

## Full Paper

## NMDA Receptor-Antagonistic Properties of Hyperforin, a Constituent of St. John's Wort

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**Abstract.** Extracts of the medicinal plant St. John's wort (*Hypericum perforatum*) are widely used for the treatment of affective disorders. Hyperforin, a constituent of St. John's wort, is known to modulate the release and re-uptake of various neurotransmitters, an action that likely underlies its antidepressive activity. We now report that hyperforin also has *N*-methyl-D-aspartate (NMDA)-antagonistic effects. Hyperforin (10  $\mu$ M) was found to inhibit the NMDA-induced calcium influx into cortical neurons. In rat hippocampal slices, hyperforin inhibited the NMDA-receptor-mediated release of choline from phospholipids. Hyperforin also antagonized the increase of water content in freshly isolated hippocampal slices, and it counteracted, at 3 and 10  $\mu$ M, the increase of water content induced by NMDA. Hyperforin was inactive, however, in two in vivo models of brain edema formation, middle cerebral artery occlusion and water intoxication in mice. In conclusion, hyperforin has NMDA-receptor-antagonistic and potential neuroprotective effects in vitro. This effect may contribute to the therapeutic effectiveness of St. John's wort extracts in some situations, for example, for relapse prevention in alcoholism.

**Keywords:** edema, fura-2, hippocampal slice, middle cerebral artery occlusion, water intoxication

## Introduction

Extracts of the medicinal herb St. John's wort (*Hypericum perforatum*) (SJW) are widely used for the treatment of affective disorders; standardized extracts have been shown to be clinically effective for mild to moderately severe depression (1–3) and may also be useful as anxiolytic agents (4, 5). Experimental and clinical studies identified hyperforin, a lipophilic constituent of SJW with a phloroglucinol structure (Fig. 1), as the major active principle for antidepressant action. Hyperforin is known to inhibit the uptake of aminergic transmitters such as serotonin and noradrenaline into synaptic nerve endings (6, 7). It also increases the extracellular levels of other transmitters including acetylcholine, glutamate, and GABA (8, 9). These effects may be secondary to an increase of the intracellular sodium concentration mediated by openings of

non-selective cation channels in the synaptosomal membrane (10). Finally, hyperforin also interacts with a variety of receptors and ion channels including glutamatergic and GABAergic receptors and calcium channels (11, 12). In the present study, we tested

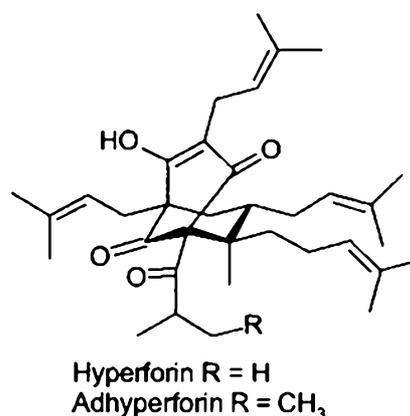


Fig. 1. Structure of hyperforin from *Hypericum perforatum*.

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hyperforin's actions on biological responses induced by *N*-methyl-D-aspartate (NMDA)-receptor activation. We demonstrate that hyperforin blocks NMDA-induced calcium influx, choline release, and edema formation in vitro. These effects, however, were not reflected in neuroprotective activities in models of brain ischemia in vivo.

## Materials and Methods

Hyperforin (sodium salt) was purified from SJW as described (13). Hyperforin solutions were freshly made every day immediately before experimentation.

