dehydrogenase and aldehyde reductase in the liver after immobilization stress depended on animal age. These shifts lead to a strain in endogenous aldehyde utilization in the aldehyde reductase reaction in 1.5-month-old animals and inhibition of utilization of these metabolites in oxidation-reduction reactions in 24-month-old rats.

Key Words: *endogenous aldehydes; stress; ontogeny; liver*

Accumulation of cytotoxic metabolites formed during free radical oxidation (*e.g.*, aldehydes) plays a role in stress-induced damage to organs and tissues [15]. These metabolites are utilized in reactions catalyzed by various enzymes. Oxidation-reduction reactions catalyzed by aldehyde reductase (AR) and aldehyde dehydrogenase (ALDH) are of considerable importance in this respect [7,12]. The role of these reactions in antistress protection remains unknown.

Here we measured activities of ALDH and AR in liver samples from rats of different age exposed to immobilization stress.

MATERIALS AND METHODS

Experiments were performed on male Wistar rats aging 1.5, 12, and 24 months (n=120). The animals were divided into 2 groups. Group 1 included intact rats. Group 2 animals were exposed to immobilization stress (fixation on the back for 30 min). The strength of stress was estimated by measuring blood concentrations of 11-hydroxycorticosteroids and epinephrine.

The animals were decapitated. The liver was immediately removed, placed in cold physiological saline, washed to remove the blood, and homogenized in a medium containing 0.25 M sucrose and 0.01 M Tris (pH 7.4). Homogenates were centrifuged at 3000g for 10 min. The supernatant was centrifuged at 10,000g for 20 min to obtain the postmitochondrial fraction.

Activities of AR [13] and ALDH [2] were measured in the postmitochondrial fraction. A special series was conducted to estimate the concentrations of fluorescent Schiff bases in frozen liver samples [11]. Protein content in samples was determined by the method of Lowry [10].

The results were analyzed by nonparametric Wilcoxon—Mann—Whitney test.

RESULTS

Activities of rat liver enzymes utilizing endogenous aldehydes in oxidation-reduction reactions progressively increased over the 1st year of life (Table 1). Activities of AR and ALDH decreased by the end of the 2nd year of life. These shifts determine age-

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TABLE 1. Activities of AR and ALDH in the Liver of Rats of Different Age after 30-min Immobilization (nmol/mg protein/min, *M*±*m*)

Enzyme	1.5 months		12 months		24 months	
	intact	stress	intact	stress	intact	stress
AR	22±3	46±4*	95±7+	50±8*	61±10+	98±9*
ALDH	3.9±0.4	6.7±1.7	4.5±0.4	4.7±0.6	2.9±0.3	4.4±0.5*

Note. $p<0.05$: *compared to intact rats; +compared to 1.5-month-old-rats.

related decrease in the efficiency of endogenous aldehyde catabolism, which is consistent with published data [4,5]. Age-related changes in the rate of carbonyl compound utilization modulate cell resistance to free radical-induced damage [4,14]. Taking these data into account, we measured activities of AR and ALDH in liver samples from rats of different age exposed to immobilization stress. Previous studies showed that this exposure is accompanied by activation of free radical processes [1,6,9].

Activities of AR and ALDH in the postmitochondrial liver fraction from rats aging 1.5 and 24 months increased after 30-min immobilization. Immobilization of 12-month-old animals was accompanied by a decrease in AR activity in the postmitochondrial liver fraction, while ALDH activity remained unchanged.

Variations in enzyme activity during immobilization stress led to changes in the concentrations of aldehydes and products of their intracellular conversion in the liver. After immobilization stress the concentration of Schiff bases in the liver increased in old rats (Fig. 1), but remained unchanged in animals aging 1.5 and 12 months ($p<0.05$).

These data suggest that variations in activities of AR and ALDH in immobilized animals of different age are followed by various changes in the rate of aldehyde utilization in liver tissue *in situ*. Activation of AR in 1.5-month-old rats prevents stress-produced accumulation of aldehydes. However, low enzyme activity in intact animals determines strain in aldehyde reduction. These rats would exhibit a lower adaptive reaction catalyzed by liver AR under stress conditions more severe than short-term immobilization.

Despite partial inhibition of AR, the concentration of Schiff bases in liver samples from 12-month-old rats remained unchanged after immobilization. This discrepancy was probably associated with acceleration of aldehyde conversion in other metabolic pathways (*e.g.*, glutathione transferase reaction).

Immobilization was followed by activation of AR and ALDH in the postmitochondrial fraction and accumulation of carbonyl metabolic products in the liver of old rats (increase in the amount of Schiff bases). These changes were probably related to deceleration of aldehyde conversion in more efficient metabolic pathways (*e.g.*, glutathione transferase reaction). Activation of AR under these conditions plays an adaptive role and contributes to increased utilization of carbonyl compounds. It should be emphasized that immobilization stress is accompanied by increased formation of these compounds. These changes can be due to acceleration of AR gene expression and direct activation of AR by the substrate [8].

Our findings indicate that functional activity of enzymes catalyzing oxidation-reduction of endogenous aldehydes in rat liver cells under conditions of immobilization stress undergoes variations during ontogeny. These changes result in the strain of endogenous aldehyde utilization by AR in 1.5-month-old animals. Moreover, oxidation-reduction of aldehydes is suppressed in 24-month-old rats. A decrease in the efficiency of carbonyl product utilization in the liver probably reflects disadaptation during late ontogeny. Therefore, the sensitivity to stress factors increases during aging [3].

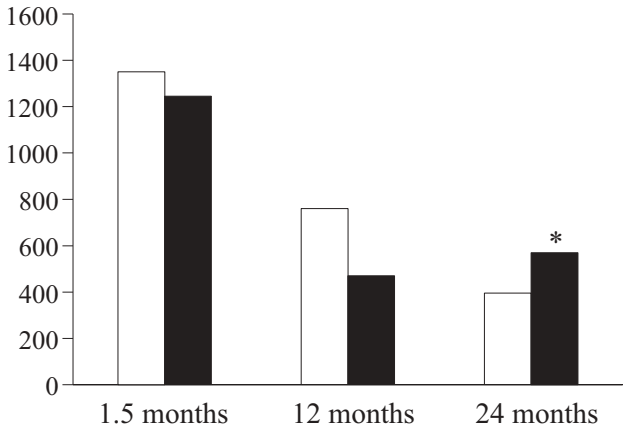

Fig. 1. Concentration of Schiff bases in the liver of rats of different age after 30-min immobilization (n=5-6). Ordinate: fluorescence units per 1 g wet tissue (430 nm). Light bars, intact rats; dark bars, stress. *$p<0.05$ compared to intact rats.

These data indicate that new methods improving stress resistance should include activation of enzymes for catabolism of carbonyl metabolic products or stimulation of enzyme biosynthesis.

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