J Pharmacol Sci **124**, 409 – 417 (2014)

Journal of Pharmacological Sciences

Full Paper

PKC-Mediated Potentiation of Morphine Analgesia by St. John's Wort in Rodents and Humans

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Received November 6, 2013; Accepted January 7, 2014

Abstract. Our purpose was to combine the use of morphine with clinically available inhibitors of protein kinase C (PKC), finally potentiating morphine analgesia in humans. Thermal tests were performed in rodents and humans previously administered with acute or chronic morphine combined or not with increasing doses of the PKC-blocker St. John's Wort (SJW) or its main component hypericin. Phosphorylation of the γ subunit of PKC enzyme was assayed by western blotting in the periaqueductal grey matter (PAG) from rodents co-administered with morphine and hypericin and was prevented in rodent PAG by SJW or hypericin co-administration with morphine, inducing a potentiation of morphine analgesia in thermal pain. The score of pain assessment in healthy volunteers were decreased by 40% when morphine was co-administered with SJW at a dose largely below those used to obtain an antidepressant or analgesic effect in both rodents and humans. The SJW/hypericin potentiating effect lasted in time and preserved morphine analgesia in tolerant mice. Our findings indicate that, in clinical practice, SJW could reduce the dose of morphine obtaining the same analgesic effect. Therefore, SJW and one of its main components, hypericin, appear ideal to potentiate morphine-induced analgesia.

Keywords: morphine, St. John's wort, protein kinase C, thermal nociception

Introduction

Morphine currently constitutes the cornerstone therapy for moderate-to-severe pain of many etiologies. Despite the high analgesic potency of the alkaloid, the number of well-known side effects that limit its use are equally important. One of the more controversial aspects relates to uncertainties about adaptations to chronic use, which include concerns over tolerance, hyperalgesia, and dependence, thus limiting morphine clinical potential. Morphine tolerance is a pharmacological phenomenon characterized by a shift of the dose–response curve to the right with a larger 50% effective dose. The conventional practice of opioid therapy in the presence of diminishing analgesic efficacy is based on dose-escalation in order to restore analgesic effect. Thus, several approaches combining other drugs with opioids to increase their potency and thus reduce the opioid doses, have been proposed. One of these is to administer an adjuvant drug with synergistic analgesic effects in order to minimize the dose of the opioid while maintaining acceptable levels of analgesia as combination of non-steroidal anti-inflammatory drugs, anticonvulsants, or sodium-channel blockers with opioids (1, 2). The administration of α2 (3), 5-HT$_7$ (4) agonists, NMDA (5), or σ antagonists (6) can potentiate morphine analgesia. However, clinical trials have sometimes found contradictory results and this approach may have some adverse clinical implications (7). Co-administration of NMDA-receptor antagonists such as ketamine resulted in the attenuation of the chronic opioid effect in animals, although clinical trials gave mixed results (7, 8).

We could previously demonstrate in rats that blockade of protein kinase C (PKC) potentiates morphine analge-

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Published online in J-STAGE
doi: 10.1254/jphs.13226FP

sia (9). In these experiments, the downregulation of the γ subunit of PKC (PKCγ) could induce a dramatic potentiation in morphine induced analgesic effect, which was observable at the earlier time and lasted longer. With these indications in mind, we have focused our attention on hypericin and pseudohypericin as potentiating agents of morphine analgesia (Italian Patent RM2006A00016). These compounds have been isolated from plants of the hypericum family. *Hypericum perforatum*, commonly called St. John's Wort (SJW), has been used for centuries as a medicinal plant. Dioscorides, the physician most important in ancient Greece, as well as Pliny in Rome and Hippocrates, were administering the herb in the treatment of many diseases. In folk medicine, SJW has always been used in the treatment of wounds, kidney, and lung diseases and in what we now call depression. Although pharmacological studies on SJW have focused on its antidepressant activity in mild depression, some studies have documented that the compound can act as PKC blocker (10), competitively binding to the regulatory domain of PKC (11). In the present study we examine the potentiating effect of SJW dried extract and its active constituents on the morphine-induced analgesic effect in rodents with the aim to establish the dose and then translate the results to humans.

Materials and Methods

Animals

Adult male Swiss albino mice and rats from the Morini (San Polo d'Enza, Italy) breeding farm were used. Ten mice and four rats were housed per cage, which were placed in the experimental room 24 h before the test for acclimatization. The animals were fed a standard laboratory diet and tap water ad libitum and kept at 23°C ± 1°C with a 12-h light/dark cycle. All experiments were carried out in accordance with the European Communities Council Directive of 24 November 1986 (86/609/EEC). All studies involving animals are reported in accordance with the ARRIVE guidelines for experiments involving animals (12). A total of 170 animals were used in these experiments. In order to reduce as much as possible the number of animals used, sample size criteria as regard to statistical significance were followed as set out in Feinstein (13). Animals were investigated by observers kept unaware of the treatment of the animals.

