

Influence of Different Antidepressant Drugs on the Effect of Chronic Variable Stress on Restraint-Induced Dopamine Release in Frontal Cortex

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The aim of this study was to evaluate the influence of an early chronic variable stress procedure (CVS) associated or not with repeated administration of various antidepressants on cortical restraint-induced dopamine (DA) release in vivo. Animals were subjected to the CVS schedule and one day after submitted to persistent administration with vehicle, desipramine (DMI, 10 mg/kg, i.p.), fluoxetine (FLU, 10 mg/kg, i.p.) or phenelzine (PHE; 10 mg/kg, i.p.) and later on exposed to a 60-min restraint period. In addition, we also explored the effect of acute administration

of these antidepressants on cortical DA overflow in response to restraint in CVS treated rats. A higher increase in cortical DA release in response to restraint was observed in CVS animals as compared with those without previous CVS. Persistent, but not acute, administration with DMI, FLU and PHE blocked the sensitized output induced by restraint following CVS exposure.

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It is well known that exposure to non-aversive arousing stimuli or activation of cognitive processes are both capable of inducing an increase in cortical dopaminergic activity (Feenstra et al. 1995; Feenstra and Botterblom 1996). In addition, a number of findings obtained from microdialysis and postmortem studies have consistently shown that mesoaccumbal and mainly mesocor-

tical dopaminergic pathways are selectively activated by diverse types of uncontrollable aversive stimuli (Thierry et al. 1976; Fadda et al. 1978; Deutch et al. 1985; Abercrombie et al. 1989). In spite of the well known activation of these dopaminergic systems following stress, the functional role of such systems in emotional behavior remains to be established. However, it has been tentatively proposed that prefrontocortical dopaminergic neurons could play a potential role in the emotional and behavioral sequelae after exposure to unavoidable aversive events (Biggio et al. 1990; Horger and Roth 1996; Espejo and Miñano 1999; Morrow et al. 1999). In addition to such neurochemical changes, uncontrollable stressors have been reported to induce behavioral aberrations analogous to important symptoms of human depression (Willner 1991). Whether these behavioral alterations are associated with the activation of cortical dopaminergic projections following stress remains unclear. However, data from recent studies may be indicative of such association. For instance, a higher cortical

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dopaminergic response to an unavoidable aversive stimulus—restraint—was found in animals previously submitted to a procedure with persistent and dissimilar stressors (Cuadra et al. 1999)—an animal model of depression—(Katz et al. 1981; Willner 1991). In line with such evidence, a higher activation of cortical DA response following uncontrollable shocks was found as compared to controllable shocks (Carlson et al. 1993; Cabib and Puglisi-Allegra 1994) using the learned helplessness paradigm—another model of depression. In a recent study, Espejo and Miñano (1999) suggested that depressive-like behaviors such as immobility during a forced swim test—a screening paradigm to evaluate antidepressive efficacy (Porsolt et al. 1978)—were linked to the enhancement of endogenous DA levels in medial prefrontal cortex. Moreover, this effect was blocked by subchronic DMI administration. The medial prefrontal cortex area among other brain centers has been postulated to participate in the alleviation of shock-induced deficit by intracranial administration of antidepressants (Petty and Sherman 1980; Sherman and Petty 1980). Studies performed in depressive patients seem to support the hypothesis that frontal cortical areas are involved in depression. For instance, functional brain imaging studies have pointed out functional changes in medial prefrontal cortex in depression (Buchsbaum et al. 1986) as well as an increased blood flow in the medial prefrontal cortex and ventral orbital cortex in patients with unipolar depression (Drevets et al. 1992). In addition, changes in blood flow were normalized after antidepressant drug therapy (Drevets and Raichle 1992).

A growing number of studies have revealed that very much like the antidepressive effect on depressed patients, prolonged antidepressant treatment but not acute administration reverses stress-induced behavioral disturbances. Noradrenergic and serotonergic projections have been long thought to be involved in the pathophysiology of mood disorders and in the mechanism of action of antidepressant drugs. However, and in addition to these neurotransmitters, a role for DA as part of the biochemical basis of depression has also been suggested (Willner 1983; Zacharko and Anisman 1991; Kapur and Mann 1992). Moreover, and despite the fact that antidepressant drugs have traditionally been reported to exert their primary action on noradrenergic and serotonergic neurons, a role for dopaminergic processes in the central effects of such drugs has also been suggested. For instance, acute and chronic antidepressants can influence dopaminergic activity on frontocortical areas (Tanda et al. 1994; Tanda et al. 1996). However, a significant drawback in extrapolating such findings to the human condition is that most of them were obtained from “normal” or unstressed rats. A more relevant approach is to evaluate the effect of these drugs on cortical dopamine responses in para-

digms designed and currently used to mimic symptoms of human depression. Therefore, the present study attempts to explore the effect of different types of antidepressant agents on cortical dopamine response (extracellular DA overflow) to an acute stressful event in animals previously exposed to a chronic variable stress procedure (CVS), a validated animal model of depression (Willner 1991). One day after the last stressful event of the CVS, animals were either submitted to repeated administration with desipramine (DMI), a tricyclic antidepressant with high affinity to block NA uptake, fluoxetine (FLU), a selective serotonin reuptake inhibitor, or phenelzine (PHE), an MAO inhibitor followed by acute restraint. Furthermore, since antidepressants must be administered chronically for therapeutic benefit (Klein and Davis 1970), we also compared the effect of acute administration with that following chronic antidepressants on cortical DA release induced by stress in previously CVS animals.

MATERIALS AND METHODS

Animals

Male Wistar rats, from our own colony, weighing 280–300 g were used in this study. Rats were housed in groups of four rats per cage with food and water freely available, under a 12-hour light-dark cycle (lights on at 7 A.M.).

