
Flos Calendulae

Definition

Flos Calendulae consists of the dried ligulate florets or composite flowers of *Calendula officinalis* L. (Asteraceae) (1–3).

Synonyms

Asteraceae are also known as Compositae.

Selected vernacular names

Atunjaq, calendula, Chinese safflower, cuc kim tiên, djamir, djomaira, feminell, flamenquilla, fleur de calandule, fleur de souci, fleur de souci officinal, fleurs de tous les mois, garden marigold, gold-bloom, Goldblume, gole hamisheh bahar, hen and chickens, Körömvirag, lellousha, maravilla, marigold, mary-bud, ok-hhawan, pot marigold, qaraqus, qawqhan, quaqahan, ringflower, Ringelblüten, saialill, sciure'e Sant'antonio, souci, souci des jardins, tabsoult, toukinsenka, tousslat, uchu k'aspa, virreina, xu xi, zergul zerzira, zobeida, zubaydah (4–7).

Geographical distribution

Indigenous to central, eastern and southern Europe. Cultivated commercially in North America, the Balkans, Eastern Europe and Germany (6, 8).

Description

An annual herb, much branched from the base, very aromatic, up to 0.3–0.6m high; stem angular, hairy and solid. Leaves sessile, light green, with semi-amplexicaul base; entire, undulate or remotely denticulate; glandular hairs on both surfaces; lower leaves spatulate, obtuse, sometimes acute at the apex, 10–20cm long and 1–4cm wide; higher leaves oblong and mucronate, 4–7 cm long. Involucral bracts 7–15mm long, covered with long, glandular hairs; inner involucral bracts with pellucid, scarious margin; marginal flowers in cultivated plants often multi-seriate; corolla oblong-spatulate, bright yellow or orange, 15–25 mm long and 3mm wide, 1–3-toothed with 4 or 5 veins, marginally entire, covered at the base with patent, long, thick hairs; corolla of disc flowers rounded, 3-dentate top, 1.5–2.5 cm long and 4–7 mm in diameter, 5 mm long

tube and moderately widened limb. Stigma short, thick, hairy; ovary oblong, 0.5 mm in length, pubescent, shrivelling after anthesis. Achenes narrowly oblong, strongly curved, faintly ribbed, thinly pubescent or glabrous, 10–12 mm long, outer achenes warty-ribbed outside, inner achenes prickly-warty, often with broad, thick margins (2, 7, 9).

Plant material of interest: dried ligulate florets and composite flowers

General appearance

Ligulate florets consist of a yellow, orange or orange-yellow ligule, 3–5 mm wide and about 7 mm in the middle part, with 3-toothed apex and hairy, partly sickle-shaped, yellowish-brown to orange-brown tube with projecting style and 2-lobed stigma; occasionally with a partly bent yellowish-brown to orange-brown ovary. Tubular florets about 5 mm long, consist of yellow, orange-red or red-violet 5-lobed corolla and yellowish-brown or orange-brown tube, hairy in its lower part, mostly with a bent yellowish-brown to orange-brown ovary (1).

Organoleptic properties

Odour: faint, pleasantly aromatic (10, 11); taste: bitter (2).

Microscopic characteristics

Inner epidermal cells of ray floret elongated, rectangular and almost straight-walled, cuticle faintly striated; stomata absent; outer epidermal cells similar, but with 3 or 4 anomocytic stomata; trichomes very numerous on the tube, biseriate; stigma epidermal cells straight-walled, polygonal. In disc floret, outer epidermal cells elongated, straight or slightly sinuous-walled, stomata absent; abundant trichomes on area below point of insertion of the stamens, mainly glandular, uniseriate or biseriate. Within the upper part of the anthers, a layer of isodiametric to elongated, moderately thick-walled, lignified and pitted cells; pollen grains spherical, up to 45 µm in diameter, with 3 germinal pores, exine finely granular with numerous short spines; apex of stigma covered by short, bulbous papillae (2).

