INTERNATIONAL STANDARD

ISO 9306

First edition
2023-11

Traditional Chinese medicine — *Ephedra sinica*, *Ephedra intermedia* and *Ephedra equisetina* herbaceous stem

Reference number
ISO 9306:2023(E)

ISO copyright office
CP 401 • Ch. de Blandonnet 8
CH-1214 Vernier, Geneva
Phone: +41 22 749 01 11
Email: copyright@iso.org
Website: www.iso.org

Published in Switzerland

Contents

Page

Foreword

ISO (the International Organization for Standardization) is a worldwide federation of national standards bodies (ISO member bodies). The work of preparing International Standards is normally carried out through ISO technical committees. Each member body interested in a subject for which a technical committee has been established has the right to be represented on that committee. International organizations, governmental and non-governmental, in liaison with ISO, also take part in the work. ISO collaborates closely with the International Electrotechnical Commission (IEC) on all matters of electrotechnical standardization.

The procedures used to develop this document and those intended for its further maintenance are described in the ISO/IEC Directives, Part 1. In particular, the different approval criteria needed for the different types of ISO document should be noted. This document was drafted in accordance with the editorial rules of the ISO/IEC Directives, Part 2 (see www.iso.org/directives).

ISO draws attention to the possibility that the implementation of this document may involve the use of (a) patent(s). ISO takes no position concerning the evidence, validity or applicability of any claimed patent rights in respect thereof. As of the date of publication of this document, ISO had not received notice of (a) patent(s) which may be required to implement this document. However, implementers are cautioned that this may not represent the latest information, which may be obtained from the patent database available at www.iso.org/patents. ISO shall not be held responsible for identifying any or all such patent rights.

Any trade name used in this document is information given for the convenience of users and does not constitute an endorsement.

For an explanation of the voluntary nature of standards, the meaning of ISO specific terms and expressions related to conformity assessment, as well as information about ISO's adherence to the World Trade Organization (WTO) principles in the Technical Barriers to Trade (TBT), see www.iso.org/iso/foreword.html.

This document was prepared by Technical Committee ISO/TC 249, *Traditional Chinese medicine*.

Any feedback or questions on this document should be directed to the user's national standards body. A complete listing of these bodies can be found at www.iso.org/members.html.

Introduction

Ephedra herbaceous stem is a commonly used Chinese herbal medicine with a long history. It is recorded in the classics of Chinese medicine: *Treatise on Febrile Diseases*, *Synopsis of the Golden Chamber*, *Shen Nong's Materia Medica* and *Compendium of Materia Medica*.[1] Its medicinal materials are derived from the dried herbaceous stem of *Ephedra sinica* Stapf, *Ephedra intermedia* Schrenk *et* C. A. Mey. and *Ephedra equisetina* Bge. (Fam. Ephedraceae).

Clinically, *Ephedra* herbaceous stem is mainly used to treat wind-cold cold, chest tightness, asthma and cough and feng shui oedema. Modern scientific research has found many types of chemical components of *Ephedra* herbaceous stem, including volatile oil, alkaloids, flavonoids and polysaccharides. Among these, alkaloids ephedrine and pseudoephedrine have the highest content and are also the most effective ingredients for medicinal value and drug manufacturing. Modern pharmacological studies have shown that the methanol extract of *Ephedra* herbaceous stem has an anti-inflammatory effect, ephedra decoction has the effect of anti-pathogenic microorganisms, the volatile oil in *Ephedra* herbaceous stem has a sweating effect and has an inhibitory effect on influenza viruses and ephedrine has the effect of raising blood pressure. It can excite the heart, constrict blood vessels and excite the central meridian. Pseudoephedrine has a noticeable diuretic effect and relieves bronchial smooth muscle spasms. *Ephedra* herbaceous stem is also a Chinese medicine frequently used in the Chinese medicine treatment plan to fight COVID-19. In addition, ephedrine alkaloids are known to induce palpitation, excitation, insomnia and dysuria as side effects. Prolonged use of *Ephedra* herbaceous stem can lead to addiction, and athletes should not use it as a stimulant. Ephedrine alkaloids and products containing ephedrine alkaloids are strictly regulated in China, Europe and the United States of America. Therefore, ion-exchange column chromatography was investigated by Japanese researchers to prepare an ephedrine alkaloid-free *Ephedra* herb extract by eliminating ephedrine alkaloids from *Ephedra* herb extract. The ephedrine alkaloid-free *Ephedra* herb extract was reported as a safer alternative to crude *Ephedra* herb with comparable analgesic, anticancer and anti-influenza activities.[2] Furthermore, *Ephedra przewalskii* Stapf. (see Annex D) is also used as *Ephedra* herbaceous stem in ethnic herbal medicine with low ephedrine and pseudoephedrine content.[3,4] The ephedrine alkaloid-free *Ephedra* herb extract and *Ephedra przewalskii* can be considered as an alternative to *Ephedra* herbaceous stem in the clinical application in some areas.

