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Phenylethylamine, Norepinephrine and Mounting Behavior in the Male Rat

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SEGAL, M., E. SHOHAMI AND D. M. JACOBOWITZ. *Phenylethylamine, norepinephrine and mounting behavior in the male rat.* PHARMACOL BIOCHEM BEHAV **20**(1) 133–135, 1984.—Phenylethylamine, which induces mounting behavior in naive adult male rats when administered chronically, was shown to selectively raise brain norepinephrine levels in the medial preoptic nucleus, a region known to be implicated in the regulation of sexual behaviors. It is suggested that the catecholamine alteration is a secondary response to the primary influence of phenylethylamine on the preoptic nucleus.

Phenylethylamine Norepinephrine Mounting behavior Preoptic nucleus

THE tryptophan hydroxylase inhibitor p-chlorophenylalanine (PCPA) has long been known to induce mounting behavior in male rats [26, 27, 31, 36]. This behavior has been postulated to be due to an altered balance between the serotonergic and dopaminergic systems [6, 7, 8]. Enhanced mounting activity by p-methoxyphenylethylamine (PMPEA) as well as by a large number of other phenylethylamine (PEA) derivatives, when administered in combination with a subliminal quantity of PCPA or when administered alone chronically over a 2-week period, has recently been reported [20,21]. Since some of the effective derivatives, in particular, orthomethoxyphenylethylamine (OMPEA) do not alter serotonergic mechanisms [5], it was concluded that some additional mechanism was required to explain this behavioral phenomenon [20,22].

The monoamine oxidase-Type B inhibitor deprenyl has been reported to increase sexual activity in sluggish male rats to a degree 10 times greater than the monoamine oxidase-Type B [13]. In addition, the chronic administration of selective naturally occurring substrate for monoamine oxidase-Type B. In addition, the chronic administration of d-phenylalanine, a direct precursor of PEA [10], was reported to induce mounting behavior in male rats [22]. The amino acid d-tyrosine, pharmacologically active on the cardiovascular system [24], and l-phenylalanine, active in a variety of behavioral models [2, 4, 9] and a precursor of catecholamines [3], were totally ineffective in inducing this behavior [22,23]. This led Segal and Dikstein to postulate that, in addition to existing theories relating to 5-HT, dopamine, endogenous peptides and sexual behaviors, PEA itself may also play an effective role in male rat mounting behavior [25].

Since evidence exists that central catecholamines are involved in some sexual behaviors [1,29], it was our intention to determine whether the chronic administration of PEA, which itself induces sexual behavior(s) (male mounting for example [23]), could possibly alter catecholamine levels in discrete brain areas. Walen *et al.* [35] have already outlined the complexities of assessing the effects of pharmacological agents on sexual behaviors (e.g., mounting, lordosis, soliciting, intromission, ejaculation and so on).

As we have already observed in our laboratory that PEA, as well as a number of other PEA derivatives which induce mounting behavior in male rats [22], also effectively induce female lordoses [22] and enhance PCPA-induced male rat mounting behavior [20], as well as estrogen-induced lordosis in the female [21]. The male rat mounting behavior model was, therefore, selected as the basic model in order to introduce a modicum of simplicity in trying to outline what, if any, role that PEA may have in mechanism(s) underlying induced male rat mounting behavior [24].

This report presents data that PEA, which induces mounting behavior in sexually naive rats, specifically alters norepinephrine (NE) levels in the nucleus preopticus medialis (POM) of the brain.

METHOD

Behavioral

Adult male "Sabra" (strain of the Hebrew University of Jerusalem) rats weighing between 180–220 g were placed in separate holding cages and allowed food and water ad lib during a normal 12 hr light:12 hr dark schedule. After 48 hr of acclimatization, PEA (Sigma) in a dose of 1.0 mg/kg (a dose which effectively induced adult male rat mounting behavior [23]) was orally administered (1.0 cc/100 g body weight as a suspension in 0.1% Tween 80) daily (at 24 hr intervals) for 14 days. Control rats were administered the vehicle in the same