Powdered plant material

Yellow-green; fragments of corollas containing light yellow oil droplets; some corollas with fairly large anomocytic stomata, others containing prismatic and very small clusters of calcium oxalate crystals. Covering trichomes biseriate, multicellular and conical; glandular trichomes with a uniseriate or biseriate, multicellular stalk and a large, ovoid, biseriate, multicellular head. Spherical

pollen grains up to 45 µm in diameter, exine finely granular with numerous short spines and with 3 germinal pores; occasional fragments of stigmas with short, bulbous papillae (1).

General identity tests

Macroscopic and microscopic examinations, and thin-layer chromatography for flavonoid content (1, 2).

Purity tests

Microbiological

Tests for specific microorganisms and microbial contamination limits are as described in the WHO guidelines on quality control methods for medicinal plants (12).

Foreign organic matter

Not more than 5% bracts and not more than 2% other foreign matter (1, 2).

Total ash

Not more than 10% (1, 2).

Acid-insoluble ash

Not more than 2% (2).

Water-soluble extractive

Not less than 20% (2).

Loss on drying

Not more than 10% (1).

Pesticide residues

The recommended maximum limit of aldrin and dieldrin is not more than 0.05 mg/kg (13). For other pesticides, see the *European pharmacopoeia* (13), and the WHO guidelines on quality control methods for medicinal plants (12) and pesticide residues (14).

Heavy metals

For maximum limits and analysis of heavy metals, consult the WHO guidelines on quality control methods for medicinal plants (12).

Radioactive residues

Where applicable, consult the WHO guidelines on quality control methods for medicinal plants (12) for the analysis of radioactive isotopes.

Other purity tests

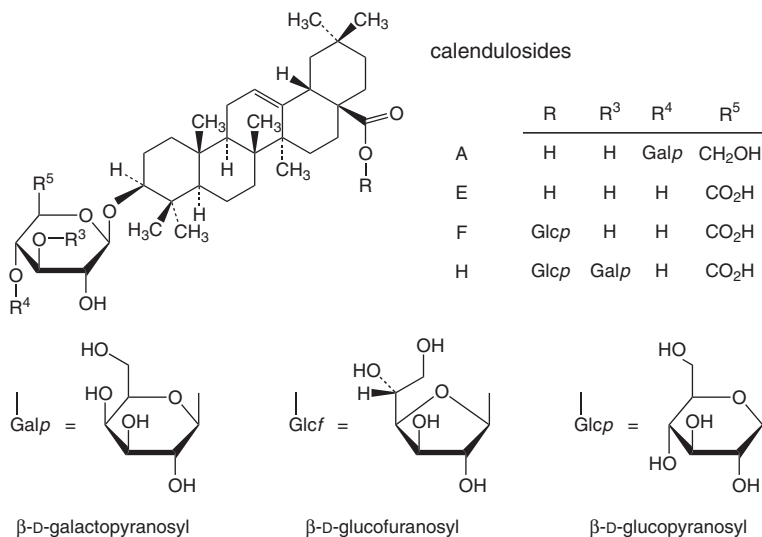
Chemical, sulfated ash and alcohol-soluble extractive tests to be established in accordance with national requirements.

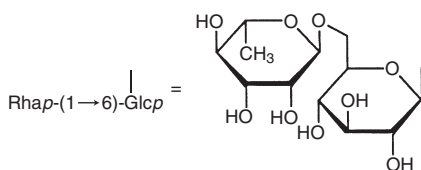
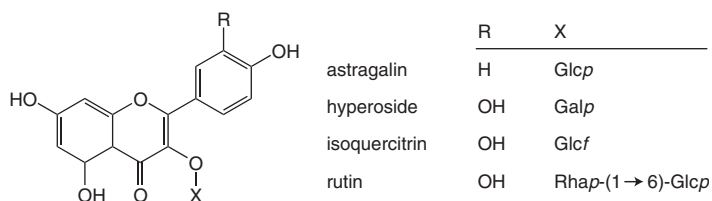
Chemical assays

Contains not less than 0.4% flavonoids, calculated as hyperoside, by spectrophotometry (1). A high-performance liquid chromatography method is also available (15).

