Journal of Ethnopharmacology 211 (2018) 394–399

Contents lists available at ScienceDirect

Journal of Ethnopharmacology

journal homepage: www.elsevier.com/locate/jethpharm

Protective effect of oleane-12-en-3β-ol-28-oic acid 3β-D-glucopyranoside in ethanol induced gastric ulcer by enhancing the prostaglandin E2 level

Imran Kazmi[a], Shakir Saleem[b], Tanveer Ahmad[b], Muhammad Afzal[c,*], Fahad A. Al-Abbasi[d], Vikas Kumar[e], Firoz Anwar[d,*]

[a] Glocal School of Pharmacy, Glocal University, Mirzapur Pole, Saharanpur, Uttar Pradesh, India
[b] Siddhartha Institute of Pharmacy, Near I.T. Park, Sahastradhara Road, Dehradun, India
[c] Department of Pharmacology, Faculty of Pharmacy, Aljouf University, Sakaka 72388, Kingdom of Saudi Arabia
[d] Department of Biochemistry, College of Science, King Abdulaziz University, Jeddah, Kingdom of Saudi Arabia
[e] Natural Product Drug Discovery Laboratory, Department of Pharmaceutical Sciences, Faculty of Health Sciences, Sam Higginbottom Institute of Agriculture, Technology and Sciences, Allahabad, Uttar Pradesh, India

ARTICLE INFO

Keywords:
Antiulcer
OAG
Aspirin
Ranitidine
Ethanol
Gasoprotective
Anti-secretary activity

ABSTRACT

Ethnopharmacological relevance: *Lantana camara* is a popular invasive weed utilized in the management of ulcer in different part of world. Study of specific compound present in this plant responsible for their antulcer activity is main topic of concern. Current study designed for evaluation of the antiulcer activity of oleane-12-en-3β-ol-28-oic acid 3β-D-glucopyranoside (OAG) from *Lantana camara* L.

Materials and methods: Antiulcer activity was carried out on NSAID's (Aspirin) and ethanol induced ulcer model. The efficacy of the OAG on ulcer index, percentage protection and gastric acid secretion were evaluated.

Results: Ulcer protection percentage (38.37%) was significant ($P < 0.001$) higher in the groups treated with the higher OAG dose (50 mg/kg), it also recover the mucosa with no redness, no inflammation, mild dilation of blood vessels. OAG significantly ($P < 0.01$ and $P < 0.001$) reduce acidity, free acidity and gastric acid volume. It also significantly ($P < 0.01$) increases the pH of stomach.

Conclusion: On the basis of results, it can be concluded that OAG shows significant gastroprotective activity by gastric acid secretion inhibition and afford protection against gastric mucosal damage. Further increase of prostaglandin E2 level establishes the mechanism of antiulcer activity of OAG.

1. Introduction

In 1800s peptic ulcer was not regarded as a cause of death, hospitalization and disability but at the middle of 20th century it covered large population and became burden on national economy (Duggan and Duggan, 2006; Chan and Leung, 2002). The prevalence and incidence of peptic ulcer unexpectedly decreased in last four decades (van Leerdam, 2008). It is a damaged duodenal or gastric mucosa area induced by the gastric juice action. It is a chronic and recurrent disease and is the most common and prevalent among the various diseases of gastrointestinal track (Guyton and Hall, 1998). Peptic ulcer is induced by an imbalance of equilibrium between mucosal defensive factors and gastric aggressive factors (Rao et al., 2000). Peptic ulcer is very common condition with an annual incidence of more than 2.5% worldwide (Everhart et al., 1998). Different class of pharmaceutical drugs like anticholinergics, antacids, PPIs, H_2-receptor antagonists, cholecystokinin-2 receptor antagonists, K^+ therapy, sucralfate, prostaglandin analogues, rebamipide, carnenoxolone, bismuth compounds, ecabet, antimicrobial drugs single or along with the combination of other drugs found efficient in the prevention and treatment of peptic ulcer but none of the drug is free from side effects (Stolte et al., 2011). Now a day, herbal medicine is extensively used worldwide for the recovery of different types of gastrointestinal diseases. Now a day, we have an option of herbal drugs in comparison to synthetic one with lesser side effect and very good potential for peptic ulcer treatment and other gastric disorders both from scientific data and traditional knowledge.

