MORPHOLOGICAL, MICROMORPHOLOGICAL AND ANATOMICAL STUDIES OF CHENOPODIUM ALBUM COMPLEX IN IRAN

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In Flora Iranica *Chenopodium album* L. includes two subspecies as *Ch. album* subsp. *album* and *Ch.. album* subsp. *iranicum*. In Flora of Iran *Chenopodium album* includes three subspecies as *Ch. album* subsp. *album, Ch. album* subsp. *iranicum* and *Ch. album* subsp. *striatum.* In this research *Chenopodium album* subsp. *iranicum* is transferred to a higher level as *Chenopodium iranicum*. This combination is made on the base of results of morphological studies, and SEM studies of the seed coat surface, ornamentation of pollen grain, leaf and stem anatomy.

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ریخت‌شناسی، ریزریخت‌شناسی و بررسی‌های تشریحی گروه آرایه‌های متعلق به گونه Chenopodium album

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در فلور ایرانیکا گونه *Chenopodium album* دارای دو زیر گونه *Ch. album* subsp. *album* و *Ch. album* subsp. *iranicum* است. در فلور ایران *Chenopodium album* دارای سه زیر گونه *Ch. album* subsp. *album* ، *Ch. album* subsp. *iranicum* و *Ch. album* subsp. *striatum* است. زیر گونه *Ch. album* subsp. *striatum* مترادف *Ch. strictum* می‌باشد. در این تحقیق زیر گونه *Chenopodium album* subsp. *iranicum* به سطح گونه ارتقاء می‌یابد. این ترکیب بر اساس نتایج حاصل از مطالعات مورفولوژی، میکرومورفولوژی سطح دانه، تزئینات دانه گرده، ساختار تشریحی برگ و ساقه صورت گرفته است.

Introduction

The genus *Chenopodium* as an annual herb has 15 species in Iran (Assadi 2001). The genus in Flora of the USSR (Iljin 1936) includes 30 species and 4 subspecies, 14 of them occur in Iran. In Flora of Turkey (Aellen 1967) it contains 11 species of which 9 species occur in Iran. In Flora Iranica (Uotila 1997) 16 species are recorded from Iran. In this Flora, *Chenopodium album* L. has two subspecies as follows: *Ch. album* subsp. *album* and *Ch. album* subsp. *iranicum* Aellen, *Chenopodium strictum* Roth is also regarded as a distinct species. While Assadi (2001) in Flora of Iran reduces the last species *Ch. strictum* to subspecific level as *Ch. album* subsp. *striatum* (Krasan) Murr in Urban & Graebner aside *Ch. album* subsp. *album* and *Ch. album* subsp. *iranicum*. The aim of this research is to study the *Chenopodium album* complex on the base of morphology, anatomy, seed micromorphology (Malekloo & al. 2008) and pollen characters (Hamdi & al. 2008). *Ch. opulifolium* Schrader ex Koch & Ziz is included in the studies as a close affinity.

Table 1: List of *Chenopodium* specimens used in the studies.

Taxa	Localties
Chenopodium iranicum (Aellen) Hamdi & Malekloo	Tehran prov.: Lar area, 2400 m , Malekloo, 2432 ; Tehran, between Tehran and Karaj, Botanical Garden, 1320 m, Assadi 76806; Karaj, Kalak, 1600 m, Mousavi, 2774 (IRAN). Semnan prov.: Shahroud, Rechinger, Allen and Esfandiari 5377 (IRAN).
Chenopodium album L. subsp. *album*	Tehran prov.: East-north of Tehran, Sorkheh Hesar, 1400 m, Malekloo, 5702 (IAUGH); Tehran, East, Ahar, Tangeh deh, Malekloo, 5703 (IAUGH); Tehran, Chitgar Park, 1320 m, Malekloo 5704 (IAUGH); Tehran, Shahr Park, 1150 m, Malekloo 5705. Mazandaran prov.: Kiasar forest, 75 km from Sari toward Semnan, 120 m, Malekloo 5709 (IAUGH).
Chenopodium album subsp. *striatum* (Krasan) Murr in Urban & Graebner.	Mazandaran prov.: Miankaleh, Ashorzadeh 2229 (IAUGH); Amol, Baladeh To Amol, 1.5 km east of Razen to Taker, 1610 m, Butttler and Botmer 22886 (TARI); Pole-Sefid, Sangdeh, 1280 m, Assadi and Azadi, 76000 (TARI). Tehran prov.: Karaj, Chalous road, Dareh-Nesa, Malakloo 2807(IAUGH).
Chenopodium opulifolium Schrader ex Koch & Ziz	Tehran prov.: between Tehran and Karaj, Botanical Garden, 1320 m, Assadi 76802 (TARI); Tehran, East, Lavasan, 1900 m, Malekloo 5707 (IAUGH); Tehran, East-north of Tehran, Sorkheh Hesar, 1400 m, Malekloo 5706 (IAUGH); Tehran, 30 km Tehran to Qom, 1150 m, Malekloo, 5708 (IAUGH).

