



AGENCIJA ZA REŠAVANJE
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EXPERT RESEARCH PROTOCOL

from 08/22/2015

Code: 15-08-20-1 (105)
Customer:
Number of samples: Qualitative analysis of the sample, determination of purity
Methods: Agilent 1200, High-performance liquid chromatography (HPLC);
Column: Zorbax SB-C18 150 mm×2.1 mm, 3 mkm;
Detector – DAD, wavelength – 215, 240 nm;
Detector – MSD, ionization method APCI Positive/Negative, SCAN (100 - 500 m/z)
Number of samples: 1
Subject: Anavar powder (Oxandrolone)

The solvent for the sample: Chloroform.

Mobile phase: A - MeOH (65%), B - H₂O (45%).

The elution mode is isocratic.

The flow rate through the column: 0.3 ml/min. Thermostat: 30°C.

Single quadrupole mass analyzer is used for identification of the chemical elements. The samples were ionized at the electrostatic spraying (ESI) and atmospheric pressure with chemical ionization (APCI) mode with fixed positive and negative ions.

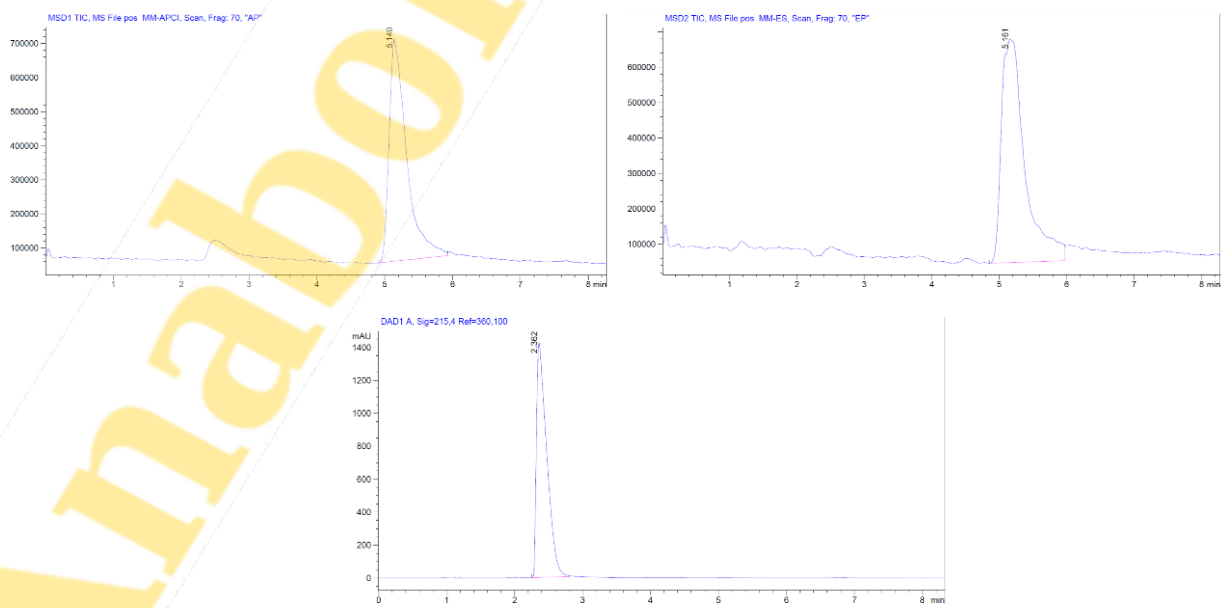


Fig.1. The component output chromatogram of the sample, detector DA, MS in EP, MS in AP

Tbl. 1. The calculation results of the peak areas on Fig. 1

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                          Area Percent Report
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Sorted By : Signal
Multiplier : 1.0000
Dilution : 1.0000
Use Multiplier & Dilution Factor with ISTDs

Signal 1: MSD1 TIC, MS File

Peak #	RetTime [min]	Type	Width [min]	Area	Height	Area %
1	5.140	BB	0.2385	1.16684e7	6.55984e5	100.0000
Totals :				1.16684e7	6.55984e5	

