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Effects of *Hypericum perforatum* Extract on Ethanol Intake, and on Behavioral Despair: A Search for the Neurochemical Systems Involved

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PANOCKA, I., M. PERFUMI, S. ANGELETTI, R. CICCOCIOPPO AND M. MASSI. *Effects of* Hypericum perforatum *extract on ethanol intake and on behavioral despair: A search for the neurochemical systems involved.* PHARMACOL BIOCHEM BEHAV **66**(1) 105–111, 2000.—The present study investigated the possible involvement of σ receptors and of serotonergic mechanisms in the effects of *Hypericum perforatum* extract (HPE) on immobility time in the forced swimming test (FST) and on ethanol intake in Marchigian Sardinian alcohol-preferring rats. The HPE employed was a dry extract containing 0.3% hypericin and 3.8% hyperforin. Intraperitoneal pretreatment with 20 mg/kg of the σ receptor antagonist rimcazole (RIM), 30 min prior to HPE, completely suppressed the antiimmobility effect of HPE (3 intragastric injections of 250 mg/kg). Intracerebroventricular pretreatment with 5,7-dihydroxytryptamine (5,7-DHT), which produced a marked depletion of brain serotonin, reduced the antiimmobility effect, although this reduction was not as pronounced as that of RIM. On the other hand, the inhibitory effect of HPE on 10% ethanol intake was modified neither by 5,7-DHT nor by RIM pretreatment. These results suggest that the antidepressant-like effect of HPE in the FST may be mediated by interaction with σ receptors and to some extent by increased serotonergic neurotransmission. On the other hand, these mechanisms appear to be unimportant for the effect of HPE on ethanol intake. © 2000 Elsevier Science Inc.

Hypericum perforatum Forced swimming test Ethanol intake Alcohol-preferring rats
5,7-Dihydroxytryptamine Sigma receptor Sigma receptor antagonist Rimcazole

SEVERAL kinds of extract of *Hypericum perforatum* (HPE), the common plant usually called St. John's wort, have antidepressant properties in humans (25,31,62) and exert an antidepressant-like action in laboratory animals (6,7,50), in experimental models such as the forced swimming test (FST) (4,5,53,66). Moreover, recent reports (20,21,50,55) that HPE reduces ethanol intake in alcohol-preferring rats have raised interest for the potential use of HPE in the pharmacotherapy of alcohol abuse.

At present, the mechanism of action for these effects of HPE is unknown. A variety of neurochemical and biochemical effects have been reported for HPE. It reduces serotonin reuptake (40,45,51), and causes MAO inhibition (17,43). Components of HPE show affinity for 5-HT_{1A} receptors (17). HPE has been shown to reduce noradrenaline, dopamine and L-glutamate reuptake (3,8,29). Butterweck et al. (6) showed that the effects of HPE in the FST may be in part mediated by activation of dopaminergic and opioid mechanisms. Cott (17) reported affinity of crude HPE for $GABA_A$, $GABA_B$, adenosine, and benzodiazepine receptors. Recently, hypericin, which is believed to be an important component for the antidepressant action of HPE (6,7,40), has been reported to bind with high affinity to σ receptors (54), which may be involved in the relief of behavioral despair in the FST (36).

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Several reports indicate comorbidity between depression and ethanol abuse (23,2,35,39,46,59), and there are data suggesting a relationship between high ethanol intake and a depression-like condition in some lines of alcohol-preferring rats (30,47,61). Moreover, it has been shown that depressive disorders and ethanol abuse may imply similar changes in the activity of central neurotransmitters (35). In Marchigian Sardinian alcohol-preferring (msP) rats, HPE has been shown to inhibit ethanol intake and to produced an antidepressant-like effect in the FST (50), comparable to that exerted by ethanol itself (15). The antidepressant-like action of ethanol was not observed in alcohol nonpreferring rats. Thus, it seemed interesting to investigate in msP rats whether reduction of ethanol intake and the antidepressant-like effect in the FST induced by HPE may involve similar mechanism(s).

Serotonin (5-HT) is one of the most important neurotransmitters involved both in ethanol intake control (38,42, 47,57,67) and in the pathogenesis of depressive disorders (3,34). To check whether ethanol intake inhibition and/or the antidepressant-like effect of HPE may depend on enhancement of 5-HT synaptic levels, we investigated the effects of HPE in msP rats depleted of serotonin by intracerebroventricular (ICV) infusion of the neurotoxin 5,7-dihydroxytryptamine (5,7-DHT). Moreover, because hypericin shows high affinity for σ receptors (54), the σ receptor antagonist RIM (9,24) was used to investigate whether these receptors are involved in the effects of HPE.