Materials and methods

This study was mainly based on plant material deposited in different Iranian herbaria, namely: the herbarium of Islamic Azad University of Garmsar, FUMH, TARI and IRAN (abbreviations according to Holmgren & al. 1990). Several field trips have also been conducted in different parts of Iran and the specimens collected were similarly deposited in the same herbaria as above. Measurements of vegetative and floral parts as well as from the seeds were carried out under a stereomicroscope (Olympus SZH). Pollen grains and seeds of four taxa of the genus *Chenopodium* were studied by scanning electron microscope (SEM). Samples were obtained mostly from fresh collected herbarium specimens. The voucher specimens and part of the studied materials are deposited in above mentioned herbaria and they are listed in table 1. The names of taxa are according to the results of this study.

For SEM, we used the protocol explained by Davies (1999) with some modifications. The specimens were mounted on 12.5 mm diameter stubs and attached with sticky tabs and then coated in a sputter coater with approximately 25 µm of Gold- Paladium. The specimens were examined and photographed by a LEO scanning electron microscope (SEM) model 440 I, at an accelerating voltage of 10 15 kv. Cross-section of exine and seeds was also examined. The number of tecta perforations (according to Punt et al. 2007) per 25 µm² and length of larger perforations in proximal face and distal face were measured. The terminology used for describing the pollen grains features followed in general Moore & al. (1991), Mc Andrews & Swanson (1967), Tsukada (1967) and Punt & al. (1994, 1999). For Anatomy studies, 5 samples used for every species and we used the protocol explained by Metcalfe (1950). The preparates were studied using an optical microscope Nikon modle ALPHAPHOT- 2YS2 and photographed with a Canon A 630 camera.

Results

Based on the studies, characters of the taxa are explained as follows and summarized in table 2.

Chenopodium iranicum (Aellen) Hamdi & Malekloo, **comb. nov.**

Syn.: *Chenopodium album* subsp. *iranicum* Aellen, Notes Roy. Bot. Gard. Edinb. 28: 30 (1967).

Stem TS: pentagonal, pustulate, vascular bundle 25, (Fig. 3). *Leaf:* Lamina of lower leaves 6 cm long; lamina shape orbicular-obtuse at the apex, irregularly toothed at the margin; petiole 10-20 mm long, having 4 vascular bundles. *Flowers:* Number of flowers in leaf axillary inflorescences 5-10. *Seed:* elliptic, semi-papillate in ornamentation; cells concave (Fig. 2). *Pollen*: Shape spherical; *Pollen* grains diameter 19.27 µm in equatorial view; Exine surface view perforate (diameter of holes on the exine is less than 1 µm²), scabrate; 5-6 conical tubercles on holes; pore numbers 100, 5-6 per 5 µm² (Fig. 1).

Table 2. Morphological and micromorphological characteristic features of Iranian representatives of *Chenopodium album* complex.

	Ch. iranicum	*Ch. album* subsp. *album*	*Ch. album* subsp. *striatum*	*Ch. opulifolium*
Stem	branched and prostrate	branched and erect	erect, simple	erect, simple
Blades length of lower leaves (cm)	6	less than 5	2-2.5	2.5-3.0
Blades shape of leaves	Orbicular, obtuse at the apex	elliptic-lanceolate, acute at the apex	elliptic-ovate, acute at the apex	deltoid
Leaf margin teeth	regular	irregular	irregular	Irregular
Petiole (mm)	10-20	15-30	20-50	maximum to 20
Number of flowers	5-10	10	5	5
Stem shape in cross section	pentagonal	pentagonal	pentagonal	orbicular
Number of vascular bundles of stem	25	19	18	15
Number of vascular bundles of petiole	4	5	5	3
Size of seed (mμ)	1070-1074×1029-1032	1056-1058×923-925	1036-1040×989-990	1076-1079×880-882
Size of surface cells of seed (mμ)	35-36×20-22	27-28×18-19	65-66×15-16	57-58×44-45
Ornamentation of seed	semi-papillate	papillate	papillate	papillate-smooth
Ornamentation of surface cells of seed	smooth-concave	granular	granular	smooth-convex
Shape of seed	elliptic	elliptic	elliptic	ovate
Shape of surface cells of seed	polygonal	irregularly pentagonal-hexagonal	irregularly pentagonal	irregularly polygonal
Seed hillum	central	central	axillary	central
Exine of pollen types	perforate	perforate	perforate	faveolate
No. of conical tubercles (5μm) on exine surface	25-30	90-95	60-65	45-50
Pores diameter of pollen surface (μm)	0.96-0.97	0.94-0.95	0.710-0.725	0.9-1.0
Equatorial polar pollen diameter (μm)	19.30	23.36	15.90	20.20
Pores number on pollen surface	100	92	60	88

