

ACTION OF SELECTIVE SEROTONIN REUPTAKE INHIBITOR ON AGGRESSIVE BEHAVIOR IN ADULT RAT SUBMITTED TO THE NEONATAL MALNUTRITION

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ABSTRACT- The effect of the malnutrition during suckling on the aggressiveness was investigated in adult rats treated or not with citalopram, a selective serotonin reuptake inhibitor (SSRI). The animals were divided into two groups according to the diet used: nourished group– the rats received the control diet with 23% protein during the life; and malnourished group– the rats had its mothers submitted to diet with 7.8% protein during suckling. At 120 days of age, each group was sub-divided according to the treatment: acute – consisting a single i.p. injection of saline solution or 20-mg/Kg citalopram; chronic – consisting the single injections (1 per day during 14 days) of saline or 20 mg/Kg citalopram. The acute or chronic treatment with SSRI reduces aggressive response in nourished rats, but not in malnourished ones. Thus, the malnutrition during the critical period of brain development seems to induce durable alterations in the function of the serotonergic neurotransmission

KEY WORDS: aggressive behavior, malnutrition, serotonin.

Ação de inibidor seletivo da recaptação de serotonina sobre comportamento agressivo em rato adulto submetido à desnutrição neonatal

RESUMO - O efeito da desnutrição durante a lactação sobre a agressividade foi investigado em ratos adultos tratados ou não com citalopram, um inibidor seletivo da recaptação de serotonina (ISRS). Os animais foram divididos em dois grupos de acordo com a dieta: grupo nutrido– ratos que receberam toda a vida dieta controle (23% de proteína); e grupo desnutrido– ratos que tiveram suas mães submetidas a dieta com 7,8% de proteína na lactação. Aos 120 dias de idade, cada grupo foi sub-dividido conforme o tratamento: agudo – consistindo de injeção única i.p. de solução salina ou 20mg/Kg de citalopram; crônico - consistindo de injeções únicas (1 por dia durante 14 dias) de salina ou 20mg/Kg de citalopram. O tratamento agudo ou crônico com ISRS reduziu a resposta agressiva nos ratos nutridos, mas não nos desnutridos. Assim, a desnutrição durante o período crítico de desenvolvimento do cérebro parece acarretar alterações duradouras na função da neurotransmissão serotoninérgica.

PALAVRAS-CHAVE: comportamento agressivo, desnutrição, serotonina.

The role of serotonin in the control of the aggressive behavior has been demonstrated through the use of pharmacological instruments¹. There are drugs that act through the selective serotonin (5-HT) reuptake inhibition; these substances increase the availability of 5-HT in the synapse and, consequently, the action of this monoamine². Among these substances are the citalopram, one

of the most selective serotonin reuptake inhibitors (SSRI)³. The growth of the central nervous system (CNS) and its developmental processes (gliogenesis, neuronal differentiation, migration, synaptogenesis, etc) occur with great intensity during the suckling period in the rat⁴. The brain is more vulnerable to several types of the aggressions in that phase⁴. Thus, nutritional insults can cause irrevers-

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ible alterations⁵. This period is called "brain growth spurt"⁶. In the man, it begins in the prenatal period (last quarter of gestation) continuing until the first years of life⁴.

The malnutrition during the neonatal period in rats results in major neurochemical alterations including those in the neurotransmitter systems⁷. The effects of nutritional insults on the neurotransmitter systems, in particular the serotonergic one, deserves special attention. This system participates in a wide variety of the functions of the CNS⁸. There are several experimental evidences about the effects of malnutrition on the serotonergic system⁹⁻¹¹. However, there are few works on the effects of manipulations of this system in undernourished subjects, particularly concerning the behavioral expression^{12,13}.

Thus, the present work investigated the effects of the malnutrition during the critical period of brain development and the effects of the treatment with SSRI, on the aggressive behavior in adult rats.

METHOD

Male Wistar rats maintained at room ($23 \pm 1^\circ \text{C}$) and on a light (6:00 a.m. to 6:00 p.m.) –dark (6:00 p.m. to

6:00 a.m.) cycle was used. The male offspring of rats were kept with 6 animals. The animals were divided according to the diet employee during suckling: nourished group – the rats received the control diet with 23% of protein (Purina of Brazil Ltd.); and malnourished group– the rats had its mothers submitted to diet with 7.8% protein ("Regional Basic Diet" - RBD) during suckling. The composition of RDB is shown in Table 1¹⁴. After weaning (24 days after birth), all rats received the control diet *ad libitum* until the day of the experiment. Body weights were determined on the 1st, 24th, 60th and 120th day. On the 120th day after birth, each group was subdivided according to the paradigm used for drug treatment: acute – consisting a single i.p. injection (1ml/kg) of saline (0,9% NaCl solution) or citalopram (20 mg/Kg, Lundbeck); chronic – consisting the single injections (1ml/kg; 1 per day during 14 days) of saline or 20 mg/Kg citalopram. The citalopram was dissolved in saline. It was formed 8 experimental groups, each one containing 20 animals. This way, the groups of adult rats were constituted, into nourished or malnourished ones in the suckling period, acute or chronically treated or not with citalopram. During the 14 days of the chronic treatment the animals were housed individually in cage. The animals were submitted to the aggressiveness tests 1-h after the acute treatment or 24h after the chronic treatment. The aggressiveness tests were accomplished

Table 1. Centesimal composition of "Regional Basic Diet" (RBD).

