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Alterations of Central Norepinephrine, Dopamine and Serotonin in Several Strains of Mice Following Acute Stressor Exposure

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SHANKS, N., S. ZALCMAN, R. M. ZACHARKO AND H. ANISMAN. *Alterations of central norepinephrine, dopamine and serotonin in several strains of mice following acute stressor exposure.* PHARMACOL BIOCHEM BEHAV **38**(1) 69–75, 1991.—Exposure to inescapable footshock provoked region-specific alterations of norepinephrine (NE), dopamine (DA) and serotonin (5-HT) activity across six strains of mice (A/J, BALB/cByJ, C3H/HeJ, C57BL/6J, DBA/2J and CD-1). The stressor provoked reductions of hypothalamic NE and increased MHPG accumulation in all strains. In contrast, the effects of the stressor on NE activity in the hippocampus and locus coeruleus varied appreciably across strains. In the mesocortex and nucleus accumbens shock induced an increase of DOPAC accumulation and pronounced reductions of DA in some strains, while in others these variations were less pronounced or entirely absent. Stressor-provoked alterations of 5-HT and 5-HIAA were most evident in the mesocortex. Strain-specific neurochemical alterations following footshock are discussed relative to stressor-induced behavioral disturbances and animal models of depression.

Strain Footshock Norepinephrine Dopamine Serotonin

IT has been repeatedly demonstrated that uncontrollable stressors provoke a wide array of behavioral disturbances (5,31). However, considerable interindividual variability exists in this respect, and it has been suggested that genetic factors may play a role in determining whether stressor effects are evident. In accordance with this view, it was demonstrated that the behavioral effects of an uncontrollable stressor will vary across strains of mice. While inescapable footshock provoked marked deficits of shuttle escape performance in some strains of mice (e.g., A/J, BALB/cByJ, C3H/HeJ), in other strains this behavior was hardly affected (e.g., DBA/2J) (25). Interestingly, the occurrence of a stressor-induced deficit in one task in a particular strain of mouse was not predictive of whether behavioral disturbances would be evident in a second task. For instance, in DBA/2J mice, in which shuttle escape performance was not affected by inescapable shock, responding for electrical brain stimulation from the nucleus accumbens was retarded after exposure to the stressor. Conversely, self-stimulation from this region in BALB/cByJ mice was actually enhanced following exposure to inescapable shock, although this treatment severely retarded escape performance in this particular strain (35). These data provisionally suggest that the two behaviors are subserved by different mechanisms and that the impact of the stressor on these mechanisms differs across strains of mice. In effect, in assessing the impact of a stressor it is inappropriate to consider some animals as being vulnerable and others as being hardy.

Considerable attention has been devoted to the proposition that the behavioral consequences of stressors may be related to the neurochemical changes elicited by the environmental insults. Thus, stressor-related variations of central norepinephrine (NE), dopamine (DA) and serotonin (5-HT) have all been implicated as factors in subserving behavioral deficits associated with stressors. It has been argued that upon exposure to an uncontrollable stressor the utilization of these amines may exceed their synthesis, resulting in a net decline of the transmitter levels, hence rendering the animal less capable of dealing with environmental demands (6,31). Given that the impact of a stressor on behavior varies across strains of mice, as well as with the specific behavior under consideration, it has been suggested that strain-specific differences may exist with respect to relative vulnerability of different transmitters, as well as the brain regions in which these occur (6,34). To be sure, it has been shown that strains of mice differ in basal turnover rates of NE (13), DA (10,15) and 5-HT (33). Not surprisingly, neurochemical alterations following stressor exposure have also been reported to vary across strains of mice. For example, it was demonstrated that footshock provoked higher mesocortical DA turnover in BALB/c strain than in C57BL/6 mice (15), and that the utilization of DA was greater in DBA/2 mice that had been exposed to restraint than in similarly treated C57BL/6 mice (10). The present investigation was conducted to determine the concentrations of NE, DA and 5-HT and their metabolites following exposure to acute inescapable footshock in six strains

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of mice which differ in their behavioral responses to the stressor (25, 26, 30, 35).

METHOD

Subjects

Forty-four naive, male, mice of each of 5 inbred strains (A/J, BALB/cByJ, C57BL/6J, C3H/HeJ, DBA/2J) and one noninbred strain (CD-1) were employed in the present study. The 5 inbred strains were obtained from the Jackson Laboratory, Bar Harbor, ME, while the noninbred CD-1 mice were obtained from Charles River Inc., St. Constance, Que. Mice were obtained at 35 days of age and were permitted approximately two weeks to acclimatize prior to serving as experimental subjects. The animals were housed five per cage with free access to food and water and maintained on a 12-hour light-dark cycle (light 0700–1900). The noninbred CD-1 mice were included in the experiment for comparative purposes, since most of the work of a nongenetic nature conducted in this laboratory over the past decade has involved this strain.