METHOD

Animals

Male genetically selected msP rats were employed. They were bred in the Department of Pharmacological Sciences and Experimental Medicine of the University of Camerino (Marche, Italy) for 26 generations from Sardinian alcohol-preferring rats (sP) of the 13th generation, provided by the Department of Neurosciences of the University of Cagliari (16). At the age of 2 months, msP rats were selected for 10% (v/v) ethanol preference, offering them free choice between water and 10% ethanol 24 h a day for 15 days. Water and 10% ethanol were offered in graduated drinking tubes equipped with metallic drinking spouts. The rats employed in the following experiments had a 24-h ethanol intake of 6–7 g/kg with a percent ethanol preference [ml of ethanol solution/ml of total fluids (water + 10% ethanol) ingested in 24 h × 100] higher than 90. At this stage of selective breeding, such a high ethanol preference is usually shown by 90–95% of the msP rats.

At the time of the experiments the body weight of rats ranged from 400 to 450 g. They were kept in individual cages in a room with a reverse 12 L:12 D cycle (lights off at 1000 h), temperature of 20–22°C and humidity of 45–55%. Rats were offered free access to tap water and food pellets (4RF18, Mucedola, Settimo Milanese, Italy). All animal testing was carried out according to the European Communities Council Directive of 24 November 1986 (86/609/EEC).

Drugs

The HPE employed in the present study was a generous gift of Indena S.p.A., Milan, Italy. It was a dry extract (cod. n. 4149040, prepared in October 1997) containing 0.3% hypericin and 3.8% hyperforin. It was given by intragastric (IG) infusion at the dose of 250 mg/kg, as a fine suspension in tap water prepared just before treatment. This dose of HPE was chosen on the basis of the results of our previous study (50).

5,7-DHT and desipramine (RBI, Natick, MA) were dissolved in 0.9% NaCl. Ascorbic acid 0.1% was added to the solution of 5,7-DHT as an antioxidant. Nomifensine (RBI, Natick, MA) was suspended in distilled water (5 mg/ml) immediately before intraperitoneal (IP) injection. Fluoxetine (a gift of Lilly Research Laboratories, Indianapolis, IN) and rimcazole (Sigma-Aldrich, Milan) were dissolved in distilled water immediately prior to intraperitoneal (IP) injection. The RIM dose of 20 mg/kg was chosen on the basis of data from other studies (9,52), in which this and a similar dose (25 mg/kg) was shown to influence the activity of the mesolimbic dopaminergic (DA) system.

Intragastric Surgery

All animals used in the present study have been implanted with intragastric (IG) catheters before experiments began. This procedure was adopted to avoid any possible disturbance to the animal during HPE administration.

Rats were anesthetized by IP injection of 100–150 μl/100 g body weight of a solution containing ketamine (86.2 mg/ml) and acepromazine (1.3 mg/ml). A polyethylene catheter (PE-50, Clay Adams) was permanently implanted into the stomach, according to the method of Lukas and Moreton (33). The PE tubing was run subcutaneously to reach the skin between the scapulae, where it was exteriorized. Prior to ICV surgery or other treatments rats were allowed a week to recover from IG surgery. Before the experiments, they were made familiar with the administration procedure.

Pretreatment with 5,7-DHT

Under ketamine/acepromazine anesthesia rats were implanted with a stainless steel cannula for the infusion of 5,7-DHT into the lateral ventricle. Coordinates taken from the stereotaxic atlas of Paxinos and Watson (49) were: 1 mm posterior and 2 mm lateral to bregma, 2 mm ventral from the surface of the skull.

After a week of recovery animals were slightly anesthetized and ICV infused through a stainless steel injector (2.5 mm longer than the guide cannula) with 5,7-DHT, 150 μg/rat, or with an equal volume of its vehicle. To protect dopaminergic and noradrenergic neurons, rats received IP pretreatment with the DA reuptake blocker nomifensine (15 mg/kg, divided into two doses, 50 and 30 min before infusion) and the noradrenaline (NA) reuptake blocker desipramine (15 mg/kg, divided into two doses, 60 and 40 min before infusion) (32). Such a treatment has been shown to protect DA and NA neurons and to enhance the selectivity of 5,7-DHT neurotoxicity (1,2). The ICV infusion was carried out at a rate of 4 μl/min (total volume 20 μl) by means of a pump (Precidor Infors, AG Basel 4103 Bottmingen, Switzerland). To reduce backflow of the infusate, the injector was left in the cannula for 3 min after the end of the infusion.

