

Review Article

Medicinal uses, phytochemistry, pharmacology and toxicological properties of *Elaeodendron croceum*

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Abstract

Elaeodendron croceum is a shrub or a small tree used as an emetic, purgative and herbal medicine for cough, sore throat, fever, tuberculosis, blood in sputum, chest congestion, human immunodeficiency virus (HIV) opportunistic infections and to clean the digestive tract. Phytochemical compounds identified from the species include alkaloids, cardiac glucoside, flavonoids, phenols, proanthocyanidins, saponins, sugars, tannins and triterpenoids. Pharmacological studies revealed that *E. croceum* extracts and compounds have antimycobacterial, anti-arthritic, antibacterial, anti-HIV, antioxidant, anti-inflammatory, antifungal and cytotoxicity activities. Future studies should focus on ethnopharmacological safety and efficacy of *E. croceum* extracts and compounds isolated from the species using in vivo preclinical studies and clinical trials.

Keywords: Celastraceae, *Elaeodendron croceum*, Herbal medicine, Southern Africa, Traditional medicine

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INTRODUCTION

Elaeodendron croceum (Thunb.) DC. is a shrub or a small evergreen tree species which belongs to the Celastraceae family. The species is commonly referred to as forest saffron, saffron or wood saffron. *E. croceum* has in the past classified under several scientific names such as *Cassine crocea* (Thunb.) Kuntze, *C. papillosa* (Hochst.) Kuntze, *Crocoxylon croceum* Thunb. N. Robson, *C. excelsum* Eckl. & Zeyh., *E. capense* Eckl. & Zeyh., *Salacia zeyheri* Planch. ex Harv., *Ilex crocea* Thunb., *E. croceum* Thunb. DC. and *E. papillosum* Hochst. [1,2]. *E. croceum* is among the top 15 medicinal plants that are becoming

scarce in KwaZulu Natal province in South Africa based on the information provided by traditional healers [3-5]. This could be as a result of over-collection as the species is widely sold in informal herbal medicine markets in both rural and urban areas of Gauteng [6] and KwaZulu Natal provinces in South Africa [4]. Over-collection of wild populations of *E. croceum* as a source for medicinal plant trade and other anthropogenic activities such as habitat destruction and land clearing for agricultural purposes have caused a severe reduction in the population size of the species [7]. *E. croceum* is regarded as Least Concern on the IUCN Red List of Threatened Species [8] but due to local

conservation concern in South Africa as the population of the species is declining, the species has been flagged as declining following recommendations made by Williams *et al* [9], Von Staden *et al* [10] and Victor and Keith [11]. It is against this background that the medicinal uses, phytochemistry, pharmacology and toxicological properties of *E. croceum* were reviewed.

METHODS

Data on medicinal uses, botany, phytochemistry, pharmacological and toxicological properties of *E. croceum* were collected from multiple internet sources including Science Direct, SciFinder, Pubmed, BMC, Elsevier, Google Scholar, Scopus and Web of Science. Complementary information was gathered from pre-electronic sources such as books, book chapters, theses, scientific reports and journal articles obtained from the University library. The search for this information was carried out between May 2017 to August 2018. The keywords used in the search included “ethnobotany”, “ethnomedicinal uses”, “medicinal uses”, “phytochemistry”, “biological activities”, “pharmacological properties”, “toxicological properties”, “*Elaeodendron croceum*”, the synonym of the species and the English common names “forest saffron”, “saffron” or “wood saffron”. The internet search generated 562 articles in total. After duplicate articles and those with limited raw data were excluded, 36 articles were included in this study. These articles included 21 journal articles, books (nine), scientific report (three), websites (two) and a single book chapter.

FINDINGS AND DISCUSSION

Botanical profile and medicinal uses of *E. croceum*

E. croceum (Figure 1) grows up to a height of 15 metres with a trunk diameter of 250 mm [12]. The species has been recorded in montane forest

margins and along the escarpment, mainly on seepage zones. The main stem is red-brown to grey in colour and may have some dark lenticels when young characterized by pale grey and thin bark, peeling off to reveal bright orange and yellow under-bark [13]. The leaves are opposite to sub-opposite in arrangement and oblong to elliptic in shape. The apex and base of the leaves is broadly tapering, leaf margins are strongly serrated and sometimes spined. The leaves are glossy green above and little paler below with prominent venation on both sides. The flowers are small, whitish or pale green in colour, occurring axillary clusters. The fruits are fleshy, ovoid in shape and creamy to yellowish in colour [14]. The species has been recorded in Mozambique, South Africa, Swaziland, Zimbabwe and naturalized in St Helena [1,14].



