SHORT REPORT

A C G publications

Rec. Nat. Prod. 5:4 (2011) 328-331

records of natural products

Analgesic Activity of *Salvia wiedemannii* Boiss. Used in Turkish Folk Medicine

Osman Ustun* **and Ekrem Sezik**

Department of Pharmacognosy, Faculty of Pharmacy, Gazi University, 06330, Ankara, Türkiye

(Received February 26, 2011; Revised April 12, 2011; Accepted April 14, 2011)

Abstract: The aerial part of *Salvia wiedemannii* Boiss. (Lamiaceae) has been used for treatment of peptic ulcers and relieving pain in Turkish folk medicine. To evaluate the analgesic effect of *S. wiedemannii*, tail flick and acetic acid-induced writhing tests were used in mice. The chloroform extract (500 mg/kg, i.p.) obtained from *S. wiedemannii* showed significant analgesic activity on tail flick assay, while water, ethanol and butanol extracts of the plant had no activity on the same test. Chloroform extract (500 mg/kg, i.p.) also inhibited number of writhings induced by acetic acid. Chloroform extract provided analgesic effects similar to morphine. Its effect was quick and durable. This *in vivo* study demonstrates that *S. wiedemannii* has strong analgesic effect as the public believed.

Key words: *Salvia wiedemannii*; analgesic activity; folk medicine.

1. Plant Source

The aerial parts of *S. wiedemannii* were collected in the vicinity of Yunak, Polatlı in Ankara, Türkiye, in May 2003. The identity of the plant was confirmed by Pharmacognosy Department, Gazi University Faculty of Pharmacy, and Ankara, Türkiye. These specimens were stored in the same institution (GUEF 2379).

2. Previous Studies

Salvia species belongs to the Lamiaceae are widely distributed in Turkey, 50% of the 89 *Salvia* species is endemic [1]. Various parts of some *Salvia* species have been reported to have traditional values. Many *Salvia* species have been used in various medical issues as follow: *S. verticillata* in catarrh and cold [2]. *S. grandiflora* in strenghten teeth. *S. cryptantha* in stomach disorders and sterilizing wounds. *S. triloba* in gastrointestinal tract (GIT) symptoms such as stomachache, flatulence, and constipation as well as cold and cough. [3]. *S. tomentosa* in abdominal and rheumatic pains [3,4]. *S. aethiopsis* in wound healing [4]. *S. russellii*, *S. dichroantha* and *S. verticulata* in abdominal pain and stomachache [5]. *S. chrysopylla* is used against rheumatism [6]. *S. sclarea* in treatment of wards and sunstroke [6,7]. *S. nemorosa* in hemorrhage and wounds [8].

* Corresponding author: E-Mail: ustun@gazi.edu.tr, oustun@yahoo.com; Phone: +90-312-202-31-82; Fax: +90-312-223-50-18.

The article was published by Academy of Chemistry of Globe Publications
www.acgpubs.org/RNP © Published 04/15/2011 EISSN: 1307-6167

Salvia species have been used for similar symptoms or disorders by public of other countries [9-13]. The common indications include GIT symptoms/disorders (colic, diarrhea, indigestion, and abdominal pain), respiratory tract symptoms/disorders (colds, sore throat, and cough), infections (tuberculosis, bacterial infections, influenza, and parasitic infections), pain (headache and arthralgia), and miscellaneous disorders (diabetes mellitus, liver diseases, barrenness, urticaria, and hemorrhage).

Studies have demonstrated that *Salvia* species have important biological activities including antimicrobial (e.g., against *Helicobacter pylori*) [14,15], antioxidant [14,16-18], antiinflammatory [17,19,20], analgesic and antipyretic [21], and antiangiogenic [22]. Although a few chemical studies have been done on *S. wiedemannii* [23-25] there is no activity studies on its extracts [25]. This study reports *in vivo* analgesic activity of the prepared extracts from *S. wiedemannii*.