Experiments in which these animals were used began 10 days following ICV infusion. After the end of the experiments (5 weeks after 5,7-DHT infusion) rats were sacrificed, brains were removed and frozen at −80°C until analysis. The tissue concentration of 5-HT and 5-hydroxyindoleacetic acid (5-HIAA) in the medial prefrontal cortex, striatum, and hippocampus was performed by high-pressure liquid chromatography (HPLC) with electrochemical detection.

Forced Swimming Test

The swimming sessions were conducted placing the rat in individual glass cylinders 60 cm high and with a diameter of

30 cm, containing water at the temperature of 23–25°C. However, water was 30 cm deep rather than 18, as reported in the original method of Porsolt et al. (53). This change was adopted according to the suggestions of recent studies by Detke et a. (22), who showed that a deeper water level offers the advantage of eliminating the false negative of the selective serotonin reuptake inhibitors. At this water depth, rats could touch the bottom of the jar with their tail, but they could not support themselves with their hindlimbs.

The first 15-min swimming session (pretest) was conducted between 1000 and 1200 h; 24 h later, rats were placed again in water for the 5-min test. Following each swimming session, rats were removed from the cylinder, dried with paper towels, placed in a heated chamber for 20 min, and then returned to their home cage. Test sessions were videotaped (Canon VC-20 color videocamera) and analyzed by means of a Panasonic (NV-HD650EG) videocassette recorder. The duration if immobility (i.e., time in which rats were making only small movements necessary to keep their head above water) was measured by an experienced observer, who was blind to the treatment conditions.

The rats employed in this experiment did not have access to ethanol for 2 weeks before the FST, because in msP rats ethanol itself produces an antiimmobility effect in this test (15).

Experimental Procedure

All the experiments were performed during the first 2 h of the dark phase of the light cycle.

Experiment 1. Effect of IG Administration of HPE on Immobility Time in the FST in msP Rats Pretreated with 5,7-DHT or with RIM

Thirty-five rats were employed to test the effect of ICV 5,7-DHT pretreatment. Eighteen of them were pretreated with 5,7-DHT, the other 17 were pretreated with 5,7-DHT vehicle, 10 days before the beginning of the experiments. Animals were divided into four groups and treated as follows: 1) 10 rats, pretreated with 5,7-DHT, received IG infusion of 250 mg/kg of HPE; 2) nine rats, pretreated with 5,7-DHT vehicle, received IG infusion of 250 mg/kg of HPE; 3) eight rats, pretreated with 5,7-DHT, were infused with IG water; 4) eight rats, pretreated with 5,7-DHT vehicle, received IG infusion of water (controls).

Thirty-three rats were employed to test the effect of IP pretreatment with RIM. Animals have been divided into four groups and treated as follows: 1) eight rats were injected with 20 mg/kg of RIM, 30 min prior to each IG infusion of 250 mg/kg of HPE; 2) seven rats were injected with 20 mg/kg of RIM, prior to each IG infusion of water; 3) eight rats were injected with RIM vehicle, 30 min prior to each IG infusion of 250 mg/kg of HPE; 4) 10 rats were injected with RIM vehicle, 30 min prior to each IG infusion of water (controls).

All IG infusions were given three times: 24, 12, and 1 h before the test. This dosing regime was employed, because our previous study (50) revealed that a single infusion with HPE is not enough to exert an antidepressant-like effect.

Experiment 2. Effect of IG Administration of HPE on 10% Ethanol Intake in msP Rats Pretreated with 5,7-DHT or with RIM

In all the experiments concerning ethanol intake, tap water and food were removed from the rats's cage immediately before the IG treatment with either HPE or vehicle, and they were offered again 1 h later together with 10% ethanol.

Twenty-five rats were employed to test the effect of ICV 5,7-DHT pretreatment. Twelve of them were infused with 5,7-DHT, the other 13 were infused with 5,7-DHT vehicle 10 days before the beginning of the experiments. Rats were divided in four groups that were treated as follows: 1) six rats, pretreated with 5,7-DHT, received IG infusion of 250 mg/kg of HPE; 2) six rats, pretreated with 5,7-DHT, received IG infusion of water; 3) seven rats, pretreated with 5,7-DHT vehicle, received IG infusion of 250 mg/kg of HPE; 4) six rats, pretreated with 5,7-DHT vehicle, were infused with IG water (controls).