Ingredients	Centesimal composition						
	g%	Proteins	Carbohydrates	Fats	Ash	Fibers	Kcal %
Beans ^a	18.34	3.99	10.66	0.24	0.57	1.09	60.76
Manioc flour	64.81	0.84	48.59	0.12	0.43	5.64	198.80
Poor fat-dried and salted	3.74	2.74	-	0.06	0.06	-	11.50
Dried and salted meat fat	0.35	-	-	0.35	-	-	3.15
Sweet potato ^a	12.76	0.30	9.99	0.03	0.20	0.48	41.43
	100	7.87	69.24	0.80	1.26	7.21	315.6

a, cooked and dried.

Table 2. Body weight of rats nourished or malnourished during suckling.

Experimental groups	Weight (g)			
	1st day	24th day	60th day	120th day
Nourished groups	7.53 \pm 0.13	44.35 \pm 1.55	214.9 \pm 6.76	322.75 \pm 9.77
Malnourished groups	7.29 \pm 0.12	23.75 \pm 1.07*	173.3 \pm 8.71*	275.25 \pm 5.34*

Rats nourished received the control diet with 23% protein during the life; and rats malnourished had its mothers submitted to diet with 7.8% protein during suckling. The animals were weighed on the 1st, 24th, 60th and 120th day. The data are reported as mean \pm SD. * $p < 0.01$ compared to the nourished group at the same age (two-tailed Student t-test).

in an acoustic isolated room, by using a box (20 x 20 x 20 cm) with the floor consisting of parallel metallic bars (inter-bar distance: 1.3cm), connected to an electric scrambled current source. The test consisted of placing a pair of rats of the same group (matched by weight) in the box, where they received a session of stimuli to induce aggressive responses. Each stimulus (an electric foot-shock) was represented by a 1.6 mA - 2 s current pulse. Each session lasted 20 min and was composed by 5 stimuli separated by a 4-min interval. During the first 3 min of this interval, the duration of the aggressive response was quantified by using a digital chronometer. So the total time for obser-

vation of aggressive behavior was 900 s. The annotations and the verification of the equipment were preceded in the last minute of each interval. The aggressive response was defined as the presentation of, at least, one of the two following behaviors: a) the animals stayed lifted up on the hind paws, facing one to the other, in a threatening attitude but without direct contact, or b) they maintained evident physical contact (besides being scratched, exhibition of the teeth and emission of characteristic vocalization). The data were compared by the two-tailed Student t-test (body weight) or by the Mann-Whitney U-test (aggressiveness) with the level of significance set at $P \leq 0.05$.

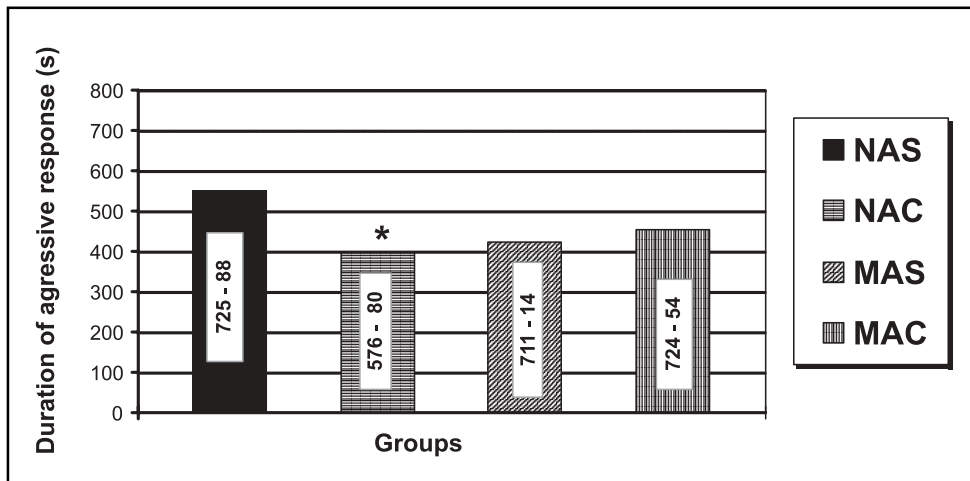


Fig 1. Aggressive responses of rats nourished or malnourished during sucking period, submitted or not to acute treatment with citalopram. The groups nourished acute saline (NAS), malnourished acute saline (MAS) nourished acute citalopram (NAC), malnourished acute citalopram (MAC) were obtained. For each group (20 rats/group), the data are reported as median of time of aggression in seconds, with a range reported inside the columns. * $P < 0.05$ compared to saline-treated nourished animals (Mann - Whitney two-tailed U-test).

