precise duplexes. Therefore, at least some of the highly conservative repeats persist for an extremely long time in the eukaryotic genome.

The next task is to create a clone library of the conservative fraction of repetitive sequences of the human genome and to analyze in this fraction the presence of repeats common to the genomes of diverse eukaryotic species.

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EXPERIMENTAL BIOLOGY

Individual Differences in Responses to Acute Stress Associated with Type of Behavior (Prediction of Stress Resistance)

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On the basis of the initial parameters of behavior in the "open field," "forced swimming," and "emotional resonance" tests, the main behavioral parameters - the number of squares crossed, the number of standing postures, and the time of passive swimming - are shown to be predictable for stress in rats with different types of behavior.

Key Words: *acute stress; specificities of behavior; prediction; stress resistance*

Prediction of resistance to stress and to other pathogenic effects, based on specificities of behavior, is of not only theoretical, but also practical importance. Specifically, this makes it possible to preliminarily choose individuals with different degrees of resistance to certain factors, which is necessary both for studying the mechanisms of individual resistance to these factors and for developing methods of individual prophylaxis and treatment of disturbances caused by these factors. For instance, a correlation between the resistance of the cardiovascular system to emotional stress and

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the specificities of behavior in the "open field" [8] and "emotional resonance" [9] tests has been revealed in rats of various strains.

In a previous study [5] we showed that acute stress causes behavioral disturbances: suppression of exploratory activity in the "open field" test and an increase of the level of depressiveness in the "forced swimming" test. The strength of this effect proved different in rats of different groups. The aim of the present study was to determine: 1) the most important characteristics of initial behavior for predicting stress-induced behavioral effects; 2) the accuracy of prediction (the maximal one and that of prediction based on the most significant parameters of behavior); 3) possible differences in the accuracy of predicting stress-induced behavioral effects for groups of rats with different types of behavior.

MATERIALS AND METHODS

The experiments were carried out on 99 male albino rats. Individual specificities of the animals' behavior were successively determined in the "open field" [3], "forced swimming" [12], and "emotional resonance" [7] tests. The first two tests lasted 10 min and the third one 5 min. The following behavioral parameters were recorded: the number of squares crossed (NSQ), the number of standing postures (NSP), the number of groomings (NWASH), the number of times when animals entered the center of the field (NCEN), the time of extinction of motor activity (TEXT), the time of passive swimming (TPS), the time of the first episode of active swimming (TFIR), the time of staying in the light compartment of the chamber without a "victim" (TSV1), and the same with a "victim" (TSV2), and the number of movements from the light compartment to the dark one without a "victim" (NPER1), and that with a "victim" (NPER2). The strength of the "emotional resonance" responses was assessed as the calculated coefficient: CTSV-2[TSV2/(TSV1+TSV2)], and the strength of the "anxiety" response as the coefficient:

$$\mathrm{CNPER}=2[\mathrm{NPER2}/(\mathrm{NPER1}+\mathrm{NPER2})].$$

At first, on the basis of behavioral parameters in the "open field" and "forced swimming" tests, three groups of rats (20 rats with an active type of behavior, 18 rats with a passive type of behavior, and 19 rats of an intermediate group) were chosen from the initial rat population. The rats were divided in groups as described previously [4,5]. Since the three main parameters (NSQ, NST, and TPS) with respect to which the animals were assigned to the groups correlated with each other [5], the assignment to groups was refined by factor analysis. For this purpose, we determined the factor loadings of the first main component, 33% quantiles of loading distribution were calculated on their basis, and the classifying of an animal to a particular group was refined according to these values. The rats were then maintained under standard conditions in the vivarium for 1 month to rule out sequelae of the stress caused by behavioral testing. After that, one half of the rats, chosen randomly within each group, were subjected to repeated behavioral testing in the "open field" and "forced swimming" (control subgroup), and the remaining rats were exposed to acute stress prior to repeated behavioral testing (experimental subgroup). Acute stress was induced by unpredictable and inescapable painful electrical stimulation of the paws, which was performed at randomly varied intervals (a variety of "waiting stress") [6]. The main behavioral characteristics following stress were predicted by calculating equations of stepwise multiple linear regression. The following behavioral parameters were chosen as independent variables: NSQ, NST, TPS, TSV1, NPER1, CTSV, and CNPER. As a result, we discovered the combinations of parameters which ensured the best prediction of the three main behavioral indexes (NSQ, NST, and TPS) following stress. Involvement of the rest of the behavioral parameters in the model did not markedly change the accuracy of prediction.