To behaviorally validate the 5,7-DHT pretreatment, after completion of the HPE experiment, the same rats were tested for the effect of IP treatment with the selective serotonin reuptake inhibitor fluoxetine, 5 mg/kg. Such a behavioral validation of 5-HT depletion was not possible in the FST, because in this experimental paradigm the rat can be used just once. In fact, the immobility time observed in repeated FST decreases probably reflecting effects of learning and memory (1,65). Moreover, repeated preexposure to a stressful situation may change also the effect of antidepressant drugs (4).

Forty rats have been used to test the effect of IP RIM pretreatment. They were divided into four groups treated as follows: 1) 10 rats were injected with 20 mg/kg of RIM, prior to IG infusion with 250 mg/kg of HPE; 2) 10 rats were pretreated with RIM vehicle, prior to IG infusion with 250 mg/kg of HPE; 3) 10 rats were pretreated with 20 mg/kg of RIM, prior to IG infusion with water; 4) 10 rats were pretreated with RIM vehicle, prior to IG infusion with water (controls). All RIM or vehicle injections were made 30 min before IG infusions of HPE or water.

The intake of water and of 10% ethanol, offered in graduated drinking tubes, was measured 15, 30, 60, 90, and 120 min following access to them. Food intake was measured 30, 60, 90, and 120 min after the beginning of the experiment.

Statistical Analysis

The results of Experiments 1 were analyzed by one-way analysis of variance (ANOVA), followed by Newman–Keuls test. The results of Experiment 2 were analyzed by split-plot ANOVA with between-group comparisons for drug treatment and within-group comparisons for time. Pair-wise comparisons were made by means of the Dunnett's test. Statistical significance was set at $p < 0.05$.

TABLE 1

LEVELS OF 5-HT AND 5-HIAA IN THE BRAIN OF RATS INFUSED WITH 5,7-DHT OR ITS VEHICLE (CONTROLS). 5-HT AND 5-HIAA CONTENT IS EXPRESSED AS PICOMOL/MG TISSUE, AND AS PERCENT DEPLETION IN COMPARISON TO CONTROLS

		5-HIAA	%	5-HT	%
Frontal cortex	Vehicle	2.46 ± 0.71	92.6*	4.48 ± 0.02	89.0*
	5,7-DHT	0.18 ± 0.01		0.49 ± 0.01	
Striatum	Vehicle	4.59 ± 0.18	63.8*	3.17 ± 0.02	85.8*
	5,7-DHT	1.66 ± 0.01		0.45 ± 0.01	
Hippocampus	Vehicle	1.51 ± 0.09	87.4*	1.07 ± 0.09	94.3*
	5,7-DHT	0.19 ± 0.01		0.01 ± 0.00	

Data are the mean ± SEM of 6 rats. *$p < 0.01$.