All procedures were in agreement with the standards for the care and use of laboratory animals as outlined in the NIH Guide for the Care and Use of Laboratory Animals.

Surgery and Microdialysis Procedure

Rats were anesthetized with sodium pentobarbital (62.5 mg/kg, i.p.) and placed in a Stoelting stereotaxic frame with the incisor bar at –3.3 mm below interaural zero. Microdialysis was performed as previously described (Cuadra et al. 1994). Briefly, AN69 HF dialysis fiber, inner diameter 200 μm and outer diameter 340 μm (Hospal, Meyzieu, France), were transversally implanted in the frontal cortex (coordinates versus bregma, A: +3.2; V: –2.6; atlas of Paxinos and Watson 1986). Dialysis was confined to the frontal cortex by covering the dialysis fiber with epoxy resin throughout its whole extension except for 6 mm of the portion in contact with cortical tissue (Figure 1). The probe was fastened to the skull with screws and dental cement and the skin was sutured. Rats were then placed into individual acrylic bowls and left to recover for at least 24 hours.

Experiments began the following day. The dialysis membrane was perfused with Ringer's solution (NaCl 147 mM; KCl 4.0 mM; CaCl₂ 1.5 mM, MgCl₂ 0.8 mM) at a constant flow rate of 1.1 $\mu\text{l}/\text{min}$. Following equilibra-

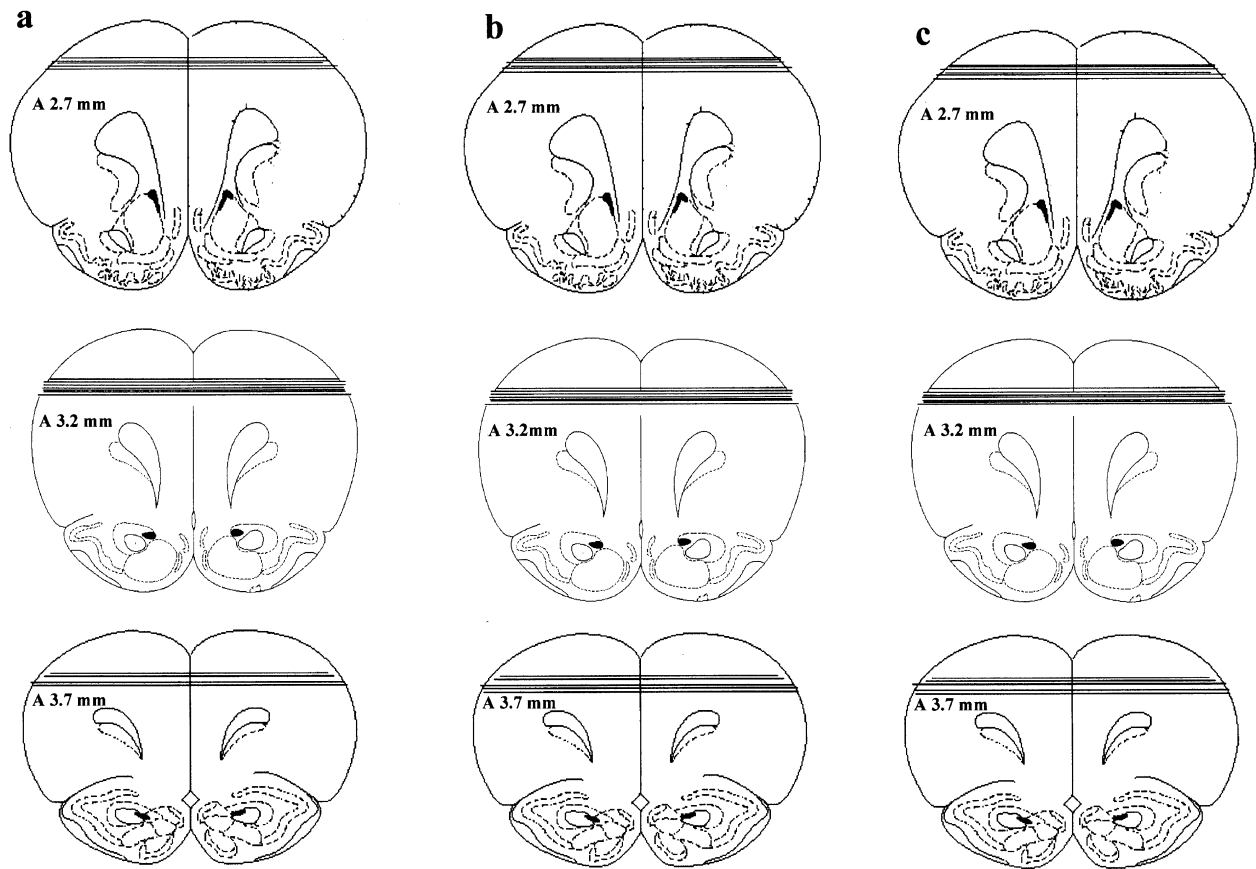


Figure 1. Schematic drawings of coronal sections of the rat brain adapted from the atlas of Paxinos and Watson (1986), showing the location of the dialysis surface of probes implanted in frontal cortex. **Panel a:** Probe locations in DMI group; **Panel b:** Probe locations in FLU group; **Panel c:** Probe locations in PHE group.

tion, samples of the dialysate were collected every 30 minutes into vials containing 1 μ l acetic acid (0.1 N) to prevent dopamine oxidation. Vials were kept at 4°C in a refrigerated fraction collector. Rats were immobilized for 60 min starting at time 0 after collection of six baseline samples (six consecutive samples differing no more than 10%). Control levels were defined as an average of these baselines. Corrections were made to account for dead volume. At the end of the experiments animals were sacrificed, their brains were removed and the position of the dialysis probe track was histologically verified.

