Pharmacology Biochemistry & Behavior, Vol. 21, pp. 671–673, 1984. © Ankho International Inc. Printed in the U.S.A. 0091-3057/84 $3.00 + .00

BRIEF COMMUNICATION

Apomorphine-Induced Stereotypy: Function of Age and Rearing Environment

B. CHITKARA, M. J. DURCAN AND I. C. CAMPBELL

Genetics Section, Institute of Psychiatry, DeCrespigny Park, London, SE5 8AF ENGLAND

Received 23 December 1984

CHITKARA, B., M. J. DURCAN AND I. C. CAMPBELL. *Apomorphine-induced stereotypy: Function of age and rearing environment.* PHARMACOL BIOCHEM BEHAV 21(4) 671–673, 1984.—Rats of different age groups ("young" vs. "old"), reared in isolation and social groups were tested for differences in apomorphine-induced stereotypy. Sensitivity to apomorphine (2 mg/kg SC) was found to be a function of an interaction between rearing environment and age. The "younger" isolated animals displayed greater stereotypy than group-reared and "older" isolated animals. Rearing environment and age, as main factors, did not have significant influence on drug-induced stereotypy.

Social isolation Age Apomorphine Stereotypy

STUDIES on the effect of early experiences in laboratory animals have consistently found that rearing in social isolation and restricted environment can produce behavioural changes in adults such as heightened emotionality, hyperactivity, poor social interaction and increased stereotypy [2, 12, 18]. It has also been suggested that social isolation affects central neurotransmitter monoamines at crucial stages of development [13,19]. Isolation has been reported to increase the concentration of brain noradrenaline (NA) and to decrease its turnover and synthesis [23,24]. Furthermore, lower dopamine turnover has been implicated in the limbic areas of isolated rats [17,24].

Apomorphine is believed to stimulate directly dopamine receptors in the CNS, and thus apomorphine-induced stereotypy is used as a behavioural measure of central dopaminergic function [9]. Numerous factors have been reported to affect apomorphine-induced stereotypy, such as species or strain [7,14]; site of injection [11]; temporal factors such as chronic vs. acute administration [3]; novel contexts and events [8]; age [5,16]; and stress [1].

The authors are not aware of any study that has investigated the sensitivity to apomorphine in rats as a combined function of age and differential social rearing conditions. Previous studies have examined dopaminergic sensitivity either as a function of age or as a function of rearing environments. Carlsson and Almasi [5] observed apomorphine-induced stereotypy (0.1, 0.2 and 0.4 mg/kg) in 4 to 10 weeks old, social reared male guinea pigs and found that stereotypy was more intense and persisted longer in the older groups. Sahakian *et al.* [15] reported more enhanced stereotyped behaviour in socially deprived rats than the controls at both 0.5 and 1.5 mg/kg apomorphine, IP. Davenport *et al.* [6] observed stereotypy induced by severely restricted environments in chimpanzees from birth to 40 months of age and found that the phenomena of stereotypy was unique to the isolated infants. In view of the above-mentioned literature, we have examined apomorphine-induced stereotypy in rats as a combined function of age and different rearing environments.

METHOD

Forty-eight adult male rats from a randomly bred strain were equally divided into 4 experimental groups according to age ("young" vs. "old") and social rearing condition (individually housed vs. group reared). The "younger" groups were aged 26 weeks (182±2 days) and the "older" groups 32 weeks (224±2 days) respectively. The individually reared animals were isolated at weaning (25 days) and housed one rat per cage (25.4×22.7×21.5 cm). Control animals were housed in groups of three per cage of the same size. All animals were maintained at constant temperature (22±2°C) and humidity (55±5%) in 17–7 hr light-dark cycle with free access to food and water.

The apparatus consisted of 6 Plexiglas circular boxes, 45.7 cm high with aluminum lids and grid floors measuring 28 cm in diameter. Each rat was rated for the degree of stereotypy on a scale of 0 to 5 (modified from that used by Creese and Iversen [4]). The ratings used were as follows: 0=absence of stereotyped behaviour, 1=discontinuous sniffing and exploratory behaviour, 2=continuous sniffing and exploratory behaviour, 3=continuous sniffing, discontinuous licking or mouthing with or without exploratory activity, 4=continuous sniffing, licking or mouthing, discontinuous gnawing with or without exploratory activity, and 5=continuous gnawing, biting and licking, no exploratory ac-

TABLE 1

SUMMARY OF ANOVA STEREOTYPY-RATING SCORES OF THE FOUR TREATMENT GROUPS, ISOLATED YOUNG, ISOLATED OLD, CONTROL YOUNG, AND CONTROL OLD

Source of Variance	*df*	F Ratio	Level of Significance
Between Subjects			
Housing	1/44	0.3304	NS
Age	1/44	2.7446	0.10
Housing × Age	1/44	4.2535	0.04*
Within Subjects			
Linear	4/41	10.8362	0.002†
Quadratic	4/41	12.0084	0.001†
Housing	4/41	0.5388	NS
Linear	4/41	0.0683	NS
Quadratic	4/41	0.0996	NS
Age	4/41	0.4736	NS
Linear	4/41	0.0027	NS
Quadratic	4/41	0.0517	NS
Housing × Age	4/41	0.5472	NS
Linear	4/41	0.0027	NS
Quadratic	4/41	1.2087	NS

*Significant at <0.05 level.
†Significant at <0.01 level.
NS=Not significant.

tivity. The stereotypy ratings were made by two observers on a blind basis between 12.00 to 17.00 hr.