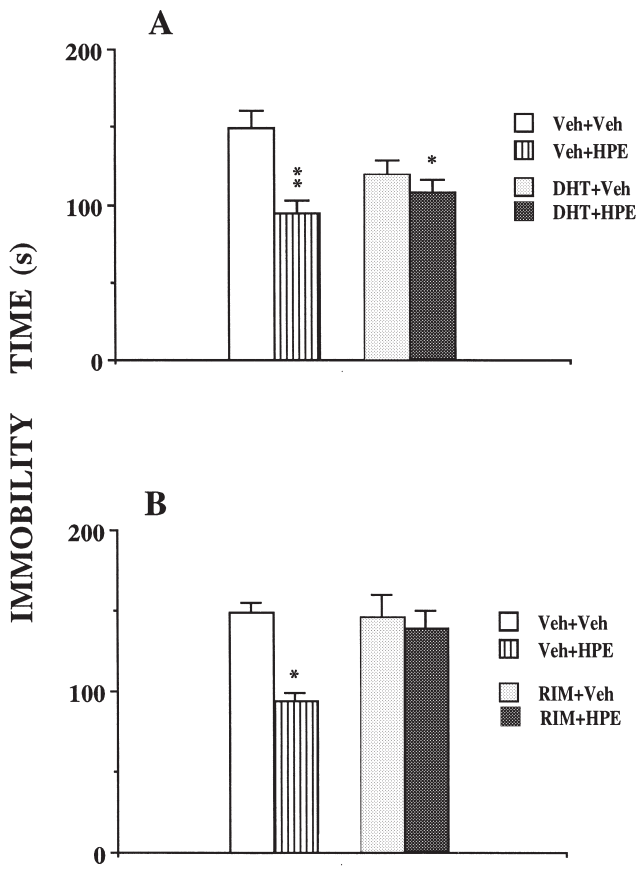

FIG. 1. (A) Immobility time in the FST in msP rats treated with IG injection of HPE or vehicle, following ICV pretreatment with 5,7-dihydroxytryptamine (DHT) or vehicle. (B) Immobility time in the FST in msP rats treated with IG injection of HPE or vehicle, following IP pretreatment with RIM, 20 mg/kg, or vehicle. Values are means ± SEM. Difference from the respective controls (IG vehicle injection): $*p < 0.05$; $**p < 0.01$; where not indicated, the difference was not statistically significant.

RESULTS

Effect of 5,7-DHT on Brain Serotonin

As shown in Table 1, the ICV infusion of 5,7-DHT produced a marked reduction of 5-HT and 5-HIAA levels in the brain areas investigated (medial frontal cortex, striatum, and hippocampus).

Experiment 1. Effect of IG Administration of HPE on Immobility Time in the FST in msP Rats Pretreated with 5,7-DHT or RIM

Figure 1A shows the effect of HPE on the immobility time in the FST in rats pretreated with 5,7-DHT. The ANOVA revealed a statistically significant treatment effect, $F(3, 31) = 4.8$, $p < 0.01$. In rats that did not receive 5,7-DHT pretreatment, HPE markedly and significantly ($p < 0.01$) reduced the immobility time to 94.2 ± 8.4 s from 149.2 ± 11.6 s in controls, receiving IG vehicle. In 5,7-DHT pretreated rats, the immobility time after HPE was 108.0 ± 7.7 s vs. 119.5 ± 12.5 s in rats receiving IG vehicle; post hoc comparisons revealed no significant effect of the HPE treatment ($p > 0.05$). The immobility of 5,7-DHT pretreated rats infused with HPE was shorter than that of controls ($p < 0.05$), but this difference was less pronounced than that observed between controls and rats without 5-HT depletion receiving HPE ($p < 0.01$).

5,7-DHT did not significantly change the time spent in immobility in comparison to controls ($p > 0.05$), although the neurotoxin induced a slight reduction in immobility time.

Figure 1B shows the effect of HPE on the immobility time in rats pretreated with RIM. The ANOVA revealed a statistically significant treatment effect, $F(3, 29) = 8.2$, $p < 0.001$. HPE produced a pronounced and statistically significant reduction in the immobility time (from 149.0 ± 5.8 to 93.7 ± 4.7 s, $p < 0.01$). Pretreatment with RIM completely abolished the effect of IG HPE, and the immobility time in rats receiving HPE after RIM pretreatment was not significantly different from that of controls ($p > 0.05$). RIM itself did not modify immobility time compared to controls ($p > 0.05$).

Experiment 2. Effect of IG Administration of HPE on 10% Ethanol Intake in msP Rats Pretreated with 5,7-DHT or with RIM

Figure 2A shows the influence of ICV 5,7-DHT on the effect of HPE on ethanol intake. The overall ANOVA revealed a highly significant effect for the IG treatment $F(1, 21) = 25.5$,

