

Behavioral and neurochemical changes produced in rats by developmental treatments with psychotropic drugs

Raffaele CAGIANO, Maria A. De SALVIA, Arcangela GIUSTINO, Giovanni SIRO BRIGIANI and Vincenzo CUOMO

Istituto di Farmacologia, Facoltà di Medicina e Chirurgia, Università degli Studi, Bari, Italy

Summary. - The timing of developmental administration of psychotropic drugs affecting dopaminergic and GABAergic neurotransmission is crucial for the induction of specific neurobehavioral and neurochemical changes in rodents. Compensatory mechanisms occurring in response to a prolonged treatment with some neuroleptic and anxiolytic agents during development seem to be markedly different from those occurring in response to a prolonged administration in adult animals.

Key words: rat, behavior, neurochemistry, psychotropic drugs.

Riassunto (*Alterazioni comportamentali e neurochimiche prodotte nel ratto dal trattamento prenatale e/o postnatale con farmaci psicotropi*). - Il particolare periodo di somministrazione durante lo sviluppo del sistema nervoso centrale (SNC) di farmaci attivi sulla trasmissione dopaminergica e GABAergica è un fattore critico nel determinismo di specifiche alterazioni neurocomportamentali e neurochimiche. Recenti ricerche suggeriscono che i meccanismi compensatori in risposta a trattamenti prolungati con farmaci neurolettici ed ansiolitici durante la vita pre- e/o postnatale differiscono significativamente da quelli che si manifestano in seguito ad una loro somministrazione nell'animale adulto.

Parole chiave: ratto, comportamento, neurochimica, farmaci psicotropi.

Introduction

The developing nervous system is extremely susceptible to various chemicals; the blood-brain barrier is absent or incomplete in the neonate (rat), so that animals prenatally or neonatally treated with chemical agents seem to be at greatest risk from substances which have potential neurotoxic effects. Interactions with specific mechanisms of neurotransmission during development alter the course of neural development and may induce short- and long- term neurochemical and behavioral alterations [1-3].

In the present paper, examples of neurochemical and behavioral changes produced by the developmental administration of some psychotropic drugs affecting central dopaminergic and GABAergic neurotransmission are reported. In particular, the results of experiments carried out in our laboratory on the neurochemical and behavioral consequences of prenatal and/or neonatal exposure to some neuroleptic and anxiolytic agents are discussed.

Neuroleptics

The period of administration of dopamine receptor blocking agents plays a critical role in producing short- and long-term behavioral changes in rats. We have

recently demonstrated that the administration of haloperidol during gestation induces, in rat offspring, behavioral changes markedly different from those produced by the exposure to this neuroleptic during early postnatal life or during adulthood.

We have shown, in fact, that the administration of haloperidol during gestation (from day 4 to day 15 of gestation, at dose level of 0.5 mg/kg s.c.) produced only a short-lasting change in the rate of ultrasonic calls in 4-day-old male rat pups removed from the nest whereas treatment during early postnatal life produced profound and long-lasting alterations in several parameters of ultrasonic vocalization [4, 5]. In fact, the neonatal administration of this dopamine receptor antagonist produced a significant and prolonged decrease in the rate of calling from the 8th up to the 14th day after birth, whereas a significant increase in the duration of calls was found from the 4th to the 16th day of age in rat pups exposed to haloperidol. Furthermore, notable changes in the frequency of calls were induced by early postnatal treatment with haloperidol with a decrease in the minimum and maximum frequency values.

Moreover, the prolonged administration of haloperidol during gestation markedly affects the behavioral responsiveness of adult rats to a dopamine receptor agonist such as apomorphine.

Benzodiazepines

In particular, the intensity of stereotyped behavior as well as the effects on locomotor activity induced by apomorphine in haloperidol pretreated rats were significantly reduced with respect to controls. These data parallel neurochemical results showing a significant decrease in the number of dopamine receptors in the striatum of animals exposed to haloperidol during prenatal life [6, 7].

On the contrary, the prolonged exposure to this neuroleptic during the first three weeks of postnatal life, which seem to be vulnerable periods for the functional maturation of the central dopaminergic system in the rat, produced an opposite response pattern: behavioral supersensitivity to apomorphine which was correlated with neurochemical results showing an increased number of striatal dopamine receptors [7, 8]. Furthermore, the administration of a challenge dose of haloperidol produced smaller increases in dopamine turnover (striatal levels of HVA and DOPAC) of adult rats treated with this neuroleptic during early postnatal life [8]. Even though dopamine receptor supersensitivity to apomorphine as well as an attenuated response to a challenge of haloperidol on dopamine turnover after prolonged administration of this neuroleptic in adult rats has been shown, there is no evidence that these effects persist up to 40 days after haloperidol withdrawal.

