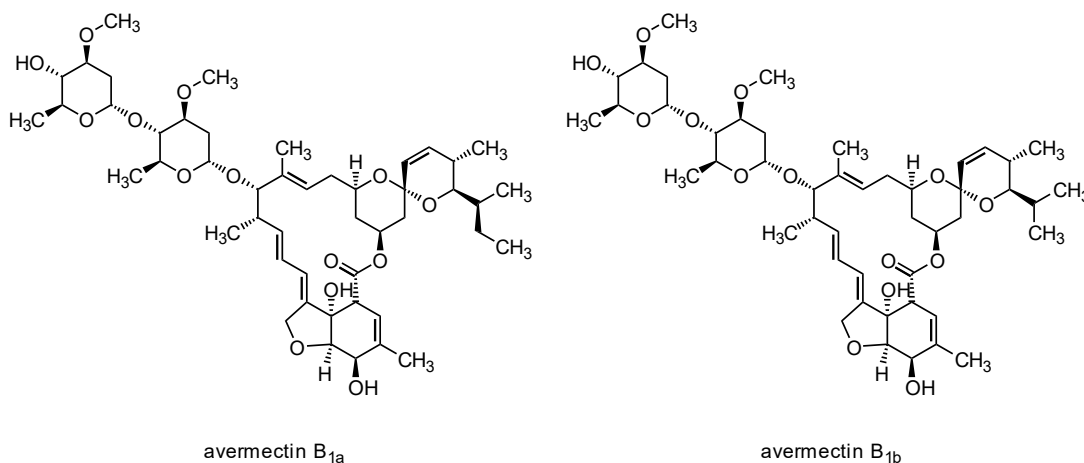


ABAMECTIN

495



ISO Common Name Abamectin

Chemical Name Mixture of Avermectin B_{1a} (at least 80%) and Avermectin B_{1b} (CA)
mixture of 80–100% (10*E*,14*E*,16*E*)-
(1*R*,4*S*,5'*S*,6*S*,6'*R*,8*R*,12*S*,13*S*,20*R*,21*R*,24*S*)-6'-[(*S*)-*sec*-butyl]-21,24-dihydroxy-5',11,13,22-tetramethyl-2-oxo-(3,7,19-trioxatetracyclo[15.6.1.1^{4,8}.0^{20,24}]*pentacos*-10,14,16,22-tetraene)-6-spiro-2'-(5',6'-dihydro-2'*H*-pyran)-12-yl 2,6-dideoxy-4-*O*-(2,6-dideoxy-3-*O*-methyl- α -L-*arabino*-hexopyranosyl)-3-*O*-methyl- α -L-*arabino*-hexopyranoside
and 20–0% (10*E*,14*E*,16*E*)-
(1*R*,4*S*,5'*S*,6*S*,6'*R*,8*R*,12*S*,13*S*,20*R*,21*R*,24*S*)-21,24-dihydroxy-6'-isopropyl-5',11,13,22-tetramethyl-2-oxo-(3,7,19-trioxatetracyclo[15.6.1.1^{4,8}.0^{20,24}]*pentacos*-10,14,16,22-tetraene)-6-spiro-2'-(5',6'-dihydro-2'*H*-pyran)-12-yl 2,6-dideoxy-4-*O*-(2,6-dideoxy-3-*O*-methyl- α -L-*arabino*-hexopyranosyl)-3-*O*-methyl- α -L-*arabino*-hexopyranoside (IUPAC)

CAS Number Abamectin: 71751-41-2
Avermectin B_{1a}: 65195-55-3
Avermectin B_{1b}: 65195-56-4

Empirical formula Avermectin B_{1a}: C₄₈H₇₂O₁₄
Avermectin B_{1b}: C₄₇H₇₀O₁₄

Molecular mass Avermectin B_{1a}: 873.1

Avermectin B_{1b}: 859.1

m.p. 161.8-169.4°C (decomp.)

v.p <0.0037 mPa at 25°C

Solubility In water 1.21 mg/l, acetone: 72 g/l, dichloromethane: 470 g/l, ethyl acetate: 160 g/l, hexane: 0.11 g/l, methanol: 13 g/l, octanol: 83 g/l, toluene: 23 g/l; all at 20-25°C

Stability Stable to hydrolysis in aqueous solutions at pH 5, 7, and 9 (25°C). Sensitive to stronger acid and base. UV irradiation causes conversion first to the 8,9-Z-isomer, then to unidentified decomposition products.

