ONTOGENESIS OF REARING AND STEREOTYPY RESPONSES TO APOMORPHINE IN THE RAT

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The ontogenesis of rearing and stereotypy behavior in response to apomorphine were investigated in the developing rat. The rearing was rarely observed in 15-day-old rats but distinctly in 18-day-old ones. A subcutaneous injection of apomorphine in dose of 1 mg/kg or 2 mg/kg decreased the frequency of rearing in older animals than day 20. On the other hand, 15-day-old rats were already responsive to apomorphine in stereotypy behavior. There was a reciprocal relationship between rearing and stereotypy responses to apomorphine in rat older than 18 days. Functional relationship between rearing and stereotypy in response to apomorphine were discussed in view of the central dopaminergic mechanisms in developing rats.

Keywords—rearing; stereotypy; apomorphine; development; dopaminergic mechanism

INTRODUCTION

Several behavioral studies have shown that central dopaminergic system probably at the neostriatum or mesolimbic area reaches functional maturity at an early stage in the developing rat.^{1,2)} A high dose of apomorphine (10 mg/kg), a dopamine (DA) receptor agonist,^{3,4)} induces rudimentary stereotypy behavior in the 2-day-old rat⁵⁾ and sniffing in the 7-day-old rat.⁶⁾ A low dose of the drug (0.05 mg/kg or 0.2 mg/kg) produces the locomotor stimulation in the 10-day-old rat.^{6,7)} The 10-day-old rat is already responsive to amphetamine.^{8–10)}

The rearing, one of behavioral arousal, has been investigated as an indicator for studying the development of central cholinergic system.¹¹⁾ There is, however, little report concerning influence of apomorphine on the ontogenesis and the development of rearing in the rat. To obtain an information on the functional maturation of the central DA system which controls behavioral arousal, the present study was arranged to

investigate the developmental change of the rearing and its relationship to the stereotypy behavior induced by apomorphine in the rat.

MATERIALS AND METHODS

Wistar rats of both sexes were used. Pups were kept with a mother under normal daylight conditions at 23 ° and weaned on day 22.

Animals at 15 days, 18 days, 20 days, 23 days, 30 days and 60 days were placed singly into a plastic cage, 24 × 17 cm and their rearing and stereotypy behavior were measured. Before a *s. c.* injection of apomorphine, the general behavior was observed for 10 min in the same cage used for further experiment. The animal was considered rearing if his forelimbs touch neither floor nor wall of the cage. The rearing was counted 10 min after a *s. c.* injection of apomorphine for 5 min every 10 min for a period of 35 min and accumulated for 3 consecutive observation periods.

The stereotypy behavior was assessed by means of a rating scale modified from that of Kelly and

Iversen (1976).¹²⁾Student's *t*-test was used for statistical analysis.

Score	Behavior
0	Sleep or stationary
1	Alert or grooming
2	Discontinuous sniffing
3	Sniffing over a large area of the cage
4	Continuous sniffing, licking or compulsive head movement
5	Continuous sniffing or biting in one location

Apomorphine hydrochloride purchased from Takeda Chemical Ind. Ltd., were dissolved in saline containing 0.01 N acetic acid.

RESULTS

1. Development of Rearing

The rearing behavior was seldom observed in 15-day-old rats. As shown in Fig. 1, the behavior obviously appeared in 18-day-old rat and increased with age until day 30. The rearing in the adult animal was less than in the 30-day-old. Apomorphine in doses of 0.5 mg/kg, 1.0 mg/kg and 2.0 mg/kg reduced the number of the rearing at each developing stage except 18 days. Apomorphine-induced reduction of the rearing was dose-dependent in 20-day-old, 23-day-old, 30-day-old and 60-day-old rats.

FIG. 1. Apomorphine-induced Rearing in the Developing Rat

Animals at each stage was placed into a plastic cage and the frequency of rearing was measured. Before an injection of the drug, the animals were adapted in the same cage used for further observation. The rearing was measured at 10 min after a s. c. injection of the drug for 5 min every 10 min for a period of 35 min and accumulated for 3 consecutive observation periods. The rearing/15 min is shown as the mean with S. E. of 5 to 8 determinations. Significance, *** $p < 0.01$ vs. control.

FIG. 2. Apomorphine-induced Stereotypy in the Developing Rat

Stereotypy behavior was observed between 10 min and 35 min after a s. c. injection of apomorphine. Stereotypy was assessed according to the scoring system described in the Materials and Methods section. Each point indicates the mean with S. E. of 5 to 8 determinations. Significance, * $p < 0.05$ vs. control; ** $p < 0.01$ vs. control.

FIG. 3. Relationship between the Apomorphine-induced Rearing and Stereotypy in the Developing Rat

Mean rearing and stereotypy scores at 18, 20, 23, 30 and 60 days of age were plotted.

▲ 18-day-old, △ 20-day-old, ■ 23-day-old, ○ 30-day-old, ● 60-day-old.

2. Development of Stereotypy Behavior induced by Apomorphine

Fig. 2 shows the developmental change of the apomorphine-induced stereotypy behavior. Apomorphine (0.5 mg/kg, 1.0 mg/kg and 2.0 mg/kg) elicited the stereotypy in each developing rat aged 15 days to the adult in a dose-dependent manner.

The rearing was correlated with stereotypy score during the development. This was done by comparing the rearing with stereotypy score at specified ages from 18 to 60 days. The results indicate (Fig. 3) that the rearing and stereotypy score closely exhibit a reciprocal correlation ($r=0.98$ ($p < 0.05$) at 23 days, $r=0.95$ ($p < 0.05$) at 30 days, and $r=0.98$ ($p < 0.05$) at 60 days), although there were no correlation in 18-day-old

and 20-day-old rats.