Major chemical constituents

The major constituents are triterpene saponins (2–10%) based on oleanolic acid (i.e. calendulosides) and flavonoids (3-*O*-glycosides of isorhamnetin and quercetin), including astragalin, hyperoside, isoquercitrin and rutin. Other constituents include essential oil, sesquiterpenes (e.g. caryophyllene) and triterpenes (e.g. α - and β -amyrins, lupeol and lupenone) (5, 6, 16). Polysaccharides have also been reported (17). The structures of the characteristic triterpene saponins and flavonoids are presented below.



O-6-deoxy- α -L-mannopyranosyl-(1→6)- β -D-glucopyranosyl

Medicinal uses

Uses supported by clinical data

None.

Uses described in pharmacopoeias and in traditional systems of medicine

External treatment of superficial cuts, minor inflammations of the skin and oral mucosa, wounds and ulcus cruris (2, 18, 19).

Uses described in folk medicine, not supported by experimental or clinical data

Treatment of amenorrhoea, angina, fevers, gastritis, hypotension, jaundice, rheumatism and vomiting (2, 5, 6).

Pharmacology

Experimental pharmacology

Phagocytosis

Three polysaccharides isolated from an aqueous extract of Flos Calendulae enhanced phagocytosis in human granulocytes in vitro in the colloidal carbon clearance test (17). Intraperitoneal injection of a polysaccharide fraction isolated from an aqueous extract of the flowers to mice (10 mg/kg body weight) enhanced phagocytosis (20). Intraperitoneal administration of an unsaponifiable fraction (0.5 ml) of a hydroalcoholic extract of the flowers weakly stimulated phagocytosis in mice inoculated with *Escherichia coli*. However, the hydroalcoholic extract was not active (21).

Antimicrobial activity

The essential oil of the flowers inhibited the growth in vitro of *Bacillus subtilis*, *Escherichia coli*, *Staphylococcus aureus*, *Pseudomonas aeruginosa* and *Candida albicans* (22). A flavonoid fraction isolated from the flowers inhibited the growth in vitro of *S. aureus*, *Sarcina lutea*, *E. coli*, *Klebsiella pneumoniae* and *Candida monosa* (23). However, chloroform, ethanol, methanol or water extracts of the flowers did not inhibit bacterial growth in vitro (24–26). Acetone, ethanol or water extracts inhibited the growth in vitro of the fungus *Neurospora crassa* (27). Extracts of the flowers inhibited the growth in vitro of *Trichomonas vaginalis* (28). Oxygenated terpenes appear to be responsible for the antimicrobial activity (29).

Antiviral activity

A tincture of the flowers suppressed the replication of herpes simplex, influenza A2 and influenza APR-8 viruses in vitro (30). However, an aqueous extract of the flowers was not active (31). A chloroform extract of the flowers inhibited the replication of HIV-1 in acutely infected lymphocytic MOLT-4 cells in vitro (IC₅₀ 0.4 mg/ml) (32). A chloroform extract also inhibited HIV-1 reverse transcriptase activity in a dose-dependent manner (ED₅₀ 51.0 µg/ml) (32). A 5% hot aqueous extract of the flowers (2 ml) inhibited the replication of encephalitis virus after intraperitoneal administration to mice (33).