Study protocol in healthy volunteers

The study protocol was approved by the appropriate institutional ethics committee and health authorities of the Italian National Health Service and was conducted in accordance with the 1964 Declaration of Helsinki and Good Clinical Practice Guidelines. Eight male healthy asymptomatic volunteers between the ages of 25 and 35 years were recruited among the staff. They reported that they did not consume drugs. Furthermore, subjects were asked not to undertake physical exertion and to refrain from consuming caffeine, alcohol, or nicotine on the days of the experiment. All subjects gave informed consent prior to study commencement. Before their involvement in the study, the participants were provided with an information leaflet explaining the procedure. Possible risks such as redness after the immersion, the guarantee on anonymity, and the possibility to stop study participation, were mentioned.

Healthy volunteers were given a morphine oral solution (10 mg/5 ml) (Oramorph; MOLTENI & C. F.LLI ALITTI SpA, Florence, Italy) in the presence or absence of SJW (300 mg) tablets containing 0.3% hyperycin (Quiens; Mario Antonetto Farmaceutici, Torino; AIC No. 034870028). The dose of SJW administered to volunteers was obtained according to a study in rodents and was normalized to kg considering an individual weighting 70 kg.

Immersion procedure

A session consisted of the gradual immersion of the dominant arm in circulating noxious hot water (46°C). Water temperature was maintained at 46°C using a thermostat and was constantly monitored with a digital thermometer. The water temperature of 46°C was specifically chosen in accordance with the study of Staud et al. (14). The subjects, after a period of acclimatization in the test room, were asked to immerse the forearm including elbow of his arm in circulating nociceptive hot water for 2 min and to rate the intensity and unpleasantness of pain perceived every 30 s during that time. Perceived pain was rated with a score between 0, indicating no pain at all, and 100 intolerable pain. Morphine was administered 30 min before starting of the immersion procedure according to morphine pharmacokinetic/pharmacodynamic studies after oral administration (15). SJW was administered 60 min before morphine.

Drug administration to rodents

Animals were randomly assigned to treatment groups. SJW dried extract containing 0.32% of total hypericin (Indena Research Laboratories, Milan, Italy), hyperforin, hyperoside, quercetin, and amentoflavone (Sigma, Milan, Italy) were dissolved in 1% carboxymethylcellulose solution immediately before use and administered by oral gavage. The doses of hypericin (0.016 mg/kg), hyperforin (0.21 mg/kg), quercetin (0.0415 mg/kg), amentoflavone (0.0029 mg/kg), and hyperoside (0.3175 mg/kg) approximate the amount of each component

present in a 5 mg/kg preparation of SJW dried extract. SJW and components were administered 60 min before morphine (morphine hydrochloride; SALARS, Como, Italy) according to a previous established protocol (22). Morphine was administered s.c. to rodents at a dose of 5 and 7 mg/kg. Hypericin (0.15 μg per mouse) and calphostin C were administered intracerebroventricularly (i.c.v.) immediately before morphine. Rats were administered intra–periaqueductal grey matter (PAG) as previously reported (16) with hypericin (0.4 μg per rat). The PKC-blocker calphostin C (Calbiochem, Milan, Italy) was dissolved in 0.5% DMSO and used at 0.2 μg per mouse. Vehicles used to dissolve drugs were tested for the absence of any effect on pain threshold in comparison with saline-treated and naïve mice.

Morphine nociceptive tolerance

Morphine tolerance was induced in mice with a 4-day scheme. The animals received 2 daily injections of morphine at 10:00 am and 8:00 pm. The individual doses were 10 mg/kg s.c. on day 1, 15 mg/kg s.c. on day 2, 20 mg/kg s.c. on day 3, and 30 mg/kg s.c. on day 4 according to a previous validated scheme (17).

Hot plate

Mice were placed inside a stainless steel container, which was set thermostatically at 52.5°C ± 0.1°C in a precision water-bath from KW Mechanical Workshop, Siena, Italy. We used a lower temperature (52.5°C instead of 54°C) to reveal potential, subtle alterations that may occur in basal thermal nociception. Reaction times (s) were measured with a stopwatch before and after administration of the investigated compounds. The endpoint used was the licking of the fore or hind paws. An arbitrary cut-off time of 60 s was adopted.