Fluorescence imaging was carried out in cortical neurons that were prepared from E15–18 rat pups as described (14). Briefly, neurons were dissociated from freshly prepared cortices by mechanical disruption and then seeded onto coverslips precoated with poly-DL-lysine. Cultures were maintained in serum-free (Neurobasal<sup>®</sup>; Gibco/Invitrogen, Carlsbad, CA, USA) medium at 37°C in a humidified atmosphere of 95% air and 5% CO<sub>2</sub>. Experiments were carried out at 9–12 days in vitro. Cultures were washed and then loaded with fura-2 AM (2 μM). After gently removing the fura-2 AM-containing buffer, washing, and equilibrating, the coverslips were placed in a thermostatted (37°C) stage on an Olympus IX70 (Olympus America, Center Valley, PA, USA) inverted microscope. Excitation wavelengths of fura-2 AM were at 340 and 380 nm with emission light monitored at 510 nm. Cell-derived fluorescent images were visualized using a 40×/1.35 oil-immersion objective and were captured by an OlymPix CCD camera (Olympus America). In each experiment 8 to 10 single neurons in the microscopic field were analyzed. Each experiment was repeated at least four times in neuronal preparations from different animals.

For the choline release assay, male Wistar rats (250–350 g; Charles River, Wilmington, MA, USA) were decapitated, and hippocampal slices (400 μm) were prepared as previously described (15, 16) and superfused (0.7 ml/min) at 35°C with Tyrode solution of the following composition: 137 mM NaCl, 2.7 mM KCl, 1.8 mM CaCl<sub>2</sub>, 1.2 mM MgCl<sub>2</sub>, 0.2 mM NaH<sub>2</sub>PO<sub>4</sub>, 11.9 mM NaHCO<sub>3</sub>, and 5.6 mM glucose. All superfusion solutions were continuously gassed with carbogen (95% O<sub>2</sub>, 5% CO<sub>2</sub>). The slices were first incubated with 0.1 mM di-isopropyl-fluorophosphate (DFP) for 30 min in order to prevent choline release from acetylcholine by the action of acetylcholinesterase. Subsequently, the slices were washed for 40 min with magnesium-free Tyrode solution, and basal choline efflux was determined. Then, the superfusion solutions were switched to those containing NMDA and/or

hyperforin in magnesium-free Tyrode solution. The superfusates were collected at 10-min intervals and analyzed for choline content. Choline was determined by a chemoluminescence assay. Briefly, 10-μl aliquots of the superfusates were given to a reaction mixture consisting of 20 mM Tris buffer pH 8.6, 1 μg luminol, 10 μg peroxidase, and 1.25 U choline oxidase, and the chemiluminescence resulting from oxidation of choline to betaine was measured at 425 nm in a LKB-Wallac luminometer (Pharmacia, Freiburg, Germany). The assay was linear from 1–5 pmol choline. The data for choline efflux (Fig. 2) are expressed in % of basal choline efflux which was  $71.6 \pm 5.4$  pmol/min per mg protein (n = 24).

To measure edema formation in vitro, rat hippocampal slices were prepared and superfused with Tyrode solution as described above. During the equilibration period, all superfusion solutions were continuously gassed with carbogen (95% O<sub>2</sub>, 5% CO<sub>2</sub>). When NMDA (300 μM) was used to induce tissue edema, the concentration of magnesium chloride in the Tyrode solution was lowered to 0.12 mM. Hyperforin, when used, was added together with NMDA. Four lanes of slices were superfused in parallel for 30 min. At the end of the superfusion period, slices from each lane were collected, superficially dried, transferred to aluminum foil, and weighed ("wet weight"). They were then dried overnight at 105°C in a desiccating oven and weighed again ("dry weight"). Total tissue brain water was calculated according to  $[(\text{wet weight} - \text{dry weight}) / \text{wet weight}] \times 100$ .