Figure 1: *E. croceum* branch showing a fruit and leaves (photo: D Becking)

E. croceum bark is used as an emetic and purgative [15,16] as herbal medicine against human immunodeficiency virus (HIV) opportunistic infections [17]. The bark is also traditionally used to treat and manage tuberculosis [18] and other related diseases such as blood in sputum [18], chest congestion [12,16,19-21], cough [18] and sore throat [16]. The bark is used to manage and treat fever [18] and bark, roots and leaves of the species as herbal medicine to clean the digestive tract [12,16,19-21].

Table 1: Medicinal uses of *E. croceum*

Medicinal use	Plant part(s) used	Country	Reference(s)
Blood in sputum	Bark infusion taken orally	South Africa	[18]
Chest congestion	Bark infusion taken orally	South Africa	[12,16,19-21]
Clean digestive tract	Bark, leaf and root infusion taken orally	South Africa	[12,16,19-21]
Cough	Bark decoction taken orally	South Africa	[18]
Emetic	Bark infusion taken orally	Swaziland	[15]
Fever	Bark decoction taken orally	South Africa	[18]
HIV	Bark decoction taken orally	South Africa	[17]
Purgative	Bark infusion taken orally	South Africa	[16]
Sore throat	Bark decoction taken orally	South Africa	[16]
Tuberculosis	Bark infusion taken orally	South Africa	[18]

Phytochemistry and pharmacological properties of *E. croceum*

Reports on the phytochemical screening of *E. croceum* leaf, root and stem bark extracts revealed the presence of alkaloids, flavonoids, phenols, proanthocyanidins, saponins and tannins [22,23]. Drewes and Mashimbye [24] isolated flavonoids, sugar and triterpenoids and these compounds included (+)-6R,13R-11,11-dimethyl-1,3,8,10-tetrahydroxy-9-methoxy-peltogynan, (-)-4'-O-methoxyepigallocatechin, ouratea proanthocyanidin A, galactitol, tingenone, tingenin B, canophyllol, 3-hydroxylupeol and 30-hydroxylup 20(29)-en-3-one from the stem bark of *E. croceum* (Figure 2). Prinsloo *et al* [20] isolated a cardiac glycoside, digitoxigenin-glucoside from *E. croceum* bark extracts (Figure 2). Yelani *et al* [21] isolated triterpenoids such as 20-hydroxy-20-epitingenone, tingenone, tingenine B, 11 α -hydroxy- β -amyrin and naringenin from the leaves of *E. croceum* (Figure 2).

Prinsloo *et al* [20] evaluated anti-HIV activities of the compound digitoxigenin-glucoside isolated from *E. croceum* extracts by assessing the inhibition of recombinant HIV strain in an MT-2 VSV-pseudotyped recombinant virus assay. The compound showed significant inhibition of 90% at 0.2 μ M [20]. Prinsloo *et al* [20,25] evaluated the cytotoxicity of the compound digitoxigenin-glucoside using the colourimetric 3-(4,5-dimethylthiazol-2-yl)-2, 5-diphenyltetrazolium bromide (MTT) assay on Vero cells and scored as percentage reduction at 540 nm of treated culture versus untreated control culture. The isolated compound showed toxicity of only 20% at a concentration of 25 μ g/ml on Vero cells with a therapeutic index of 250 [20,25]. Yelani *et al* [21] evaluated the cytotoxicity of hexane, dichloromethane, and acetone leaf extracts of *E. croceum* and compounds 20-hydroxy-20-epitingenone, tingenone, tingenine B, 11 α -hydroxy- β -amyrin and naringenin isolated from the leaves of the species using the XTT (sodium 3'-[1-(phenyl amino-carbonyl)-3,4-tetrazolium]-bis-[4-methoxy-6-nitro] benzene sulfonic acid hydrate) colorimetric assay against four human cancer cell lines, Vero (monkey kidney), HeLa (human cervix cancer), SNO (human oesophagus cancer) and MCF-7 (human breast cancer) with doxorubicin and zearalenone as positive controls. All extracts showed activities, but significant activities were demonstrated by compounds 20-hydroxy-20-epitingenone and tingenone which showed activities against Vero cells with half maximal inhibitory concentration (IC₅₀) values of 2.7 nM and 8.2 μ M, respectively. The cytotoxicity of compounds 20-hydroxy-20-

epitingenone and tingenone against three human cancer cell lines, HeLa, MCF-7, and SNO showed IC₅₀ values ranging between 2.5 μ M to 0.4 μ M [21].