Description Colorless to pale yellow crystals

ABAMECTIN TECHNICAL

*495/TC/(M)/-

1 Sampling. Take at least 100 g.

2 Identity tests

2.1 HPLC. Use the HPLC method below. The relative retention time of avermectin B_{1a} and B_{1b} for the sample solution should not deviate by more than 1.5% from that for the calibration solution.

2.2 Infrared. Prepare potassium bromide discs for the technical sample and abamectin reference substance. Scan the discs from 4000 to 400 cm⁻¹. The spectrum from the sample should not differ significantly from that of the reference substance.

3 Abamectin

OUTLINE OF METHOD

Abamectin is determined by reversed phase high performance liquid chromatography using UV detection at 245 nm and external standardization.

REAGENTS

Abamectin: reference standard of known purity

Methanol: HPLC grade

Acetonitrile: HPLC grade

Water: Ultrapure water

Calibration solutions. Weigh in duplicate (to the nearest 0.1 mg) 50 mg of abamectin reference standard (*s* mg) into separate volumetric flasks (100 ml). Dilute to volume with methanol. Mix thoroughly and filter the solution through a 0.45 µm filter membrane prior to analysis (calibration solutions C_A and C_B).

APPARATUS

High performance liquid chromatograph, equipped with a UV detector capable for operation at 245 nm, a constant-temperature column compartment and an injection system capable of injecting 5µl.

Column stainless steel, 250 ×4.6 mm (i.d), packed with C₁₈ 5.0 μm, or equivalent with the same selectivity.

Filtering apparatus disposable plastic syringes (or equivalent) fitted with 0.45 μm filters.

Electronic integrator or data system

PROCEDURE

(a) Liquid chromatographic conditions (typical):

<i>Column</i>	stainless steel, 250 ×4.6 mm (i.d), packed with C ₁₈ 5.0 μm, or equivalent
<i>Mobile phase</i>	methanol: acetonitrile: water, 55:30:15 (v/v)
<i>Column temperature</i>	30°C ± 2°C
<i>Flow rate</i>	0.8 ml/min
<i>Detector wavelength</i>	245 nm
<i>Injection volume</i>	5 μl
<i>Retention times</i>	avermectin B _{1a} about 25.2 min avermectin B _{1b} about 19.7 min.
<i>Run time</i>	35 min

(b) System equilibration. Inject 5 μl portions of calibration solution C_A until the response factors (*f_i*) obtained for two consecutive injections differ by less than 1.5%. Then inject 5 μl portions of calibration solution C_B. The response factor (*f_i*) for two consecutive injections should not deviate by more than 1.5% from that of solution C_A, otherwise prepare new calibration solutions.

(c) Sample preparation. Prepare solutions in duplicate for each sample. Weigh (to the nearest 0.1 mg) sufficient sample (*w* mg) to contain about 50 mg of abamectin into a volumetric flask (100 ml). Make up to volume with methanol. Mix thoroughly and filter the solution through a 0.45 μm filter membrane prior to analysis (sample solutions S₁ and S₂).