Oleane-12-en-3β-ol-28-oic acid 3β-D-glucopyranoside (OAG), pentacyclic triterpenoid extracted from *Lantana camara* L. (family: verbanaceae) leaves (Kazmi et al., 2012). Oleanolic acid derivatives like disodium 3β-(3-carboxypropionuloxy) olean-9(11)-en-28-oate and disodium 3β-(carboxypropionyloxy) ursa-9, 12-dien-28-oate reported for antiulcer activity (Farina et al., 1988). Traditionally *L. camara* mainly used in different part of world for the management of ulcers, chickenpox, tumors, eczema, high blood pressure, carcinoma, catarrhal

* Corresponding authors.
E-mail addresses: afzalgufran@gmail.com (M. Afzal), firoz_anwar2000@yahoo.com (F. Anwar).

http://dx.doi.org/10.1016/j.jep.2017.09.012
Received 19 May 2017; Received in revised form 10 September 2017; Accepted 11 September 2017
Available online 21 September 2017

infections, bilious fevers, rheumatism, tetanus, asthma, abdominal viscera ataxy, measles and malaria (Ghisalberti, 2000; Day et al., 2003), epilepsy (Ganesh et al., 2010), weakness of memory, enhance intellect and cognition (Adams et al., 2007). The plant has reported as anticonvulsant (Adesina, 1982), anticancer (Bisi-Johnson et al., 2011; Gomes de Melo et al., 2010), antiulcer (Sathish et al., 2011), antioxidant (Gomes de Melo et al., 2010), anti-diabetic (Venkatachalam et al., 2011; Garg et al., 1997), antifungal, antibacterial (Sinha, 1987; Rwangabo, 1988; Barreto, 2010), anti-feedant, larval mortality/repellency (Pandey, 1977; Sagar, 2005; Pandey, 1983), anti-motility (Sagar, 2005) analgesic and anti-inflammatory (Ghosh et al., 2010) activities. However OAG, an new oleanolic acid derivative has not been tested for antiulcer properties, though the plant is traditionally used against such disorders.

Hence, in the present study OAG from *L. camara* leaves was assessed for antiulcer effect usingaspirin induced ulcerogenic model.

2. Materials and methods

Current study was approved by the Ethical Committee of Animal, Siddhartha Institute of Pharmacy, Dehradun, Uttarakhand, India (Ref. SIP/IAEC.CLEAR/55/2010-11).

2.1. Plant material

Leaves of *Lantana camara* plant were collected from nearby areas of Siddhartha Institute of Pharmacy, Dehradun and identified by Dr. I. Kazmi, Siddhartha Institute of Pharmacy, Dehradun (SIP/DPP/consult/−15-05-13/156/27).

2.2. Isolation of oleane-12-en-3β-ol-28-oic acid 3β-D-glucopyranoside

4 kg leaves of plant was collected, dried and powdered. This powdered leaves was extracted with 12 L of methanol at 50 °C for 24 h. The slurry extracted was dissolved in minimum volume of methanol, and further, adsorbed on 60–120 mesh silica gel. The slurry was chromatographed over silica gel in column for sepatation of phytoconstituents by using CHCl3/MeOH gradient system (49:1; 2.0 L for gradient system); it yielded colorless crystals of OAG (yield 16.1 g, 0.40%) (Kazmi et al., 2012). It was found to be 100% pure by HPTLC by using solvent system chloroform and methanol (99:1).

2.3. Chemicals

Ranitidine obtained from Orchev Pharma PVT. LTD. All the chemicals and solvents used in experimental protocol and chromatographic isolation were of standard analytical grade and purchased from Himgiri Traders of Dehradun, India. OAG is obtained (Kazmi et al., 2012), as a gift sample from Siddhartha Institute of Pharmacy, Dehradun. The molecular structure of OAG is shown in Fig. 1.