In vivo ischemia in the brain was induced as described in detail previously (17). Briefly, female CD-1 mice (26–32 g, Charles River) were anesthetized with 1% isoflurane in 30% O<sub>2</sub> / 70% N<sub>2</sub>O. The skin was incised, and the left occipital and superior thyroid artery, branches of the external carotid artery (ECA), as well as the pterygopalatine artery, a branch of the internal carotid artery, were exposed, electrocoagulated, and cut. After occlusion of the common carotid artery by a microclip, the left ECA was ligated, coagulated and cut distally to the cranial thyroid artery. A 21-mm monofilament nylon suture (5-0; Harvard Apparatus, Holliston, MA, USA) (diameter of the heat-rounded tip: 0.2–0.3 mm) was inserted into the ECA and gently advanced through the internal carotid artery until its tip occluded the origin of the middle cerebral artery (MCA). Correct placement of the suture was indicated by a sudden drop of the local cortical blood flow in the left MCA territory to 10%–15% of basal flow as monitored by laser-Doppler flowmetry (Moor Instruments, Wilmington, DE, USA). After successful occlusion, the monofilament was secured in place with ligature, and the skin

incision was closed by surgical clips. Throughout surgery, temperature was maintained at 37°C by a thermostatic blanket (rectal thermometer). Hyperforin (or saline for controls) was injected intraperitoneally 60 min before induction of ischemia at a dose of 10 mg/kg. Middle cerebral artery occlusion (MCAO) was sustained for a period of 24 h, after which the animals were deeply anesthetized with isoflurane and euthanized by decapitation. The brains were quickly removed, sectioned coronally into 1-mm slices, and stained with 2,3,5-triphenyl-tetrazolium chloride (TTC). Images were acquired by a digital camera, and areas of both hemispheres and the infarcted regions were quantified for each slice using Image J 1.30 (NIH, Bethesda, MD, USA). Brain edema (brain swelling) were quantified by comparing the area of the ipsilateral (ischemic) hemisphere to the contralateral (non-ischemic) hemisphere, as described previously (17–19).

As another *in vivo*-model of brain edema formation, we used water intoxication in mice (20, 21). Briefly, male CD-1 mice ( $n=32$ ) were given distilled water (20% of body weight) by rapid intraperitoneal infusion. Desmopressin (DDVP) was added in a dose of 3  $\mu\text{g}/\text{kg}$  to prevent renal elimination of excess fluid. Following water intoxication, the mice showed a decrease of spontaneous motility after 15 min which was accompanied by uncoordinated movements. After 30 min, the mice were rapidly anesthetized with isoflurane (4%) in an induction chamber, and decapitated. The brain hemispheres were superficially dried, transferred to aluminum foil, weighed (“wet weight”) and dried over night 105°C in a desiccating oven. The dried slices were weighed again (“dry weight”), and total brain water was calculated according to the following equation:  $[(\text{wet weight} - \text{dry weight}) / \text{wet weight}] \times 100$ .

All animal studies were conducted in accordance with the NIH Guide for the Care and Use of Animals and were approved by the Animal Care and Use Committee at TTUHSC.

## Results

### Fluorescence imaging

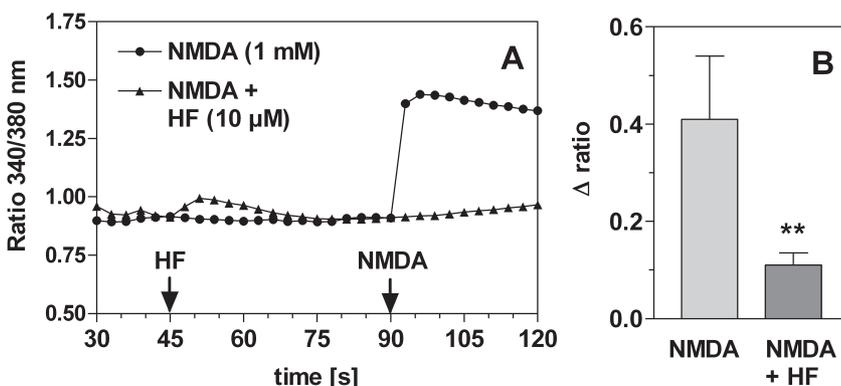
Basal calcium concentrations in primary cortical neurons were  $142 \pm 16$  nM ( $n=6$ ) (for calculation, see ref. 22). Addition of 1 mM NMDA to the neuronal cultures induced a calcium influx as measured by fluorescence microscopy (Fig. 2A). Addition of hyperforin (10  $\mu\text{M}$ ) caused a small increase of the fluorescence ratio, which was probably due to perturbation of the medium (cf. ref. 14). Importantly, hyperforin strongly reduced the NMDA-induced calcium signal (Fig. 2A). In four experiments with 8 to 10 single neurons in the microscopic field in each experiment, the NMDA-induced calcium fluorescence was reduced by an average of 73% in the presence of hyperforin (Fig. 2B).