(d) Determination. Inject in duplicate 5µl portions of each sample solution bracketing them by injections of the calibration solutions as follows:

$C_A, S_1, S_1, C_B, S_2, S_2, C_A, \dots$

(e) Calculation. Calculate the mean value of each pair of calibration response factors f , bracketing the two injections of a sample, and use this value for calculating the abamectin contents of the bracketed sample injections.

$$f_i = \frac{s \times P}{H_s}$$

$$\text{Content of abamectin} = \frac{H_w \times f}{w} \text{ (g/kg)}$$

where:

f_i = individual response factor

f = mean response factor

H_s = peak area of abamectin (peak $B_{1b}+B_{1a}$) in the calibration solution

H_w = peak area of abamectin (peak $B_{1b}+B_{1a}$) in the sample solution

s = mass of abamectin reference standard in the calibration solution (mg)

w = mass of sample taken (mg)

P = purity of the abamectin reference standard (g/kg)

Calculate the ratio of B_{1a} to B_{1b} of avermectin in the sample:

$$\alpha(B_{1a}/B_{1b}) = \frac{H_{WB_{1a}}}{H_{WB_{1b}}}$$

where:

$\alpha(B_{1a}/B_{1b})$ = ratio of B_{1a} to B_{1b} of avermectin in the sample

$H_{WB_{1a}}$ = peak area of avermectin (peak B_{1a}) in the sample solution

$H_{WB_{1b}}$ = peak area of avermectin (peak B_{1b}) in the sample solution

Repeatability r = **XXX-XXX g/kg at an active ingredient content of**
 XXX – XXX g/kg