Anti-inflammatory activity

Topical application of a 70% ethanol extract of the flowers to mice at a dose of 1.2 mg/ear (corresponding to 4.16 mg crude drug) reduced croton oil-induced ear oedema by 20% (34). External application of a carbon dioxide extract of the flowers (300 µg/cm²) suppressed croton oil-induced ear oedema in mice (34). The triterpene fraction of an extract of the flowers had marked anti-inflammatory activity in mice (1 µg/ear) against ear oedema induced by 12-O-tetradecanoylphorbol-13-acetate (35). Faradiol esters isolated from the flowers (240 µg/cm²) inhibited croton oil-induced ear oedema in mice (36). Intra-gastric administration of an aqueous extract of the flowers (100 mg/kg body weight) inhibited carrageenan-induced footpad oedema in rats (37). However, an 80% ethanol extract of the flowers was weakly active (11% inhibition) at a concentration of 100 mg/kg body weight administered orally 1 hour prior to induction of oedema (38). Isorhamnetin glycosides isolated from the flowers inhibited rat lung lipoxygenase in vitro (39).

Wound-healing activity

External application of a hydroalcoholic extract accelerated the rate of contraction and epithelialization of excision wounds in rats (40). A 3% freeze-dried aqueous extract of the flowers induced vascularization in the chick chorioallantoic membrane assay. Histological sections of the treated chorioallantoic

membranes also indicated the presence of hyaluronan, a tissue glycosaminoglycan associated with neovascularization (41).

Clinical pharmacology

Although no randomized, controlled clinical trials have been performed, two case reports in the early medical literature support the traditional use of *Flos Calendulae*. The reports describe the use of a strong tincture of the flowers applied on compresses to reduce inflammation and suppuration, and to accelerate the healing of wounds (42, 43). These reports may be of historical value only.

Contraindications

Flos Calendulae is contraindicated in cases of known allergy to plants of the Asteraceae (Compositae) family (18).

Warnings

No information available.

Precautions

Carcinogenesis, mutagenesis, impairment of fertility

Saponins isolated from *Flos Calendulae* were not mutagenic at a concentration of 400 µg/ml in the *Salmonella*/microsome assay using *S. typhimurium* strain TA98, with or without S9 metabolic activation (44). Extracts of the flowers were not carcinogenic after daily intragastric administration of 0.15 g/kg body weight to rats (for 22 months) or hamsters (for 18 months) (45). Mutagenicity testing of the fluidextract in the *Salmonella*/microsome assay (using *S. typhimurium* strains TA98, TA100, TA1535 and TA1537) was negative at concentrations of up to 5 mg/plate. The mouse bone marrow micronucleus test was also negative after daily administration of up to 1 g/kg body weight for 2 days (46). A fluidextract of the flowers (100 mg/ml, 60% ethanol) was genotoxic in both mitotic crossing-over and chromosome segregation when assayed for mitotic segregation in the heterozygous diploid D-30 of *Aspergillus nidulans* (46).

Other precautions

No information available on general precautions or precautions concerning drug interactions; drug and laboratory test interactions; teratogenic and non-teratogenic effects in pregnancy; nursing mothers; or paediatric use. Therefore, *Flos Calendulae* should not be administered during pregnancy or lactation or to children without medical supervision.

Adverse reactions

Weak skin-sensitization has been reported (47).

Dosage forms

Infusion for topical use; aqueous and alcohol extracts, tinctures and ointment for external use (2, 18, 19). Store in a well-closed container, protected from light (1).

Posology

(Unless otherwise indicated)

Topical application: an infusion of 1–2 g/150 ml (18). External use: a 40% alcohol extract (1:1), or tincture (1:5) in 90% alcohol (2). For the treatment of wounds, the tincture is applied undiluted; for compresses, the tincture is usually diluted at least 1:3 with sterile water (18, 48, 49). Ointment: 2–5% (48, 50).