Ephedra herbaceous stem has a lot of international attention and demand, and as a special medicinal material its quality should be strengthened and controlled at the international level. Searching the published papers using ephedrine as the subject term in the Web of Science database (https://www.webofscience.com/wos) for nearly a decade revealed that 57 countries and regions have carried out relevant research, of which the top 10 countries were China, the United States of America, India, the United Kingdom, Japan, South Africa, Germany, Turkey, France and Canada. According to Chinese customs data, the average export trade value of medicinal *Ephedra* herbaceous stem from 2012 to 2016 reached US$ 3 million per year. Japan, South Korea and Taiwan are the main importers.

In terms of quality supervision of medicinal materials, the quality and safety of *Ephedra* herbaceous stem are regulated in the *Pharmacopoeia of the People's Republic of China*,[5] the *European Pharmacopoeia*,[6] the *Japanese Pharmacopoeia*,[7] the *Korean Pharmacopoeial*[8] and the *Hong Kong Chinese Materia Medica Standards*.[9] However, the requirements are not all the same among these pharmacopoeias and standards. The *Ephedra* herbaceous stem standards have not yet been unified at the international level, leading to the supervision being different among the regulatory authorities in many countries.

In addition, *Ephedra* herbaceous stem ranks second only to ginseng in ISO/TR 23975. This means that *Ephedra* herbaceous stem is a high priority in ISO/TC 249 in terms of developing International Standards.

In summary, it is essential to develop an International Standard for *Ephedra* herbaceous stem to ensure its consistency of quality and safety and regulate its international trade, and also to explore the international supervision and control of particular types of Chinese medicinal materials.

As national implementation can differ, national standards bodies are invited to modify the values given in 5.5, 5.6 and 5.7 in their national standards. Examples of national and regional values are given in Annex C.

Traditional Chinese medicine — *Ephedra sinica*, *Ephedra intermedia* and *Ephedra equisetina* herbaceous stem

1 Scope

This document specifies the quality and safety requirements of *Ephedra* herbaceous stem, which is the dried herbaceous stem of *Ephedra sinica* Stapf, *Ephedra intermedia* Schrenk et C. A. Mey. or *Ephedra equisetina* Bge. (Fam. Ephedraceae).

This document applies to *Ephedra* herbaceous stem that is sold and used as natural medicine in international trade, including Chinese materia medica (whole medicinal materials) and decoction pieces derived from the plant.

2 Normative references

The following documents are referred to in the text in such a way that some or all of their content constitutes requirements of this document. For dated references, only the edition cited applies. For undated references, the latest edition of the referenced document (including any amendments) applies.

ISO 18664, *Traditional Chinese Medicine — Determination of heavy metals in herbal medicines used in Traditional Chinese Medicine*

ISO/TS 21310, *Traditional Chinese medicine — Microscopic examination of medicinal herbs*

ISO 22217, *Traditional Chinese medicine —Storage requirements for raw materials and decoction pieces*

ISO 22258, *Traditional Chinese medicine — Determination of pesticide residues in natural products by gas chromatography*

ISO 23723:2021, *Traditional Chinese medicine — General requirements for herbal raw material and materia medica*

3 Terms and definitions

For the purposes of this document, the following terms and definitions apply.

ISO and IEC maintain terminology databases for use in standardization at the following addresses:

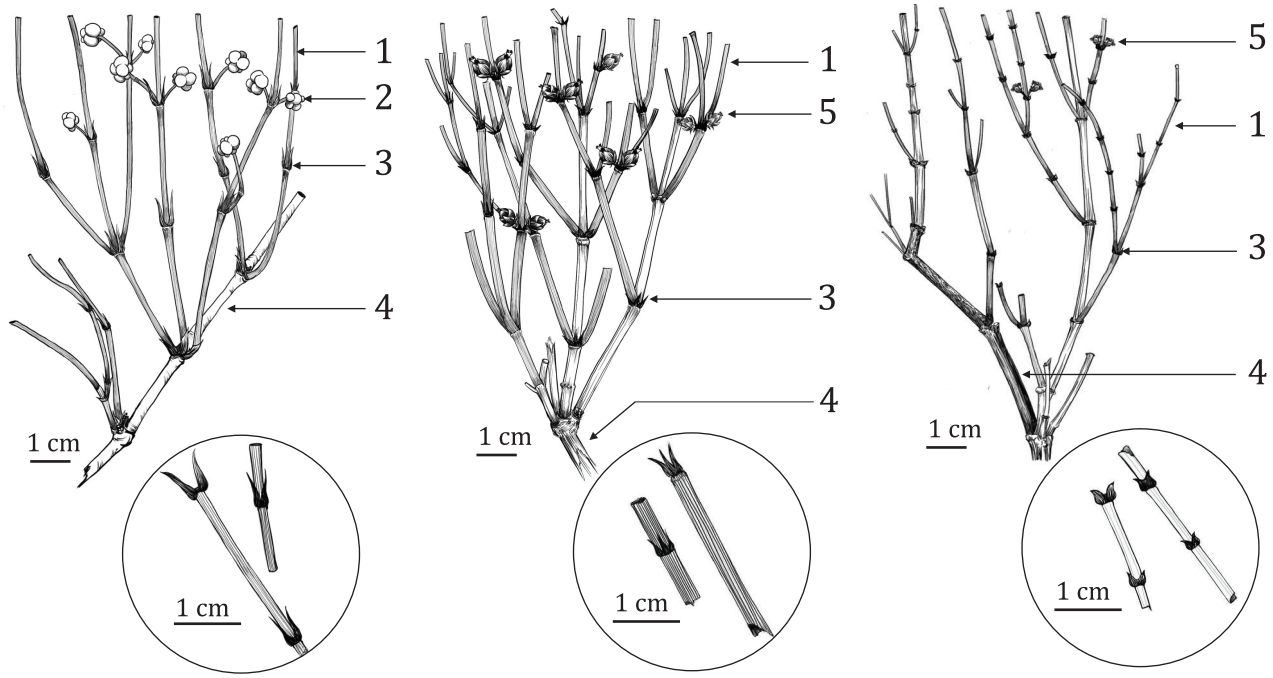
— ISO Online browsing platform: available at https://www.iso.org/obp

— IEC Electropedia: available at https://www.electropedia.org/

3.1
***Ephedra* herbaceous stem**
dried herbaceous stem of *Ephedra sinica* Stapf, *Ephedra intermedia* Schrenk et C. A. Mey. or *Ephedra equisetina* Bge

4 Description

Ephedra herbaceous stem is collected in autumn and dried in the sun (see Figure 1).



a) Plant of *Ephedra sinica* Stapf. **b) Plant of *Ephedra intermedia* Schrenk et C. A. Mey.** **c) Plant of *Ephedra equisetina* Bge.**

Key

1 herbaceous stem
2 female cones
3 scaly leaf
4 woody stem
5 male cones

Figure 1 — Structure of *Ephedra* herbaceous stem

5 Requirements

5.1 General characteristics

The following requirements shall be met before sampling.

a) *Ephedra* herbaceous stem shall be clean and free of leaf and foreign matter.

b) The presence of living insects, mouldy fruit and external contaminants which are visible to the naked eye shall not be permitted.

5.2 Morphological features

a) *Ephedra sinica* herbaceous stem is slender and cylindrical, infrequently branched and 1 mm to 2 mm in diameter. Some have a few brown wooden stems. Externally it is pale green. The nodes are distinct. The internodes are 2 cm to 6 cm long. Scaly leaves are membranous on the nodes, 3 mm to 4 mm long, with two lobes (occasionally three) and acutely triangular. The apex is greyish white and reversed. The base is tubular and reddish-brown. The texture is light, fragile and easily broken. The fractures are slightly fibrous with a greenish-yellow edge and subrounded reddish-brown pith. The odour is slightly aromatic and the taste is astringent and slightly bitter.

b) *Ephedra intermedia* herbaceous stem is frequently branched, 1,5 mm to 3 mm in diameter and rough. Membranous scaly leaves are 2 mm to 3 mm long, with three lobes (occasionally two). The apex is acute. The fracture shows a triangular and rounded pith.