**Reproducibility R = XXX-XXX g/kg at an active ingredient content of
XXX – XXX g/kg**

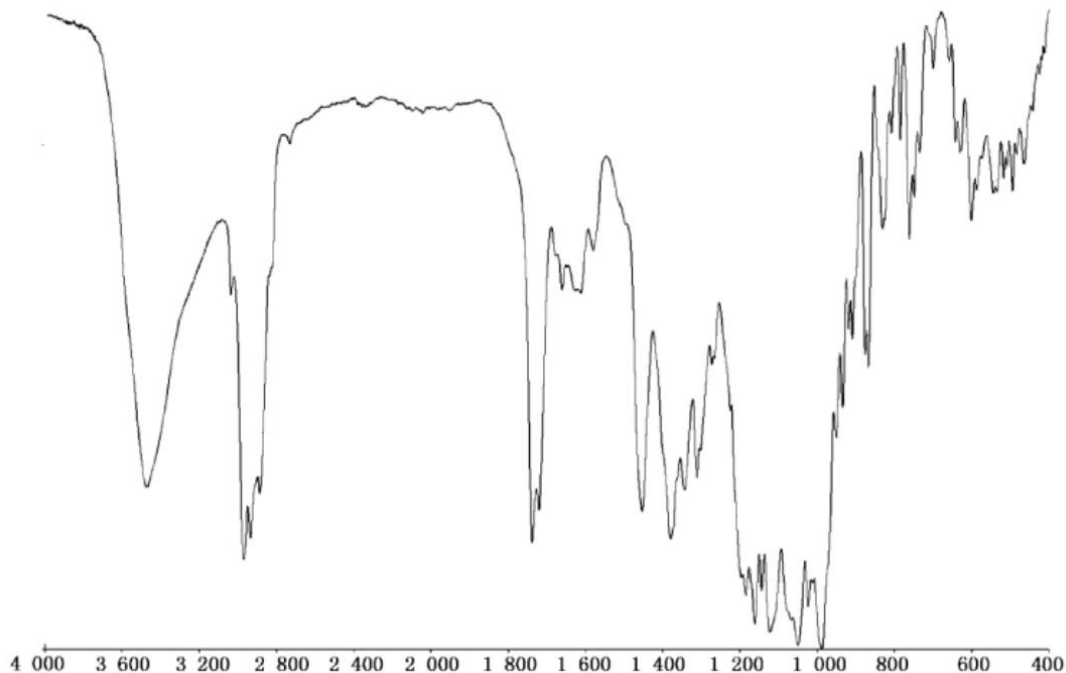


Fig. 1 FTIR spectrum of abamectin standard

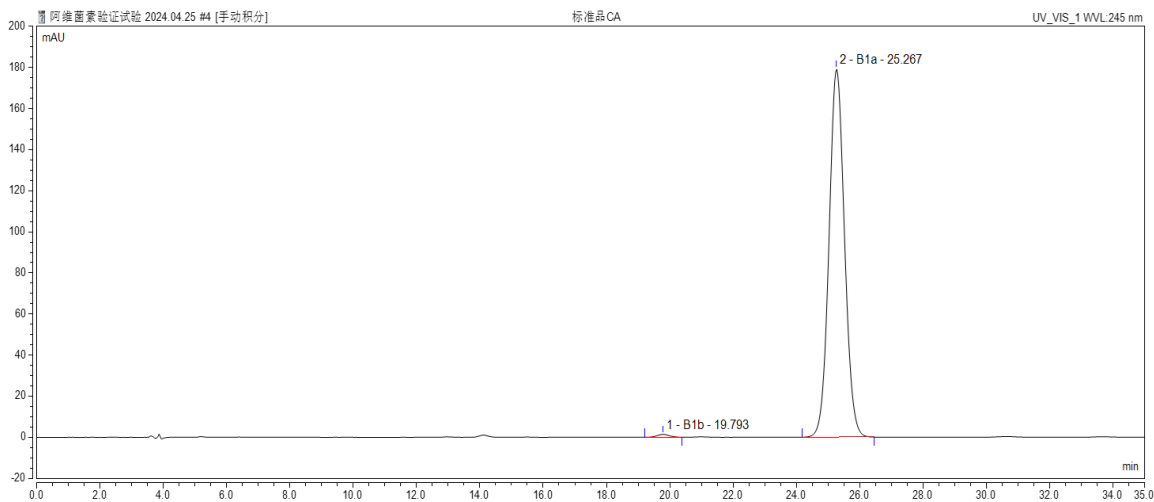


Fig. 2 HPLC Chromatogram of abamectin standard

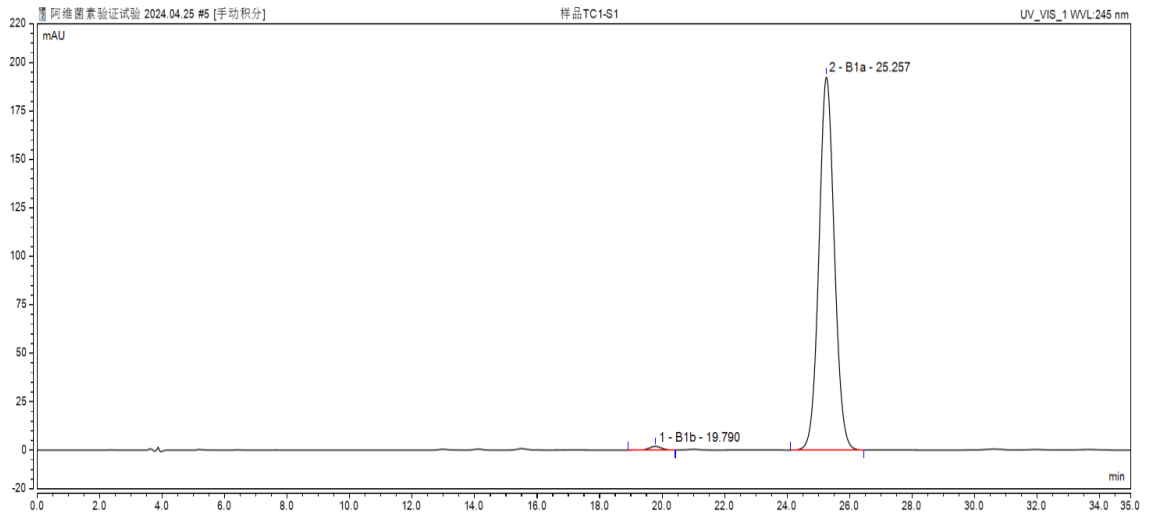


Fig. 3 HPLC Chromatogram of abamectin TC