3. Present Study

Air-dried and powdered plant was macerated in 95% ethanol for 6 h in water bath adjusted to 40°C. Then the macerate was filtrated. The entire procedure was repeated three times. Combined extracts were evaporated to dryness in vacuo using a rotary evaporator (EtOH extract). EtOH extract was re-dissolved in 90% MeOH/H_2O (1000 mL) and extracted with portions of $CHCl_3$ (6x750 mL). A precipitate was obtained through the addition of methanol to the combined $CHCl_3$ extract that was removed through filtration. The filtrate was then evaporated in a rotary evaporator ($CHCl_3$ extract). The aqueous extract was then extracted with n-BuOH saturated with distilled H_2O and evaporated to dryness (BuOH extract). The remaining aqueous part was lyophilized (remaining H_2O extract).

Swiss albino mice (20–25 g) of either sex was purchased from the Animal Breeding Laboratories of Gulhane Military Academy of Medicine (Ankara, Türkiye). The animals were left at least for 7 days to acclimatize to animal room conditions (24 °C) before experiments. They were maintained on standard pellet diet and water ad libitum. The food was withdrawn 24 h before the experiment, but allowed free access of water. To avoid coprophagy the mice were fasted in wire-bottomed cages. Animals, each group constituting of 6 mice, were used. Throughout the experiments, animals were processed according to the suggested international ethical guidelines for the care of laboratory animals. Institutional Animal Ethic Committee approval was obtained.

Acetic acid-induced writhing test: This test was accomplished according to the modified method of Koster et al. [26]. The writhes were induced by intraperitoneal administration of 0.8 % (v/v) acetic acid solution (10 mL/kg). Twenty minutes prior to the administration of acetic acid, the animals were treated intraperitonally with the extracts, vehicle (0.5% carboxymethyl cellulose) and references (morphine and aspirin). The number of writhings as an indication of pain was counted and recorded during 20 min period.

Tail flick test (D'Amour & Smith Test) [27]: Mice were held in position with the tail extending out and light heat was applied to tails of mice. A cut-off time of 10 sec. was used to avoid tissue damage. From application of heat to move the tail recorded. The mice had been administered different extracts of *S. wiedemannii*, a vehicle, or morphine via intraperitonal before light heat applied. The test repeated at the time periods of 20, 40 and 60 minutes.

All values were expressed as means ± S.E.M. The statistical analysis was performed by using one-way analysis of variance of (ANOVA) followed by the test of Dunnett's Multiple Comparison Test. $P < 0.05$ was considered significant from the control.

Considering its common dosages used among people, we prepared the extracts. *In vivo* test results of its analgesic activity were given in Tables 1 and 2. As shown in Table 1, i.p. administration of the chloroform extract of *S. wiedemannii* at a dose of 500 mg/kg inhibited tail flick response at the 20th min. in mice. This response was rapid and durable similar to that observed with morphine. The analgesic activity, although decreased, was detected 60 minutes after.

When its chloroform, ethanol, butanol, and water extracts were used in writhing test, chloroform extract was found to have the strongest analgesic activity (Table 2). Once again, its efficacy was very close to morphine. The other extracts showed analgesic activities similar to that observed with aspirin.

The results of these two *in vivo* tests indicate that chloroform extract of *S.wiedemannii* has one or more constituents with strong analgesic effect. Since the tail-flick test is considered a specific model for compounds producing central antinociceptive activity [27], these results indicates that chloroform extract also exhibits central analgesic effects in mice.

In conclusion, the present study has clearly demonstrated that *S.wiedemannii* possesses a potent analgesic activity as suggested in Turkish folk medicine. This is the first report demonstrating the analgesic activity of *S. wiedemannii in vivo*; however, further studies will be necessary to isolate the active compounds which are responsible for the analgesic effect and to understand exact mechanisms of this activity.