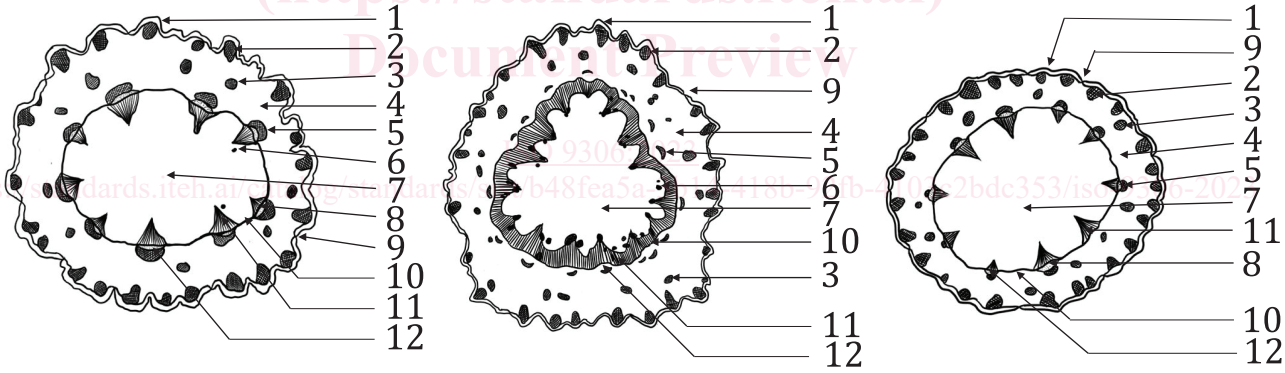
c) *Ephedra equisetina* herbaceous stem is frequently branched, 1 mm to 1,5 mm in diameter and smooth. Internodes are 1,5 mm to 3 mm in diameter, 1,5 cm to 3 cm long and smooth. Membranous scaly leaves are 1 mm to 2 mm long, with two lobes (occasionally three). The upper part is short-triangular and greyish-white. The apex is infrequently reversed. The base is brownish-red to brownish-black.

5.3 Microscopic identification

a) In the transverse section, the epidermis of *Ephedra sinica* herbaceous stem covers a thick cuticle. Ridges are relatively dense with waxy warty protruding. Sunken stomata are located between two ridges. Hypodermal fibre bundles are located in the ridges, with the thickened and unlignified wall. Cortex is relatively broad and fibre bundles are scattered. Pericycle fibre bundles are crescent-shaped. There are 8 to10 vascular bundles in *Ephedra sinica*. Cambium ring is subrounded. Xylem is triangular. Pith parenchymatous cells contain brown masses, occasionally showing perimedullary fibres. The outer walls of epidermal cells, parenchymatous cortex cells and fibres contain numerous fine sandy crystals or prisms of calcium oxalate, see Figure 2 a).

b) There are 12 to 15 vascular bundles in the transverse section of *Ephedra intermedia* herbaceous stem. Cambium is ring subtriangular. Perimedullary fibres are scattered in bundles or singly, see Figure 2 b).

c) There are 8 to 10 vascular bundles in the transverse section of the *Ephedra equisetina* herbaceous stem. The cambium ring is subrounded. Perimedullary fibres are absent, see Figure 2 c).



a) *Ephedra sinica* herbaceous stem | **b) *Ephedra intermedia* herbaceous stem** | **c) *Ephedra equisetina* herbaceous stem**

Key

1	cuticle and epidermis	7	pith
2	hypodermal fibre bundle	8	phloem
3	cortical fibre bundle	9	stomata
4	cortex	10	cambium ring
5	pericycle fibre bundle	11	xylem
6	perimedullary fibre	12	vascular bundle

Figure 2 — Transverse section of *Ephedra* herbaceous stem

5.4 Thin-layer chromatography (TLC) identification

The spot in the chromatogram obtained with the test solution should correspond in the position and colour to the spot of the reference solution.

5.5 Foreign matter

The content of foreign matter should not be more than a mass fraction of 5,0 %.

5.6 Moisture

The content of moisture should not be more than a mass fraction of 12,5 %.

5.7 Total ash

The content of total ash should not be more than a mass fraction of 11,0 %.

5.8 Heavy metals

The contents of heavy metals, such as arsenic, mercury, lead and cadmium, should be determined.

5.9 Pesticide residues

The contents of pesticide residues should be determined.

5.10 Marker compounds

The total content of ephedrine ($C_{10}H_{15}NO$) and pseudoephedrine ($C_{10}H_{15}NO$) in *Ephedra* herbaceous stem shall be determined and calculated with reference to the dried drug.

6 Sampling

Sampling shall be carried out in accordance with the method described in ISO 23723:2021, Clause 8.

7 Test methods

7.1 Macroscopic identification

Samples of not less than 500 g are taken from each batch randomly. These samples are examined with the naked eye in sunlight and for smell and taste.

7.2 Microscopic identification

The testing method specified in ISO/TS 21310 applies.

7.3 Thin-layer chromatography (TLC) identification

See Annex A for additional information.

7.4 Determination of foreign matter

The method of determination of foreign matter specified in ISO 23723:2021, 7.2.2 shall apply.