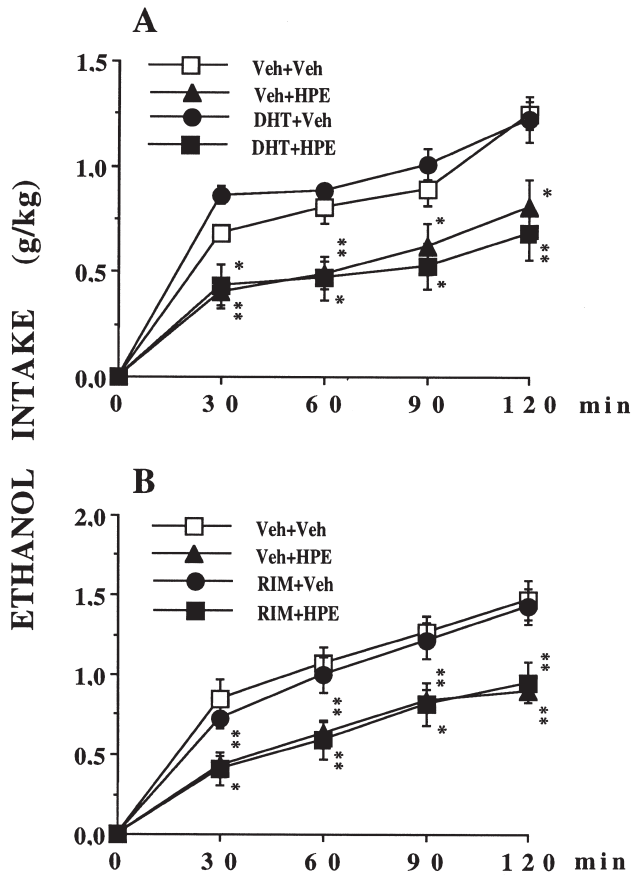

FIG. 2. (A) Cumulative 10% ethanol intake in msP rats treated with IG injection of HPE or vehicle, following ICV pretreatment with 5,7-dihydroxytryptamine (DHT) or vehicle. (B) Cumulative 10% ethanol intake in msP rats, treated with IG injection of HPE or vehicle, following IP pretreatment with RIM, 20 mg/kg, or vehicle. Values are means ± SEM. Difference from controls as in Fig. 1.

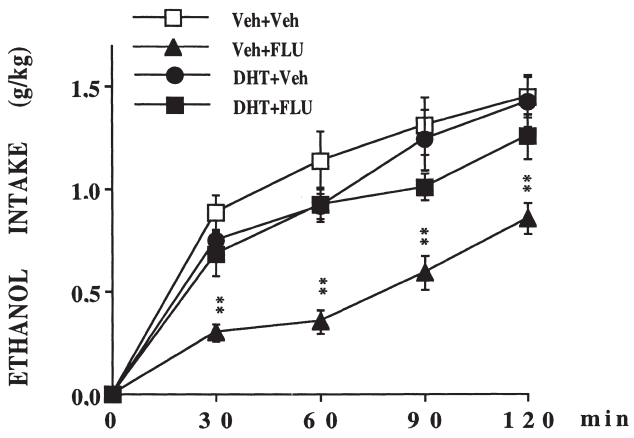

FIG. 3. Cumulative 10% ethanol intake in msP rats treated with IP injection of fluoxetine (FLU), 5 mg/kg, or vehicle, following ICV pretreatment with 5,7-dihydroxytryptamine (DHT) or vehicle. Values are means ± SEM. Difference from controls as in Fig. 1.

$p < 0.001$, and for time, $F(3, 63) = 46.6$, $p < 0.001$, but a not significant effect for the ICV treatment, $F(1, 21) = 0.06$, $p > 0.05$. 5,7-DHT pretreatment did not change ethanol intake in rats treated with IG vehicle (water) compared to controls. HPE, 250 mg/kg, reduced 10% ethanol intake in rats pretreated with 5,7-DHT vehicle to the same extent as in rats pretreated with 5,7-DHT.

Food and water intake in the 2-h test were not significantly modified either by the ICV (5,7-DHT or its vehicle) or the IG (HPE or its vehicle) treatment (data not shown).

On the other hand, the ICV pretreatment with 5,7-DHT completely abolished the reduction of ethanol intake induced by IP treatment with the selective serotonin reuptake inhibitor fluoxetine, 5 mg/kg (Fig. 3).

Figure 2B shows the influence of IP pretreatment with RIM, 20 mg/kg, on the effect of HPE on ethanol intake. RIM had no effect per se on 10% ethanol intake, in comparison to controls that received IP and IG vehicle. HPE, 250 mg/kg, markedly reduced ethanol intake in rats pretreated with IP RIM vehicle. Moreover, IP RIM pretreatment, 20 mg/kg, failed to modify the inhibition of ethanol intake induced by HPE.

Neither HPE nor RIM significantly modify water and food intake (data not shown).

DISCUSSION

The present results confirm that in msP rats IG administration of 250 mg/kg of HPE markedly reduces 10% ethanol intake and immobility time in the FST. Our previous study had shown that this HPE dose affects neither food nor water intake. Moreover, it does not influence the pharmacokinetics of ethanol and does not modify locomotor activity (50). Thus, its effects on ethanol intake and on the immobility time in the FST are behaviorally selective.