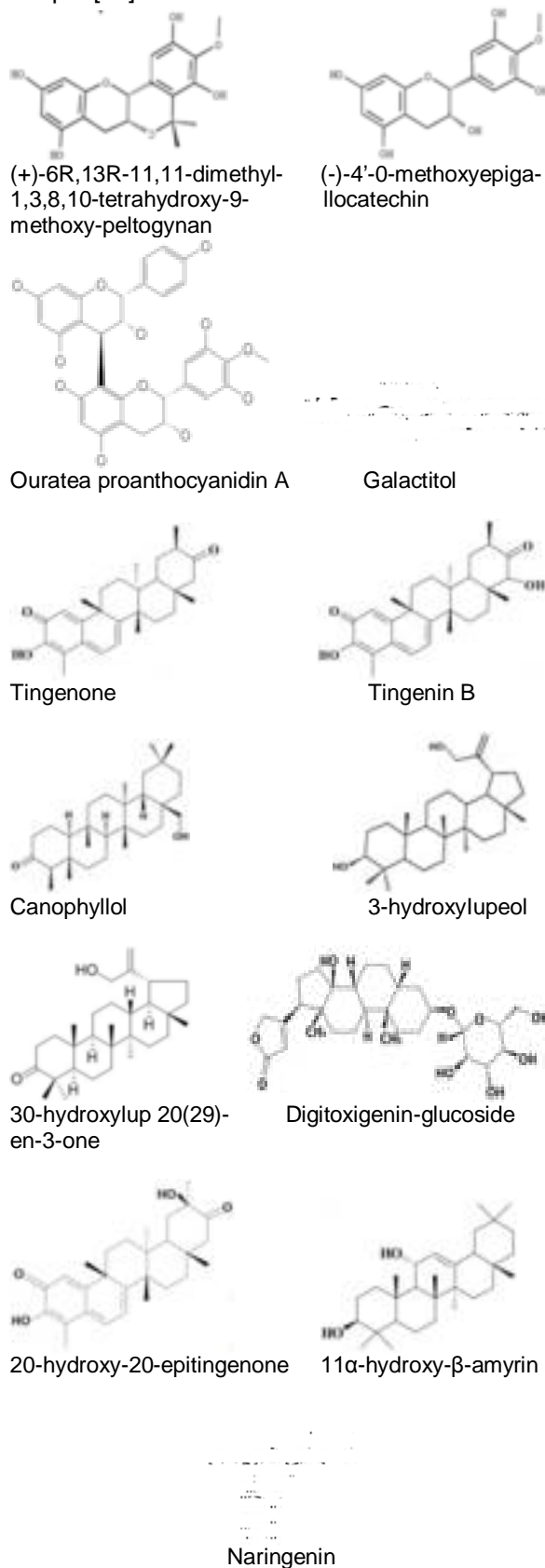


Figure 2: Chemical structures of compounds isolated from *E. croceum*

Anti-arthritic activity

Elisha *et al* [23] evaluated the anti-arthritic activities of acetone extract of *E. croceum* leaves using an anti-protein denaturation assay. The extract had a dose dependent response in the in vitro anti-arthritic test, exhibited IC₅₀ value of 80.0 µg/ml which was higher than the IC₅₀ value of 32.4 µg/ml exhibited by the positive control diclofenac sodium [23]. The promising activities of the extracts support the traditional usage of the species for inflammatory conditions.