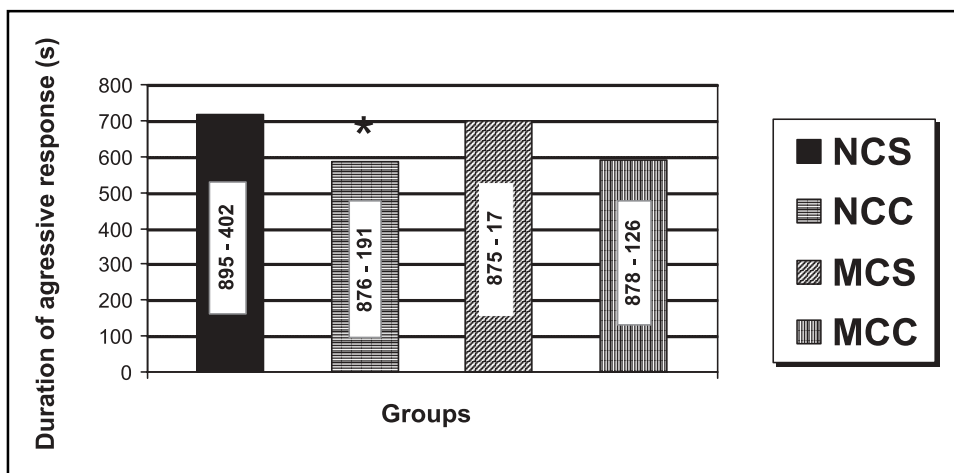


Fig 2. Aggressive responses of rats nourished or malnourished during sucking period, submitted or not to chronic treatment with citalopram. The groups nourished chronic saline (NCS), malnourished chronic saline (MCS) nourished chronic citalopram (NCC), malnourished chronic citalopram (MCC) were obtained. For each group (20 rats/group), the time of aggression this represented in the columns (median), inside the same ones they are the values maximum and minimum. * $P < 0,05$ compared to saline-treated nourished animals (Mann - Whitney two-tailed U-test).

RESULTS

Compared to the nourished (Table 2), malnourished rats showed a reduction in body weight on the 24th, 60th and 120th day. The acute or chronic treatment with citalopram reduced the aggressive responses in nourished group but not in malnourished one (Figs 1 and 2). The high values of the time of aggressiveness in the animals submitted to the chronic treatment can be due to the largest time of isolation in the cages.

DISCUSSION

The present study showed that malnutrition during the critical period of brain development impaired the weight evolution of the rats. This effect can be a consequence of the protein deficiency that was imposed the mothers during the suckling period. In this phase, the protein deficiency causes alterations in the quality of the maternal milk^{15,16} that induces damage of the growth of the body in several species of mammals⁴. The treatment with citalopram reduced the intraspecific aggressive response in the nourished rats. This anti-aggressiveness effect of the selective serotonin reuptake inhibitor (SSRI) may be a consequence of an increase in the serotonergic transmission. The SSRI increase the availability of 5-HT in the synapse and, consequently, the action of this monoamine². However, the malnutrition during brain growth spurt blocked the effect of the SSRI on aggressive behavior in adult rats.

The reduction of aggressiveness after treatment with SSRI found in the present work agrees with previous studies accomplished in humans¹⁷ and in animals^{2,18}. The serotonin has an important role on the emotional processes¹⁹. Serotonergic projections innervate cerebral areas that participate in the control of the aggressive behavior¹⁹. The reduction of the serotonergic activity seems to increase the aggressiveness²⁰. In contrast, the SSRI treatment increasing the synaptic availability of the 5-HT diminishes the aggressive behavior^{8,21}. The reduction of the aggressiveness after the treatment with SSRI may be the consequence of the action of the serotonin on the postsynaptic receptors^{20,22,23}.

In the present study, the neonatal malnutrition interfered in the effect of the SSRI on the intraspecific aggressiveness in the adult rats. This alteration could be related to the nutritional insult during the neonatal phase. The malnutrition during the neonatal period results in major neurochemical consequences, including those in the neurotransmitter systems⁷. In rats, the first serotonergic neurons ap-

pear between the 12^o and the 14^o day of gestation²⁴, but the final density and definitive location of terminals, it is established during the postnatal maturation of the central nervous system²⁵. Thus, the malnutrition imposed early in life could cause alterations in the serotonin neurotransmission system, reflecting on its functional responses to drugs. Some drugs which act in the central nervous system has its effects diminished in the malnourished animals^{12,13,26}. This suggests a sequel of malnutrition on the serotonergic system.

In conclusion, the malnutrition during the critical period of brain development renders adult rats resistant to the effect of the SSRI on the aggressive response. The nutritional insult appears to have an enduring effect upon the functioning of the serotonergic system. Though, it is not still clear which components of the serotonergic system are altered in a persistent way by the nutritional aggression.

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