Therefore, these findings further confirm that the compensatory mechanisms occurring in response to a chronic treatment with dopamine receptor blocking agents during development are markedly different from those occurring in response to their prolonged administration during adulthood.

Moreover, we have demonstrated that adult rats exposed to haloperidol during prenatal life displayed a notable deficit in the acquisition of a DRL-15 s schedule [6]. In this regard, the behavioral changes produced by prenatal treatment with haloperidol are similar to those elicited by its administration during early postnatal life [8]. On the contrary, the prolonged exposure to haloperidol in adult rats did not cause any long-lasting learning impairment in animals subjected to a DRL-15 s task. Even though the neuronal bases for learning deficits produced by both prenatal and postnatal haloperidol administration remain to be determined, these data confirm and extend the differences in long-term consequences of prenatal and early postnatal exposure to this neuroleptic, respectively.

Finally, we have recently shown that prenatal treatment with haloperidol, at dose level of 0.5 mg/kg (from day 4 to day 15 of gestation) that does not affect the typical parameters of sexual activity of male rats, markedly influences the 22 kHz post-ejaculatory vocalizations. In fact, male rats treated prenatally with haloperidol exhibited an increase in both intensity and duration of 22 kHz post-ejaculatory calls [5].

It has been shown that prenatal and/or early postnatal treatment with diazepam, the most widely used anxiolytic agent, produces in rodents specific behavioral changes that are markedly different from those occurring in response to drug administration to mature animals [9].

Recently, studies on the effects of late gestational exposure (rat) to low doses of diazepam have been carried out in our laboratory. These findings have shown a wide range of short- and long-term emotional and motivational changes in rat offspring treated with 0.1 and 1 mg/kg of diazepam during the third week of pregnancy. In fact, infant animals exhibited notable alterations in the length of ultrasonic calling elicited by the removal from their nest: in particular, the duration pattern of ultrasonic calls during the first 2 weeks of postnatal life was markedly affected by prenatal administration of diazepam [10].

In this regard, it has been suggested [11] that the benzodiazepine-GABA-receptor chloride channel complex plays a role in the physiological mediation of rat pup ultrasonic isolation calls. Therefore, the alterations in this response produced by prenatal exposure to diazepam could be due to an impaired ontogenesis of this system.

Although the biological significance of changes in the duration of ultrasonic calls remains to be explored, it is of interest to observe that infant rats prenatally treated with diazepam also displayed a decreased activity in the open field at the end of the second week of postnatal life, possibly indicating increased emotionality. Conversely, a reduced emotionality was found in rats treated with a benzodiazepine-receptor antagonist (Ro 151788) during development [12]. Since there is evidence that ultrasonic calls emitted by rat pups markedly affect some behavioral parameters in lactating dams, such as nest building and retrieval, the alterations in ultrasonic vocalization of rat pups exposed prenatally to diazepam could alter maternal behavior which, in turn, might influence the behavior of the offspring. Therefore, changes in rat pup ultrasonic calling induced by prenatal treatment with diazepam could have an important role in the production of enduring behavioral abnormalities of adult rats exposed to this benzodiazepine derivative during pregnancy. Moreover, we have demonstrated that prenatal diazepam administration influenced some parameters of sexual behavior in adult male rats. In particular, a significant increase in the latency of the first mount-intromission as well as a decrease in the length of ultrasonic postejaculatory calls were found in diazepam treated rats with respect to controls [10].

Since some studies suggest an important role of GABA in the neural control of the mating pattern as well as in the mediation of postejaculatory vocalization during sexual behavior in rats [13], the alterations produced by

prenatal diazepam could be related to an interaction of this benzodiazepine derivative with the development of the GABAergic system.

Moreover, we have also investigated the behavioral consequences of early postnatal exposure to benzodiazepine derivatives.

In the rat brain, benzodiazepine receptors proliferate rapidly during the last week of gestation and reach adult levels three weeks after birth.

In particular, we have evaluated the behavioral and neurochemical effects produced in adult rats by prolonged exposure to low doses of chlordiazepoxide during the first 3 weeks of postnatal life [14].

The results of this study indicate that, if rats are treated with chlordiazepoxide even at relatively low doses (0.22 mg/kg) during the three weeks of lactation, the offspring show altered behavioral and biochemical parameters at 60 days of age. In particular, an impaired acquisition of an active avoidance task was found in animals exposed postnatally to chlordiazepoxide. Since spontaneous locomotor activity was not affected by the treatment, these changes are probably due to alterations of motivational or associative processes.