Western blot analysis

PAG samples from mice were homogenized in an homogenization buffer containing 25 mM Tris-HCl (pH 7.5), 25 mM NaCl, 5 mM EGTA, 2.5 mM EDTA, 2 mM NaPP, 4 mM PNFF, 1 mM Na_3VO_4, 1 mM PMSF, 20 μg/ml leupeptin, 50 μg/ml aprotinin, and 0.1% SDS. The homogenate was centrifuged at 9,000 × *g* for 15 min at 4°C, and the low speed pellet was discarded. Protein concentration was quantified using Bradford's method (protein assay kit; Bio-Rad Laboratories, Milan, Italy). Membrane homogenates (10 μg) were separated on 10% SDS-PAGE and transferred onto nitrocellulose membranes (90 min at 120 V) using standard procedures. Membranes were blocked in PBST (PBS containing 0.1% Tween) containing 5% nonfat dry milk for 120 min. Following washings, blots were incubated overnight at 4°C with specific antibodies against PKCγ phosphorylated on Thr514 (pPKCγ, 1:1000 dilution) (Biosource, Camarillo, CA, USA); β-actin (1:1000 dilution) (Santa Cruz Biothechnology, Inc., CA, USA). After being washed with PBS containing 0.1% Tween, the nitrocellulose membrane was incubated with goat anti-rabbit horseradish peroxidase–conjugated secondary antisera (1:10,000) and left for 1 h at room temperature. Blots were then extensively washed according to the manufacturer's instruction and developed using an enhanced chemiluminescence detection system (Pierce, Milan, Italy). Exposition and developing time used was standardized for all the blots. Optical density measurements were performed by dividing the intensity of the bands by the intensity of the house-keeping protein β-actin, used as loading control. Measurements in control samples were assigned a relative value of 100%.

Statistical analyses

All experimental results are given as the mean ± S.E.M. One-way and two-way analysis of variance, followed, respectively, by the Tukey and Bonferroni post hoc test, were used for statistical analyses.

Results

SJW dose study in rodents

In the mouse hot plate test, morphine showed antinociceptive properties starting from the dose of 5 mg/kg, whereas lower doses were ineffective. The morphine-induced antinociceptive effect reached its maximum at 30 min after morphine administration, and thermal nociceptive thresholds returned to baseline values 60 min post-drug injection (Fig. 1A). Morphine elicited a peak antinociceptive response at 30 min post-drug injection, and thermal nociceptive thresholds returned to baseline values by 60 min post-drug injection (Fig. 1A). SJW was endowed with antinociceptive properties as well. Oral administration of a 10 mg/kg dose of SJW increased the pain threshold 60 min after injection, peaked at 90 min, and was still significant after 120 min. Lower doses were devoid of any effect (Fig. 1B). The co-injection of morphine with an ineffective SJW dose (1 mg/kg) potentiated morphine antinociceptive activity. SJW increased the licking latency values following the administration of an ineffective dose of morphine (2 mg/kg) (Fig. 1C) and potentiated the antinociceptive effect induced by morphine at 5 mg/kg (Fig. 1D). The response of the combination of morphine with SJW remained elevated significantly above that produced by morphine alone until the end of the test period. A shift to the left of the dose–response curve of morphine was observed after co-administration with a low dose of SJW, drastically reducing the dose of morphine necessary to