Monoamine assay

The amount of dopamine in the collected fractions was analyzed on a High-Pressure Liquid Chromatography (HPLC) equipped with a reverse-phase column (ultrasphere C18, 15 cm, 5 μ m particle size, Beckman) coupled with electrochemical detection. The HPLC system consisted of a BAS LCD-4 electrochemical detector with a glass-carbon electrode and a pump (SpectraSeries P200). The potential was set at 650 mV (vs Ag\AgCl reference electrode). The mobile phase containing 120

mM citric acid; 110 mM sodium acetate; 1.2 mM 1-octane sulfonic acid; 0.39 mM EDTA; 12% methanol (pH 3.6) was filtered and pumped through the system at a flow rate of 1.0 ml/min.

Under these conditions, the limit of detection was 5 femtomoles per sample. Peaks from HPLC system were displayed, integrated and stored with PeakSimple II Data System (SRI Instr, Torrance, CA, USA). Quantification of all substances was made by comparing peak heights of the samples to a standard curve.

Chronic and variable stress

The chronic variable stress schedule used was similar to that previously described (Murua et al. 1991; Molina et al. 1994). Animals assigned to the chronic variable stress procedure were exposed to the following stressors: 90 minutes of intermittent (on 5 min/off 1 min) shaker (high speed); 30 unpredictable shocks (1 mA, 3 sec duration, one shock/30 sec); intermittent white noise (90 dB) with bright light (200–250 lx) for 90 minutes (on 5 min/off 1 min); crowding by placing four un-

familiar animals into a standard individual cage for 16 hours. One stressor was applied per day and the order of stress administration was as follows: 1) shaker, 2) shock, 3) white noise, 4) crowding, 5) shaker, 6) shock, 7) white noise.

Experimental Procedures

Experiment 1: Effect of prior chronic variable stress exposure on restraint-induced dopamine release in frontal cortex. Effect of repeated administration of three different antidepressants. Animals were randomly assigned to four groups: vehicle-control (VH-CON, unstressed animals administered with vehicle), vehicle-chronic variable stress (VH-CVS, chronically stressed animals administered with vehicle), antidepressant-control (DMI-CON, FLU-CON, PHE-CON, unstressed animals administered with either DMI, FLU or PHE) and antidepressant-chronic variable stress (DMI-CVS, FLU-CVS, PHE-CVS, chronically stressed animals administered with either DMI, FLU or PHE). During seven consecutive days animals were exposed or not to the CVS schedule, both groups (CVS or CON) were then administered DMI, FLU, PHE or vehicle as follows: on day 7 a single administration of antidepressant (10 mg/kg, i.p.) or vehicle, day 8 to day 10 rats were injected with DMI, FLU or PHE (5 mg/kg, i.p. each administration) or vehicle twice a day. On day 11, dialysis fibers were implanted. On day 12, following assessment of baseline values, all animals were submitted to a 60-min restraint session. This stressor was carried out in acrylic tubes, fitted closely to body size, leaving the head of the rat outside of the restraint device. Samples were collected during 360 minutes. The dose of the antidepressant drugs used was selected based on previous findings which showed that at this dose, antidepressant drugs were highly effective to normalize those behavioral aberrations suggested to resemble symptoms of human depression (Murua and Molina 1991; Lacerra et al. 1999).

Experiment 2: Effect of prior chronic variable stress exposure on restraint-induced dopamine release in frontal cortex. Effect of a single administration of three different antidepressants. Animals were submitted to a chronic variable stress regime and assigned to four groups as previously described. After seven days of treatment animals remained undisturbed until day 10, when a single dose of DMI, FLU, PHE (5mg/kg, i.p.) or vehicle was administered. On day 11, dialysis fibers were implanted and the response to restraint was assessed by microdialysis 24 hours later.

Drugs

Drugs were obtained from the following sources (in parentheses): sodium pentobarbital (Abbott Lab., N.

Chicago, IL, USA), desipramine hydrochloride (Prest Laboratories, Buenos Aires, Argentina), fluoxetine hydrochloride (Prest Laboratories, Buenos Aires, Argentina) and phenelzine sulfate (Prest Laboratories, Buenos Aires, Argentina). Drugs were dissolved in water. Water was used for control injections (vehicle). All injections were given in a volume of 1 ml/kg.

Statistics

Values are expressed as mean \pm S.E.M. Data were analyzed by a 2-way ANOVA for repeated measures with treatment as between-subjects, factor and time as within-subjects factor. Treatments were defined as the combination of stress (presence or absence) and drug (presence or absence) factors. Degrees of freedom for time and time X treatment hypotheses were corrected using Greenhouse and Geisser Epsilon (Lindsey 1993) because of lack of sphericity to comply with the Mauchly's criterion ($p < .0001$), for the variance-covariance matrix of the observations coming from the same individual (rat).

In addition, to explore between-within factor interaction in the repeated measured analysis, we used 1-way ANOVAs and subsequent treatment (VH-CON; VH-CVS; DRUG-CON; DRUG-CVS) mean contrasts at any single time point ($\alpha = .05$).

Statistical analysis was performed using the software SAS/STAT version 6.12 (SAS Institute, Cary, NC, USA, 1997).

RESULTS

Experimental Procedures

Experiment 1: Effect of early chronic variable stress exposure on restraint-induced dopamine release in frontal cortex. Effect of repeated administration of three different antidepressants. As shown in Figure 2, the different experimental conditions had no effect on basal extracellular dopamine in frontal cortex. Baseline levels of dopamine were: vehicle-control group: 0.68 ± 0.05 nM ($n = 6$); vehicle-chronic variable stress group: 0.73 ± 0.08 nM ($n = 6$); DMI-control group: 0.68 ± 0.08 nM ($n = 5$); DMI-chronic variable stress group: 0.71 ± 0.02 nM ($n = 6$). All concentrations are mean \pm S.E.M.