Apparatus and Procedure

Inescapable footshock was administered in five black Plexiglas chambers that measured 30 × 14 × 15 cm. The chamber floors consisted of 0.32 cm stainless steel rods spaced 1.0 cm apart (center to center) and were connected in series by neon bulbs. The end walls of the chambers were lined with stainless steel plates and were connected in series to the grid floor. Shock was delivered to the floor through a 3000-V source. A red Plexiglas roof served to reduce illumination of the chambers. Chambers of the same characteristics and dimensions, but not connected to a power source, were used for pretreatment of the no shock groups. Between 0800 and 1200 h mice were individually placed in the shock boxes and exposed to 360 footshocks of 2 s duration at 300 microamps (9-s intertrial intervals) or placed in the shock apparatus without shock being delivered. Immediately thereafter, mice were decapitated and brains were sectioned and quickly frozen for subsequent determinations of central amines and metabolites using a slight modification of the HPLC procedure of Seegal, Brosch and Bush (23).

Brain Microdissection

Brain regions were sectioned on a dissecting block fashioned from 0.5 mm thick aluminum templates and separated by brass plates (0.25 mm). The slots created by the brass plates in the block served as guides for single-edged razor blades. The brain sections were teased from the blades onto glass slides and frozen on dry ice. A petri dish filled with powdered dry ice served as a cold stage for sectioning and was situated under a stereomicroscope, which was illuminated by a fiber optic cold light source. Under low magnification, the mesocortex, nucleus accumbens (Nas), caudate nucleus, dorsal hippocampus, substantia nigra (SN), ventral tegmentum (VTA) and locus coeruleus (LC) were punched with hollow microdissection needles, which ranged in diameter from 22 to 16 gauge, while the hypothalamus was taken in its entirety prior to microdissection. The punched sections were placed in round-welled titer trays and were frozen at −70°C. High performance liquid chromatography (HPLC) with coulometric detection following the prcedure of Seegal et al. (24) was employed to determine amine and metabolite concentrations in the various brain regions.

HPLC Procedure

Frozen tissue were sonified in 0.3 M monochloroacetic acid containing 0.1 mM disodium EDTA. The protein content of each sample was measured using the method of Lowry et al. (21). The homogenate was then centrifuged and the supernatant filtered through a 0.2 μm regenerated cellulose filter. An aliquot of this filtrate was injected onto a C_{18} reversed-phase column (Chromatography Sciences Company Inc.) in a high performance liquid chromatography (HPLC) system (Waters) equipped with a coulometric detector (ESA). The ouput from the detector was plotted and measured using a Hewlett-Packard HP-3390A plotting integrator. The mobile phase used with this aliquot (0.1 M phosphate buffer with 20% acetonitrile) allowed for the separation of 3-methoxy,4-hydroxyphenylethylene glycol (MHPG) and 3,4-dihydroxyphenylacetic acid (DOPAC), the metabolites of NE and DA, respectively. Sodium octyl sulphate (0.2 mM) was added as an ion-pairing agent and EDTA (0.05 mM) was added as an antioxidant. A separate analysis of the samples using 50% acetonitrile added to the phosphate buffer was performed to obtain homovanillic acid (HVA), serotonin (5-HT) and its metabolite 5-hydroxyindoleacetic acid (5-HIAA).

RESULTS

Analyses of variance were conducted independently for each of the amines and metabolites. A number of samples were lost during the course of taking the tissue samples or in the HPLC analyses, and hence the degrees of freedom for the analyses varied across brain regions. In the present investigation it was of particular interest to identify those strains of mice in which the stressor influenced amine activity, as well as the brain regions in which these occurred. Indeed, it was expected that strain-specific amine changes induced by the stressor might correspond with several behavioral changes which were previously demonstrated to occur in these strains (25, 26, 35). Accordingly, Newman-Keuls multiple comparisons ($\alpha = 0.05$) were conducted of the simple effects comprising the Strain × Shock treatments interactions irrespective of the significance of the interaction (33). In addition, since considerable data are available concerning those regions in which stressors generally affect amine turnover, the present report focusses on these regions. Analyses that have been deemed relatively less important (e.g., dopamine within the hypothalamus, as opposed to specific nuclei) were omitted.