### NMDA-receptor activation and choline efflux

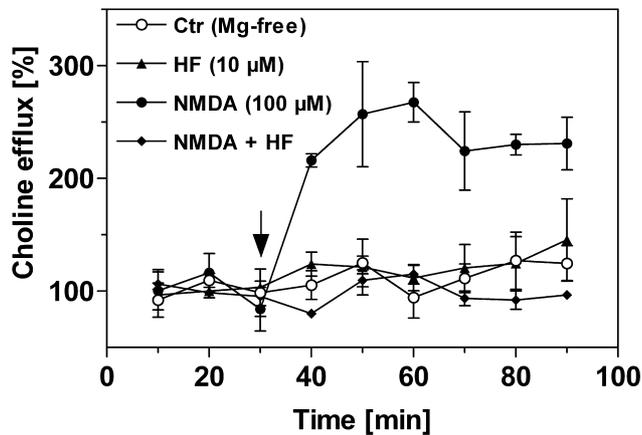
Release of choline upon NMDA-receptor activation reflects hydrolysis of phosphatidylcholine in cellular membranes (23). In experiments on rat hippocampal slices, basal efflux of choline was  $1.5 \pm 0.1$  pmol/10  $\mu\text{l}$  ( $n=24$ ) and was constant throughout the experimental period (Fig. 3). Basal choline efflux was unchanged in the absence or presence of magnesium (data not shown). Upon addition of NMDA (100  $\mu\text{M}$ ) in magnesium-free solution, we observed an immediate release of choline that reached a maximum of  $267 \pm 17\%$  after 30 min (Fig. 3). Hyperforin (10  $\mu\text{M}$ ) did not affect the basal release of choline. The NMDA-induced release of choline, however, was completely suppressed by hyperforin (Fig. 3).

### Brain edema induced by *in vitro* ischemia

To test a potential effect of hyperforin on edema formation *in vitro*, we prepared rat hippocampal slices and superfused them with Tyrode solution (24–26). Slice water contents were  $78.7 \pm 1.6\%$  at the time of



**Fig. 2.** Calcium imaging of rat cortical neuronal cultures preloaded with fura-2 AM. A: Representative experiment demonstrating the effect of NMDA (1 mM) in the absence and presence of hyperforin (10  $\mu\text{M}$ ). B: Effect of hyperforin (HF) on calcium influx induced by NMDA (mean  $\pm$  S.E.M.,  $n=4$  for each compound). \*\* $P<0.01$  vs NMDA (*t*-test). Each  $n$  represents the average of 8–10 single neurons from one preparation.