A significant time X treatment interaction was observed among all DA profiles expressed at each time point as percentage of DA baseline, [$F(11,209) = 4.94$, $p = .022$]. Additionally, a significant drug X stress interaction was detected [$F(1,19) = 8.28$, $p = .009$]. Exposure to restraint induced a significant increase in dopamine levels in cortical dialysate. This effect reached a maximum at 120 minutes (139%) and was still evident after 360 minutes (Figure 2, Panel A). A subsequent restraint exposure in chronically stressed rats induced a signifi-

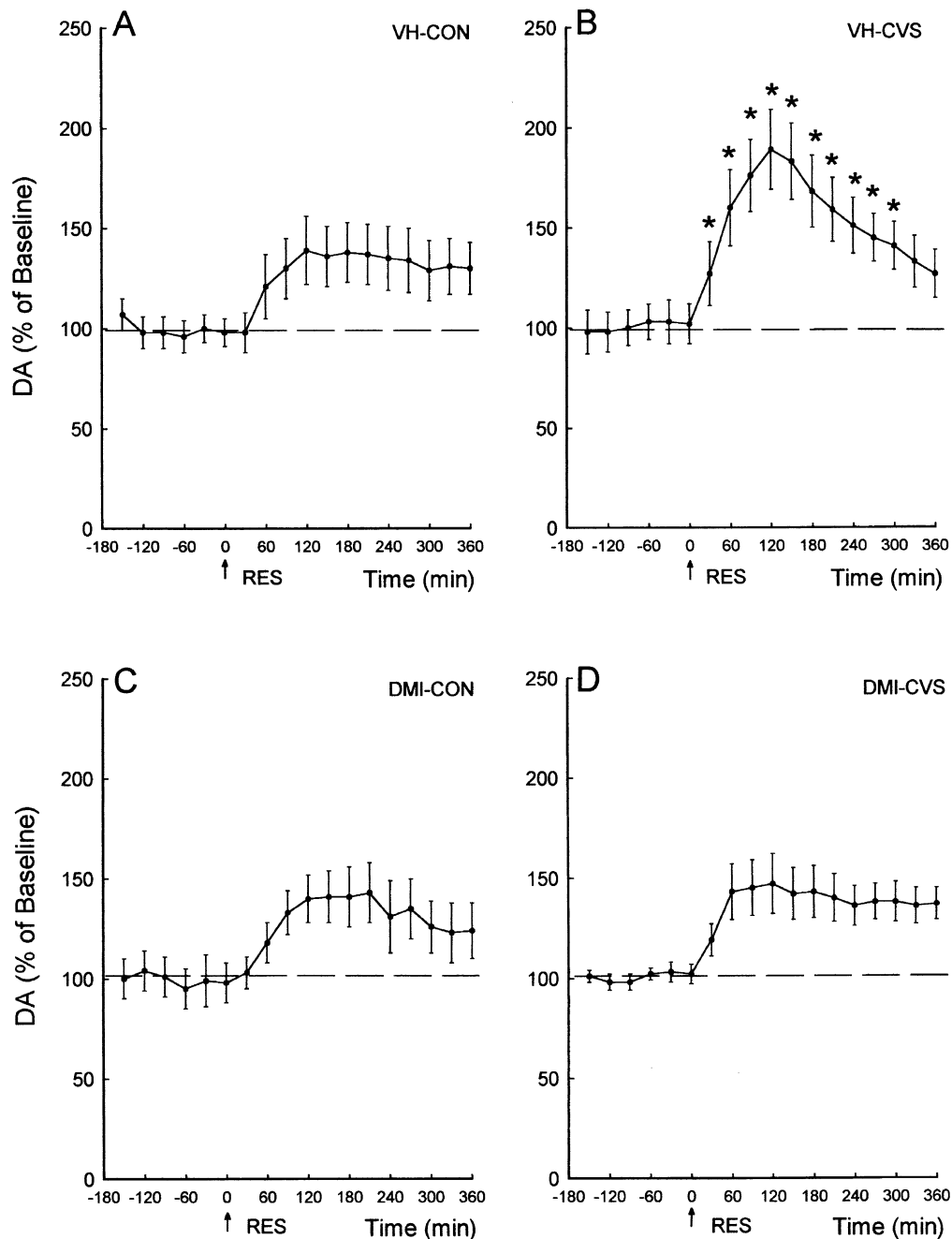


Figure 2. Effect of restraint on extracellular levels of dopamine in microdialysis from frontal cortex in animals submitted or not to CVS, and repeatedly administered with DMI or vehicle: **Panel A:** vehicle-control (VH-CON, $n = 6$), **Panel B:** vehicle-chronic variable stress (VH-CVS, $n = 6$), **Panel C:** Desipramine-control (DMI-CON, $n = 5$) and **Panel D:** desipramine-chronic variable stress (DMI-CVS, $n = 6$) treated groups. Restraint started at time 0 \uparrow . Data are expressed as percentage of control levels (average of six samples before restraint = 100%), mean \pm S.E.M. $*p < .05$ compared with the same time sample of VH-CON, DMI-CON and DMI-CVS groups by means contrasts.

cant increase in extracellular dopamine, 189% at 120 min (Figure 2, Panel B). Therefore, a stronger dopamine release in response to restraint, mainly in peak values between 30 and 300 minutes, was observed in chronically stressed rats as compared to controls. Restraint also induced the characteristic increase in extracellular

dopamine in DMI-control animals. This effect was similar to that observed in the vehicle-control group exposed to restraint, reached a maximum of 143% over basal levels at 210 min and lasted until 360 min (Figure 2, Panel C). However, DMI administered after the chronic variable stress procedure prevented the higher

effect on dopamine release shown by chronically stressed rats administered with vehicle and subsequently submitted to restraint. Exposure to subsequent restraint in chronically stressed subjects administered with DMI induced a 147% increase in dopamine levels at 120 min and remained above baseline levels for at least 360 minutes (Figure 2, Panel D). Furthermore, ex-

tracellular dopamine levels in DMI-chronic variable stress group were similar to those observed in DMI-control group.

