Two Chromatographic Methods for the Determination of Some Antimigraine Drugs

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Abstract: Two stability indicating chromatographic methods were proposed for the determination of almotriptan, eletriptan, and rizatriptan, in presence of their acid degradation products. The first method is a quantitative densitometric thin layer chromatography. The developing systems were; acetonitrile: methanol: dichloromethane: ammonia (10:6:3:1 v/v), ethyl acetate: methanol: ammonia (15:4:1 v/v), and methanol: acetonitrile: ammonia (9:4:1 v/v) for almotriptan, eletriptan and rizatriptan respectively. The TLC plates were scanned at 235 nm. Linear relationships were obtained over concentration ranges (5–50 µg/spot) for almotriptan and rizatriptan, and (5–60 µg/spot) for eletriptan. The second method based on the separation and determination of the studied drugs, using RP-HPLC technique. The separation was achieved on C18 Hypersil column, elution was carried out using phosphate buffer pH 3: methanol: acetonitrile (2:1:1 v/v) at flow rate 2 mL/min and UV detection at 235 nm. Linear relationships were obtained over concentration ranges (10–200 µg/mL) for almotriptan and eletriptan, and (10–180 µg/mL) for rizatriptan. The chromatographic methods were successfully applied for the determination of each of the studied drugs in pure form, tablet form, and in laboratory prepared mixtures with their acid degradation products.

Keywords: chromatography, antimigraine, almotriptan, eletriptan, rizatriptan, high performance liquid chromatography, thin layer chromatography

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Introduction

Migraine is described as neurovascular headache, and it is characterized by recurrent attacks of headache which typically last from 4 to 72 hours. Simple analgesics and non steroidal anti-inflammatory drugs are effective if taken at the earliest signs of the attack. Attacks not responding to simple analgesics or non steroidal anti-inflammatory drugs may be treated with some selective serotonin (5HT1) agonists, eg, almotriptan, eletriptan and rizatriptan (Fig. 1).

Few methods have been reported for the determination of almotriptan, eletriptan and rizatriptan in pure, pharmaceutical dosage forms and in biological fluids. The methods include RP-HPLC using UV detector. Method4,7,8 were used as reference methods for almotriptan, eletriptan and rizatriptan respectively. They are HPLC methods in which almotriptan and rizatriptan were determined on hypersil C18 column. A mixture of 20% acetonitrile, 80% buffer pH 4 and 0.2% triethylamine was used as a mobile phase for almotriptan, while the used mobile phase in determination of rizatriptan was 20% acetonitrile and 80% potassium dihydrogen phosphate buffer pH 3.4,8 Eleddetriptan was determined using micro bondaback C18 column and mobile phase of 55% phosphate buffer pH 2.5 and 45% acetonitrile. HPLC methods using fluorimetric detector were used for the determination of rizatriptan in presence of its impurities or in plasma.11–12 HPLC coupled with MS method was described for the determination of rizatriptan either in plasma or in serum.13–15 Stability indicating method for the determination of mixture of rizatriptan and naproxen was established using microemulsion electrokinetic chromatography (MEEC).16

Experimental

Apparatus
1. Dual wavelength, flying scanning densitometer, Shimadzu CS-9301 PC, Japan.
2. HPLC system, Hewlett Packard (1050 series) with quaternary pump, multiple wave length detector, auto-sampler, Germany.
3. Hypersil C18, 5 mm column, (25 × 4 cm) I.D Merck, Darmstadt, Germany.

Materials and reagents
1. Almotriptan working standard (purity 100.16% ± 0.163%),4 provided by European Egyptian Pharma. Ind. Co., Cairo, Egypt.
2. Almotrip forte tablets, European Egyptian Pharma. Ind. Co., Cairo, Egypt, labeled to contain 17.5 mg Almotriptan malate.
3. Eletriptan working standard (purity 99.95% ± 0.166%),7 provided by Pfizer Co., Cairo, Egypt.
4. Relpax tablets, Pfizer Co. Cairo, Egypt, labeled to contain 40 mg eletriptan HBr.
5. Rizatriptan working standard (purity 100.19% ± 0.315%),8 provided by Epico Co., Cairo, Egypt.
6. Migratec tablet, Epico Co., Cairo, Egypt, labeled to contain 10 mg rizatriptan benzoate.