DISCUSSION

The results presented here show that 15-day-old rats were already responsive to a low dose of apomorphine in the stereotypy behavior and that the response resembled adult ones, although Kellogg and Lundborg¹³⁾ have reported that the marked stereotypy was not observed in response to apomorphine until 21 days of rats. Our results is rather consistent with those of Lal and Sourkas (1973).⁵⁾

The rearing behavior was rarely observed in 15-day-old rats but distinctly in 18-day-old ones. The frequency of the rearing increased with age until day 30 followed by the decrease in the adult. The systemic administration of apomorphine decreased the frequency of the rearing in the animal older than 20 days compared to that of control. Therefore, the rearing response to dopaminergic agonists could not be a more valid indicator than the drug-induced stereotypy or hyperactivity for studying the functional development of the central dopaminergic nervous system because the response appeared much later than those of stereotypy or hyperactivity.^{3,5-7)} In addition, since there was a reciprocal relationship between rearing and stereotypy response to apomorphine in the developing rat, it is concluded that the higher stereotypy is induced by apomorphine, the lower frequency of the rearing is produced.

The present results show that a low dose (0.5 mg/kg, s. c.) of apomorphine which has been known to induce locomotor stimulation⁷⁾ did not increase the frequency of rearing in the developing rat. It seems that the rearing behavior dissociates from hyperactivity. In addition, with regard to the relationship between rearing and stereotypy, a reciprocal relationship was found. There is a data which indicates differences in the dopaminergic mechanisms mediating stereotypy and hyperactivity within the extrapyramidal caudate-putamen or mesolimbic nucleus accumbens and between these areas.¹⁴⁾ Rearing response is not a simple reflex but composed from multiple

factors. Further studies will be required to clarify the involvement of dopaminergic mechanisms in the CNS in rearing and its relationship with hyperactivity or stereotypy. In the case it appears also important to consider that a physiological function of the central cholinergic mechanism could be responsible to the rearing behavior, since atropine increased the frequency of rearing in the developing rat, in particular on day 23.^{11,15,16)}

REFERENCES

- 1) J. T. Coyle: Biochemical Aspect of Neurotransmission in the Developing Brain. in "International Review of Neurobiology," vol. 20, ed. by J. R. Smythies and R. J. Bradley, Academic Press, New York, 1977, pp. 65-103.
- 2) L. A. Baez, N. K. Eskridge and R. Schein: Postnatal development of dopaminergic and cholinergic catalepsy in the rat, *Eur. J. Pharmacol.*, **36**, 155-162 (1976).
- 3) N. -E. Andén, A. Rubensen, K. Fuxe and T. Hökfelt: Evidence for dopaminergic receptor stimulation by apomorphine, *J. Pharm. Pharmacol.*, **19**, 627-629 (1967).
- 4) A. M. Ernst: Mode of action of apomorphine and dexamphetamine on gnawing compulsion in rats, *Psychopharmacologia(Berl.)*, **10**, 316-323 (1967).
- 5) S. Lal and T. L. Sourkas: Ontogeny of stereotyped behavior induced by apomorphine and amphetamine in the rat, *Arch. int. Pharmacodyn.*, **202**, 171-182 (1973).
- 6) D. K. Reinstein, D. McClearn and R. L. Isaacson: The development of responsiveness to dopaminergic agonists, *Brain Res.*, **150**, 216-223 (1978).
- 7) Y. Nomura and T. Segawa: Apomorphine-induced locomotor stimulation in developing rats treated with 6-hydroxydopa, *Eur. J. Pharmacol.*, **50**, 153-156 (1978).
- 8) B. A. Campbell, L. D. Lytle and H. C. Fibiger: Ontogeny of adrenergic arousal and cholinergic inhibitory mechanisms in the rat, *Science*, **166**, 635-637 (1969).
- 9) R. Miller and B. Sahakian: Differential effects of neuroleptic drugs on amphetamine-induced stimulation of locomotor activity in 11-day-old and adult rats, *Brain Res.*, **81**, 387-392 (1974).
- 10) B. C. White and W. N. Tapp: Unilateral catecholamine depletion of the corpus striatum and amphetamine-induced turning: An ontogenic study, *Psychopharmacol.*, **53**, 211-212 (1977).
- 11) D. Blozovski and J. Bachevalier: Effect of atropine on behavioral arousal in the developing rat, *Developmental Psychobiol.*, **8**, 97-102 (1975).
- 12) P. H. Kelly and S. D. Iversen: Selective 6-OHDA-

- induced destruction of mesolimbic dopamine neurons: Abolition of psychostimulant-induced locomotor activity in rats, *Eur. J. Pharmacol.*, **40**, 45–56 (1976).
- 13) C. Kellogg and P. Lundborg: Ontogenic variations in responses to L-DOPA and monoamine receptor-stimulating agents, *Psychopharmacologia*, **23**, 187–200 (1972).
- 14) B. Costal, R. J. Naylor, J. G. Cannon and T. Lee: Differentiation of the dopamine mechanisms mediating stereotyped behavior and hyperactivity in the nucleus accumbens and caudate putamen, *J. Pharm. Pharmacol.*, **29**, 337–342 (1977).
- 15) Y. Nomura and T. Segawa: Muscarinic hyposensitivity in the developing rat pretreated with 6-hydroxydopa, *Eur. J. Pharmacol.*, **50**, 431–435 (1978).
- 16) Y. Nomura and T. Segawa: Developmental change in rearing response to atropine in the rat treated with 6-hydroxydopa, *Jpn. J. Pharmacol.*, **30**, 117–120 (1980).