As shown in Figure 3, the different experimental conditions had no effect on basal extracellular dopamine in frontal cortex. Baseline levels of dopamine were: vehicle-control group: 0.71 ± 0.06 nM ($n = 6$); ve-

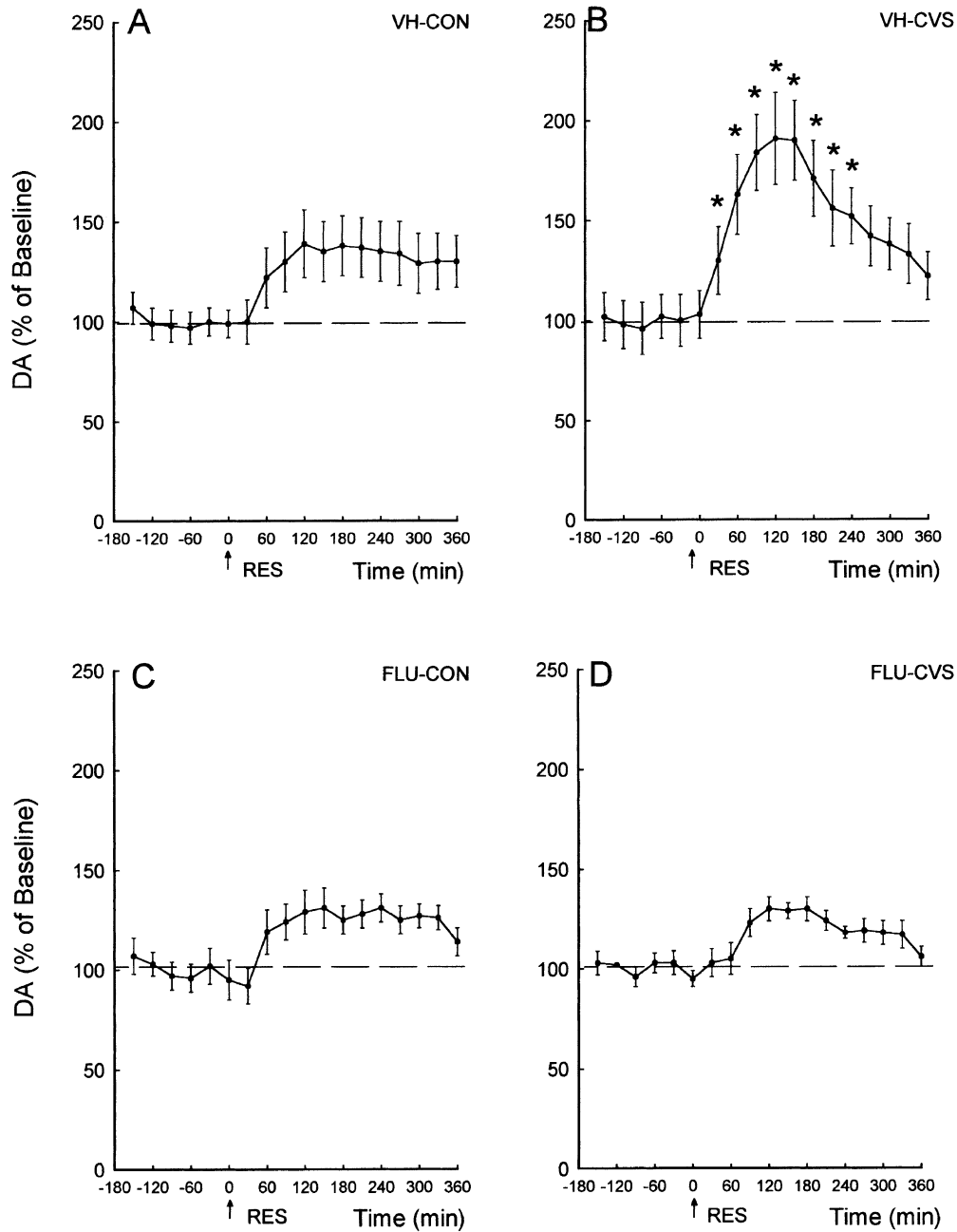


Figure 3. Effect of restraint on extracellular levels of dopamine in microdialysis from frontal cortex in animals submitted or not to CVS, and repeatedly administered with FLU or vehicle: **Panel A:** vehicle-control (VH-CON, $n = 6$), **Panel B:** vehicle-chronic variable stress (VH-CVS, $n = 6$), **Panel C:** Fluoxetine-control (FLU-CON, $n = 6$) and **Panel D:** fluoxetine-chronic variable stress (FLU-CVS, $n = 6$) treated groups. Restraint started at time 0 \uparrow . Data are expressed as percentage of control levels (average of six samples before restraint = 100%), mean \pm S.E.M. * $p < .05$ compared with the same time sample of VH-CON, FLU-CON and FLU-CVS groups by means contrasts.

hicle-chronic variable stress group: 0.70 ± 0.09 nM ($n = 6$); FLU-control group: 0.70 ± 0.06 nM ($n = 6$); FLU-chronic variable stress group: 0.71 ± 0.03 nM ($n = 6$). All concentrations are mean \pm S.E.M.