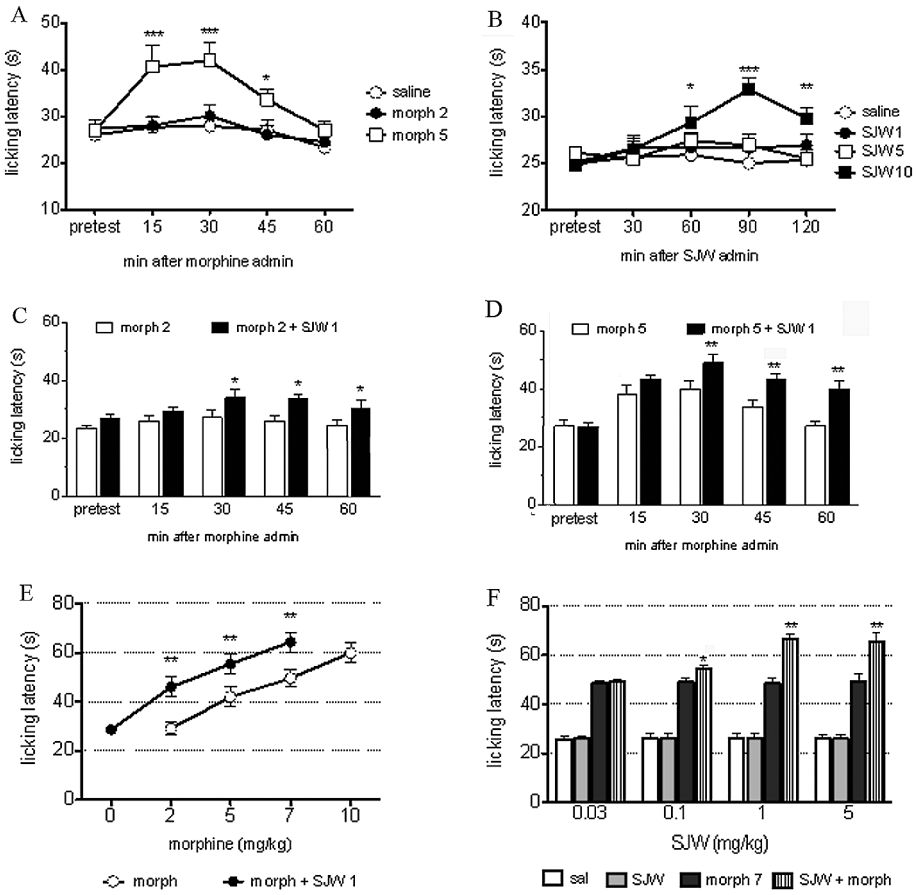

Fig. 1. Low doses of SJW increased morphine antinociception in the hot plate test. Antinociceptive activity of morphine [A: $F(2,135)49.19$, $P < 0.0001$] and SJW [B: $F(3,179)27.67$, $P < 0.0001$]; significant difference is presented as difference from the saline group at the corresponding time point: $^{*}P < 0.05$, $^{**}P < 0.01$, $^{***}P < 0.001$, compared with the treated group. In panel C, an ineffective dose of SJW (1 mg/kg) rendered effective a dose of morphine (2 mg/kg) devoid of antinociceptive properties [$F(1,90)41.97$, $P < 0.0001$] and increased the antinociception induced by a morphine (5 mg/kg) effective dose [D: $F(1,90)42.78$, $P < 0.0001$]. Shift to the left of the dose–response curve of morphine by a very low dose of SJW is shown in panel E [$F(1,99)141.30$, $P < 0.0001$]. Dose–response curve of the morphine potentiating effect of SJW is represented in panel F [$F(3,144)662.30$, $P < 0.0001$]. Nociceptive testing in panels E and F was performed 30 min after morphine administration. Post hoc comparison in C – F: $^{*}P < 0.05$, $^{**}P < 0.01$, $^{***}P < 0.001$ vs. corresponding morphine-treated group.

reach the maximal antinociceptive effect (Fig. 1E). Dose–response experiments showed that lower doses of SJW (0.03 mg/kg) were unable to modify morphine antinociception and that no further potentiating activity was detected by increasing the SJW concentration (5 mg/kg) (Fig. 1F).

Co-administration of morphine and SJW did not alter motor coordination and spontaneous mobility (data not shown).

Human test

Eight health volunteers immersed forearm in circulating nociceptive hot water for 2 min and rated the intensity and unpleasantness of perceived pain. The mean pain intensities of all scores, registered every 30 s during 2-min arm immersion, are shown in Fig. 2 for each subject after morphine administration in presence or absence of SJW at low dose. The score was assessed on different days for the baseline, treatment with morphine alone and morphine co-administration with SJW. A post hoc power analysis was performed for the significant differences in pain scores. Total mean pain intensity score

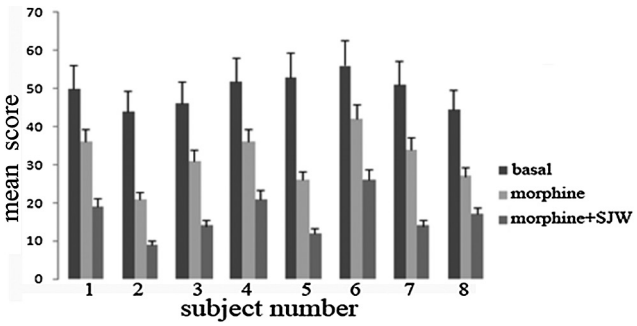

Fig. 2. Reported pain intensities in morphine-treated subjects co-administered with SJW. Mean pain score from repeated assessment of nociceptive response are reported for each single evaluated healthy volunteer as regards different treatment (no treatment, morphine, morphine/SJW co-treatment). One-way ANOVA for repeated measures yielded a significant main effect for morphine co-treatment with SJW as compared to morphine alone in decreasing the nociceptive score [$F(2, 24)21.7$, $P < 0.0001$].

was significantly different between single morphine and morphine co-administration with SJW indicating a better score when healthy subjects were administered with morphine in the presence of SJW.