Norepinephrine

The mean concentrations of hypothalamic NE and MHPG as a function of the Shock Treatment in the six strains of mice are shown in Fig. 1. Analysis of variance indicated that the MHPG accumulation did not differ across strains of mice ($F<1$), while the shock treatment markedly increased the metabolite accumulation, $F(1,242) = 95.08$, $p<0.01$. Concentrations of NE were found to vary across strains of mice, $F(5,250) = 5.68$, $p<0.01$. Newman-Keuls multiple comparisons revealed that NE levels were lower in A/J mice than in the remaining strains, while the levels in C57BL/6J were lower than in BALB/cByJ and DBA/2J mice. The concentration of the amine did not differ in the remaining strains. Levels of NE were reduced by the shock treatment, $F(1,250) = 29.61$, $p<0.01$, but this effect did not interact with the strain of mouse. However, as seen in Fig. 1 and confirmed by Newman-Keuls comparisons, the reduction of NE induced by the shock treatment in C3H/HeJ did not reach statistical significance, while in the remaining strains this reduction was significant.

In contrast to the hypothalamus, MHPG accumulation within the locus coeruleus varied as a function of the Shock treatment × Strain interaction, $F(5,201) = 2.91$, $p<0.05$. Newman-Keuls mul-

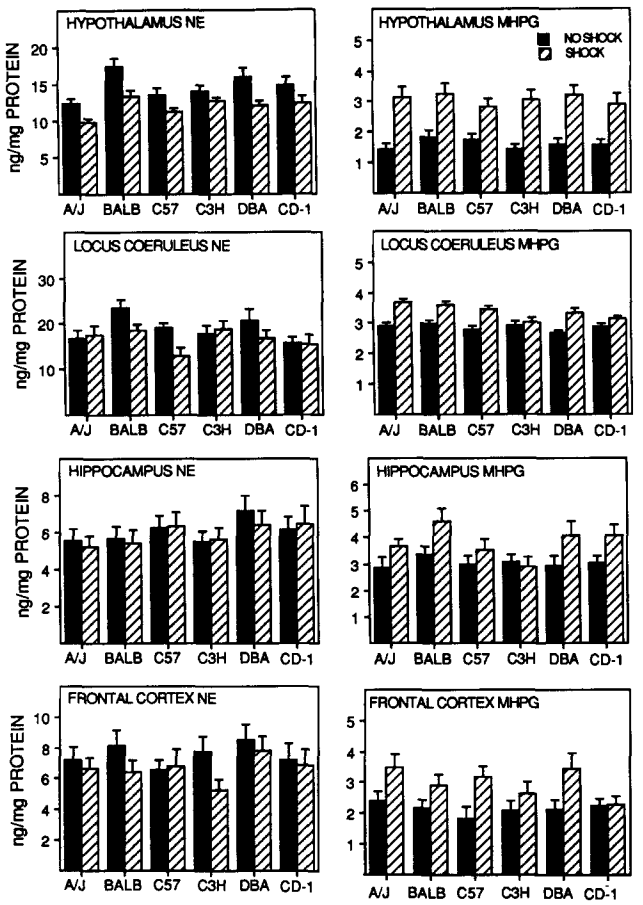

FIG. 1. Mean (±S.E.M) concentrations of NE (left panels) and MHPG (right panels) in the hypothalamus, locus coeruleus, hippocampus and mesocortex in six strains of mice (A/J, BALB/cByJ, C57BL/6J, C3H/HeJ, DBA/2J and CD-1) that had been exposed to either footshock or no shock treatment.

tiple comparisons of the simple effects comprising this interaction indicated that in the absence of the shock treatment, the accumulation of MHPG was comparable across strains. Stressor exposure increased MHPG concentrations in A/J, BALB/cByJ, C57BL/6J and DBA/2J mice, but not in the two remaining strains. Analysis of the NE concentrations revealed that levels of this amine varied across strains, $F(5,206)=2.42$, $p<0.05$, and were reduced by the Shock treatment, $F(1,206)=4.26$, $p<0.05$. The Shock treatment × Strain interaction did not reach statistical significance. Newman-Keuls multiple comparisons that were nevertheless conducted confirmed that in the BALB/cByJ, C57BL/6J and DBA/2J mice the levels of NE were reduced by the stressor. In the remaining strains, however, NE levels were unaffected by footshock (see Fig. 1). The fact that NE levels remained stable in A/J mice despite the increased MHPG accumulation is probably attributable to an increase in synthesis of the amine.

As reported by other investigators (22), only limited alterations of hippocampal NE activity were induced by the stressor. The analysis of variance revealed that hippocampal MHPG concentrations were increased by the shock treatment, $F(1,213)=12.14$, $p<0.01$. As seen in Fig. 1 and confirmed by the Newman-Keuls comparisons, the increase of MHPG induced by the stressor was significant only in the BALB/cByJ, DBA/2J and CD-1 mice. Modest, nonsignificant increases occurred in the A/J and C57BL/6J strains, while in C3H/HeJ there was no indication of a stressor-provoked increase of MHPG accumulation. In contrast to the stressor-induced NE variations seen in the hypothalamus and locus coeruleus, concentrations of the amine in hippocampus did not vary as a function of the Strain or the Shock treatment mice received.