A significant time \times treatment interaction was observed among all DA profiles expressed at each time point as percentage of DA baseline, [$F(11,220) = 3.50$, p

$= .010$]. Additionally, a significant drug \times stress interaction was detected [$F(1,20) = 8.97$, $p = .007$]. Exposure to restraint induced a significant increase (139% at 120 min) in dopamine levels in cortical dialysate (Figure 3, Panel A). A subsequent restraint exposure in chronically stressed rats induced a significant higher increase (191% at 120 min) in extracellular dopamine (Figure 3,

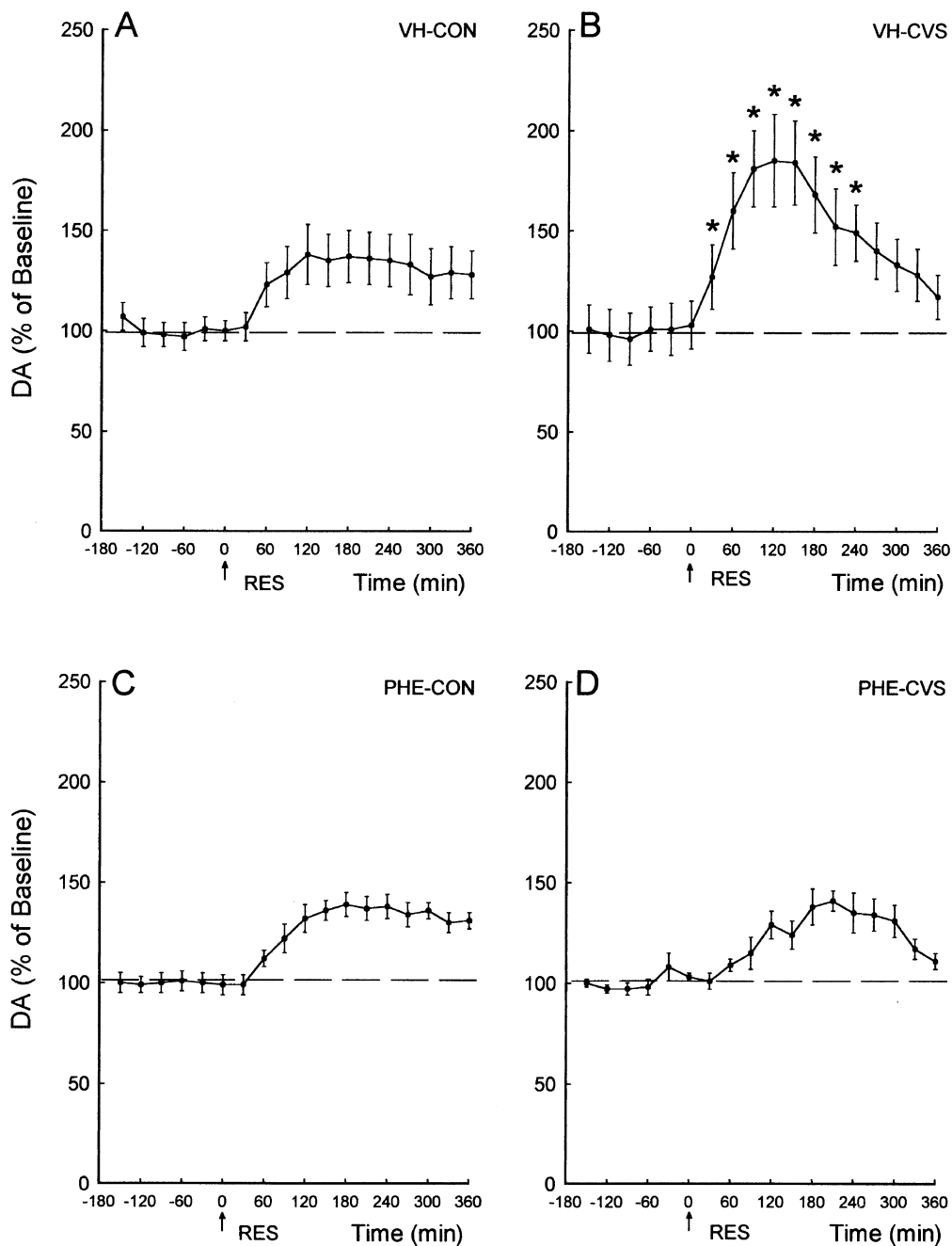


Figure 4. Effect of restraint on extracellular levels of dopamine in microdialysis from frontal cortex in animals submitted or not to CVS and repeated administered with PHE or vehicle: **Panel A:** vehicle-control (VH-CON, $n = 6$), **Panel B:** vehicle-chronic variable stress (VH-CVS, $n = 6$), **Panel C:** phenelzine-control (PHE-CON, $n = 6$) and **Panel D:** phenelzine-chronic variable stress (PHE-CVS, $n = 6$) treated groups. Restraint started at time 0 \uparrow . Data are expressed as percentage of control levels (average of six samples before restraint = 100%), mean \pm S.E.M. $*p < .05$ compared with the same time sample of VH-CON, PHE-CON and PHE-CVS groups by means contrasts.

Panel B) compared with control animals. Therefore, a similar pattern to those described above was observed after CVS exposure. Restraint also induced the characteristic increase in extracellular dopamine in FLU-control animals. This effect was similar to that observed in the vehicle-control group, reached a maximum of 131% over basal levels at 150 min (Figure 3, Panel C). How-

ever, FLU administered after the chronic variable stress procedure prevented the higher effect on dopamine release shown by chronically stressed rats administered with vehicle. Exposure to subsequent restraint in chronically stressed animals administered with FLU induced a significant 130% increase in dopamine levels at 120 min which remained above baseline levels for at least