Biochemical studies have shown that [^3H] flunitrazepam binding sites in cerebral cortex and hippocampus were decreased by the treatment, whereas no change was detected in cerebellum. [^3H] flunitrazepam labels both benzodiazepine type 1 and type 2 receptors, but since its binding was more affected in those areas (cerebral cortex and hippocampus) which contain a higher percentage of type 2 receptors, and did not change in cerebellum (which contains 90% type 1 receptors) it has been suggested that the postnatal treatment with chlordiazepoxide selectively affects type 2 receptors.

This study also showed that except for the cerebral cortex (where no changes in GABAergic receptors were found following postnatal chlordiazepoxide administration) there may be some correlation between the effects induced by this benzodiazepine on [^3H] flunitrazepam and on [^3H] muscimol binding in 60 day old rats. In fact, in hippocampus where the major decrease in benzodiazepine binding sites was observed, a concomitant increase in GABAergic receptors was found, and similarly, no difference was detected in the cerebellum.

These data indicate that both prenatal or early postnatal treatment with some benzodiazepine derivatives produces behavioral and neurochemical abnormalities that may persist beyond the first phase of postnatal life. These findings may have implications for the exposure of the human fetus or neonate to benzodiazepines.

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REFERENCES

1. LEONARD, B.E. 1983. Behavioral teratology and toxicology. In: *Preclinical psychopharmacology*. D.G. Grahame-Smith & P.J. Cowen (Eds). Excerpta Medica, Amsterdam. pp. 248-299.
2. ZBINDEN, G., CUOMO, V., RACAGNI, G. & WEISS, B. 1983. *Application of behavioural pharmacology in toxicology*. Raven Press, New York, USA.
3. RILEY, E.P. & VORHEES, C.V. (Eds). 1986. *Handbook of behavioural teratology*. Plenum Press, New York, USA.
4. CAGIANO, R., SALES, G.D., RENNA, G., RACAGNI, G. & CUOMO, V. 1986. Ultrasonic vocalization in rat pups: effects of early postnatal exposure to haloperidol. *Life Sci.* 38: 1417-1423.
5. CAGIANO, R., BARFIELD, R.J., WHITE, N.R., PLEIM, E.T., WEINSTEIN, M. & CUOMO, V. 1988. Subtle behavioural changes produced in rat pups by in utero exposure to haloperidol. *Eur. J. Pharmacol.* 175: 45-50.
6. CUOMO, V., CAGIANO, R., RENNA, G., SERINELLI, A., BRUNELLO, N. & RACAGNI, G. 1985. Comparative evaluation of the behavioural consequences of prenatal and early postnatal exposure to haloperidol in rats. *Neurobehav. Toxicol. Teratol.* 7: 489-492.
7. ROSENGARTEN, H. & FRIEDHOFF, A.J. 1979. Enduring changes in dopamine receptor cells of pups from drug administration to pregnant and nursing rats. *Science* 203: 1133-1135.
8. CUOMO, V., CAGIANO, R., COEN, E., MOCCHETTI, I., CATTABENI, F. & RACAGNI, G. 1981. Enduring behavioural and biochemical effects in the adult rat after prolonged postnatal administration of haloperidol. *Psychopharmacology* 74: 166-169.
9. CUOMO, V. 1987. Perinatal neurotoxicology of psychotropic drugs. *Trends Pharmacol. Sci.* 8: 346-350.
10. CAGIANO, R., DE SALVIA, M.A., PERSICHELLA, M., RENNA, G., TATTOLI, M. & CUOMO, V. 1990. Behavioural changes in the offspring of rats exposed to diazepam during gestation. *Eur. J. Pharmacol.* 177: 67-74.
11. INSEL, T.R., HILL, J.L. & MAYOR, R.B. 1986. Rat pup ultrasonic isolation calls: possible mediation by the benzodiazepine receptor complex. *Pharmacol. Biochem. Behav.* 24: 1263-1267.
12. CAGIANO, R., DE SALVIA, M.A., GIUSTINO, A., LACOMBA, C. & CUOMO, V. 1993. Behavioral changes produced in rats by developmental exposure to flumazenil, a benzodiazepine receptor antagonist. *Prog. Neuro-Psychopharmacol. Biol. Psychiatry* 17: 151-159.
13. CAGIANO, R., BARFIELD, R.J., WHITE, N.R., PLEIM, E.T. & CUOMO, V. 1989. Mediation of rat postejaculatory 22 kHz ultrasonic vocalization by dopamine D2 receptors. *Pharmacol. Biochem. Behav.* 34: 53-58.
14. COEN, E., ABBRACCHIO, M.P., BALDUINI, W., CAGIANO, R., CUOMO, V., LOMBARDELLI, G., PERUZZI, G., RAGUSA, M.C. & CATTABENI, F. 1983. Early postnatal chlordiazepoxide administration: permanent behavioural effects in the mature rat and possible involvement of the GABA-benzodiazepine system. *Psychopharmacology* 81: 261-266.