Pharmacology Biochemistry & Behavior, Vol. 3, pp. 723–726.

BRIEF COMMUNICATION

Brain Glycine and Aggressive Behavior

PAVAO STERN AND SABRIA ĆATOVIĆ

Department of Pharmacology and Toxicology, University of Sarajevo, 71001 Sarajevo, Yugoslavia

(Received 14 August 1974)

STERN, P. AND S. ĆATOVIĆ. *Brain glycine and aggressive behavior.* PHARMAC. BIOCHEM. BEHAV. 3(4) 723–726, 1975. – Intraperitoneal glycine reduced aggressiveness caused by water deprivation or forebrain septal lesion in the rat. Nalorphine and mephenesin, drugs previously shown to elevate central glycine levels, acted in the same way as systemically administered glycine. In mice made aggressive by prolonged isolation, glycine and mephenesin acted as tranquilizers, but nalorphine failed to act. Aggressiveness in mice, induced by L-dopa or clonidine was enhanced by nalorphine and mephenesin, but was left unaffected by systemically administered glycine. Behavioral effects of glycine extend to other forms of excitation.

Glycine Nalorphine Mephenesin Aggressiveness Tranquillization

NOT long ago the idea was put forward that glycine most likely plays the role of an inhibitory neurotransmitter [2], and this notion is now largely accepted upon evidence accumulated in the meantime, especially insofar as the function of glycine in the central nervous system is concerned [2, 4, 5]. We suggest that, owing to these circumstances, an alteration of central glycine levels might produce behavioral effects. Some evidence along this line recently emerged from studies on synthetic substance P, a naturally occuring brain polypeptide, carried out in our laboratory [11,12]. Intramuscular administration of the polypeptide raised glycine levels in brain and spinal cord of mice [11], and this capability of substance P may be related to its tranquilizing action reported in an earlier paper [14]. A raise of central glycine levels could also be obtained with nalorphine [10], and mephenesin (to be published). The well-known behavioral effects of these drugs might, therefore, be due – at least in part – to glycine-mediated action. On the other hand, one should expect that adding glycine to a mobile central pool, either by systemic administration, or by liberation in situ, will counteract behavioral effects of drug depleting glycine from the mobile pool. This expectation was borne out by our finding substance P capable of abolishing the psychomotor effect [11] of moderate (but not excessive) central glycine depletion by β,β-iminodipropionitrile (IDPN) [6], which may be regarded as a behavioral effect of substance P due to restoring lost glycine.

The present communication is primarily concerned with studies of glycine activity towards aggressiveness as a particular kind of behavior. These studies were based, firstly, on direct administration – glycine having been shown to readily penetrate the hemo-encephalic barrier, thus raising both spinal and supraspinal glycine levels [9] – and secondly, on indirect elevation of central glycine levels by using nalorphine and mephenesin. Following up a collateral line not directly related to aggressive behavior, we attempted to confirm our assumption that substance P counteracts the effect of IDPN by a glycine-mediated mechanism. The possibility of abolishing psychomotor unrest caused by IDPN [14] by systemic administration of glycine was, therefore, investigated, and for obvious reasons this examination was extended to include the effects of nalorphine and mephenesin.

METHOD

Animals

Models of aggressive behavior were induced in two species, namely adult Wistar rats of either sex weighing 250–300 g, and male general-purpose albino mice, weighing 20–30 g.

Procedure

In rats, aggressiveness was induced either by water deprivation (A. Delini-Stulla, personal communication) or by inflicting a lesion to the *septum pellucidum* (septal rats) [3]. Mice were made aggressive by imposing an extended period of isolation [15], or by administration of chemical substances, namely L-dopa [6] or clonidine [7]. Pertinent details are given on Tables 1 and 2.

Water-deprived rats were classed as aggressive or non-aggressive, depending on their behavior when being given free access to a drinking tube. The aggressive group, subsequently used in experiments, consisted of animals who drove each other away from the drinking tube at least three

TABLE 1

INFLUENCE OF SYSTEMICALLY ADMINISTERED GLYCINE AND CENTRAL GLYCINE RELEASING DRUGS ON AGGRESSIVE BEHAVIOR IN RATS AND MICE