Antibacterial activity

Eloff *et al* [26] evaluated antibacterial activities of acetone extract of *E. croceum* bark using the agar diffusion method against *Staphylococcus aureus*, *Enterococcus faecalis*, *Escherichia coli* and *Pseudomonas aeruginosa*. The extract showed activities against the tested pathogens with total activity values ranging from 8.0 ml/g to >275.0 ml/g [26]. Kaikabo *et al* [27] evaluated antibacterial activities of acetone extract of *E. croceum* bark using a two-fold serial microdilution method against *Staphylococcus aureus*, *Enterococcus faecalis*, *Escherichia coli* and *Pseudomonas aeruginosa*. The minimum inhibitory concentrations (MIC) screening was conducted at concentrations of 0.02 mg/ml to 2.5 mg/ml. The extracts had activity against at least one of the test organisms over a period of 1, 2 or 24 hours with average MIC values of the extracts against the tested pathogens ranging from 0.2 mg/ml to 1.8 mg/ml [27].

Elisha *et al* [28] evaluated antibacterial activities of acetone extract of *E. croceum* leaves against *Bacillus anthracis* Sterne strain using the microplate serial dilution method with gentamicin as a positive control. The extract showed activities with MIC value of 0.3 mg/ml and total antibacterial activity of 290.0 ml/g [28]. Elisha *et al* [29] evaluated the antibacterial activities of acetone extract of *E. croceum* leaves against *Stenotrophomonas maltophilia*, *Klebsiella pneumoniae*, *Salmonella typhimurium*, *Proteus mirabilis*, *Enterobacter cloacae* and *Escherichia coli* using a serial microdilution method with gentamicin as a positive control. The extract exhibited activities with MIC values ranging from 0.08 mg/ml to 0.31 mg/ml and total antibacterial activity values ranging from 290.2 ml/g to 1124.6 ml/g [29].

Elisha *et al* [30] evaluated antibacterial activities of acetone extract of *E. croceum* leaves against *Escherichia coli*, *Staphylococcus aureus*, *Enterococcus faecalis*, *Bacillus cereus*, *Salmonella typhimurium* and *Pseudomonas aeruginosa* using

microplate serial dilution technique with gentamicin as a positive control. The extract showed activities with MIC values ranging from 0.08 mg/ml to 0.4 mg/ml and total antibacterial activity ranging from 214.2 ml/g to 1124.6 ml/g [30]. Mamba *et al* [17] evaluated antibacterial activities of ethanol extract of *E. croceum* bark against *Gardnerella vaginalis*, *Neisseria gonorrhoeae* and *Oligella ureolytica* using the broth microdilution method with ciprofloxacin as a positive control. The extract exhibited activities against tested pathogens with MIC values ranging from 1.6 mg/ml to 12.5 mg/ml while ciprofloxacin exhibited MIC value of <0.01 mg/ml [17]. Odeyemi and Afolayan [24] evaluated antibacterial activities of aqueous extract of *E. croceum* leaves and stem bark against *Enterococcus faecalis*, *Klebsiella pneumoniae*, *Proteus vulgaris*, *Salmonella typhimurium*, *Shigella flexneri* and *Staphylococcus aureus* using the agar well diffusion and broth microdilution methods with amoxicillin (0.0125 mg/mL) as the positive control. The extracts exhibited activities against all the tested pathogens with zone on inhibition ranging from 13.0 mm to 15.7 mm. The control, amoxicillin was active against *Enterococcus faecalis*, *Klebsiella pneumoniae*, *Shigella flexneri* and *Staphylococcus aureus* with zone on inhibition ranging from 12.5 mm to 26.5 mm. The MIC values exhibited by the extracts and the control against all the pathogens ranged from 0.6 mg/ml to >5.0 mg/ml [24]. The documented antibacterial activities exhibited by *E. croceum* extracts show the potential of the species as herbal medicine against bacterial pathogens and also corroborate the traditional uses of the species as herbal medicine against sore throat in South Africa [16].

Antimycobacterial activity

Lall and Meyer [18] evaluated antimycobacterial activities of aqueous and acetone extracts of *E. croceum* bark against drug-resistant and drug-sensitive strains of *Mycobacterium tuberculosis* H37Rv using the agar plate method. The acetone extract exhibited activities with MIC value of 1.0 mg/ml [18]. Elisha *et al* [31] evaluated the antimycobacterial activities of acetone extract of *E. croceum* leaves against *Mycobacterium aurum*, *Mycobacterium fortuitum* and *Mycobacterium smegmatis* using serial microdilution method with rifampicin and streptomycin as positive controls. The extracts showed activities against tested pathogens with MIC value of 1.3 mg/ml against *Mycobacterium fortuitum* and MIC value of 2.5 mg/ml against both *Mycobacterium aurum* and *Mycobacterium smegmatis* and total antimycobacterial activities ranged from 36.0 ml/g to 72.0 ml/g. The positive

controls, rifampicin and streptomycin showed MIC values ranging from 0.01 mg/ml to 0.06 mg/ml [31]. These findings corroborate the traditional uses of the species as herbal medicine against tuberculosis [18] and related diseases such as blood in sputum [18], chest congestion [12,16,19-21], cough [18] and sore throat [16].

