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The Role of Neurotransmitters in Stress-Induced Antinociception (SIA)

ANNE E. SNOW, SUSAN M. TUCKER AND WILLIAM L. DEWEY

Department of Pharmacology, Medical College of Virginia
Virginia Commonwealth University, Richmond, VA 23298

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SNOW, A. E., S. M. TUCKER AND W. L. DEWEY. *The role of neurotransmitters in stress-induced antinociception (SIA).* PHARMAC. BIOCHEM. BEHAV. **16**(1) 47–50, 1982.—The role of the neurotransmitters, norepinephrine, dopamine and serotonin in stress-induced antinociception (SIA) was examined by altering neurochemical tone with appropriate pharmacological tools. Quipazine (15.0 mg/kg, IP) a serotonin agonist, increased the peak and duration of antinociception following stress and BC-105 (3.0 mg/kg, IP), a serotonin antagonist, blocked the increase of tail-flick latency following stress. Clonidine (0.1 mg/kg, SC) an α_2 agonist, markedly decreased SIA whereas phenoxybenzamine (2.5 mg/kg, IP), an α_1 antagonist, increased the peak and duration of SIA. When dopaminergic tone was increased with apomorphine (0.05 mg/kg, SC) the increase of tail-flick latency after stress was markedly attenuated whereas blockage of dopamine receptors with haloperidol (2.5 mg/kg, IP) increased the peak and duration of SIA. Alterations of serotonergic, but not noradrenergic or dopaminergic, tone had similar effects on increased latency in tail-flick test produced by brain stimulation produced analgesia (SPA), morphine and SIA. These data support the hypothesis that alterations in tail-flick latency involves a serotonergic system.

Stress Antinociception Serotonin Norepinephrine Dopamine

STRESS-induced antinociception (SIA) is an intriguing phenomenon whereby exposure to stressful stimuli or events decreases an animal's response to other painful stimuli. These stressful procedures elicit antinociception comparable to a moderate dose of morphine. Stimuli which produce this type of antinociception include inescapable foot shock, classical conditioning, cold, swim stress, 2-deoxy-d-glucose, restraint and food deprivation, and conditioned emotional fear [2, 4, 5, 14, 20]. Decreases of response to painful stimuli have been quantitated by the tail-flick test and by response to dental pulp stimulation, flinch jump, hot plate, paw pinch and tail pinch tests [4, 9, 13, 15].

Antinociception appears to involve activation of a supraspinal endogenous pain inhibitory system [16, 17, 18]. Observations that spinal transection at T_3-T_4 obviate behaviorally-activated antinociception provide additional support for the role of a supraspinal influence on antinociception [6]. Output from this system must then descend to the spinal cord to inhibit transmission of nociceptive information. This corticofugal transmission may involve a specific neurotransmitter system or several neurotransmitter systems. Consequently, if a specific neural pathway is involved in SIA, then blockade or stimulation of the neurotransmitter of that pathway should lead to alterations in SIA.

The possible role of neurotransmitters and neurohumoral modulators on opiate-induced antinociception and stimulation-produced analgesia (SPA) have been examined in great detail. No systematic study of the role of neurotransmitters in stress-induced antinociception has been carried out. Consequently, with the use of various agonists and antagonists of purported neurotransmitters, we sought to examine the role of norepinephrine, dopamine and serotonin neurotransmitter systems in the antinociceptive response produced by stress and to compare these results with those previously reported for the role of these neurotransmitter systems in other types of antinociception.

METHOD

White male Sprague Dawley rats weighing 250–350 grams were used. Rats were provided with water and food ad lib. The room was kept at a constant temperature of 23°±1°C and light and dark cycles were maintained on a 12 hr on 12 hr off schedule.

Antinociception was measured by the tail-flick response [10,13]. The lamp intensity was adjusted so that control tail-flick latencies were between 2–4 seconds and a 10 second cutoff limit was used to prevent damage to the tail. Prior to any experimental manipulation or treatment, a control tail-flick latency was determined for each animal.

Stress was provided by placing the rat on an electrified grid measuring 16×23 cm with rods placed 17 mm apart. Current to the rods was supplied by a Lafayette Master Shocker A615 C and the duration of the shock was controlled by a Lafayette Bank Timer 1431 A. The current was intermittent at one pulse/second. Intensity of the shock was 0.8–1.0 mA and the duration of the shock session was 20 seconds.