Species[a]	Induction of Aggressive Behavior	Control Value[b] of Observed Parameter[c]	Effect[b,d] of Drugs (mg/kg IP) Observed 60 Min (First Row) and 120 Min (Second Row) After Administration		
			Glycine (200)	Nalorphine (100)	Mephenesin (50)
Rat 8	Lesion of septum pellucidum	11.25 ± 0.59	3.37 ± 0.50[d]	2.87 ± 0.58[d]	2.87 ± 0.58[d]
			4.25 ± 0.88[d]	3.50 ± 0.63[d]	6.37 ± 0.66[d]
Rat 16	Water deprivation for 24 hr before exp.	5.00 ± 0.33	0.87 ± 0.22[d]	1.00 ± 0.18[d]	1.12 ± 0.15[d]
			0.81 ± 0.21[d]	0.87 ± 0.18[d]	0.14 ± 0.02[d]
Mouse 20	Isolation for 21 days before exp.	3.05 ± 0.43	14.85 ± 0.95[d]	3.70 ± 0.37[g]	15.20 ± 0.76[d]
			16.30 ± 0.84[d]	4.00 ± 0.44[g]	15.90 ± 0.60[d]
Mouse 20	L-Dopa IV 850 mg/kg	2.25 ± 0.24	2.15 ± 0.18[g]	1.30 ± 0.11[e]	1.30 ± 0.13[f]
			1.80 ± 0.17[g]	1.10 ± 0.07[d]	1.10 ± 0.01[e]
Mouse 20	Clonidine IP 50 mg/kg	4.45 ± 0.42	4.85 ± 0.22[g]	1.55 ± 0.22[d]	1.75 ± 0.22[d]
			4.20 ± 0.47[g]	1.95 ± 0.35[e]	1.50 ± 0.20[d]

[a]Number of animals in groups given in parentheses

[b]Mean ± s.e.m.

[c]For septal rats: Henderson-Stark score (see [14]; seven parameters, such as startle reactions to different stimuli, and attempts to bit an object in contact with, or approaching, various parts of the body were rated 0 to 2 points. A score of 2 and above, up to 11, is considered normal; above 11 it means hyperreactivity). For water-deprived rats: number of attempts per minute to drive another animal away from a drinking tube. For isolated mice: length of delay (sec) before attacking a non-aggressive animal placed into the cage. For mice with drug-induced aggressiveness: number of attacks launched per minute against a non-aggressive mouse.

[d]Differences between observed and control values for all parameters are statistically significant (Student's t-test) at $p<0.001$, unless stated otherwise. Parameters given in the table can only be compared across rows, but not down the columns.

[e]Significant at $p<0.01$

[f]Significant at $p<0.02$

[g]Non significant

times within an interval of 60 sec. Other animals were discarded as non-aggressive. In septal rats the intensity of aggressive behavior was graded. We used the Stark-Henderson [13] scale for this purpose, utilizing only animals scoring at least eight points on this scale. The aggressiveness of isolated mice was assessed by observing the promptitude of their attacking a non-aggressive fellow. Animals were classed as strongly, moderately, and weakly aggressive, or non-aggressive, if they launched an attack within 5, 7, 15 or more than 15 sec, respectively. Strongly and moderately aggressive mice were pooled and used as one group, the other animals were discarded. The strength of drug-induced aggressiveness was judged by the number of attacks launched per minute against a non-agressive animal.

Psychomotor unrest, a condition lacking any aggressive component, was induced in general-purpose albino mice of either sec, weighing 20–30 g, by administration of IDPN according to the procedure (route, dosage scheme) outlined in Table 2.

The drugs under examination – glycine, nalorphine and mephenesin – were administered intraperitoneally at the peak of aggressiveness, in doses specified on Tables 1 and 2, and their effects were determined twice, at 60 and 120 minutes after administration. The action of a drug was judged successful if: (1) it completely abolished the aggressiveness of water-deprived rats; (2) it reduces the Stark-Henderson score of septal rats to 4 points or less, and (3) if it delayed the attack of an aggressive mouse to over 15 sec

TABLE 2

INFLUENCE OF SYSTEMICALLY ADMINISTERED GLYCINE AND CENTRAL GLYCINE RELEASING DRUGS ON PSYCHOMOTOR SYMPTOMS PRODUCED WITH IDPN* IN MICE

Control Values† of Observed Parameter‡	Effect†§ of Drugs (mg/kg IP) Observed 60 Min (First Row) and 120 Min (Second Row) After Administration		
	Glycine (200)	Nalorphine (100)	Mephenesin (50)
1.60 ± 0.36	114.2 ± 1.8	115.3 ± 1.8	118.1 ± 1.4
	116.7 ± 1.1	118.9 ± 0.7	118.7 ± 0.9

*β, β-iminodipropionitrile, administered IP during five days. Dosage scheme: 1st and 2nd days, 15 mg/kg; 3rd day, no treatment; 4th day, 15 mg/kg; 5th day and 6th day, 15 mg/kg.

†Mean ± s.e.m.

‡Total period of resting (in sec) within a 2-min observation interval

§All differences between observed and control values were statistically significant (Student's t-test) at $p<0.001$.

on the average; when tried against psychomotor unrest, a drug was deemed successful only if it achieved complete suppression.

RESULTS AND DISCUSSION

A summary of results is given in Tables 1 and 2, showing that the three drugs under examination were throughout successful against aggressiveness in rats, regardless of the kind of model, but they gave inconsistent results in experiments with mice. Glycine and mephenesin successfully abolished the isolation-induced aggressiveness, but nalorphine was unsuccessful. In contrast, the models induced by either L-dopa or by clonidine were enhanced by nalorphine and mephenesin, while directly administered glycine did not affect these types of aggressiveness.