2.4. Animals

Albino Wistar rats of 150–200 g were procured from DIPSAR Institute, New Delhi. Maintained in animal house facility of Siddhartha Institute of Pharmacy (Dehradun) and kept at 55 ± 5% humidity, 25 ± 1 °C along with 12 h dark/light cycle. Food given to the rats was standard pellet and normal distill water during the complete experimental study. The experimental study was approved by Ethics Committee of Animals. OAG and Ranitidine both orally administered.

2.5. Behavioral and toxic study

OAG was given to the test groups animals in graded doses up to 100 mg per kg body weight per day and the animals were inspected for any behavioral changes and mortality sign for ten days afterwards.

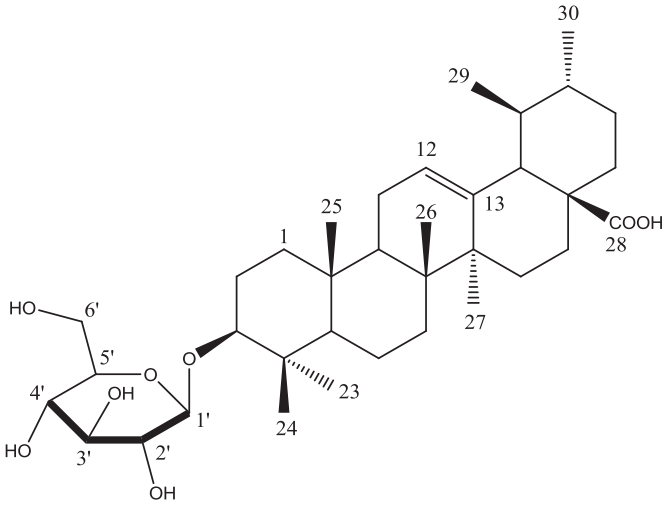

Fig. 1. Structure of oleane-12-en-3β-ol-28-oic acid 3β-D-glucopyranoside.

Administration of dose was increased up to 50 mg per kg and again inspected for behavioral changes and mortality sign for next ten days. LD50 value of animals was found to be on higher dose than 50 mg per kg body weight in animals. 25 mg per kg and 50 mg per kg body weight per day, dose of OAG was used for the further experimental protocol.

2.6. NSAID's induced ulcer (Aspirin induced ulcer)

150–200 g of female Wistar rats of were segregated into four groups and all the group containing six rats.

Group I: Aspirin control group (200 mg/kg, p.o. aspirin)
Group II: Standard group (Ranitidine 20 mg/kg in 2% gum acacia, p.o).
Group III: OAG group (25 mg/kg, p.o.).
Group IV: OAG group (50 mg/kg, p.o.).

On 5th day, aspirin with 200 mg per kg dose (20 mg per ml) was given to the rats of different groups (I to IV) after one hour of the last dose of the OAG/ Ranitidine. After 4 h of the aspirin administration, animals sacrificed, the stomach isolated and cut along the greater curvature area, then washed properly with 0.9% NaCl (5 ml) and ulcers scored in the stomach glandular portion. Index of ulcer was measured by cumulative total number of ulcers and the severity of total ulcers. Ulcers total severity was measured by recording each ulcer severity and sample further used for histopathological examination (Berenguer et al., 2006).

Number of ulcers/ stomach was recorded and ulcers severity scored by microscope with the help of 10X hand lens and scoring was measured as follows.

0.0 = normal stomach, 0.5 = red color, 1.0 = spot ulcers, 1.5 = streaks of hemorrhage,
2.0 = ulcer > 3 but < 5, 3.0 = ulcer > 5

Mean score of ulcer for each rats was expressed as a ulcer index. Protection percentage was calculated by using the formula,

$$\text{Percentage protection} = 100 - \frac{Ut}{Uc} \times 100$$

Where, Ut = Treated group ulcer index.
Uc = Control group ulcer index.

2.6.1. Measurement of total acidity and free acidity

Gastric juice (1 ml) was transferred into 100 ml of conical flask. 2–3 drop topfer reagent mixed and then titrated with 0.01N of NaOH till red coloration of solution disappears. The solution color changes to yellow