The overall analysis of variance of MHPG accumulation in the anterior mesocortex revealed that stressor exposure increased accumulation of the metabolite, $F(1,147)=17.37$, $p<0.01$. Multiple comparisons of the simple effects confirmed that in the absence of the shock treatment MHPG concentrations were comparable across the strains; however, in A/J, BALB/cByJ, C57BL/6J and DBA/2J mice the stressor significantly increased MHPG accumulation, while in C3H/HeJ the increase was modest and not significant. In CD-1 mice there was no indication of an increase in the metabolite concentrations.

In contrast to the hypothalamus and locus coeruleus, NE levels in the anterior mesocortex were not significantly altered by the shock treatment, $F(1,150)=2.94$, $p=0.088$. While there was a trend towards reduced NE in stressed mice, Newman-Keuls multiple comparisons confirmed that NE concentrations were not significantly altered in those strains in which MHPG accumulation was increased. Only in C3H/HeJ mice, where MHPG was only slightly increased by the stressor, was the concentration of NE significantly reduced following shock exposure.

Dopamine

The concentrations of ventral tegmental DA and DOPAC as a function of the strain and shock treatments are shown in Fig. 2. Analysis of variance indicated that the strains did not differ with respect to DOPAC accumulation, while the shock treatment increased the metabolite accumulation, $F(1,183)=7.47$, $p<0.01$. The Strain × Shock treatment interaction did not reach statistical significance, $F(5,183)=1.82$, $p=0.11$. Newman-Keuls multiple comparisons of the simple effects, however, confirmed that DOPAC concentrations in nonstressed CD-1 mice were significantly lower than in C3H/HeJ and BALB/cByJ mice, while the remaining strains exhibited intermediate concentrations of the metabolite. The shock treatment provoked a very marked increase of DOPAC accumulation in the CD-1 strain (142%), and a marginal increase ($0.05<p<0.10$) in C57BL/6J mice. In none of the other strains was DOPAC affected by the stressor. Analyses of HVA concentrations were conducted in only 2 of 4 replications of this study, and hence the sample size was relatively small (6–11 samples per group). The analysis of the HVA concentrations revealed variable results with neither the strain nor the shock treatment yielding significant effects, although it should be noted that in C3H/HeJ and BALB/cByJ mice there was a tendency towards an increase of HVA concentrations in shocked animals ($0.05<p<0.10$).

Concentrations of DA in the VTA were found to vary as a function of the Shock treatment × Strains interaction, $F(5,161)=2.30$, $p<0.05$. In the absence of the shock treatment DA concentrations were higher in DBA/2J, C3H/HeJ and BALB/cByJ mice than in A/J and C57BL/6J mice. The shock treatment provoked a marked reduction of DA concentrations in BALB/cByJ mice, but not in any of the other strains. Indeed, in the C57BL/6J strain the shock treatment provoked an increase of DA concentrations, but this effect did not reach statistical significance.

The accumulation of DOPAC in the nucleus accumbens was increased by the shock treatment, $F(1,215)=13.13$, $p<0.01$, but this effect did not vary with the strain of mouse. Likewise, the analysis of DA concentrations yielded only a main effect of the shock treatment, $F(1,211)=3.99$, $p<0.05$, indicating a stressor-