Signal 2: MSD2 TIC, MS File

Peak #	RetTime [min]	Type	Width [min]	Area	Height	Area %
1	5.161	VV	0.2976	1.52575e7	6.31117e5	100.0000
Totals :				1.52576e7	6.31117e5	

Signal 3: DAD1 A, Sig=215,4 Ref=360,100

Peak #	RetTime [min]	Type	Width [min]	Area [mAU*s]	Height [mAU]	Area %
1	2.362	BB	0.1456	1.49574e4	1423.54687	100.0000
Totals :				1.49574e4	1423.54687	

The analysis results of the received peaks by detector DA are shown in Fig. 2

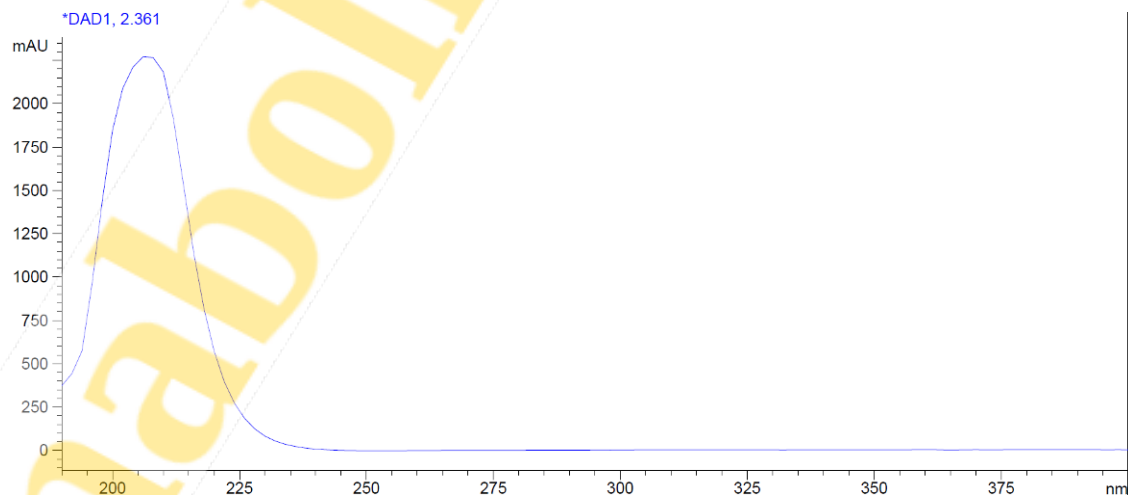


Fig. 2. The analysis of the peak 1, DA detector

The analysis results of the received peaks by detector MS is shown on Fig. 3

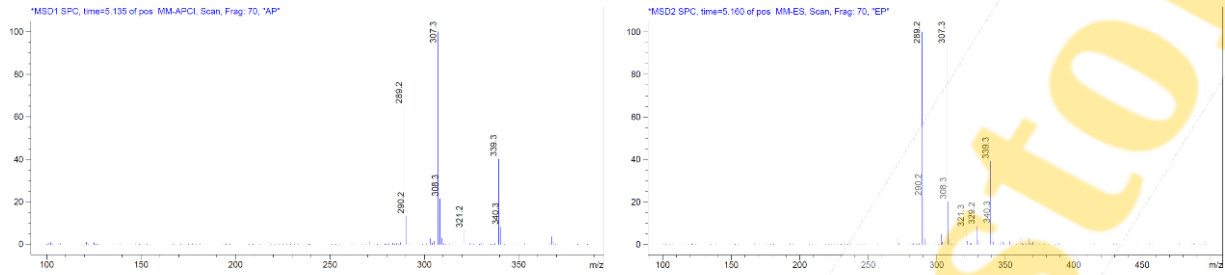


Fig. 3. The analysis of the peak 1, MS detector (modes EP and AP)

The received data of the analytical comparison results of MS detectors with the calculated data on the test substance allow us to state that peak 1 refers to Oxandrolone (mw 306.43).

The chromatographic purity of Oxandrolone by MS and DA detectors is 99%.

Remarks:

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