Rats were randomly assigned to a treatment group. Drugs were made up in saline and were administered either subcutaneously (SC) or intraperitoneally (IP). A dose of the neurochemical modulator, which had little to no activity itself, was given at a time in relation to stress so that its peak effect

of the central neurochemicals was present when stress was applied. These peak times corresponded to previously reported maximum effects on the neurotransmitter system. Following exposure to stress we determined tail-flick latencies (TFL) every 5 minutes for 30 minutes. For every neurotransmitter treatment control groups consisted of stress-no drug animals and drug-non stressed animals. The non-stressed drug groups were held on the shock grid for 20 seconds without the shock stimulus turned on.

Drug Regimens

For noradrenergic systems, clonidine (0.1 mg/kg, SC) in a dose which potentiated morphine was administered 20 minutes prior to stress [21]. Phenoxybenzamine (2.5 mg/kg, IP) was administered 20 minutes prior to stress.

A dose of 15.0 mg/kg of quipazine is reported to be effective for stimulating serotonin receptors [12]. For this study we used 15.0 mg/kg, IP of quipazine 15 minutes before stress. The serotonin blocker BC-105 (3.0 mg/kg, IP) was administered one hour before stress [23].

In order to examine the role of dopaminergic systems in SIA we used apomorphine a dopamine agonist and haloperidol a dopamine antagonist [3,8]. Apomorphine (0.05 mg/kg, SC) was administered 20 minutes prior to stress. Haloperidol (2.5 mg/kg, IP) was administered 30 minutes before stress.

Apomorphine was purchased from Merck and Co. and Haldol was obtained from McNeil. Phenoxybenzamine was a gift from Merck Sharpe and Dohme, quipazine from Miles Laboratories, BC-105 from Sandoz and clonidine was obtained from Boehringer.

For data analysis, each animal served as its own control for baseline tail-flick latencies and subsequent tail-flick latencies following stress were plotted versus time. For a given neurotransmitter, differences from baseline in tail-flick latency were combined and the data analyzed for the entire group. Statistical manipulations included Dunnett's t and ANOVA. Each F value was less than 3.0 with degrees of freedom of 2,23 or 2,29. Significant differences were noted for values less than $p=0.05$.

RESULTS

Results are described under the subheading of the neurotransmitter investigated.

Norepinephrine

Phenoxybenzamine has been shown to potentiate the action of morphine [8]. The effect of phenoxybenzamine on SIA was to prolong the duration of SIA with tail-flick latencies significantly increased from 10–30 minutes (Fig. 1). Phenoxybenzamine (2.5 mg/kg, IP) without stress had no effect on tail-flick latency over the 30 minute test period (Table 1). Clonidine reduced SIA by 82% at 5 minutes and at 10 minutes by 63% when compared to stressed non-drug controls. These results indicate that clonidine is able to antagonize the sharp increase of tail-flick latency following stress. Clonidine did not alter tail-flick latency in non-stressed rats (Table 1).

Dopamine

Apomorphine pretreatment slightly reduced the peak response at 5 minutes and significantly attenuated the response

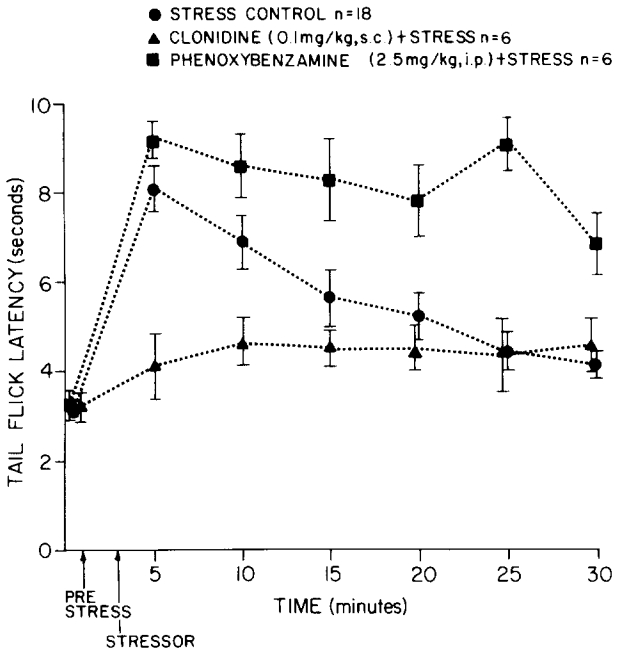

FIG. 1. Effects on SIA by pretreatment with phenoxybenzamine (2.5 mg/kg, IP) and clonidine (0.1 mg/kg, SC). Stress controls are also shown for comparison.