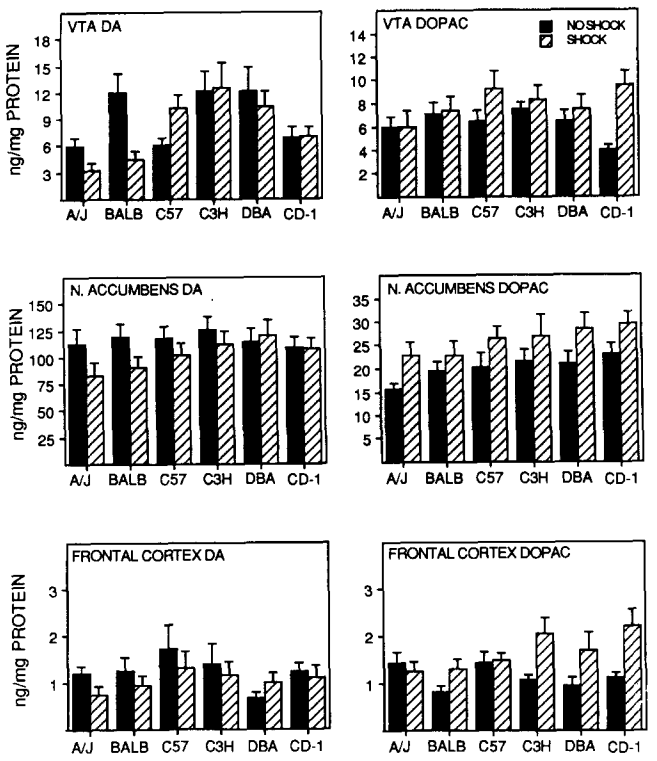

FIG. 2. Mean (±S.E.M) concentrations of DA (left panels) and DOPAC (right panels) in the ventral tegmentum (VTA), nucleus accumbens and mesocortex in six strains of mice (A/J, BALB/cByJ, C57BL/6J, C3H/HeJ, DBA/2J and CD-1) that had been exposed to either footshock or no shock treatment.

induced reduction of the amine. In fact, however, it is clear from Fig. 2 that in some of the strains, notably DBA/2J and CD-1, the shock treatment did not influence DA concentrations. Indeed, despite the absence of a significant Strain × Shock interaction, the variance associated with the shock effect was largely attributable to the effects observed in A/J and BALB/cByJ mice.

Accumulation of DOPAC in the anterior mesocortex varied as a function of the Shock treatment × Strains interaction, $F(5,155) = 2.29$, $p<0.05$. Newman-Keuls multiple comparisons confirmed that in the C3H/HeJ, DBA/2J and CD-1 mice DOPAC accumulation increased after shock exposure, while in BALB/cByJ mice the increase induced by shock approached, but did not reach statistical significance ($0.05<p<0.10$). In neither A/J nor C57BL/6J mice was there any indication of an increase of DOPAC accumulation. Concentrations of DA in the mesocortex were found to be relatively variable, and did not vary as a function of either the Shock or Strains main effects, or as a function of the interaction of these variables. Other investigators (1), as well as earlier work conducted in this laboratory (3), indicated that stressors provoke marked DA reductions in this region, and thus the lack of effect in the present investigation was surprising.

Consistent with earlier reports (1,29), analyses of DOPAC and DA in the substantia nigra and caudate revealed that the stressor treatment was without effect. However, both DA and DOPAC concentrations within the caudate were found to vary across strains of mice, F's(5,234; 5,231) = 4.76 and 3.77, $p<0.05$. Multiple comparisons revealed that DA concentrations in A/J mice were lower than in CD-1, DBA/2J and C3H/HeJ, while the levels in BALB/cByJ were lower than in the CD-1 strain. Likewise, DOPAC accumulation in A/J and BALB/cByJ mice were lower than in CD-1. Similarly, in the substantia nigra, DA and DOPAC concentrations varied across strains of mice, F's(5,115; 5,131) = 3.15, $p<0.05$ and 3.58, $p<0.01$. Multiple comparisons confirmed that DA levels were higher in the C3H/HeJ than in the remaining strains, and DOPAC accumulation was significantly lower in the A/J strain, but higher in the DBA/2J mice relative to the other four strains.

Serotonin

The mean concentrations of 5-HT and 5-HIAA in each of the strains as a function of the shock treatment are shown in Fig. 3. The accumulation of hypothalamic 5-HIAA was found to vary as a function of the Shock × Strain interaction, $F(5,138) = 2.95$, $p<0.05$. Multiple comparisons revealed that in the absence of the shock treatment 5-HIAA concentrations were lower in DBA/2J mice than in any of the remaining strains, except for C57BL/6J mice. Following the shock treatment, 5-HIAA accumulation was increased significantly in C57BL/6J mice, and marginally in the C3H/HeJ mice relative to nonshocked animals of these strains. As a result, the metabolite concentrations in the latter two strains significantly exceeded those of the remaining strains. As previously observed (28), the stressor treatment produced less profound changes of 5-HT than those of NE. There were no statistically significant stressor-induced alterations observed in the hypothalamic concentrations of 5-HT, although the amine levels varied across strains of mice, $F(5,135) = 5.79$, $p<0.01$. Multiple comparisons revealed that 5-HT levels were higher in the C3H/HeJ strain relative to the CD-1 and DBA/2J strains, while concentrations of the amine in C57BL/6J mice exceeded those of the CD-1 strain. The Strain × Shock treatment interaction did not reach statistical significance, $F(5,135) = 2.08$, $p = 0.072$, although multiple comparisons confirmed that footshock provoked a significant rise of 5-HT levels in C57BL/6J mice.