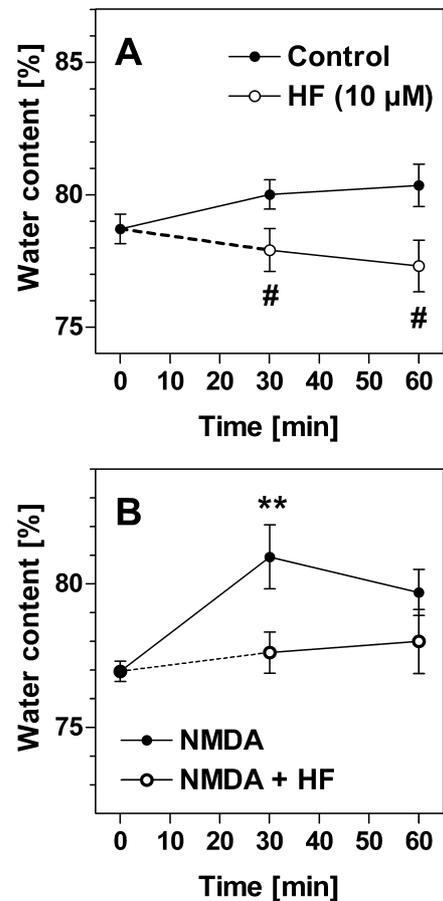


**Fig. 3.** Choline efflux from hippocampal slices evoked by NMDA: inhibition by hyperforin. Hippocampal slices were superfused with Mg-free KHB, and choline efflux was measured using a chemoluminescence assay. NMDA (100  $\mu$ M) was added at 30 min (arrow). Hyperforin, when present, was added together with NMDA. Data are given as relative changes of the basal choline efflux (1.51 pmol per 10  $\mu$ l) and are means  $\pm$  S.E.M. of 5 experiments.

preparation (Fig. 4A). The slices took up water spontaneously during a 60-min period, a process that was inhibited in the presence of hyperforin (Fig. 4A). In a second series of experiments, the slices were exposed to NMDA (300  $\mu$ M) in Tyrode solution containing 0.12 mM of magnesium (Fig. 4B). The excitotoxic agent, NMDA, caused a significant increase of slice water content within 30 min. In the presence of hyperforin (10  $\mu$ M), this increase was almost completely prevented (Fig. 4B). In separate experiments (not illustrated), hyperforin (3  $\mu$ M) also significantly reduced NMDA-induced water gain by 39% ( $P < 0.05$ ), whereas hyperforin (1  $\mu$ M) reduced NMDA-induced increases of slice water contents by less than 5% ( $P > 0.3$ ).

#### Effect of hyperforin in vivo

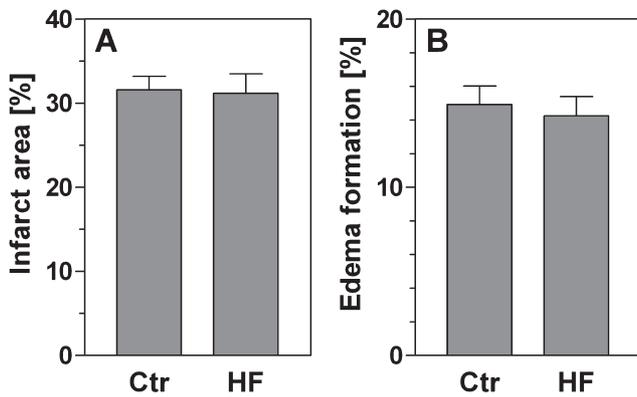
The potential neuroprotective effects of hyperforin were tested in the model of MCAO in the mouse. In this in vivo-model of focal brain ischemia, the infarct area after stroke in control mice was  $31.6 \pm 1.4\%$  ( $n = 8$ , Fig. 5A) as calculated with reference to the total brain area. Pretreatment of the mice with hyperforin (10 mg/kg) did not affect the size of the infarct area ( $31.1 \pm 2.3\%$ ,  $n = 7$ ). Formation of brain edema was calculated by comparing the size of infarcted and contralateral hemisphere ("hemispheric enlargement", see the Materials and Methods section). At 24 h after MCAO, the ischemic side of the brain showed significant swelling compared with the control side which caused a  $14.9 \pm 3.1\%$  increase of the hemispheric area (Fig. 5B). Slices from treated mice (hyperforin,