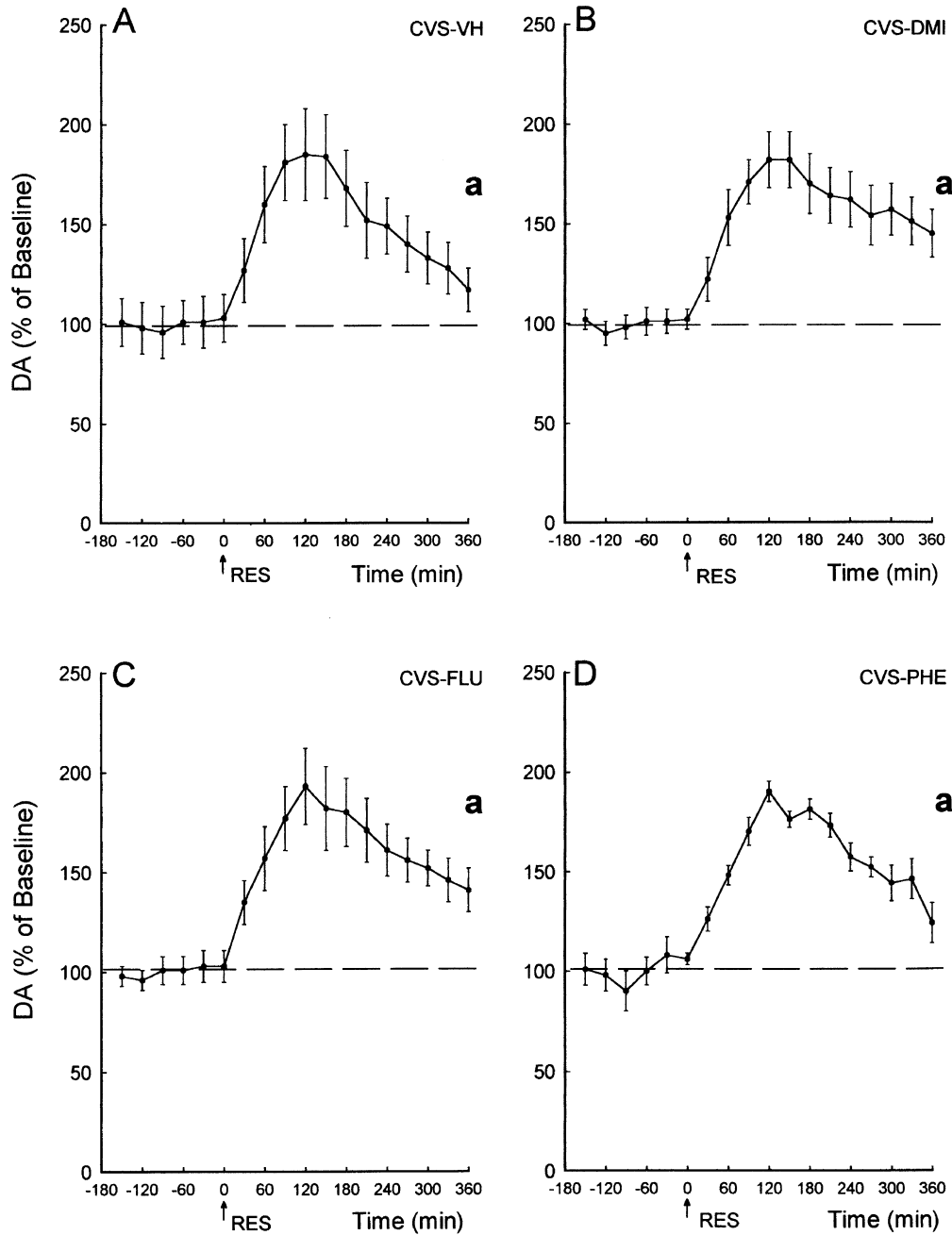


Figure 5. Comparison of the effect of a single administration of vehicle and different antidepressants drugs (VH, DMI, FLU, PHE) on restraint-induced dopamine release in frontal cortex in rats with chronic variable stress pre-exposure: **Panel A:** chronic variable stress-vehicle (CVS-VH, n = 6), **Panel B:** chronic variable stress-Desipramine (CVS-DMI, n = 6), **Panel C:** chronic variable stress-Fluoxetine (CVS-FLU, n = 6) and **Panel D:** chronic variable stress-Phenelzine (CVS-PHE, n = 6). Restraint started at time 0 ↑. Data are expressed as percentage of control levels (average of six samples before restraint = 100%), mean ± S.E.M. Treatment profiles with the same lower-case letters are not statistically different.

360 minutes (Figure 3, Panel D). Furthermore, extracellular dopamine levels in FLU-chronic variable stress group were similar to those observed in FLU-control group.

As shown in Figure 4, the different experimental conditions had no effect on basal extracellular dopamine in frontal cortex. Baseline levels of dopamine were: vehicle-control group: 0.73 ± 0.05 nM ($n = 6$); vehicle-chronic variable stress group: 0.72 ± 0.09 nM ($n = 6$); PHE-control group: 0.69 ± 0.03 nM ($n = 6$); PHE-chronic variable stress group: 0.69 ± 0.02 nM ($n = 6$). All concentrations are mean \pm S.E.M.

A significant time \times treatment interaction was found among all DA profiles expressed at each time point as percentage of DA baseline, [$F(11,220) = 4.45, p = .0011$]. Additionally, a significant drug \times stress interaction was detected [$F(1,20) = 8.19, p = .009$]. Exposure to restraint induced a significant increase (138% at 120 min) in dopamine levels in cortical dialysate (Figure 4, Panel A). A subsequent restraint exposure in chronically stressed rats induced a significant increase (185% at 120 min) in extracellular dopamine (Figure 4, Panel B) compared to control animals. Therefore, a similar pattern to those depicted above was observed after CVS exposure. Restraint also induced the characteristic increase in extracellular dopamine in PHE-control animals. This effect, similar to that observed in the vehicle-control group, reached a maximum of 139% over basal levels at 180 min and lasted at least for 360 minutes (Figure 4, Panel C). However, PHE administered after the chronic variable stress procedure prevented the higher effect on dopamine release shown by chronically stressed rats administered with vehicle. Exposure to subsequent restraint in chronically stressed animals administered with PHE induced a significant 138% increase in dopamine levels at 180 min (Figure 4, Panel D). Furthermore, extracellular dopamine levels in PHE-chronic variable stress group were similar to those observed in PHE-control group.