The accumulation of 5-HIAA in the anterior mesocortex was found to vary as a function of the Shock treatment, $F(1,140) = 8.82$, $p<0.01$. However, as evident in Fig. 3, although the shock treatment induced relatively marked increases of the metabolite in BALB/cByJ, C57BL/6J and C3H/HeJ mice, only a moderate increase was evident in mice of the A/J strain, while there was no indication of such an elevation in either the DBA/2J or CD-1 mice. Mesocortical 5-HT levels did not vary across the strains, but the stressor was found to produce marked reductions of the amine, $F(1,135) = 10.60$, $p<0.01$. Once again, however, a posteriori comparisons revealed that the stressor provoked 5-HT reductions in excess of 50% in some strains (e.g., DBA/2J, A/J) even though the increase of 5-HIAA was moderate or not evident in these strains. In other strains (e.g., BALB/cByJ, C57BL/6J and C3H/HeJ) which had displayed appreciable increases in metabolite accumulation, the 5-HT reduction, although statistically significant, was less marked. Finally, 5-HT levels, like the accumulation of 5-HIAA, was not affected by the stressor in CD-1 mice.

The stressor treatment did not appear to affect hippocampal 5-HT and 5-HIAA appreciably in that no significant Shock main effects were indicated. However, the analysis did reveal a statistically significant Strain main effect for hippocampal 5-HIAA levels, $F(5,187) = 2.47$, $p<0.05$. The multiple comparisons indicated that metabolite levels were higher in the C57BL/6J strain as compared to the DBA/2J strain. None of the remaining four strains differed from one another.

DISCUSSION

Consistent with earlier reports, exposure to acute footshock elicited region-specific alterations of amine activity and concen-

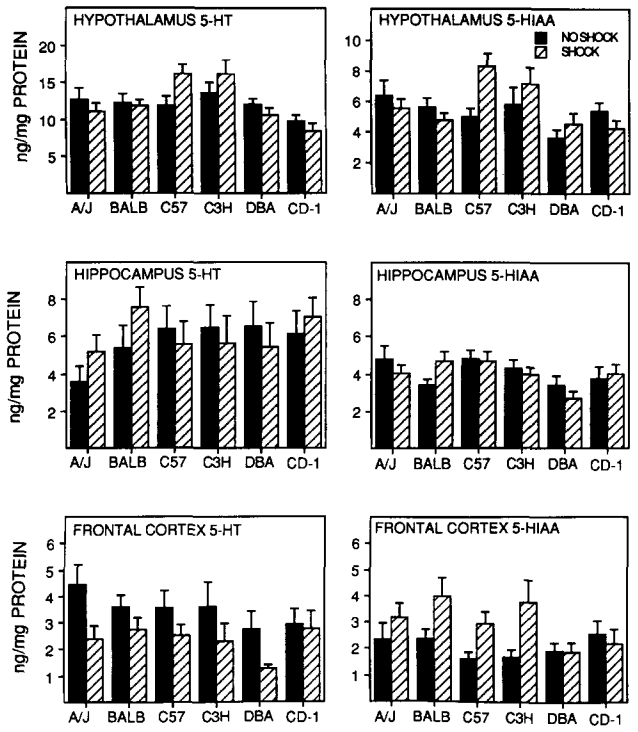

FIG. 3. Mean (±S.E.M) concentrations of 5-HT (left panels) and 5-HIAA (right panels) in the hypothalamus, mesocortex and hippocampus in six strains of mice (A/J, BALB/cByJ, C57BL/6J, C3H/HeJ, DBA/2J and CD-1) that had been exposed to either footshock or no shock treatment.

trations (6,22), but these effects appeared to vary in a strain-dependent fashion. The accumulation of hypothalamic MHPG increased appreciably in each of the strains, and NE concentrations were reduced in all but the C3H/HeJ mice. The stressor-induced variations of NE in other brain regions, most notably those comprising the dorsal bundle, could readily be distinguished from those of the hypothalamus. Furthermore, the strain profile for the NE alterations in the locus coeruleus, the site of origin of the dorsal bundle, was distinguishable from that seen at terminal regions (e.g., the hippocampus and mesocortex). For instance, the largest accumulation of MHPG in the locus coeruleus was evident in A/J and C57BL/6J mice, followed by BALB/cByJ and DBA/2J. In the latter three strains the increased MHPG accumulation was accompanied by a decline of NE concentrations, while this was not the case in A/J mice. Evaluation of NE concentrations in terminal sites indicated that the strain profile for the effects of the stressor did not entirely parallel that observed within the locus coeruleus. In particular, the greatest MHPG accumulation in mesocortex occurred in A/J, DBA/2J, C57BL/6J and BALB/cByJ mice; however, reductions of NE were most pronounced in BALB/cByJ and C3H/HeJ mice, even though the latter strain exhibited a relatively small increase of MHPG concentrations in the mesocortex and also displayed the smallest alterations of NE activity within the locus coeruleus. Finally, as previously observed (22), the acute stressor treatment elicited smaller hippocampal NE variations relative to those seen in other regions. Although MHPG accumulation was apparent in some of the strains, NE concentrations were not reduced. Thus while locus coeruleus activity, and the influence of somatodendritic receptor stimulation, may regulate NE release in terminal regions (2, 7, 17), the effects of stressors on NE release may be determined by factors in addition to activity at the site of origin. Moreover, it is likely that the regulatory mechanisms influencing NE activity in one strain are different from those in a second strain.