Several data indicate that HPE can increase serotonergic neurotransmission by reducing 5-HT reuptake (8,29,40,44,51) and inhibiting MAO activity (17,43). On the other hand, hypofunction of the serotonergic system is thought to play an important role in the pathogenesis of depressive disorders (3,34) and in ethanol abuse (38,42,4,57). However, the results of the present study suggest that inhibition of 5-HT reuptake or inhibition of 5-HT enzymatic degradation is not of importance for reduction of 10% ethanol intake induced by HPE, because this effect was not modified even following subtotal 5-HT depletion caused by 5,7-DHT. On the other hand, 5,7-DHT pretreatment influenced the results obtained in the FST. The antiimmobility effect of HPE was markedly lower in rats pretreated with 5,7-DHT, in comparison to rats with unchanged 5-HT levels. The results obtained in the FST suggest, therefore, that inhibition of 5-HT reuptake or inhibition of 5-HT enzymatic degradation may contribute to the antidepressant-type effect of HPE.

On the other hand, because it has been shown that different components of HPE bind to several subtypes of 5-HT receptors, such as 5-HT_{1A} and 5-HT_{2A} (17,60), 5-$HT_{3/5}$, 5-HT_4 (12), 5-HT_6 and 5-HT_7 (58), we cannot exclude that HPE effects on ethanol intake or/and on immobility in the FST could be mediated by interaction with postsynaptic 5-HT receptors.

Taking into account that serotonergic drugs are well known to influence ethanol intake and depressive disorders, it may seem strange that 5,7-DHT changed neither ethanol intake nor immobility time in the FST. Nevertheless, the present results are according to other studies showing that following 5-HT depletion both immobility time in the FST (10) and ethanol intake in rats with developed ethanol preference (41,56) remain unchanged or even decreased. In regard to the FST, it may be speculated that during the period between 5,7-DHT treatment and the FST (usually 7–10 days) compensatory functional changes of other neuronal systems involved in the control of depressive states (such as the noradrenergic or opioidergic) might take place. In the case of ethanol intake, we may hypothesize a ceiling effect, because msP rats show ethanol preference of about 90%, which can hardly be enhanced. Such a high ethanol preference may result, at least in part, from 5-HT system hypofunction, reported in msP rats (13,48) and in sP rats (19). On the other hand, stimulation of 5-HT activity by fluoxetine markedly reduced ethanol intake in msP rats in the present and in our previous study (14), in keeping with other studies (26,28).

The antidepressant-like action of HPE was completely absent in rats pretreated with the σ receptor antagonist RIM, suggesting that σ receptors play a crucial role in this effect of HPE. This is consistent with reports that hypericin, which has been proposed to be an important component of HPE for its antidepressant activity (6,7,54) binds preferentially to σ receptors (54). This is also consistent with reports that σ receptors may be involved in the action of antidepressant drugs (36,37,54). Which subtype of σ receptor plays a role in the antiimmobility effect of HPE remains to be elucidated, although it has been suggested that σ_1 receptors can be involved in the relief of behavioral despair in the FST (36).

Rimcazole does not alter ethanol intake inhibition produced by HPE. Thus, σ receptors, as well as enhancement of serotonergic transmission, are apparently not important for this effect. At present, we cannot suggest which neurochemical system may mediate the inhibitory effect of HPE on ethanol intake; however, it should be considered that several studies have separately documented that pharmacological manipulations of the central dopaminergic, opioidergic, GABAergic, and glutamatergic mechanisms can influence ethanol consumption (63) and that HPE has been reported to influence these neurochemical systems (6,7,11, 17,40).

In conclusion, the present results confirm that HPE reduces both ethanol intake and the immobility time in the FST in msP rats. Apparently the effect on ethanol intake is not mediated either by an increase in serotonin neurotransmission or by interaction with σ receptors. On the other hand,

the present results suggest that σ receptors may have a crucial role in this effect of HPE in the FST and that also brain serotonergic system may at least in part mediate the antidepressant-like effect of HPE. Several reports have shown comorbidity between depression and ethanol abuse (23,27,39,46,64), and similar changes in the regulation of some central neurotransmitters (35). However, the findings of the present and of the previous study (50) apparently suggest that in alcohol-preferring msP rats the effects of HPE on ethanol intake and on the animals' behavior in the FST are mediated by different neurochemical systems.

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