Chenopodium album* L. subsp. *album

Stem TS*:* Pentagonal, pustulate; vascular bundles 19 (Fig. 3). *Leaf* TS*:* Mesophyl having 2 rows of spongy and 2 rows of palisadic parenchyma (Fig. 3). *Petiole.* Having 5 vascular bundles. *Seed:* elliptic, papillate; surface of cells concave (Fig. 2). *Pollen*: spherical shape, pollen grains diameter 15.90 μm in equatorial view size; Exine surface perforate (that diameter of holes on the exine is less than 1 μm²), 8-9 conical tubercles on holes; pore numbers 60, scabrate, 3-4 per 5 μm² (Fig. 1).

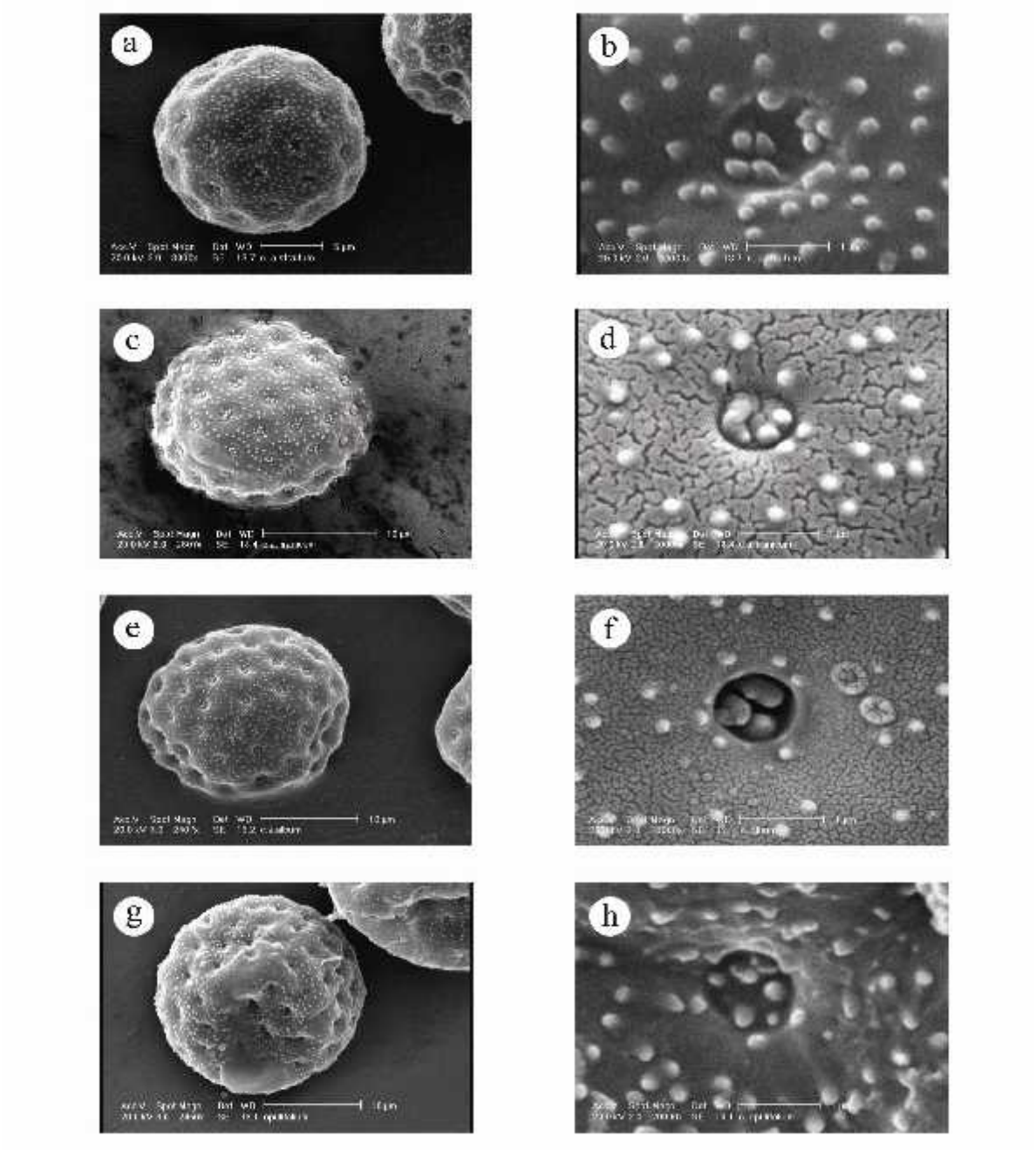

Fig. 1. Micrographs of pollen grains in *Chenopodium*. Fig. a-b, perforate tectum at the proximal face in *Ch. album* subsp. *striatum,* pollen with perforate ornamentation of exine. Fig. c-d, perforate tectum at the proximal face in *Ch. iranicum,* pollen with perforate ornamentation of exine. Fig. e-f, perforate tectum at the proximal face in *Ch. album* subsp. *album*, pollen with perforate ornamentation of exine. Fig. g-h, faveolate tectum at the proximal face in *Ch. opulifolium* pollen with faveolate ornamentation of exine. Scale bar = 10µm, Figs. c & e & g; Scale bar =1µm, Figs. b & d & f & h; Scale bar = 5µm, Fig. a.