Table 1. Effect of the extracts on tail flick test in mice

Treatment	Dose mg/kg, i.p.	Time after injection			
		0 min (basal)	20 min	40 min	60 min
Vehicle (CMC)		2.2 ± 0.1	2.2 ± 0.1	2.2 ± 0.1	2.2 ± 0.1
Morphine	10	2.1 ± 0.1	5.5 ± 0.7*	4.5 ± 0.6*	3.8 ± 0.5*
EtOH extract	500	2.3 ± 0.1	2.2 ± 0.1	2.1 ± 0.1	2.3 ± 0.1
BuOH extract	500	2.3 ± 0.4	2.6± 0.1	2.9 ± 0.3	2.9 ± 0.3
H_2O extract	500	2.3 ± 0.2	2.3± 0.2	2.6 ± 0.3	2.7 ± 0.2
$CHCl_3$ extract	500	2.4 ± 0.1	5.2 ± 0.6*	4.1 ± 0.5*	3.4 ± 0.3*

n = 6 animals; CMC: Carboxymethyl cellulose; *$p < 0.05$, relative to control group value

Table 2. Effect of the extracts on acetic acid-induced writhing test in mice

Treatment	Dose mg/kg, i.p.	Writhing (mean ± S.E.)
Vehicle (CMC)		28.87 ± 2.27
Morphine	10	0.26 ± 0.48*
Aspirin	100	18.44 ± 2.79*
EtOH extract	500	20.05±4.27
BuOH extract	500	23.04±5.57
H_2O extract	500	22.84±3.41
$CHCl_3$ extract	500	0.33 ± 0.2*

n = 6 animals ; CMC: Carboxymethyl cellulose ; $p < 0.01$ relative to control group value

Acknowledgments

This project is financially supported by the Research Fund of Gazi University (No: EF.02/2004-07).

References

[1] P.H. Davis (1982) Flora of Turkey and the East Aegean Islands, pp 241 University Press, Edinburgh.

[2] M. Tabata, E. Sezik, G. Honda, E. Yeşilada, H. Fukui, K. Goto and Y. Ikeshiro (1994). Traditional medicine in Turkey III. folk medicine in East Anatolia, Van and Bitlis provinces, *Int. J. Pharmacog*. **32** (1), 3-12.

[3] G. Honda, E. Yeşilada, M. Tabata, E. Sezik, T. Fujita, Y. Takeda, Y. Takaishi and T. Tanaka (1996).Traditional medicine in Turkey IV. Folk medicine in West Anatolia: Afyon, Kütahya, Denizli, Muğla, Aydın provinces, *J. Etnopharmacol.* **53**, 75-87.

[4] T. Fujita, E. Sezik, M. Tabata, E. Yeşilada, G. Honda, Y. Takeda, T. Tanaka and Y. Takaishi (1995). Traditional medicine in Turkey VII. Folk medicine in Middle and West Black Sea regions, *Econ Bot*. **49**(4), 406-422.

[5] E. Sezik, E. Yeşilada, G. Honda, Y. Takaishi, Y. Takeda and T. Tanaka (2001). Traditional medicine in Turkey X. Folk medicine in Central Anatolia, *J. Etnopharmacol.* **75**, 95-115.