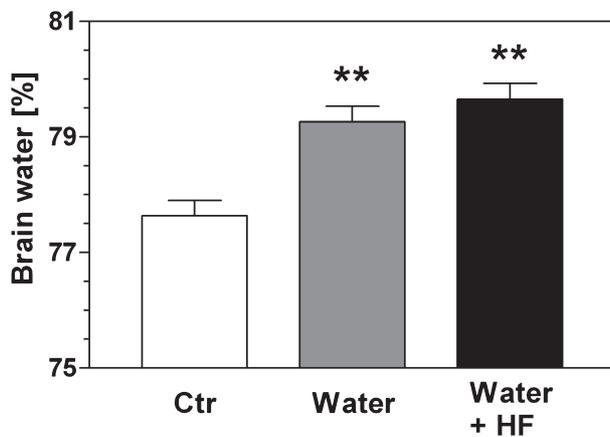
**Fig. 4.** Tissue water content in hippocampal slices. A: Freshly prepared slices were superfused with Tyrode solution ("control") or with Tyrode solution containing hyperforin ("HF", 10  $\mu$ M). B: Hippocampal slices were exposed to NMDA (300  $\mu$ M) in a low-magnesium solution for 30 min; in the data points labeled "HF", hyperforin (10  $\mu$ M) was present in the superfusion solution. Water contents were determined at the end of the superfusion procedure by differential weighing before and after drying the slices. # $P < 0.05$  vs control at same time point ( $t$ -test). \*\* $P < 0.01$  vs control at zero min (ANOVA).  $n = 5 - 8$  for each data point.

10 mg/kg, i.p.) had a similar increase due to swelling ( $14.2 \pm 1.1\%$ ); the difference was not significant versus controls ( $P > 0.2$ ).

To test the effect of hyperforin in another in vivo model of brain edema formation, we used water intoxication in which rapid intraperitoneal water infusion causes hyponatremia and hypoosmolarity in plasma (20, 21). In control mice, brain water content was  $77.6 \pm 0.6\%$  ( $n = 8$ ) (Fig. 6). Water infusion caused signs of toxicity (hunched posture, reduced spontaneous motility) after about 15 min and an increase of brain water to  $79.3 \pm 0.7\%$  (two-way ANOVA,  $P < 0.01$ ,  $n = 8$ ). Pretreatment of the mice with hyperforin (10 mg/kg, i.p.) 30 min before water injection did not affect the development of brain edema; brain water contents



**Fig. 5.** Effects of hyperforin on middle cerebral artery occlusion (MCAO) in the mouse. Infarct area (A) and edema formation (B) in the mouse brain were measured 24 h after MCAO in vehicle controls (Ctr) and animals pretreated with hyperforin (HF; 10 mg/kg, i.p.) 60 min before stroke induction. Infarct area is given as percentage of total brain area. Edema formation is given as relative increase of the brain area in the infarcted hemisphere vs the contralateral hemisphere. Results are mean  $\pm$  S.E.M.,  $n = 7$ .



**Fig. 6.** Water intoxication in mice: effect of hyperforin. Water intoxication was induced by rapid intraperitoneal infusion of distilled water (20% of body weight) plus desmopressin (3  $\mu$ g/kg). Mice were sacrificed after 30 min, and brain water was determined by differential weighing of total brain tissue before and after drying of the brains. Hyperforin (10 mg/kg) was administered 30 min before water intoxication. \*\* $P < 0.01$  vs controls (two-way ANOVA,  $n = 8$  experiments).

in hyperforin-treated mice were  $79.6 \pm 0.7\%$  ( $n = 8$ ) (Fig. 6).

## Discussion

### *Inhibitory effects of hyperforin on calcium fluxes and in the choline release assay*

The central finding of the present study is the ability of hyperforin to antagonize excitotoxic actions of NMDA in three different neurochemical assay proce-

dures of NMDA receptor activation. The study was initiated because of the serendipitous finding that hyperforin almost completely (by  $>70\%$ ) blocked the NMDA-induced calcium influx in rat cortical neurons (Fig. 2). This action resembled previous findings with NMDA-receptor blockers (22), including our observations with memantine that inhibited NMDA-induced calcium influx by 80% (14).