Antifungal activity

Mamba *et al* [17] evaluated antifungal activities of ethanol extract of *E. croceum* bark against *Candida albicans* using the broth microdilution method with ciprofloxacin as a positive control. The extract exhibited activities with MIC value of 1.6 mg/ml while ciprofloxacin exhibited MIC value of <0.01 mg/ml [17]. The documented antifungal activities exhibited by *E. croceum* extracts show the potential of the species as herbal medicine against fungal and microbial pathogens.

Anti-HIV activity

Prinsloo *et al* [23] evaluated anti-HIV activities of *E. croceum* by assessing inhibition of transcription factors in the HeLa-TAT-Luc and MT-2 VSV-pseudotyped recombinant virus assays. The extracts showed potent inhibition of transcription factors at 100 ng/mL [23]. Mamba *et al* [17] evaluated anti-HIV activities of ethanol extract of *E. croceum* bark against recombinant HIV-1 enzyme using non-radioactive HIV-reverse transcriptase (RT) colorimetric assay with doxorubicin as a positive control. The extract exhibited low inhibitory activity of 30.2% while the positive control doxorubicin showed 96.5% inhibitory activity [17]. The documented anti-HIV activities exhibited by *E. croceum* extracts and the compound digitoxigenin-glucoside isolated from the species corroborate the traditional usage of the species against HIV opportunistic infections in South Africa [17].

Anti-inflammatory activity

Elisha *et al* [25] evaluated the anti-inflammatory activities of acetone extract of *E. croceum* leaves by determining the inhibition of nitric oxide production in lipopolysaccharide activated RAW 264.7 macrophages as well as 15-lipoxygenase enzyme inhibition. The extracts inhibited nitric oxide production in a dose-dependent manner in the LPS-stimulated RAW 264.7 macrophages with *E. croceum* inhibiting NO production by 71.1 % at a concentration of 30.0 µg/ml. The extract exhibited good activities against 15-lipoxygenase activity with IC₅₀ value of 26.2 µg/ml which was better than IC₅₀ value of 53.7 µg/ml exhibited by the positive control quercetin [25].

Mamba *et al* [17] evaluated anti-inflammatory activities of ethanol extract of *E. croceum* bark by assessing the inhibitory effects on the pro-inflammatory enzyme, 15-lipoxygenase (15-LOX) with quercetin as a positive control. The extract exhibited activities with IC₅₀ values of 80.2 µg/ml while quercetin, the positive control exhibited IC₅₀ value of 48.9 µg/ml [17]. Odeyemi and Afolayan [24] evaluated anti-inflammatory activities of acetone extracts of *E. croceum* leaf and stem bark using the protein denaturation assay with diclofenac as a positive control. The extracts showed activities with leaf and stem bark extracts exhibiting IC₅₀ values of 0.9 mg/ml and 1.9 mg/ml, respectively while diclofenac, the positive control exhibited IC₅₀ value of 0.3 mg/ml [24]. Olaokun *et al* [32] evaluated the anti-inflammatory activities of acetone extracts of *E. croceum* bark by assessing the inhibitory effects on the pro-inflammatory enzyme, 5-lipoxygenase (5-LOX) with quercetin as a positive control. The extract inhibited the activity of 5-lipoxygenase with an IC₅₀ value 75.5 µg/ml [32]. These findings show the potential of *E. croceum* as herbal medicine for inflammatory conditions, pain and wounds.