[6] E. Yeşilada, G. Honda, E. Sezik, M. Tabata, K. Goto and Y. Ikeshiro (1993). Traditional medicine in Turkey IV. Folk medicine in the Mediterranean Subdivision, *J. Etnopharmacol.* **39**, 31-38.
[7] E. Yeşilada, G. Honda, E. Sezik, M. Tabata, T. Fujita, T. Tanaka, Y. Takeda, and Y. Takaishi (1995). Traditional Medicine in Turkey V. Folk medicine in the inner Taurus Mountains, *J. Etnopharmacol.* **46**, 133-152.
[8] E. Sezik, E. Yeşilada, M. Tabata, G. Honda, Y. Takaishi, T. Fujita, T. Tanaka and Y. Takeda (1997). Traditional medicine in Turkey VIII. Folk medicine in East Anatolia; Erzurum, Erzincan, Ağrı, Kars, Iğdır Provinces, *Econ Bot*. **51**(3), 195-211.
[9] G.P.P. Kamatou, N.P. Makunga, W.P.N. Ramogola and A.M. Viljoen (2008). South African *Salvia* species: A review of biological activities and phytochemistry, *J. Etnopharmacol*. **119**, 664–672.
[10] P.M. Guarrera, G. Forti, S. Marignoli (2005). Ethnobotanical and ethnomedicinal uses of plants in the district of Acquapendente (Latium, Central Italy), *J. Etnopharmacol*. **96,** 429–444.
[11] E. Lev and Z. Amar (2002). Ethnopharmacological survey of traditional drugs sold in the Kingdom of Jordan, *J. Etnopharmacol.* **82**, 131-145.
[12] V. Tene, O. Malagon, P.V. Finzi, G. Vidari, C. Armijos and T. Zaragoza (2007). An ethnobotanical survey of medicinal plants used in Loja and Zamora-Chinchipe, Ecuador, *J. Etnopharmacol.***111**, 63–81.
[13] O. Said, K. Khalil, S. Fulder and H. Azaizeh (2002). Ethnopharmacological survey of medicinal herbs in Israel, the Golan Heights and the West Bank region, *J. Etnopharmacol* .**83**, 251-265.
[14] G. Ozkan, O. Sagdic, R.S. Gokturk, O. Unal and S. Albayrak (2010). Study on chemical composition and biological activities of essential oil and extract from *Salvia pisidica*, *LWT - Food Sci Technol.* **43**, 186–190.
[15] G. Stamatis, P. Kyriazopoulos, S. Golegou, A. Basayiannis, S. Skaltsas and H. Skaltsa(2003). In vitro anti-Helicobacter pylori activity of Greek herbal medicines, *J. Etnopharmacol.***88,** 175–179.
[16] A. Kabouche, Z. Kabouche, M. Ozturk, U. Kolak and G. Topcu (2007). Antioxidant abietane diterpenoids from *Salvia barrelieri*, *Food Chem*. **102**, 1281–1287.
[17] G.P.P. Kamatou, A.M. Viljoen and P. Steenkamp (2010). Antioxidant, antiinflammatory activities and HPLC analysis of South African *Salvia* species, *Food Chem.* **119**, 684–688.
[18] I. Orhan, M. Kartal, Q. Naz, A. Ejaz, G. Yilmaz, Y. Kan, B. Konuklugil, B. Sener and M.I. Choudhary (2007). Antioxidant and anticholinesterase evaluation of selected Turkish *Salvia* species, *Food Chem*. **103**, 1247–1254.
[19] E.K. Akkol, F. Goger, M. Kosar and K.H.C. Baser (2008). Phenolic composition and biological activities of *Salvia halophila* and *Salvia virgata* from Turkey, *Food Chem.* **108,** 942–949.
[20] M. Kaileh, W.V. Berghe, E. Boone, T. Essawi and G. Haegeman (2007). Screening of indigenous Palestinian medicinal plants for potential anti-inflammatory and cytotoxic activity, *J. Etnopharmacol.* **113**, 510–516.
[21] G.J. Amabeoku, P. Eagles, G. Scott, I. Mayeng, E. Springfield (2001). Analgesic and antipyretic effects of *Dodonaea angustifolia* and *Salvia africana-lutea*, *J. Etnopharmacol.* **75**, 117–124.
[22] H.J. Jung, Y.S. Song, C.J. Lim and E.H. Park (2009). Anti-inflammatory, anti-angiogenic and anti-nociceptive activities of an ethanol extract of *Salvia plebeia* R. Brown, *J. Etnopharmacol.* **126**, 355–360.
[23] G. Topçu, A.Ulubelen (1990). Diterpenoids from *Salvia wiedemannii,Phytochemistry* **29**, 2346-2348.
[24] G. Topçu, A. Ulubelen (1991). Diterpenoids from *Salvia wiedemanni, Phytochemistry* **30**, 2412-2413.
[25] A. Ulubelen, G.Topçu, S. Chen, P. Cai, J.K. Snyder (1991). A New Abietane Diterpene from *Salvia wiedemanni* Boiss, *J. Org. Chem*. **56**, 7354-7356.
[26] R. Koster, M. Anderson and E.J. Beer (1959). Acetic acid for analgesic screening, *Fed Proc*. **18**, 412–416.
[27] F.E. D'Amour and D.L. Smith (1941). A method for determining loss of pain sensation, *J Pharmacol Exp Therap*. **72**, 74–79.