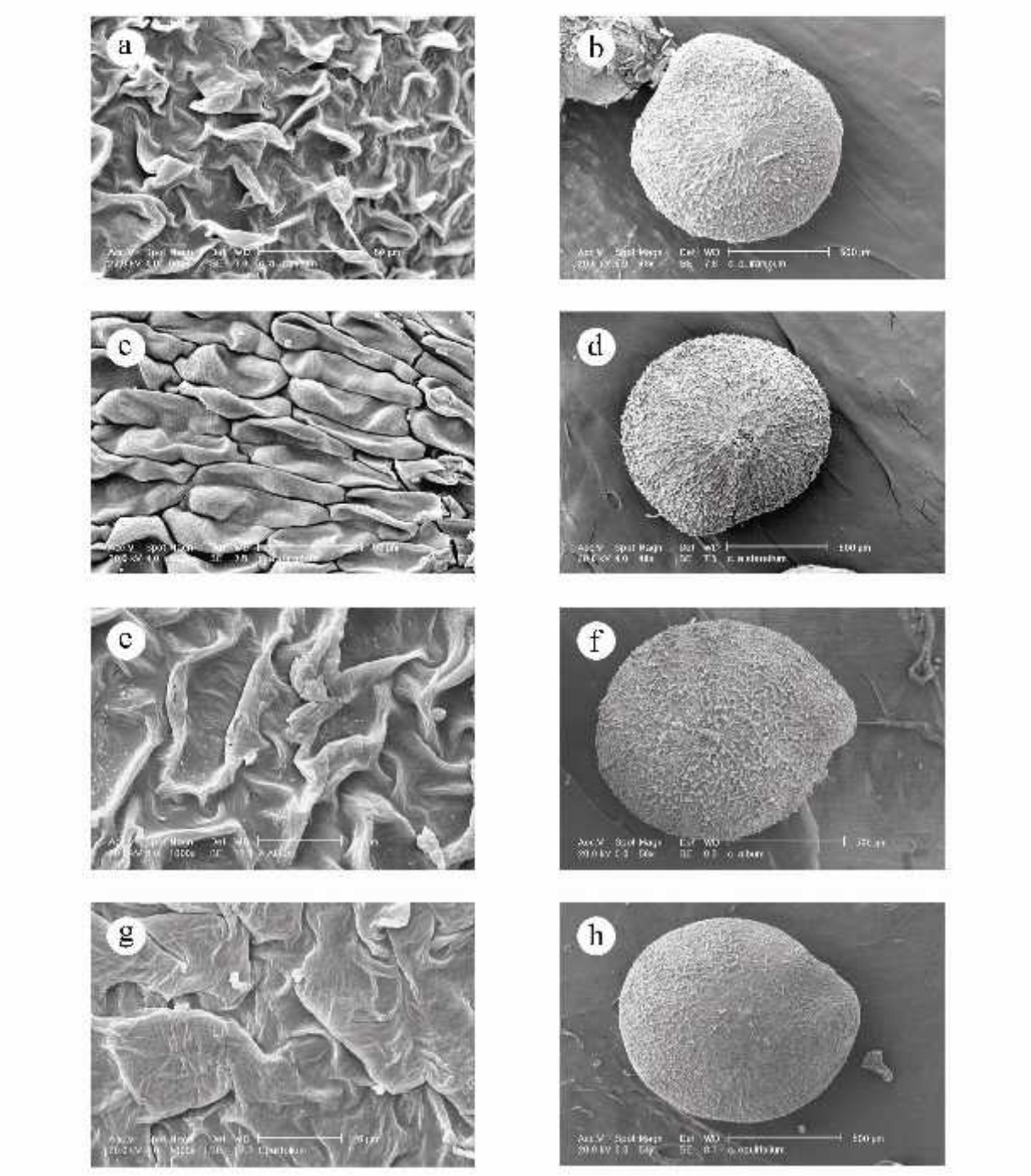

Fig. 2. Scanning electron micrograph of seeds of *Chenopodium*. a & b: *Ch. iranicum* (Malekloo 1234), overview (a), seed surface cells (b). c & d: *Ch. album* subsp. *striatum* (Malekloo 5678), overview (c), seed surface cells (d). e & f: *Ch. album* subsp. *album* (Malekloo 9012), overview (e), seed surface cells (f). g & h: *Ch. opulifolium* (Malekloo 3456), overview (g), seed surface cells (h). Scale bars: a=50µm, b=500µm, c=50µm, d=500µm, e=20µm, f=500µm, g=20µm, h=500µm.