Antioxidant activity

Elisha *et al* [25] evaluated the antioxidant activities of acetone extract of *E. croceum* leaves using the 2,2-diphenyl-1-picrylhydrazyl (DPPH) free radical scavenging, 2,2'-azinobis-3-ethylbenzothiazoline-6-sulphonate (ABTS), ferric reducing power (FRAP) and the trolox equivalent antioxidant capacity (TEAC) assays with ascorbic acid and trolox as reference drugs. The extracts showed good ABTS and DPPH activities with IC₅₀ values of 3.1 µg/mL and 7.7 µg/mL, respectively, which were comparable to IC₅₀ values ranging from 2.9 µg/mL to 5.6 µg/mL exhibited by ascorbic acid and trolox, the two positive controls. FRAP and TEAC values were 1.0 and 1.3, respectively while ascorbic acid and trolox showed values ranging from 1.0 to 3.7 [25].

Odeyemi and Afolayan [24] evaluated antioxidant activities of acetone extracts of *E. croceum* leaf and stem bark using the DPPH free radical scavenging, ABTS and FRAP assays with ascorbic acid, butylated hydroxytoluene (BHT) and rutin as reference drugs. The leaf extract IC₅₀ values for ABTS assay was 0.09 mg/ml, DPPH (0.1 mg/ml) and FRAP (2.5 mg/ml) while bark extract IC₅₀ values for ABTS assay was 0.2 mg/ml, DPPH (0.07 mg/ml) and FRAP (9.2 mg/ml). The IC₅₀ values exhibited by the reference drugs, ascorbic acid, BHT and rutin ranged from 0.02 mg/ml to 3.5 mg/ml [24]. The

documented antioxidant activities of the extracts of *E. croceum* are probably a result of flavonoids and phenolics which have been isolated from the species [24,25].

Cytotoxic activity

Prinsloo *et al* [20,23] evaluated the cytotoxicity of semi-purified stem bark extracts of *E. croceum* using the colorimetric MTT assay on Vero cells and scored as percentage reduction at 540nm of treated culture versus untreated control culture. The extract showed toxicity of only 20% at a concentration of 25 µg/ml on Vero cells with a therapeutic index of 250 [20,23]. Elisha *et al* [28] evaluated cytotoxicity activities of acetone extract of *E. croceum* leaves on Vero monkey kidney cells using the MTT reduction assay with doxorubicin as a positive control. The extract showed activities with median lethal concentration (LC₅₀) value of 5.2 µg/ml and selectivity index (SI) value of 0.02 while doxorubicin, the positive control showed LC₅₀ value of 8.3 µg/ml [28].

Elisha *et al* [30] evaluated cytotoxic activities of acetone extract of *E. croceum* leaves using MTT reduction assay on Vero cells. The extract showed activities with LC₅₀ value of 5.2 µg/ml and selectivity index (SI) value ranging from 0.01 to 0.07 while doxorubicin, the positive control showed LC₅₀ value of 8.3 µg/ml [30]. The extract exhibited high cytotoxicity, supporting findings by Yelani *et al* [21] who argued that cytotoxicity of *E. croceum* on Vero cells is due to toxic triterpenoids 20-hydroxy-20-epitingenone, tingenone, tingenine B, 11-hydroxy-amyrin and naringenin isolated from the leaves of the species.

Despite the long use of *E. croceum* as herbal medicine in South Africa and Swaziland to treat numerous human diseases, the roots of the species are reported to have been used to cause death by traditional healers [33,34]. Research by Kunene *et al* [35] showed that *E. croceum* is categorized as detrimental and causing death of cattle in KwaZulu Natal province, South Africa. Experiments upon rabbits found that 2.5 g per kg body weight, and 10 g of fresh leaves per kg body weight caused death fifteen minutes after administration [36]. Based on these toxicological reports, it can be inferred that *E. croceum* should be taken with caution when used as herbal medicine.

FINAL REMARKS

Pharmacological studies carried out so far showed that the different extracts and

constituents of the species to be promising, which calls for detailed phytochemical, pharmacological and clinical studies of different extracts and chemical constituents of the species. Determination of the active principles is very important in the extracts and compounds of the species in order to avoid life-threatening complications as the species is widely used as herbal medicine. There is lack of information about the effect of prolonged consumption of *E. croceum* extracts. Therefore, future studies should include the identification of toxic compounds, possible side effects caused by taking *E. croceum* as herbal medicine, and mechanisms of how potential toxic components of the species can be managed.

DECLARATIONS

Acknowledgement

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Conflict of Interest

No conflict of interest associated with this work.

Contribution of Authors

The authors declare that this work was done by the authors named in this article and all liabilities pertaining to claims relating to the content of this article will be borne by them.

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