Testing was carried out with groups of 6 rats per session for a period of 60 min. Each rat was weighed and injected with apomorphine (2 mg/kg SC right flank) at 1 min intervals and placed in the circular box. Ten minutes after drug administration each rat was rated for a 60 sec interval every 10 min for a period of 1 hr. While the use of nonparametric statistics for the analysis of the stereotypy rating data was more appropriate we used analysis of variance on repeated measures as this permits the determination of time course effects.

RESULTS

ANOVA revealed that neither rearing environment nor age as the main factor influenced apomorphine-induced stereotypy (Table 1). However, a significant interaction effect of Rearing Environment × Age was found on apomorphine-induced stereotypy, $F(1,44)=4.2535$, $p<0.04$. The "younger" isolated animals showed greater sensitivity to apomorphine throughout the testing phase (Fig. 1) compared to the control groups which had intermediate stereotypy scores and the "older" isolated group showed the least sensitivity.

Trend analysis on rating scores across five periods yielded significant linear, $F(4,41)=10.8362$, $p<0.001$, trends of stereotypy. Stereotypy was found to consistently increase until Period 4, reaching its peak 40 min after drug administration, and decreasing thereafter. The inter-rater reliability coefficient was 0.97.

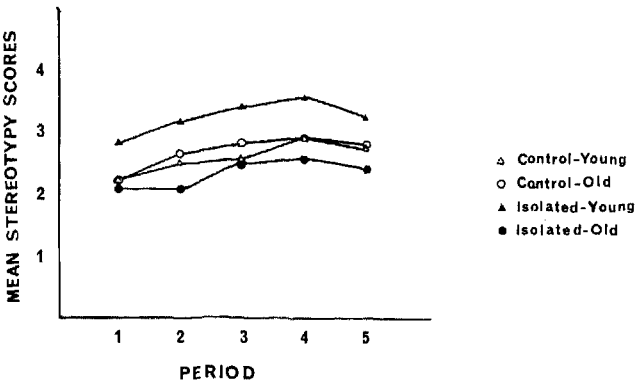

FIG. 1. Mean stereotypy scores of socially isolated rats and controls of two age groups across five periods of 10 min each.

DISCUSSION

The study indicates that sensitivity to apomorphine (2 mg/kg, SC) in rats is a function of an interaction between differential rearing environment and age. The "younger" isolated rats were more sensitive to apomorphine and displayed greater stereotypy than both age groups of socially reared rats, and the "old" isolates had minimal stereotypy. To our knowledge, this is the first study to examine the miltiplicative effect of age and different social rearing conditions on sensitivity to apomorphine in rats. Neither rearing environment nor age, as the main factor, had a significant influence on drug-induced stereotypy at dose level of 2 mg/kg, SC in rats.

Contrary to Sahakian *et al.* [15] we did not find significant primary effect of different rearing environments on stereotypy. There were methodological differences between the two studies in that Sahakian *et al.* used lower doses (0.5, 1.0 and 1.5 mg/kg) of apomorphine on female hooded rats without clear age specification. Although differential environments are known to alter cerebral measures significantly in both young and old animals, it has been suggested recently that the magnitude of certain effects may decline as a result of maturation [10, 20]. Our results confirm this view, suggesting that the influence of rearing environments is more conspicuous in the relatively "younger" animals. These results do not reflect differences in pharmacokinetics of apomorphine because the "older" animals would be expected to show greater drug-induced stereotypy.

In the present study, age did not significantly influence stereotypy in rats. This is in contrast to earlier reports which suggest that apomorphine-induced stereotypy is more intense and persists longer in "older" rodents compared to their "younger" counterparts [5, 16. 21, 22]. The discrepancy between our findings and earlier studies may be due to: (a) different duration of isolation, e.g., the present study used prolonged period of over 155 days of isolation; (b) use of different rating scales, and (c) different criterion of age groups, e.g., the present sample used relatively "older" animals with small between age-group difference.

Duration of the observation period may have a significant influence on the experimenter's observations of age-related apomorphine-induced stereotypy. Watanabe *et al.* [22] observed apomorphine-induced (3 mg/kg, SC)

stereotypy for 2.5 hr and found that the "older" rats (34 weeks) had lower stereotypy at earlier stages of the observation period, and the onset of biting was slow and of longer duration. Our findings revealed a gradual decline in stereotypy scores after 40 min for all age groups, thereby justifying our choice for one 1 hr observation period.

In summary, apomorphine-induced stereotypy in rats was found to be a function of an interaction between age and different rearing environments. The "younger" isolated animals showed greater sensitivity to apomorphine than "older" isolated animals and controls. Perhaps further studies should investigate several doses and monitor individual items of behaviour in groups of animals with larger age difference.