We then proceeded to an NMDA-receptor-mediated effect of pathological relevance, namely, the breakdown of membrane phospholipids (Fig. 3). NMDA-induced choline release from hippocampal slices reflects NMDA-receptor activation and is dependent on calcium influx and activation of phospholipase  $A_2$  as previously shown by our laboratory (15). In earlier work, NMDA-induced choline release was highly sensitive to NMDA-receptor antagonists such as MK-801 and HA-966 (16). In the present study, hyperforin did not affect basal choline release, but completely suppressed the release that was evoked by NMDA in magnesium-free buffer (Fig. 3). Hyperforin, therefore, prevents phospholipid hydrolysis and membrane breakdown under conditions of excitotoxicity, presumably by an action on the NMDA receptor.

### *Effects of hyperforin on edema formation in vitro*

In a second model of cellular injury, we measured tissue water content in hippocampal slices as an indicator of cellular edema formation. Hippocampal slices were previously introduced as an *in vitro* model of brain edema formation (24–26). Preparation of hippocampal slices is known to cause a swelling of slices over 30–60 min, which is accompanied by sodium and calcium uptake (24). In studies employing ischemia and reperfusion in hippocampal slices, increases of sodium and calcium uptake were observed that were attenuated by blockers of voltage-operated cation channels as well as glutamate-receptor antagonists (25, 26). NMDA-receptor antagonists, in particular, caused a partial (26) or almost complete (25) protection of edema formation in these studies.

In our hands, and in agreement with an earlier study (24), superfusion of freshly prepared slices was accompanied by a small but visible increase of water contents during 30–60 min (Fig. 4A). This increase of water content that reflects cytotoxic edema formation in freshly cut tissue, was prevented by hyperforin (Fig. 4A). We then found that exposure of slices to NMDA causes a similar extent of edema formation as the ischemic conditions used in earlier studies (24, 25). When slices were exposed to NMDA for 30 min, edema formation was reflected in an increase of water contents by 3%–4% (Fig. 4B). Hyperforin strongly attenuated

this response; the reduction of NMDA-induced edema formation was 82% at 10  $\mu$ M, 39% at 3  $\mu$ M, but less than 5% at 1  $\mu$ M of hyperforin. Thus, hyperforin acts against NMDA-induced toxicity in low micromolar concentrations.

The mechanism by which hyperforin interferes with NMDA-receptor-mediated signaling is a matter of speculation. As a derivative of phloroglucinol, hyperforin is redox active, and extracts of SJW are known to possess pro- and antioxidative properties (27). We speculate, therefore, that hyperforin may affect the well-known redox regulation of the NMDA receptor (28). In preliminary experiments, phloroglucin (100  $\mu$ M) showed similar activity as hyperforin in the slice edema assay, indicating that hyperforin may act as a lipophilic derivative of phloroglucinol (V. Kumar, unpublished observations), but further work is required to test this hypothesis.

#### *Lack of effects of hyperforin in vivo*

We tested hyperforin in two in vivo paradigms, a stroke model and a brain edema model. MCAO is a standard model to induce focal brain ischemia; when used in the mouse, it causes the formation of a large infarct area in cortex and striatum, with some damage also observed in the hippocampus (17). NMDA-receptor antagonists strongly suppress neuronal death after MCAO (17, 29). In the present experiments, pretreatment of the animals with hyperforin (10 mg/kg) did not affect the infarct areas measured in slices of mouse brain (Fig. 5). Hyperforin was also inactive in preventing the formation of tissue edema that is a consequence of stroke and which was measured in this study as an increase of hemispheric area (see ref. 17 for details).

We proceeded to test hyperforin in a second model of brain edema, a water intoxication model in which the infusion of a large volume of distilled water, combined with an antidiuretic vasopressin derivative, causes hyponatremia and a lowering of plasma osmolarity. As a consequence of the osmotic gradient, water enters the brain and produces a cellular (cytotoxic) edema without disrupting the blood-brain barrier. We applied this paradigm because hyperforin actually reduced water contents in superfused slices below control level (although the effect did not reach statistical significance, Fig. 4A), and we mused that hyperforin may interact with aquaporin channels, the molecular entities responsible for water crossing the membrane. Indeed, brain water uptake following water intoxication has recently been shown to depend on aquaporin-4 (20), while a role of NMDA receptors is unlikely in this assay, and to our knowledge has not been tested. In other words, we tested hyperforin in this model because blockade of aquaporins

would be an *alternative* mechanism of action for hyperforin's anti-edema effects in brain slices. The experimental results, however, did not support a blocking action of hyperforin on water transport as hyperforin was unable to inhibit the increase of brain water observed after water intoxication (Fig. 6). Therefore, NMDA antagonism remains the major hypothesis for hyperforin's anti-edema actions in the brain slice.