RESULTS

Analysis of the experimental results showed that in most cases the behavioral parameters recorded for repeated testing in the control and experimental subgroups correlated with the initial values of the same behavioral parameters. The overall regression equations were as follows:

$$\mathrm{NSQE} = 0.42\,\mathrm{NSQF} - 0.08\,\mathrm{TSV1}, \quad R^2 = 0.69,\ \mathrm{SE} = 11.0; \tag{1}$$

$$\mathrm{NSTE} = 0.08\,\mathrm{NSQF}, \quad R^2 = 0.63,\ \mathrm{SE} = 2.41; \tag{2}$$

$$\mathrm{TPSE} = 90.5\,\mathrm{CNPER} + 1.24\,\mathrm{TPSF} - 135, \quad R^2 = 0.62,\ \mathrm{SE} = 37.1, \tag{3}$$

where R^2 is the square of the coefficient of multiple correlation, determining the share of the overall variance described by the model, SE is the mean square deviation of the predicted values from the experimental values, F is the baseline value, and E is the value after stress. As is shown by

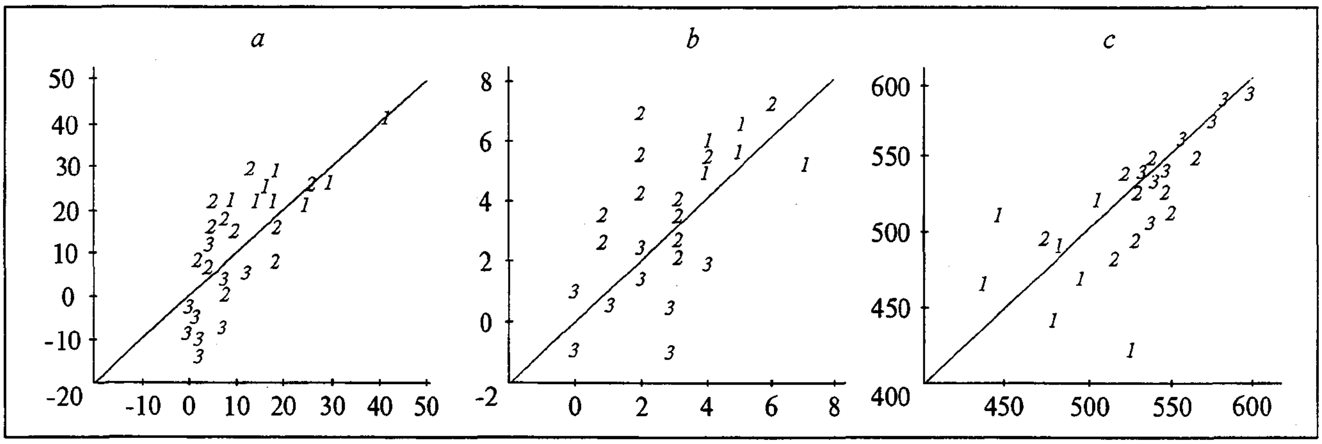

Fig. 1. Diagram of relationship between predicted (calculated by regression equation) and actual values of NSQ (*a*), NST (*b*), and TPS (*c*) after stress in rats with different types of behavior. Numbers denote groups of rats: *1*) with active behavior; *2*) intermediate group; *3*) with passive behavior. Abscissa: actual values of behavioral parameters; ordinate: values predicted from regression equations. The diagonal corresponds to full coincidence of the predicted and actual behavioral parameters.

equation (1), in the experimental subgroup of rats there is a linear dependence between the level of horizontal motor activity following stress (NSQE) and its initial value (NSQF). The fact that the coefficient is lower than 1 (0.42) indicates that the higher the initial level of horizontal activity, the lower it drops (in absolute values) after stress, although the relative drop is the same in all cases. On the other hand, if the initial level of horizontal motor activity is unchanged, an increase in the time spent in the light compartment without a "victim" (TSV1), i.e., in the intensity of the passive defense response to a novelty, also results in a drop of NSQE. These two parameters (NSQE and TSV1) foretell changes in the horizontal motor activity after stress with sufficient accuracy, accounting for 72% of the total variability of this behavioral parameter. The coefficient of correlation between the predicted and empirical NSQ values is 0.84.