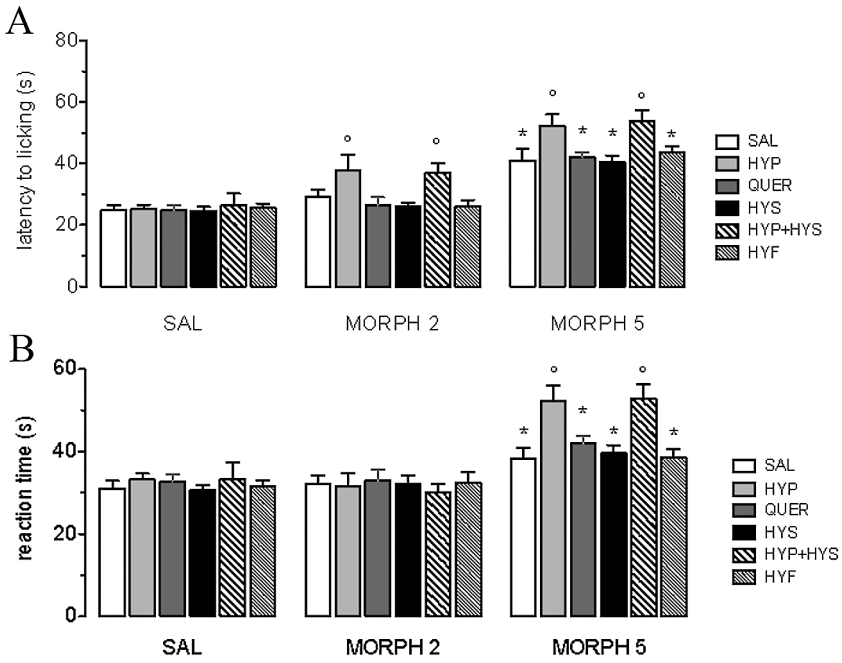

Fig. 3. Effect of oral administration of SJW components on morphine antinociception. In the hot plate [A: F(5,59)3.18, $P < 0.0001$] and in the cold plate [B: F(5,59)6.99, $P < 0.0001$] tests, hypericin (HYP) showed a potentiating profile similar to SJW. Conversely, quercetin (QUER), hyperoside (HYS), and hyperforin (HYF) were devoid of any effect. Nociceptive testing was performed 30 min after morphine injection. Post hoc Tukey test: $*P < 0.001$, compared to saline-treated mice. $°P < 0.001$ vs. morphine-treated group at the corresponding time point.

Differential involvement of SJW components in the potentiation of morphine analgesic efficacy

We investigated the effects produced by some of the main components of this herbal drug administered in a concentration corresponding to the content present in a 5 mg/kg preparation of SJW. The administration of hypericin augmented the morphine effect with a profile similar to SJW (Fig. 3A). Conversely, oral administration of the flavonoids quercetin, hyperoside, or hyperforin was devoid of any effect (Fig. 3A). Co-administration of morphine with hypericin and hyperoside did not modify the antinociceptive potentiation induced by morphine with hypericine alone (Fig. 3: A, B). Similar results were obtained in the cold plate test (Fig. 3B).

Potentiation of acute morphine thermal antinociception by hypericin component of SJW

Intracerebroventricular co-administration of hypericin to mice rendered antinociceptive an ineffective dose of morphine (2 mg/kg, Fig. 4A) and enhanced the thermal antinociception induced by morphine at antinociceptive doses of 5 mg/kg (Fig. 4B) and 7 mg/kg (Fig. 4C) in the hot plate test. Hypericin alone was devoid of any effect (Fig. 4D).

SJW and hypericin affect PKC phosphorylation

We conducted western blotting experiments *ex vivo* in the PAG area to detect how morphine co-administration with the SJW component hypericin affects the expression of the PKC isoform mainly involved in morphine potentiation. Phosphorylation of PKCγ was increased in rat PAG at 30 min after i.p. (intraperitoneally) morphine administration (5 mg/kg). Co-administration of intra-PAG hypericin with morphine completely reversed the pPKCγ overexpression in rat PAG (Fig. 5A). A similar result could be obtained by injecting calphostin C, a selective PKC-blocker, into rat PAG (Fig. 5A). Intra-PAG injection of hypericin combined with morphine at 5 mg/kg produced a thermal antinociceptive response largely above that produced by the alkaloid alone (Fig. 5B). Co-administration of intra-PAG calphostin C potentiated morphine antinociception with an antinociceptive profile similar to the hypericin one (Fig. 5B). CC and SJW alone did not induce any change on PKCγ phosphorylation (data not shown).

Effect of SJW on chronic morphine tolerance

The antinociceptive responses produced by increasing doses of morphine, administered twice daily for 4 days, in the hot plate test are shown in Fig. 6A. The administration of morphine on day 1 produced a maximal antinociceptive effect, but successive daily injections of morphine resulted in a progressive loss of the drug-induced antinociceptive response, which approached baseline value on day 4. Daily administration of hypericin alone produced a response comparable with that produced by saline (data not shown). Figure 6B illustrates the time course of the antinociceptive response on day 5. As shown, morphine did not produce any significant antinociceptive effect in comparison to the saline-treated control group. In sharp contrast, the response to the hypericin–morphine combination could maintain the