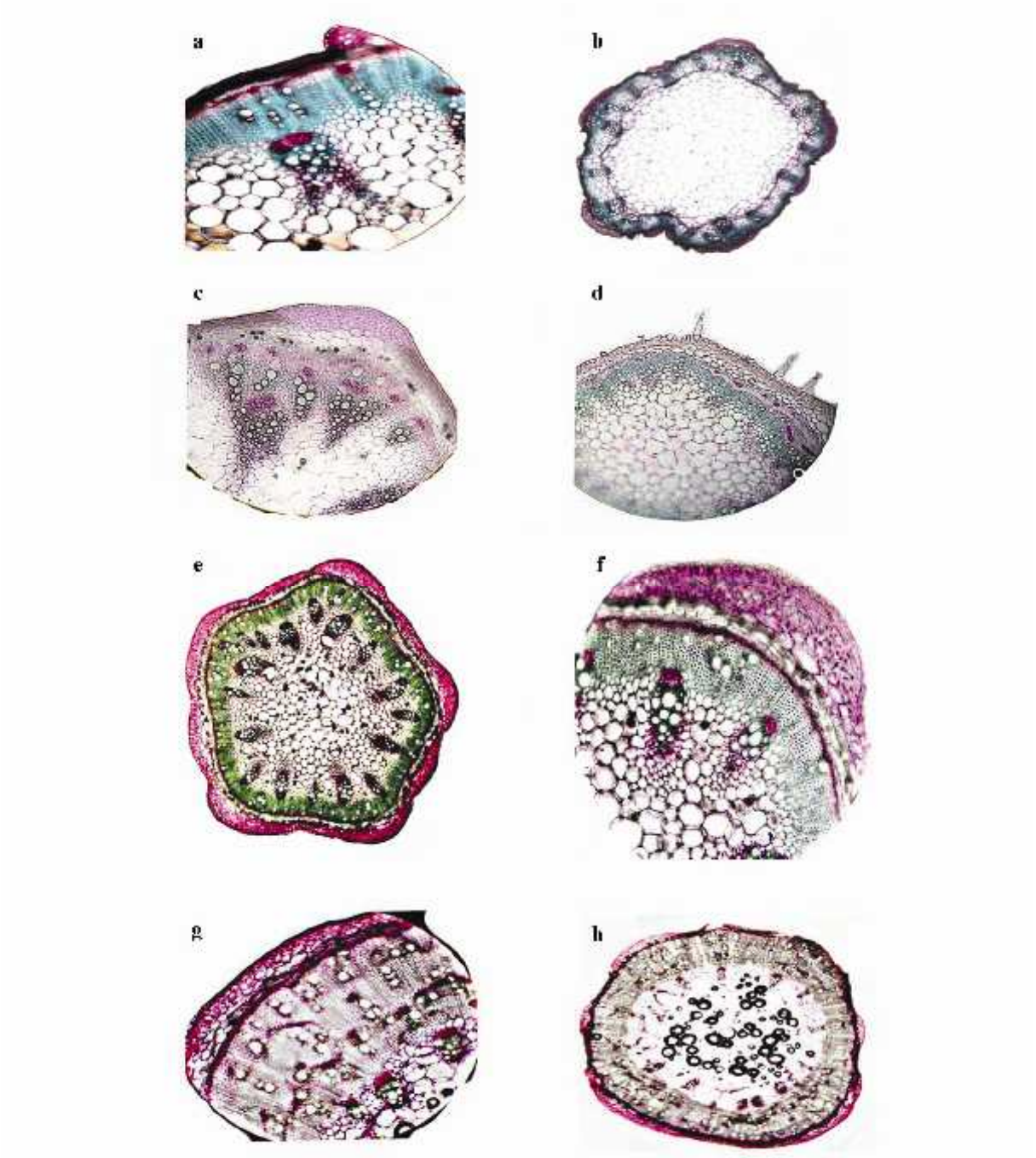

Fig. 3. Stem: a & b, *Chenopodium album* subsp *album* (2803); c &d, *Ch. iranicum* (2432); e & f, *Ch. album* subsp *striatum* (2229); g & h, *Ch. opulifolium* (2802).

Chenopodium album* subsp. *striatum (Krasan) Murr in Urban & Graebner.
Syn: *Chenopodium strictum* Roth, Nov. Pl. Praes. Ind. Or.: 180 (1821).
Stem TS: Pentagonal, pustulate; vascular bundles 18 (Fig. 3). *Leaf* TS. mesophyl having 3-4 spongy rows and 2 palisadic parenchima rows (Fig. 3). *Petiole*. Having 5 vascular bundles (Fig. 3). *Seed.*: Elliptic; hillum central, papillate; cell surface irregularly pentagonal; surface of cells granular (Fig. 2). *Pollen* : spherical shape; pollen grains diameter 20.20 µm in equatorial view size, Exine surface, Faveolate (that diameter of holes on the exine is more than 1 µm²), 5-7 conical tubercles on holes, pore numbers 88, scabrate, 3-4 pore per 5 µm² (Fig. 1).
Chenopodium opulifolium Schrader ex Koch & Ziz
Stem TS: Orbicular in cross section, vascular bundles 15 (Fig. 3). *Leaf* TS: Mesophyl having 3-4 spongy rows and 1 row palisadic parenchima. *Petiole*. Having 5 vascular bundles; middle midrib 3 vascular bundles. *Seed:* ovate, central hillum, shape of cells irregular, smooth-convex on the surface (Fig. 2). *Pollen*: spherical shape, pollen grains diameter 20.20 µm in equatorial view size, exine surface faveolate (that diameter of holes on the exine is more than 1 µm²), 5-7 conical tubercles on holes, pore numbers 88, scabrate, 3-4 pore per 5 µm² (Fig. 1).