In assessing the behavioral impairments engendered by stressors, Weiss and Goodman-Simson (31) offered the view that NE variations within the locus coeruleus were fundamental to the induction of behavioral disturbances. While not dismissing a role for the locus coeruleus, the results of the present investigation suggest that alternative mechanisms contribute to the stressor-induced behavioral impairments. It will be recalled that stressors readily increased NE utilization and reduced the concentrations of this amine within the locus coeruleus of BALB/cByJ, C57BL/6J and DBA/2J mice. Yet we previously observed that footshock greatly impaired shuttle escape performance in BALB/cByJ mice, produced only a modest disturbance in the C57BL/6J strain, and hardly affected the performance of DBA/2J mice (26).

Consistent with earlier observations (14,29), stressor exposure did not appreciably influence nigrostriatal DA turnover, but was effective in modifying mesocorticolimbic DA activity. As in the case of NE, it appeared that the extent of the DA alterations varied with the strain of mouse, as well as with the brain region examined. In the VTA the accumulation of DOPAC or HVA was increased by the stressor in CD-1, BALB/cByJ and C3H/HeJ mice, and pronounced DA reductions were apparent in some of the strains (e.g., BALB/cByJ and A/J). In other strains, however, DA levels were hardly affected by the stressor, or were even marginally increased (e.g., C57BL/6J). In the nucleus accumbens the accumulation of DOPAC was moderately increased, being most prounced in A/J, C3H/HeJ and DBA/2J mice. In the A/J and BALB/cByJ strains, levels of DA were reduced by the stressor treatment, even though the DOPAC accumulation in the latter strain was not appreciable. Finally, in the mesocortex the stressor increased DOPAC accumulation in C3H/HeJ, DBA/2J and CD-1 mice, and only marginal reductions of DA concentrations were evident in A/J and BALB/cByJ mice. As indicated earlier, DA activity within the mesocortex has typically been reported to be readily affected by stressors, and certainly more pronounced than in the nucleus accumbens (3,6). Indeed, in contrast to the results of the present investigation, we observed that 60 shocks of 150 μA (as opposed to 360 shocks of 300 μA in the present investigation) appreciably increased DOPAC accumulation and reduced DA concentrations within the prefrontal cortex of CD-1 mice (3). Thus the absence of such an effect in the present investigation was unexpected. Interestingly, it has been reported that while stressor-induced DOPAC changes occur more readily in the prefrontal cortex than in the nucleus accumbens, as the stressor session continues adaptation of DA utilization occurs in prefrontal cortex, whereas in the nucleus accumbens the accumulation of DOPAC persists (23). It is conceivable that the stressor parameters employed in the present investigation were of sufficient severity and duration to permit the adaptation of the mesocortical DA alterations, while favoring the variations of the amine in the nucleus accumbens.

The finding that the stressor treatment did not have comparable effects on DA activity within terminal regions (e.g., mesocortex and nucleus accumbens) and site of origin (VTA) is not particularly surprising. Moreover, a neurochemical distinction can be drawn with respect to the effects of stressors on mesocortical and mesolimbic activity. For instance, it has been demonstrated that mesocortical DA activity may be influenced by substance P (8,9) and by Met-enkephalin activity (19) in the VTA. It seems, however, that stressors primarily influence Met-enkephalin activity in the medial portion of the VTA, which projects to the mesocortex, without appreciably affecting the lateral and ventral VTA which largely comprises the mesolimbic DA system (18).

In contrast to the enhancement of mesocortical DA activity effected by substance P release, substance K appears to have a preferential action on A10 neurons of the mesolimbic system (11). Paralleling these findings, it was observed that exposure to footshock differentially influenced responding for self-stimulation from different aspects of the VTA, thus providing a functional differentiation for the apparent neurochemical heterogeneity of the VTA that had already been available (34).

As indicated by other investigators (12,20), 5-HT activity was influenced by stressor exposure. However, as in the case of NE and DA, it seemed that the extent of the 5-HT and 5-HIAA changes was dependent upon the strain of mouse and the brain region examined. The most pronounced serotonergic alterations were evident in the mesocortex, as evidenced by enhanced 5-HIAA accumulation in all but the DBA/2J and CD-1 strains, and reduced 5-HT concentrations in all but the CD-1 mice. It should be noted, however, that Dunn (12) reported reductions of 5-HT in the mesocortex and in hypothalamus following 15 min, but not after 30 min, of restraint. Accordingly, it should be considered that the absence of stressor-induced 5-HT alterations in the hypothalamus in the present investigation, as well as the detection of strain differences, may be related to interstrain variations in the rate of the adaptation.