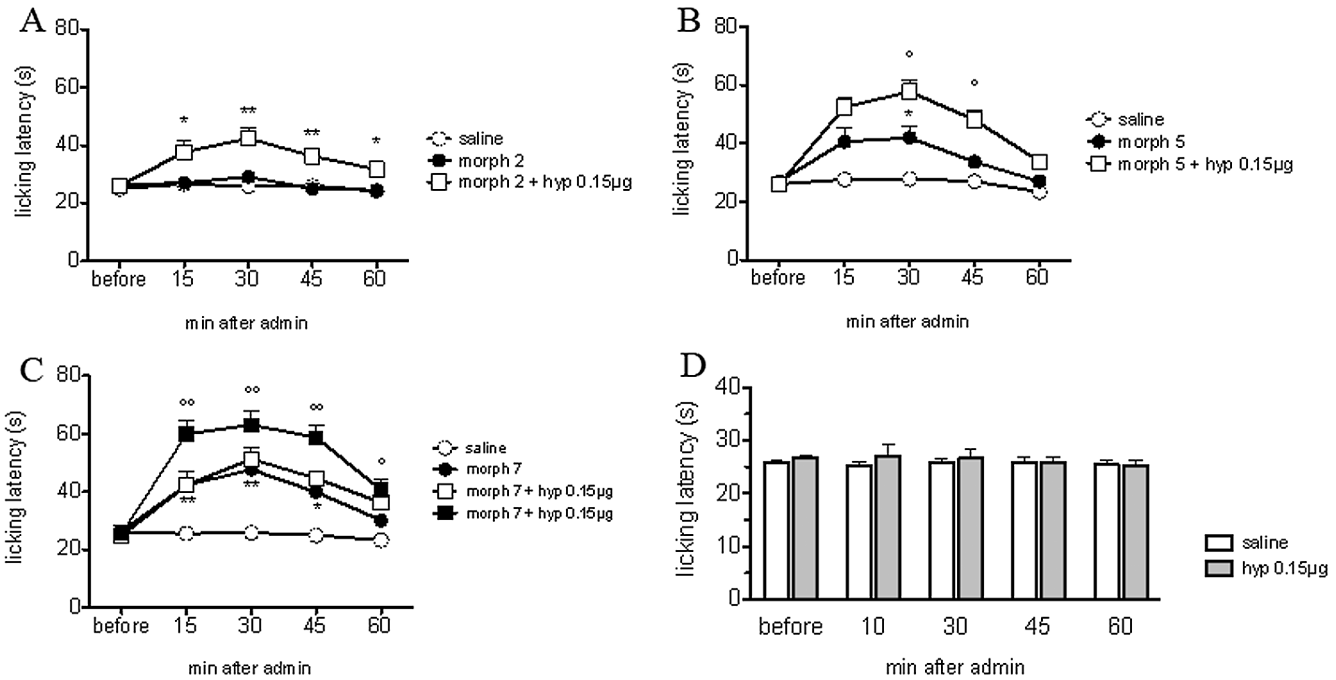

Fig. 4. Potentiating effect of i.c.v.-injected hypericin on morphine antinociception. A: i.c.v. administration of hypericin (hyp, 0.15 μg per mouse) rendered antinociceptive an ineffective dose of morphine (2 mg/kg) [F(3,180)27.98, $P < 0.001$]. Post hoc comparison: *$P < 0.05$, **$P < 0.01$ vs. morphine-treated group at the corresponding time point. B, C: i.c.v. hypericin potentiated the antinociception induced by morphine at a dose of 5 mg/kg [F(3,180)11.6, $P < 0.001$] and 7 mg/kg [F(4,225)103.8, $P < 0.001$]. Post hoc comparison: *$P < 0.05$, **$P < 0.01$ vs. saline; °$P < 0.05$, °°$P < 0.01$ vs. morphine-treated group at the corresponding time point. D: hypericin alone was devoid of any effect on mouse pain threshold.