Discussion

According to Flora Iranica (Uotila 1997) 16 species occur in Iran, in this Flora, *Chenopodium album* includes two subspecies as follows: *Ch. album* subsp. *album*, *Ch. album* subsp. *iranicum*, and *Ch. strictum* was regarded as a distinct species. In Flora of Iran (Assadi 2001) 15 species were recorded from Iran, for *Ch. album* 3 subspecies introduced as follows: *Ch. album* subsp. *album*, *Ch. album* subsp. *striatum* and *Ch. album* subsp. *iranicum*. In a taxonomical studies of the group including *Ch. opulifolium* using micro- and macromorphological characters we found that the three taxa are similar in stem cross section shape, petiole length and seed shape. *Ch. album* subsp. *iranicum* differs from the other subspecies in lower leaves length of blades, marginal teeth of leaves, blade shape of leaves, inflorescence, the number of flowers, ornamentation of seed, ornamentation of seed surface cells, shape of surface cells, seed hillum, numbers of conical tubercles on exine surface of pollen grains, pores diameter of pollen grains, equatorial polar pollen diameter and number of pores on pollen grains surface (Table 1, Figs. 1 & 2). Therefore, *Chenopodium album* subsp. *iranicum* highly differs from the other subspecies and deserve a higher rank as *Chenopodium iranicum*.

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