It was argued previously that the strain differences in response to stressors in several behavioral paradigms cannot be ascribed to some of the strains being hardier than others, or alternatively to nociceptive differences which might exist across the strains (25,30). After all, if such factors contributed to the strain-specific behavioral impairments, then it might have been expected that the hardier strains would exhibit the smallest behavioral disturbances across a variety of behavioral tasks. To the contrary, it appeared that strains which exhibited marked behavioral impairments in one paradigm (e.g., shuttle escape) displayed proficient performance in other paradigms (i.e., self-stimulation from the nucleus accumbens) (25,35). In a like fashion, the data of the present investigation suggest some strains exhibited relatively greater alterations of NE than DA activity, or vice versa. Moreover, variations of a transmitter in one brain region were not necessarily predictive of similar alterations in other regions. Nevertheless, it did appear that BALB/cByJ mice were more vulnerable to some neurochemical alterations than the remaining strains. For instance, relative to the other strains, BALB/cByJ mice were more likely to exhibit NE alterations (e.g., in hypothalamus, locus coeruleus and mesocortex), as well as DA reductions in the VTA. Moreover, in this strain moderate reductions of DA were evident in the nucleus accumbens and mesocortex, as were reductions in cortical 5-HT concentrations. It was likewise reported that stressor-induced mesocortical DA turnover is more pronounced in BALB/cByJ than in C57BL/6 mice (15), and also that footshock provokes a particularly marked rise of plasma corticosterone in BALB/cByJ mice relative to other remaining strains (27). In contrast to the vulnerability of BALB/cByJ mice, employing the stressor parameters of the present investigation the noninbred CD-1 mice appeared to be least vulnerable to amine alterations. These mice typically showed increases in the utilization of DA, as well as NE in hypothalamus, but reductions of amine concentrations were rarely apparent, suggesting that the enhanced utilization was met with adequate synthesis. This should not be misconstrued to imply that CD-1 mice are invulnerable to stressor-elicited amine variations. Indeed, we have demonstrated previously that using a somewhat different procedure, mesocortical DA reductions could readily be engendered in CD-1 mice (4).

Given the pronounced genetic variations in the behavioral changes exerted by stressors, it was of particular interest to determine whether the alterations of one or more transmitters in different brain regions might correspond with any of the stressor-induced behavioral impairments in the strains of mice. For instance, we previously observed that following exposure to inescapable shock pronounced deficits of escape performance were evident in BALB/cByJ and C3H/HeJ mice, moderate behavioral disturbances were apparent in the C57BL/6J, CD-1 and A/J strains, and hardly any disruption in performance was apparent in the DBA/2J mice. In a forced swim task the pattern of responding was quite different, in that inescapable shock resulted in reduced vigorous swimming in C57BL/6J mice, provoked a response enhancement in both the BALB/cByJ and CD-1 mice, while the performance of the remaining strains was hardly affected. Finally, evaluation of self-stimulation from the nucleus accumbens and mesocortex revealed that performance in DBA/2J mice was disrupted by inescapable shock in both regions, whereas the performance in the C57BL/6J strain was hardly affected. In BALB/cByJ mice, however, self-stimulation from the mesocortex was disrupted, whereas responding for brain stimulation from the nucleus accumbens was enhanced (34,35). None of the stressor-induced neurochemical profiles in these strains was congruent with the behavioral profiles previously observed. In general, correspondence between specific stressor-induced behavioral and neurochemical alterations could not be identified. For instance, contrary to the pattern of self stimulation from the nucleus accumbens, where BALB/cByJ mice exhibited enhanced responding and DBA/2J mice exhibited reduced responding, the most prominent DA reductions in both the nucleus accumbens and in the VTA were in BALB/cByJ mice. Separate analyses of the DOPAC/DA ratio likewise failed to reveal a parallel with the stressor-induced behavioral changes. While such a finding does not preclude a role for DA in accounting for the disruption of self- stimulation or for the stressor-related strain differences, it does suggest that other factors likely contribute to the behavioral effects of the stressor. Indeed, recent data collected in this laboratory (34) indicated that in CD-1 mice, the stressor-induced disruption of self-stimulation from the A10 region could be antagonized by administration of the opioid analogue, D-Ala^2-Met^5-enkephalinamide. Thus, as discussed earlier, it is likely that endogenous opioids contribute to the stressor-induced behavioral changes. It remains to be determined whether the endorphin changes associated with a stressor in these various strains of mice parallel the observed behavioral changes.

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