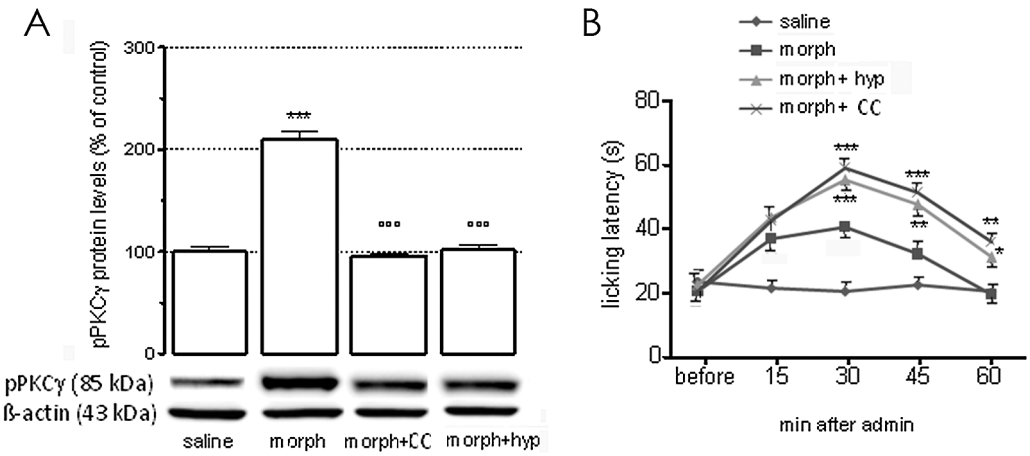

Fig. 5. Effect of hypericin on morphine induced PKCγ phosphorylation. Western blotting experiments performed in the PAG area showed that intra-PAG hypericin (hyp, 0.4 μg) prevented the increased phosphorylation of PKCγ induced by i.p. morphine (morph) administration at 5 mg/kg dose (A). Post hoc comparison: ***$P < 0.001$ vs. saline-treated group, °°°$P < 0.001$ vs. morphine-treated group. In panel B, intra-PAG hypericin and calphostin C (CC) potentiated the thermal antinociception induced by morphine [F(3,180)29.4, $P < 0.001$]. Post hoc comparison: *$P < 0.05$, **$P < 0.01$, ***$P < 0.001$ vs. morphine-treated group at the corresponding time point.

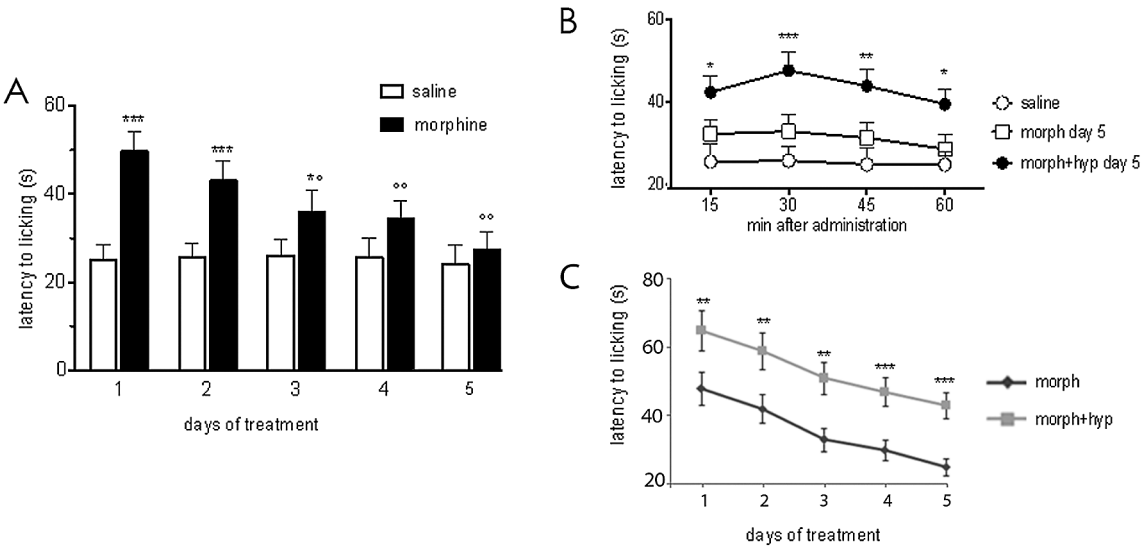

Fig. 6. Effect of hypericin on the chronic antinociceptive effect of morphine in the hot plate test. A) Time course of the antinociceptive effect of repeated administration of morphine delivered twice daily [$F(1,72)446.37$, $P < 0.0001$]. Nociceptive testing was performed 30 min after each injection. $^{*}P < 0.05$, $^{***}P < 0.001$ vs. saline-treated group; $^{\circ}P < 0.05$, $^{\circ\circ}P < 0.01$ vs. morphine effect on day 1. Time-course effect of morphine-saline and morphine-hypericin combination on day 5 (B) and is shown for each of the 5 days (at time 30 min) in animals that had received chronic treatment with these drugs [$F(2,108)119.74$, $P < 0.0001$]. Significant difference from morphine at the same time point: $^{*}P < 0.05$, $^{**}P < 0.01$, $^{***}P < 0.001$; morph = morphine, hyp = hypericin.

antinociceptive effect of morphine as shown by licking latency values comparable to those obtained by morphine acute administration (Fig. 6C).

Discussion

Our findings show that SJW increases the antinociceptive effect of morphine in both naïve and morphine-tolerant animals. As an experimental model to measure the algogenic threshold, we chose the hot plate test that produces pain without inflammation. Nociception was evaluated considering the reaction time of the animal to a thermal stimulus. A significant increase of the licking latency values could be observed after morphine co-administration with a low dose of SJW in mice submitted to this test, drastically reducing the dose of morphine necessary to reach the maximal antinociceptive effect. When SJW low dose was co-administered with repeated treatment with morphine, the antinociceptive response to morphine was not modified in the tolerant animals, establishing that SJW could decrease opioid requirement in animals exhibiting chronic nociceptive tolerance to the alkaloid. These results could be translated to humans. Consistent with ethical requirements and therefore limited to the acute administration, SJW was co-administered with the morphine analgesic dose to a small sample of healthy volunteers at the same low dose used in animal experiments. The efficacy of pain inhibition was evaluated by heat stimuli, and the results showed that efficacy was significantly increased when healthy subjects were treated with morphine in the presence of SJW as compared to morphine alone. These results indicate that in clinical practice, SJW could reduce the dose of morphine while obtaining the same analgesic effect.