Experiment 2: Effect of early chronic variable stress exposure on restraint-induced dopamine release in frontal cortex. Effect of a single administration of three different antidepressants. Figure 5 shows the effect of acute administration of VH, DMI, FLU and PHE on the day before testing on restraint induced dopamine release in animals previously submitted to a CVS regime. There was no effect of different experimental conditions on basal extracellular dopamine levels in frontal cortex. Baseline levels of dopamine were: vehicle-chronic variable stress group: 0.73 ± 0.08 nM ($n = 6$); CVS-DMI: 0.73 ± 0.04 nM ($n = 6$); CVS-FLU: 0.73 ± 0.05 nM ($n = 6$); CVS-PHE: 0.70 ± 0.03 nM ($n = 6$). All concentrations are mean \pm S.E.M.

In contrast with the results observed after repeated administration of the three antidepressant drugs a comparison of different treatment with the single dose of

CVS-DMI (Figure 5, Panel B), CVS-FLU (Fig. 5C) and CVS-PHE (Fig. 5D), showed no significant differences in extracellular increased dopamine levels compared to CVS-VH (Figure 5, Panel A) treated group. No treatment \times time interaction was observed among all groups [$F(33,220) = 1.51, p = .15$].

DISCUSSION

Consistent with a large number of findings (Thierry et al. 1976; Fadda et al. 1978; Deutch et al. 1985; Abercrombie et al. 1989), exposure to a single aversive stimulus such as a restraint session promoted a clear increase in extracellular DA in frontal cortex. As previously reported (Cuadra et al. 1999), CVS animals showed a higher increase in cortical DA release in response to the restraint event as compared with rats unexposed to a prior CVS regime. Hence, an early history with repeated dissimilar stressors sensitized the cortical dopaminergic response to a subsequent uncontrollable stressor. A similar sensitization of DA metabolism and DA efflux from frontal cortical tissue was observed following diverse chronic stress procedures (Blanc et al. 1980; Kalivas and Duffy 1989; Gresch et al. 1994). The present results also showed that none of the antidepressants used altered basal DA overflow from frontal cortex in controls. In contrast, Tanda et al. (1996) showed a significant increase in basal extracellular DA from medial prefrontal cortex following a 14-day treatment with a daily 10 mg/kg DMI dose in unstressed rats. These authors suggested that such an effect could be a consequence of a direct blockade of DA uptake in cortical noradrenergic terminals since DMI concentrations can still be high 24 hours after the last administration. Hence, the lack of effect on basal DA overflow in the present experiments following our schedule of antidepressant administration (considerable shorter than that used by Tanda et al. 1996) might point out that at the time interval between the last dose of antidepressant and DA assessment (approximately 36 hours) under our experimental conditions there is no significant drug concentration present in frontal cortical tissue. Moreover, these drugs did not modify the overflow of cortical DA induced by restraint in animals unexposed to previous chronic stress. However, repeated, but not acute, administration with DMI, FLU and PHE all blocked the sensitized DA output in response to restraint following CVS exposure. From these data it is evident that antidepressants exert an effect only on rats exposed to the experimental paradigm of stress-induced depression (CVS). Similarly, it is widely accepted that chronic antidepressant treatments are generally devoid of mood elevating properties in normal humans.

Besides, the direct acute action of these antidepressants to block monoamine reuptake, including dopa-

mine, or monoamine degradation cannot be directly associated with the reversal of the sensitized response to stress. Alternatively, such normalization could be due to adaptive changes induced by repeated administration of these psychotherapeutic agents. Nevertheless, further experiments are necessary to elucidate the precise neural changes following each of the antidepressants used and the functional association of these potential adaptive changes on frontal cortex and the reversal of this sensitization process.

Interestingly, animals exposed to a prolonged treatment with variable stressors similar to those used in the present study showed an exaggerated level of behavioral deficits when these rats were subsequently submitted to diverse types of uncontrollable aversive situations (Molina et al. 1994; Zurita et al. 1999). Based on such findings, it was concluded that past experience with persistent exposure to uncontrollable and dissimilar stressors enhances the vulnerability for the onset of "depressive-like" behaviors in subsequent exposures with an uncontrollable stressor (Zurita et al. 1999). As previously reported (Cabib and Puglisi-Allegra 1994), a higher cortical dopaminergic output was promoted when animals were submitted to uncontrollable shocks as compared to controllable shocks. It is well known that escape acquisition failure and passivity to aversive stimuli are promoted by uncontrollable shocks but not by controllable shocks, a behavioral aberration suggested to model psychomotor retardation as a symptom of depression. In fact, interference in escape acquisition either induced by CVS or uncontrollable shocks is clearly reversed by repeated antidepressants (Sherman et al. 1979; Murua and Molina 1991; Murua et al. 1991). Although the functional significance of the enhanced cortical activation in response to stress is not fully understood, it could be speculated that under certain stressful conditions an increased cortical activation might be involved in the modulation of negatively motivated states triggered by highly aversive experiences. In this line, a sensitized activation of cortical dopaminergic output in response to stress could favor the onset of depressive-like behaviors. Further experiments seem necessary to evaluate such hypothesis.

In conclusion, the present results show that regardless of the different mechanism of action of the antidepressant drugs used, all of them reversed the sensitized cortical dopaminergic output in response to stress in animals previously exposed to the CVS procedure: a validated animal model of depression.

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