Glycine was capable of suppressing psychic excitation other than aggressiveness, as shown in the separate experiment with IDPN-induced psychomotor unrest in mice [6]. Under the condition of this experiment, however, not only directly applied glycine, but also the ostensible central glycine increased by nalorphine and mephenesin, completely suppressed the psychomotor symptoms, and these results strongly support the assumption of a glycine-mediated mechanisms as the base for a similar action of substance P.

The inconsistencies observed in experiments with different models of aggressiveness might be ultimately traced to the origins of various models, and possibly to species differences. With respect to the effects observed in mice let us point out that the models induced in these animals are very likely related to entirely different biochemical brain processes [1], hence glycine, and drugs increasing central glycine levels, must act differently, depending on the kind of model. Should the aggressive behavior stem from generally or locally depressed central glycine levels, it is logical to assume that a dose of the amino acid introduced from without, or a sufficient amount released from some store in situ, will abolish the aggressiveness. The same argument holds true for other consequences of low central glycine, e.g. for the psychomotor unrest induced by IDPN. The cases where glycine failed to act, and the drugs allegedly increasing central glycine levels acted contrary to expectation, are presently beyond our speculations and must not be discussed at this moment.

To close, we repeat that central glycine levels might be somehow involved in the development of certain types of aggressive behavior, as well as in that of some other behavioral patterns. This much seems to be well supported by our results so far. It is necessary to consider the possibility that a raise in central glycine levels, primarily suppressing excitation (e.g. the action of increased central glycine against psychomotor excitation produced by IDPN), has only a secondary effect on aggressive behavior. In our experiments, however, increased central glycine did not diminish the motility of animals, but distinctly reduced aggressive behavior. In these experiments at least, the action of glycine was directed primarily against aggressiveness. Investigations are going on and eventually a more comprehensive picture may arise from further results.

ACKNOWLEDGEMENT

This work was supported by Grant No. 02/2-811-1/73 from the Republic's Scientific Research Fund of Bosna and Hercegovina.

REFERENCES

1. Aprison, M. H. and J. N. Hingtgen. Neurochemical correlates of behavior. *Int. Rev. Neurobiol.* **13**: 325–341, 1970.

2. Aprison, M. H. and R. Werman. A combined neurochemical and neurophysiological approach to the identification of central nervous system transmitters. In: *Neurosciences Research*, Vol. 1, edited by S. Ehrenpreis and O. C. Solnitzky, New York: Academic Press, 1968, pp. 143–174.

3. Brady, J. V. and W. J. H. Nauta. Subcortical mechanisms in emotional behavior: affective changes following septal forebrain lesions in the albino rat. *J. comp. physiol. Psychol.* **46:** 339–346, 1953.
4. Hösli, L. and H. L. Hass. The hyperpolarization of neurones of the medulla oblongata by glycine. *Experientia* **28:** 1057–1058, 1972.
5. Hösli, L. and A. K. Tebecis. Actions of amino acids and convulsants on bulbar reticular neurones. *Expl Brain Res.* **11:** 111–127, 1970.
6. Katz, M. R., H. C. Yon-Koo and S. Krop. The effects of psychoactive drugs on 3, 4-dihydroxyphenylalanine/DL-dopa/induced excitation in mice. *Fedn Proc.* **26:** 289, 1967.
7. Morpurgo, C. Aggressive behavior induced by large doses of 2-/2,4-di-chloro-phenylamino/-2-imidazoline hydrochloride/ST 155/in mice. *Eur. J. Pharmac.* **3:** 374–377, 1968.
8. Stern, P. Substance P and its central effects. *J. of neurovisceral Rel.* Suppl. **9:** 236–248, 1969.
9. Stern, P., S. Ćatović and N. Filipović. Erzeugung von Rigor der Rattenextremitäten durch Hemmung der Glyzin-Sinthese. *Experientia* **29:** 144, 1973.
10. Stern, P., S. Ćatović and N. Filipović. The metabolism of glycine in mice treated acutely and chronically with morphine. *Pharmacology* **10:** 97–103, 1973.
11. Stern, P., S. Ćatović and M. Stern. Mechanism of action of Substance P. *Naunyn-Schmiedebergs Arch. exp. Path. Pharmac.* **281:** 233–239, 1974.
12. Stern, P. and J. Hadžović. Pharmacological analysis of central actions of Substance P. *Archs int. Pharmacodyn.* **202:** 259–262, 1973.
13. Stark, P. and J. K. Henderson. Increased reactivity in rats caused by septal lesions. *Int. J. Neuropharmac.* **5:** 379–383, 1966.
14. Thuillier, J., A. Burger and P. Mouille. Contribution àl'étude du syndrome moteur provoqué chez la souris par l'iminodipropionitrile (souris tournantes). *C. r. hebd. Séanc. Acad. Sci., Paris* **147:** 1052–1055, 1953.
15. Valzelli, L. Drugs and aggressiveness. In: *Advances in Pharmacology, Vol. 5.* edited by S. Garattini and P. A. Shore. New York: Academic Press Inc. 1967.