REFERENCES

1. Antelman, S. M., H. Szechtman, P. Chin and A. E. Fisher. Tail pinching induced eating, gnawing and licking behaviour in rats: Dependence on the nigrostriatal dopaminergic system. *Brain Res* **99**: 319–337, 1975.
2. Ardila, R., M. Rezk, R. Polanco and F. Pereira. Early handling, electrick shock and environmental complexity: Effects of exploratory behaviour emotionality and body weight. *Psychol Rec* **27**: 219–224, 1977.
3. Browne, R. G. and D. S. Segal. Metabolic and experiential factors in the behavioural response to repeated amphetamine. *Pharmacol Biochem Behav* **6**: 545–552, 1977.
4. Creese, I. and S. D. Iversen. Blockage of amphetamine induced motor stimulation and stereotypy in the adult rat following neonatal treatment with 6-hydroxydopamine. *Brain Res* **55**: 369–392, 1973.
5. Carlson, K. G. and J. Almasi. Sensitivity to apomorphine in the guinea pig as function of age and body weight. *Psychopharmacology (Berlin)* **57**: 279–282, 1978.
6. Davenport, R. K., Jr. and E. W. Menzel, Jr. Stereotyped behaviour of the infant chimpanzee. *Arch Gen Psychiatry* **8**: 99–104, 1963.
7. Durcan, M. J., D. W. Fulker and I. C. Campbell. Differences in the stereotypy response but not the hypomotility response to apomorphine in the Roman high and Roman low avoiding strains of rats. *Psychopharmacology (Berlin)* **82**: 215–220, 1984.
8. Einon, D. F. and B. J. Sahakian. Environmentally induced differences in susceptibility of rats to CNS stimulants and CNS depressants: Evidence against a unitary explanation. *Psychopharmacology (Berlin)* **61**: 299–307, 1979.
9. Ernst, A. M. and P. G. Smelik. Site of action of dopamine on compulsive gnawing behaviour in rats. *Experientia* **22**: 837–838, 1966.
10. Fiala, B. A., J. N. Joyce and W. T. Greenough. Environmental complexity modulates growth of granule cell dendrites in developing but not adult hippocampus of rats. *Exp Neurol* **59**: 372–383, 1978.
11. Ljungberg, T. and U. Ungerstedt. Apomorphine-induced locomotion and gnawing: Evidence that the experimental design greatly influences gnawing while locomotion remains unchanged. *Eur J Pharmacol* **46**: 147–151, 1977.
12. Mason, W. A. The effects of social restriction on the behaviour of rhesus monkeys, III Dominance tests. *J Comp Physiol Psychol* **54**: 694–699, 1961.
13. Modigh, K. Effects of isolation and fighting in mice on the rate of synthesis of noradrenaline, dopamine and serotonin in the brain. *Psychopharmacologia* **33**: 1–17, 1973.
14. Randrup, A. and I. Munkvad. Stereotyped activities produced by amphetamines in several animal species and man. *Psychopharmacologia* **11**: 300–310, 1967.
15. Sahakian, B. J., T. W. Robbins, M. J. Morgan and S. D. Iversen. The effects of psychomotor stimulants on stereotypy and locomotor activity in socially deprived and control rats. *Brain Res* **84**: 195–205, 1975.
16. Smith, R. C., D. E. Leelavathi and M. A. Lauritzen. Behavioural effects of dopamine agonist increase with age. *Commun Psychopharmacol* **2**: 39–43, 1978.
17. Stolk, J. M., R. L. Conner and J. D. Barchas. Social environment and brain biogenic amine metabolism in rats. *J Comp Physiol Psychol* **87**: 203–207, 1974.
18. Syme, L. A. Social isolation at weaning: Some effects on two measures of activity. *Anim Learn Behav* **1**: 161–163, 1973.
19. Thoa, N. G., Y. Tizabi and D. M. Jacobowitz. The effect of isolation on catecholamine concentration and turnover in discrete areas of the rat brain. *Brain Res* **131**: 259–269, 1977.
20. Uphouse, L. Re-evaluation of mechanisms that mediate brain differences between enriched and impoverished animals. *Psychol Bull* **88**: 215–232, 1980.
21. Watanabe, H. S., S. Nakano and N. Ogawa. Pharmacokinetic study of apomorphine-induced stereotypy in food deprived rats. *Pharmacol Biochem Behav* **14**: 493–496, 1981.
22. Watanabe, H., S. Nakano and N. Ogawa. Age differences in apomorphine-induced stereotypy in rats: Relationship to plasma and brain concentrations. *Psychopharmacology (Berlin)* **76**: 57–61, 1982.
23. Weinstock, M., Z. Speiser and R. Ashkenazi. Changes in brain catecholamine turnover and receptor sensitivity induced by social deprivation in rats. *Psychopharmacology (Berlin)* **56**: 205–209, 1978.
24. Welch, A. S. and B. L. Welch. Reduction of norepinephrine in the lower brainstem by psychological stimulus. *Proc Natl Acad Sci USA* **60**: 478–481, 1968.