The vertical motor activity after stress (NSTE) proved to be best predicted not from its initial values (NSTF), but from the initial values of horizontal motor activity (NSQF). This indicates that the effect of stress on the vertical motor activity is not described by a linear dependence, i.e., the correlation between NSTE and NSTF is nonlinear. Nevertheless, the accuracy of prediction in this case is rather high (mean error ±2.4, R^2=0.63, which corresponds to the correlation coefficient of 0.8).

The times of passive swimming after stress (TPSE) are proportional to the initial values (TPSF), but they are 1.24 times as long. In other words, the absolute increment of the level of depressiveness following stress is higher in individuals with initially higher values of this parameter (in rats with an active behavior and in the intermediate group). In addition, as the "anxiety" parameter (CNPER) increases, TPSE becomes higher. The mean error of prediction is ±37.1 and the contribution of these two parameters (CNPER and TPSF) in combination accounts for 61% of the TPS variability, which corresponds to the correlation coefficient of 0.78.

Analysis of variance shows that the predictive accuracy with respect to TPS does not differ among rats of different groups (F_{TPS}=0.038, p=0.96), i.e., the time of passive swimming after stress can be predicted with equal accuracy for the rats of all three groups. On the other hand, with respect to other two parameters (NSQ and NST) following stress, the accuracy of prediction differs between the groups (F_{NST}=5.407, p=0.01, F_{NSQ}=3.397, p=0.04), this attesting to the presence of some neglected factor which shifts the mean error of prediction to one side in the extreme groups and to another in the intermediate group (Fig. 1). The relative error of predicting NSQ, NST, and TPS by using the regression equation is 19.95±44.38, 30.36±17.97, and 06±1.6%, respectively.

Thus, on the whole, a comparison of the predicted and empirical values demonstrated a good correlation between the predicted and actual behavioral parameters after stress (correlation coefficients of the order of 0.8). At the same time, the deviations of the experimental values of TPS from the predicted ones were virtually the same in all groups (±2, on average), whereas in the case of the other two indexes (NSQ and NST) they were different. In the two latter cases the deviations of the predicted values from the experimental ones were of the same sign for the extreme groups and of the opposite sign for the intermediate group. This shows that the intermediate group responded to stress by changing the behavioral parameters (NSQ and NST) differently than did the extreme groups. Probably, the technique of dividing the animals into groups enabled us to pinpoint not just an intermediate, but precisely the "midpoint group" with inherent specific characteristics distinguishing it from the extreme groups of animals by the absence of a predominant trend in both behavior and oxidative metabolism in the brain [5], and by enhanced sensitivity to both circulatory cerebral hypoxia [4] and stress [6].

Our findings provide evidence that the most significant parameters of initial behavior for predicting the behavioral effects of stress are as follows: NSQF, TPSF, TSV1, and CNPER. Linear functions ensure a rather high predictability for practical use (R^2=0.6-0.7). The accuracy of prediction on the basis of the most important behavioral characteristics virtually does not differ from the maximum attainable. In the case of foretelling the behavioral effects of stress from the TPS index the accuracy of prediction does not differ between the groups of rats with different types of behavior but does differ in the case of the NSQ and NST indexes. This implies that, in contrast to NSQ and NST, the behavioral parameter TPS more strictly reflects some essential factor providing for stress resistance. A tendency for there to be a change (a rise or a drop) of the tension of free oxygen in the brain in a stress situation may prove to be such a factor, which, as was previously shown by us, correlates reliably and linearly with the behavioral parameter TPS (and unreliably with NSQ and NST) [5]. Stress is known to be attended by a reduced catecholamine level in the brain [2,10] (which is lower in animals not resistant to stress than in resistant ones [2]). A subsequent change of the level of amines in the brain is determined by the rate of their resynthesis. The process of catecholamine synthesis is highly sensitive to oxygen deficiency [13], since it involves the oxygen-dependent sites at the level of tyrosine hydroxylase and dopamine β-hydroxylase [11]; therefore, the degree of decrease of the amine content in the brain (and, consequently, of the animals' resistance to stress) will depend on how the tension of free oxygen in the brain changes in a stress situation. Stress is also known to be accompanied by the development of cerebral circulatory hypoxia [1]. In addition, the behavioral parameter TPS has been shown to correlate negatively with the brain level of catecholamines [12]. Hence, it is clear why the behavioral parameter TPS, which reflects, on the one hand, the brain level of catecholamines and, on the other, the nature of the changes of oxygen tension in the brain during stress, is the most reliable index for predicting stress resistance.

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