SJW dried extract contains numerous active components (18). The effects produced by the main constituents were investigated in order to identify the component responsible for the potentiation of morphine antinociception. The doses used were estimated based on the percentages of each single constituent usually present in the SJW dose administered in our experiments. Among the numerous SJW components, only purified hypericin increased morphine antinociception in both animal models of acute thermal nociception and in nociceptive tolerance with an efficacy and time course similar to that of SJW. Conversely, hypericin alone was devoid of any effect. We here report the morphine-potentiating activity of hypericin, suggesting it as the component of SJW that underlies morphine analgesia potentiation.

Notably, in our experiments, relatively low dose of SJW significantly potentiated the analgesic effect of morphine in rodents and humans. It has been reported that tricyclic antidepressant drugs possess clinical efficacy in the treatment of chronic pain states such as

adjuvant analgesic. However, it would not be rational that SJW enhances the analgesic effect of morphine by having an antidepressant effect because the doses used in these experiments would not be sufficient to exert antidepressant action in both rodents and humans (19). SJW is also endowed with antinociceptive properties in animal models (20). The potentiating effect shown by this herbal medicine was not secondary to its antinociceptive activity, since the increase of the morphine analgesic effect was produced at doses devoid of any capability to modulate the pain threshold in rodents.

It has been reported that SJW increases the activity of cytochrome P450 3A4 (CYP3A4) enzyme (21). Several reports have documented decreased blood/plasma levels of CYP3A4 substrates, such as indinavir and cyclosporin A, in patients concomitantly taking SJW, finally reducing the drug effect considerably. In a previous study, the concentrations of morphine in plasma and brain after s.c. injection of morphine in mice pretreated with SJW were not significantly different from those in morphine-treated mice without this herbal drug (22). Therefore, it can be excluded that SJW could alter morphine pharmacodynamics.

Among the PKC family, the gamma isotype of PKC (PKCγ) is unique amongst the members of classical the PKC family in its exclusive neuronal distribution in the CNS and appears to be the isoform with a prominent role in the modulation of pain perception (23). In our previous investigations, we could show that PKCγ downregulation could potentiate morphine analgesia (9). In the present experiments, morphine administration to mice induced the phosphorylation of PKCγ in PAG, a critical brain area in processing pain signals and a primary site of action of many analgesic compounds (24). When the SJW main component hypericin was injected directly into the PAG of rats administered with morphine, PKCγ phosphorylation was prevented in this brain area and the thermal antinociceptive response was enhanced, further supporting that PAG is directly implicated in SJW-induced morphine potentiation through PKCγ activation. SJW has been shown to play also the role of a specific PKC inhibitor as previously demonstrated according to enzyme assays performed on rat brain (10) and a previous study on cultured cells supported the idea that SJW might competitively bind to the regulatory domain of PKC (11). It is therefore conceivable that SJW plays this role in morphine potentiation.

Opioids, and particularly morphine, are considered to be the "gold standard" for pain management. However, several serious side effects associated with repeated and/or increasing morphine doses, including constipation, sedation, respiratory depression and nausea, represent substantial drawbacks to its use. As far as we can discuss ethical terms, pharmacological, and clinical studies, morphine remains the most effective and efficient (in terms of cost-effectiveness) for pain therapy in cancer patients and palliative therapy in general. Nevertheless, it is still alive the sense of anxiety in patients about the effects of morphine that prevents adequate access to pain treatment. Among the main reasons for this distrust survival, the first is the fear of the phenomenon of habituation, a risk that according to the literature remains below 0.01%, but it scares both the patients and sometimes professionals. Otherwise, it is therefore a typical case of full trust that makes choosing a herbal natural cure possible because one thinks that what is natural is always good. The World Health Organization has estimated that 80% of people in the advanced nations rely on herbal medicines for some of their health care. Complement morphine with a natural substance as SJW might induce a better compliance in patients submitted to palliative therapy.

Diminished opioid efficacy during the course of opioid therapy has been considered a sign of pharmacological opioid tolerance. Thus, opioid dose escalation is a logical approach to restoring effectiveness of opioid analgesia. Our experiments offer reason to hope that this practice might be revisited using morphine in combination with SJW and its component hypericin as a safer therapeutic perspective for decreasing opioid requirements.

Acknowledgments

This work was supported by MiUR Grants, PAR project.

Conflicts of Interest

The authors of the present paper declare that no financial or personal relationship with other people or organization inappropriately influenced our work.

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