References

1. *European pharmacopoeia*, 3rd ed., Suppl. 2000. Strasbourg, Council of Europe, 1999.
2. *British herbal pharmacopoeia*. London, British Herbal Medicine Association, 1996.
3. *Pharmacopoeia Hungarica*, 7th ed. Budapest, Hungarian Pharmacopoeia Commission, Medicina Konyvkiado, 1986.
4. Boulos L. *Medicinal plants of North Africa*. Cairo, Reference Publications, 1983.
5. Farnsworth NR, ed. *NAPRALERT database*. Chicago, University of Illinois at Chicago, IL, January 28, 1998 production (an online database available directly through the University of Illinois at Chicago or through the Scientific and Technical Network [STN] of Chemical Abstracts Services).
6. Bisset NG. *Herbal drugs and phytopharmaceuticals*. Boca Raton, FL, CRC Press, 1994.
7. Youngken HW. *Textbook of pharmacognosy*, 6th ed. Philadelphia, PA, Blakiston, 1950.
8. Leung AY, Foster S. *Encyclopedia of common natural ingredients used in food, drugs, and cosmetics*, 2nd ed. New York, NY, John Wiley & Sons, 1996.
9. Backer CA, Van den Brink B. *Flora of Java. Vol. 2*. Noordflog-Groningen, NVP, 1965: 574.
10. *Pharmacopoea helvetica*, 8th ed. Berne, Département fédéral de l'intérieur, 1997.
11. *Pharmacopée française*. Paris, Adrapharm, 1996.
12. *Quality control methods for medicinal plant materials*. Geneva, World Health Organization, 1998.
13. *European pharmacopoeia*, 3rd ed. Strasbourg, Council of Europe, 1996.
14. *Guidelines for predicting dietary intake of pesticide residues*, 2nd rev. ed. Geneva, World Health Organization, 1997 (document WHO/FSF/FOS/97.7).
15. Vidal-Ollivier E et al. Dosage par CLHP. Des flavonoides majoritaires de *Calendula officinalis* L. En fonction de la variété culturale et de la date de récolte. *Pharmaceutica Acta Helveticae*, 1991, 66:318–320.
16. Bruneton J. *Pharmacognosy, phytochemistry, medicinal plants*. Paris, Lavoisier, 1995.
17. Varljen J, Lipták A, Wagner H. Structural analysis of a rhamnoarabinogalactan and arabinogalactans with immunostimulating activity from *Calendula officinalis*. *Phytochemistry*, 1989, 28:2379–2383.
18. *ESCOPE monographs on the medicinal uses of plant drugs*. Fascicule 1. Elburg, European Scientific Cooperative on Phytotherapy, 1996.
19. Blumenthal M et al., eds. *The complete German Commission E monographs*. Austin, TX, American Botanical Council, 1998.