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TABLE 2

EFFECT OF PEA (1.0 mg/kg PO, 14 DAYS) ON THE NE AND DA CONCENTRATION IN DISCRETE BRAIN REGIONS

Areas	Number of Punches	Cannula (mm size)	Approximate Coordinates‡	Control† (pg/μg Protein±SEM) NE	Control† (pg/μg Protein±SEM) DA	Experimental (pg/μg Protein±SEM) NE	Experimental (pg/μg Protein±SEM) DA
N. Interstitialis Stria Terminalis	4	0.5	A6860	22.70±5.64 (7)	6.49±1.46 (7)	21.69±3.03 (6)	4.50±0.92 (6)
Medial Preoptic n.	2	0.7	A6860	10.52±1.03 (6)	—	17.79±2.32 (7)*	—
Anterior Hypothalamic n.	4	0.7	A6060	12.49±1.62 (7)	—	9.96±0.86	—
Median Eminence	3	0.5	A4620, A4380	14.25±1.93 (7)	21.40±5.26 (7)	13.87±1.60 (7)	28.93±5.01 (6)
Arcuate n.	6	0.3	A4620, A4380	19.46±3.62 (8)	5.36±0.96 (8)	23.61±3.21 (7)	5.97±0.61 (7)
Habenula	4	0.7	A3750	2.83±0.40 (6)	1.55±0.16 (6)	2.90±0.20 (7)	1.76±0.31 (7)
Medial Amygdaloid n.	6	0.5	A3430	3.22±0.33 (7)	—	3.01±0.33 (5)	—

*$p<0.05$.
†n in parentheses.
‡Based on König and Klippel [12].

manner. On the 15th day, the rats were placed in a large open plastic container (30 cm long, 24 cm wide, 10 cm high, with the floor covered by wood shavings) in groups of 4 and mounting behavior determined over a 15-min period as outlined by Segal *et al.* [20]. As opposed to the previous study [20] in which the results were expressed as the accumulated total number of mounts per 4 animals over the 15 minute period, the results in this study are expressed as the number of mounts observed for each animal per group of 4 over the 15 minute period. A total of 16 animals were used (4 groups of 4) in the experimental design and another 16 as controls.

Biochemical

Immediately after the behavioral determination, the rats were decapitated and the brains removed rapidly and frozen with dry ice. Serial 300 μm sections were cut in the frontal plane in a cryostat at −7°C. The sections were thawed briefly and refrozen on chilled microscope slides. Brain areas were removed with stainless steel cannulae under a stereomicroscope as described by Palkovits [18], using standard neuroanatomical landmarks [12]. Table 2 presents further details of the microdissection procedure. Tissue punches were expelled into 100 μl of ice-cold 0.1 N perchloric acid containing 500 pg of an external standard dihydroxybenzylamine and sonicated for approximately 3 sec. A 10–20 μl aliquot was removed for protein assay [16]. The remaining homogenate was stored in the deep freeze until catecholamines were assayed by high pressure liquid chromatography with electrochemical detection as described by Shohami *et al.* [28].

RESULTS

As can be seen in Table 1, the chronic administration of PEA (1.0 mg/kg PO daily for 14 days) significantly induced male rat mounting behavior, a behavior not induced in vehicle-treated control animals ($p<0.005$).

The effects of this PEA-induction of mounting behavior on NE and DA levels in discrete areas of the brain are outlined in Table 2. The only significant change observed was a rise in the NE level in the POM ($p<0.05$).

TABLE 1

EFFECTS OF CHRONICALLY ADMINISTERED PEA ON MALE RAT MOUNTING BEHAVIOR

Description	No. Mounts per Rat per 15 Min*
Vehicle-treated controls	0 (16)
PEA (1 mg/kg PO for 14 days)	1.0 ± 0.33† (16)

*No. in parentheses represents the number of individual rats mounting per groups of 4.
†$p<0.005$: Mann and Witney U-test for non-parametric data.

DISCUSSION

The present study suggests an interrelationship between behavioral and NE alterations in the POM. The significance of the monoamine change is not clear. The POM is a region involved with sexual behavior [15], gonadotropin release [32] and is a target region for gonadal steroids [30]. In a prior study, a reduction in the NE level was observed in the male offsprings of stressed pregnant rats measured during adulthood [17]. It is interesting that the reduction in NE concentrations in the POM correlated with feminized sexual behavior of prenatally stressed male offsprings as adults [34]. Similarly, in the present study, PEA-induced mounting behavior in males correlated with a 70% increase in the NE concentration in the POM. The functional significance of these differences is not readily apparent. However, we are inclined to suggest that the PEA-induced change in the NE concentration of the POM reflects a secondary alteration of NE within nerve terminals of the POM rather than changes

in areas innervated by an entire monoaminergic system (e.g., ventral noradrenergic pathway) [33]. In this regard, the presynaptic release of NE is conceived to be a secondary event that occurs following a primary stimulus which emanates from the postsynaptic receptor neuron (POM) [11].

Although PEA has been identified as a normal constituent of brain, the evidence supporting the existence of a role of PEA in brain function is still controversial [19]. Nevertheless, as PEA and various derivatives have been reported to induce mounting behavior after chronic administration [20, 22, 25], the potential clinical utility of this class of chemical agents in sexual incontenance deserves further investigation.

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