from 10–30 minutes (Fig. 2). Apomorphine without stress had no effect on tail-flick latency (Table 1). These results indicate that activation of dopamine receptors by apomorphine reduces the antinociceptive response to stress. Haloperidol pretreatment before stress produced maximal antinociception (10 sec) at 5 minutes in all 6 animals tested. In addition the tail-flick latencies of haloperidol pretreated animals were significantly greater than stressed controls from 15 minutes until the end of the test period (Fig. 2). Haloperidol had no activity in the tail-flick test in the absence of stress (Table 1). These data demonstrate that blockade of dopamine receptors with haloperidol increased antinociception following stress.

Serotonin

Figure 3 shows the results of serotonergic manipulations. BC-105 administered one hour before challenge with stress almost completely antagonized SIA, the tail-flick latencies being decreased significantly from 5–25 minutes. In the absence of stress BC-105 produced a slight decrease in tail-flick latency (Table 1). These results indicate that blockade of serotonergic systems prevents the expression of stress-induced antinociception. The effect of quipazine can be seen in Fig. 3. Quipazine in combination with stress produced an increase of SIA both in the magnitude and duration of response with a significant increase of tail-flick latency from 10–30 minutes. In the absence of stress, quipazine slightly increased tail-flick latency (Table 1). These data support the hypothesis that serotonergic systems are involved in antinociceptive responses and that increasing serotonergic tone potentiates stress-induced antinociception.

TABLE 1

EFFECT OF DRUG PRETREATMENT ON TAIL-FLICK LATENCY (TFL) IN NON-SHOCKED RATS
(TFL Expressed in Seconds)

Agent	Baseline	Time (Minutes) 5	10	15	20	25	30
Quipazine	2.7 ± 0.2	4.5 ± 0.5	4.4 ± 0.3	5.4 ± 0.6	4.4 ± 0.4	3.4 ± 0.3	3.5 ± 0.3
BC105	3.2 ± 0.2	2.3 ± 0.2	2.5 ± 0.2	2.1 ± 0.2	2.0 ± 0.2	2.2 ± 0.2	1.9 ± 0.2
Apomorphine	3.4 ± 0.2	2.9 ± 0.1	3.0 ± 0.3	4.2 ± 0.1	4.4 ± 0.3	4.1 ± 0.4	4.1 ± 0.3
Haloperidol	3.1 ± 0.3	3.9 ± 0.4	4.1 ± 0.3	3.5 ± 0.3	3.8 ± 0.2	3.5 ± 0.2	2.7 ± 0.4
Clonidine	3.3 ± 0.3	3.8 ± 0.6	3.6 ± 0.4	3.8 ± 0.4	4.6 ± 0.3	3.6 ± 0.6	4.0 ± 0.3
Phenoxy-benzamine	3.2 ± 0.2	3.2 ± 0.5	3.5 ± 0.3	3.2 ± 0.3	3.1 ± 0.2	3.3 ± 0.1	3.1 ± 0.3

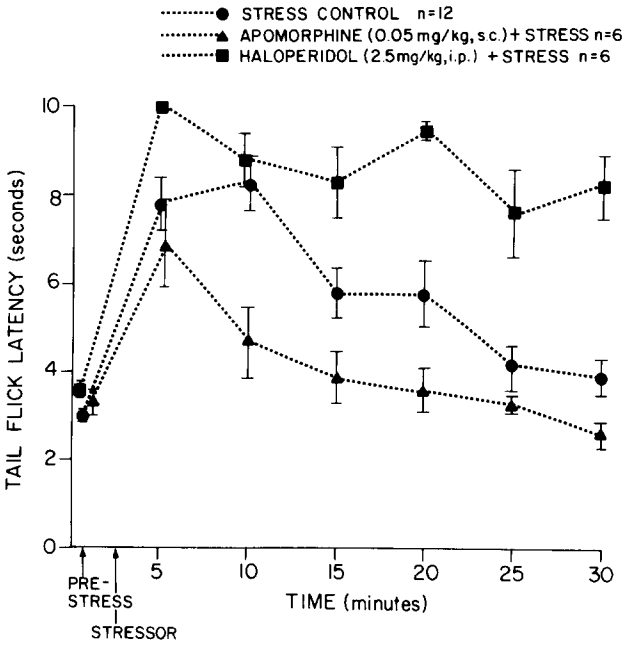

FIG. 2. Dopaminergic Systems. Effects of apomorphine (0.05 mg/kg, SC) and haloperidol (2.5 mg/kg, IP) on SIA. Stress controls are shown for comparison.