20. Wagner H et al. Immunstimulierend wirkende Polysaccharide (Heteroglykane) aus höheren Pflanzen. *Arzneimittel-Forschung*, 1985, 7:1069–1075.
21. Delaveau P et al. Drogues végétales stimulant l'activité phagocytaire du système réticulo-endothélial. *Planta Medica*, 1980, 40:49–54.
22. Janssen AM et al. Screening for antimicrobial activity of some essential oils by the agar overlay technique. *Pharmaceutisch Weekblad*, 1986, 8:289–292.
23. Tarle D, Dvorzak I. Antimicrobial substances in Flos Calendulae. *Farmaceutski Vestnik (Ljubljana)*, 1989, 40:117–120.
24. Rios JL, Recio MC, Villar A. Antimicrobial activity of selected plants employed in the Spanish Mediterranean area. *Journal of Ethnopharmacology*, 1987, 21:139–152.
25. Dornberger K, Lich H. Screening for antimicrobial and presumed cancerostatic plant metabolites. *Pharmazie*, 1982, 37:215–221.
26. Acevedo JG, Lopez JL, Cortes GM. In vitro antimicrobial activity of various plant extracts used by purepecha against some Enterobacteriaceae. *International Journal of Pharmacognosy*, 1993, 31:61–64.
27. Kubas J. Investigations on known or potential antitumoral plants by means of microbiological tests. Part III. Activity of some cultivated plant species in *Neurospora crassa* test. *Acta Biologica Cracoviensia Series Botanica*, 1972, 15:87–100.
28. Racz G et al. Trichomonocidal and anthelmintic activity of Roumanian folkloric plants. *Planta Medica*, 1980, 39:257A.
29. Gracza L. Oxygen-containing terpene derivatives from *Calendula officinalis*. *Planta Medica*, 1987, 53:227.
30. Bogdanova NS et al. Study of antiviral properties of *Calendula officinalis*. *Farmakol Toksikol (Moscow)*, 1970, 33:349.
31. May G, Willuhn G. Antiviral activity of aqueous extracts from medicinal plants in tissue cultures. *Arzneimittel-Forschung*, 1978, 28:1–7.
32. Kalvatchev Z et al. Anti-HIV activity of extracts from *Calendula officinalis* flowers. *Biomedicine and Pharmacotherapy*, 1997, 51:176–180.
33. Fokina GI et al. Experimental therapy of tick-borne encephalitis. *Soviet Progress in Virology*, 1991, 1:27–31.
34. Della-Loggia R et al. The role of triterpenoids in the topical anti-inflammatory activity of *Calendula officinalis* flowers. *Planta Medica*, 1994, 60:516–520.
35. Akihisa T et al. Triterpene alcohols from the flowers of Compositae and their anti-inflammatory effects. *Phytochemistry*, 1996, 43:1255–1260.
36. Zitterl-Eglseer K et al. Anti-oedematous activities of the main triterpenediol esters of marigold (*Calendula officinalis* L.). *Journal of Ethnopharmacology*, 1997, 57:139–144.
37. Peyroux J et al. Anti-oedemic and anti-hyperhaemic properties of *Calendula officinalis* L. *Plantes médicinales et Phytothérapie*, 1981, 15:210–216.
38. Mascolo N et al. Biological screening of Italian medicinal plants for anti-inflammatory activity. *Phytotherapy Research*, 1987, 1:20–31.
39. Bezakova L et al. Inhibitory activity of isorhamnetin glycosides from *Calendula officinalis* L. on the activity of lipoxygenase. *Pharmazie*, 1996, 51:126–127.
40. Rao SG et al. *Calendula* and *Hypericum*: two homeopathic drugs promoting wound healing in rats. *Fitoterapia*, 1991, 62:508.
41. Patrick KFM et al. Induction of vascularisation by an aqueous extract of the flowers of *Calendula officinalis* L., the European marigold. *Phytomedicine*, 1996, 3:11–18.
42. Livezey A. Some observations on our indigenous medical flora. *Medical and Surgical Reporter*, 1868, 19:85.
43. Reynolds RG. *Calendula*. *Pacific Medical and Surgical Journal*, 1886, 29:720.
44. Elias R et al. Antimutagenic activity of some saponins isolated from *Calendula officinalis* L., *C. arvensis* L. and *Hedera helix* L. *Mutagenesis*, 1990, 5:327–331.
45. Avramova S et al. Source of new products for the cosmetic industry. *Medical and Biological Information*, 1988, 4:28–32.

WHO monographs on selected medicinal plants

46. Ramos A et al. Genotoxicity of an extract of *Calendula officinalis* L. *Journal of Ethnopharmacology*, 1998, 61:49–55.
47. Bruynzeel DP et al. Contact sensitization by alternative topical medicaments containing plant extracts. *Contact Dermatitis*, 1992, 27:278–279.
48. Willuhn G. Pflanzliche Dermatika, eine kritische Übersicht. *Deutsche Apotheker Zeitung*, 1992, 132:1873–1883.
49. Van Hellemond J. *Fytotherapeutisch compendium*, 2nd ed. Utrecht, Bohn, Scheltema & Holkema, 1988:113–114.
50. Hänsel R et al., eds. *Hagers Handbuch der pharmazeutischen Praxis. Bd. 4: Drogen A–K*, 5th ed. Berlin, Springer-Verlag, 1994.