Summarizing, our findings indicate that hyperforin antagonizes NMDA-receptor-mediated responses in the low micromolar range. Its potency in the slice assay was similar to the inhibition of NMDA currents observed in hippocampal pyramidal neurons ( $IC_{50} = 3.2 \mu$ M). In a similar concentration range, hyperforin has been shown to inhibit AMPA receptors ( $IC_{50} = 4.6 \mu$ M), while inhibition of neuronal voltage-dependent calcium channels of the P-type was observed in the submicromolar range (11, 12). In contrast, N- and L-type calcium channels, as well as sodium channels, were not significantly affected in the presence of hyperforin (30). NMDA-receptor antagonism remains the most likely mechanism of action for hyperforin's anti-edema effects in vitro.

#### *Therapeutic relevance*

The pharmacological relevance of NMDA-receptor channel blockade relates to the prominent role of NMDA-receptor activation in excitotoxicity associated with ischemia, stroke and traumatic brain injury (31–33). The present results show that hyperforin, a constituent of SJW, antagonizes NMDA-receptor-mediated responses in the low micromolar range (Figs. 2–4). This action likely contributes to the previously described neuroprotective properties of SJW extracts, for example, those reported in neuronal cell cultures exposed to hydrogen peroxide (34, 35). NMDA-antagonistic activity may also be beneficial in a chronic neurodegenerative disease such as Alzheimer's disease (36). Hyperforin was, however, not sufficiently active to reduce edema formation or cell death in vivo (Figs. 5 and 6). Thus, our results do not support a therapeutically relevant neuroprotection by hyperforin in brain ischemia. It should be noted, however, that other constituents of SJW extracts may have neuroprotective activities (37).

One possible explanation for hyperforin's lack of effect in the in vivo-models is that the compound, given systemically, does not reach a sufficient concentration in the brain to effectively block NMDA receptors. Hyperforin is clearly brain-permeable as illustrated by a myriad of behavioral effects of the compound (6, 7). Recently, LC-MS was applied to measure hyperforin in the brain of mice dosed orally with 15 mg/kg of pure hyperforin (38, 39). The authors report a hyperforin

brain level of 28 ng/g wet wt., which corresponds to a concentration of 0.1  $\mu$ M hyperforin if it were distributed homogeneously in brain water. This situation is unlikely, however; hyperforin is a highly lipophilic compound that is expected to accumulate in brain membranes and may reach much higher concentrations in the vicinity of its targets (e.g., amine transporters, ion channels, or NMDA receptors). For the present study, we cannot rule out insufficient brain levels of hyperforin as an explanation for its lack of effect *in vivo*.

Can there be additional therapeutic relevance of NMDA-receptor antagonism in the therapeutic context of SJW extracts? Recent work has suggested that SJW extracts are effective for relapse prevention of alcoholism. SJW extracts reduce alcohol intake in alcohol-preferring rats (40–42), and one study has suggested that hyperforin may be the active principle for this effect (43). Interestingly, the effects of SJW extracts on alcohol intake were found to be independent of serotonergic or GABAergic mechanisms (44). Acamprosate, an NMDA-receptor antagonist, is an FDA-approved drug to prevent relapse in alcoholic patients. As hyperforin and hyperforin-rich SJW extracts evidently have antidepressant as well as NMDA-receptor-antagonistic properties, they combine two useful therapeutic principles for the treatment of alcoholism, and further investigations in this area may be warranted.

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