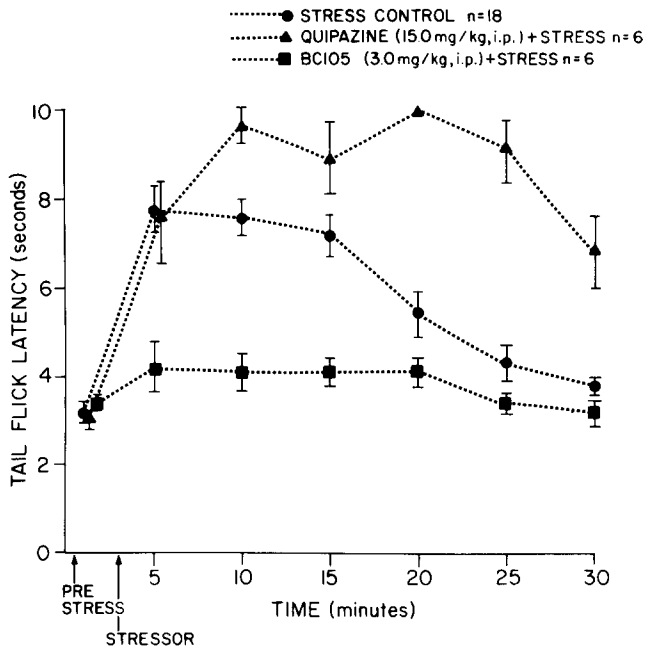

FIG. 3. This figure shows the effect on SIA of a serotonin agonist quipazine (15.0 mg/kg, IP) 15 min before stress and a serotonin antagonist BC-105 (3.0 mg/kg, IP) 60 minutes before stress. A stress control is also shown for comparison.

DISCUSSION

Not surprisingly, manipulation of neurotransmitters via neurohumoral modulators altered stress-induced antinociception. Agents which block dopaminergic and noradrenergic receptors enhance SIA whereas agonists diminish the antinociceptive response to stress. On the other hand, a serotonin receptor agonist increased antinociception in response to stress whereas an antagonist decreased antinociception (Table 2). Although systemic administration of neurotransmitter agonists and antagonists does not define specific sites of interaction, the results from these type of experiments do permit a comparison of the effects of these neuromodulators on tail-flick latency increased by stress with those previously reported for morphine and brain stimulation induced antinociception.

When neurotransmitter alterations were compared for their effects on morphine antinociception, stimulation-produced analgesia and stress-induced antinociception some similarities emerge. The results show that enhancing serotonergic tone increases antinociception and decreasing serotonergic tone decreases antinociception [1,19]. Decreasing noradrenergic tone with phenoxybenzamine increased morphine and SIA induced antinociception while tetrabenazine, another agent which decreases noradrenergic

TABLE 2

SUMMARY OF NEUROTRANSMITTER INTERACTIONS AND SIA

Neurotransmitter	Agent	Effect on SIA
Dopamine	antagonist-haloperidol	↑
	agonist-apomorphine	↓
Norepinephrine	antagonist-phenoxybenzamine	↑
	agonist-clonidine	↓
Serotonin	antagonist-BC 105	↓
	agonist-quipazine	↑

TABLE 3

EFFECT OF NEUROTRANSMITTER ALTERATIONS

	SIA	MSO4	SPA
Norepinephrine			
Agonist	↓	↑	↓
Antagonist	↑	↑	↑
Dopamine			
Agonist	↓	↓	↑
Antagonist	↑	↑	↓
Serotonin			
Agonist	↑	↑	↑
Antagonist	↓	↓	↓

tone by a different mechanism increased brain stimulation induced antinociception [1,7]. However, other reports indicate that morphine antinociception is increased by agents which increase noradrenergic tone [11]. For dopaminergic systems antinociception induced by SIA is similar to that induced by morphine since they are antagonized by apomorphine and potentiated by haloperidol [23] (Table 3). Brain stimulation induced antinociception differs since apomorphine increases and pimozide, a dopamine receptor blocker decreases its effect [1].

Although neurohumoral modulators may block or enhance SIA, it is unclear whether these manipulations interfere with the stress response or act directly on antinociceptive systems. Our results do support the hypothesis that for stress-induced antinociception as for morphine and brain stimulation, serotonergic systems are involved.

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