![Chemical structure of rizatriptan, almotriptan and eletriptan.](image-url)
Chromatographic determination of almotriptan, eletriptan and rizatriptan

7. Topiramate working standard (purity 99.84% ± 0.405%), provided by Cairo, Co., Cairo, Egypt, used as internal standard.
8. Acetonitrile, Methanol, Dichloromethane, Ammonia, and ethylacetate, orthophosphoric acid, Hydrochloric acid, analytical grade (Analar).
9. Methanol, Acetonitrile, HPLC grade (Analar).
10. Potassium dihydrogen orthophosphate powder, (Elnasr).

Developing systems
For almotriptan: a mixture of acetonitrile: methanol: dichloromethane: ammonia (50:30:15:5 v/v),
For eletriptan: a mixture of ethyl acetate: methanol: ammonia (15:4:1 v/v),
For rizatriptan: a mixture of methanol: acetonitrile: ammonia (18:8:2 v/v).

Procedure
Preparation of degradation products solutions
An accurate weight of each of almotriptan or eletriptan or rizatriptan (100 mg) was introduced into a 50 mL round bottom flask, and dissolved in least amount of acetonitrile. Five mL of hydrochloric acid was added for each flask and the mixture was refluxed for five hours. The solution was evaporated under pressure to dryness to constant weight. The residue was weighed then dissolved in methanol to get concentration of 1 mg/mL of degradation products.

Preparation of working standard solutions
For densitometric method: An accurate weight (200 mg) of almotriptan or eletriptan or rizatriptan working standards was transferred into a 50 mL volumetric flask. Acetonitrile (25 mL) was added and the solution was shaken in ultrasonic shaker for 10 minutes, then the volume was completed with the same solvent to the mark to form 4 mg/mL working standard stock solutions.

For HPLC: Ten mL aliquot of the working standard stock solutions was diluted to 100 mL with the same solvent to form 0.4 mg/mL working standards stock solutions.

Preparation of internal standard solution
A solution of 100 µg/mL topiramate in acetonitrile was prepared.

Preparation of pharmaceutical dosage form solutions
Accurate weights of the powdered Almotrip forte, Relpax, and Migratec tablets equivalent to 100 mg of almotriptan, eletriptan and rizatriptan respectively, were transferred into three different 150 mL conical flasks. Acetonitrile (40 mL) was added for each flask and the solutions were shaken for 15 min. using ultrasonic shaker. The solutions were filtered and the filter papers and the residues were washed three times each with 10 mL acetonitrile. The combined filtrates and washings were collected into three 100 mL volumetric flasks and the volumes were completed with the same solvent to produce the pharmaceutical dosage form solutions of 1 mg/mL.

Preparation of laboratory prepared mixtures
For densitometric method
Different aliquots of each of almotriptan, eletriptan and rizatriptan working standard solution equivalent to (2–20 mg) were transferred into three series of 10 mL volumetric flasks. Aliquots of the degradation product solutions representing (10%–90%) of the intact drugs were added and the volumes were completed with the same solvent.

For HPLC method
Different aliquots of each of almotriptan, eletriptan and rizatriptan working standard solution equivalent to (0.1–2 mg) were transferred into three series of 10 mL volumetric flasks. Aliquots of the degradation product solutions representing (10%–90%) of the intact drugs were added and 1 mL aliquots of 100 µg/mL topiramate internal standard solution were added to each flask then volumes were completed using acetonitrile.

General procedure and linearity
Densitometric method
Different aliquots of almotriptan and rizatriptan stock standard solutions (0.5–5 mL) equivalent to (2–20 mg), and different aliquots of eletriptan stock standard solution (0.5–6 mL) equivalent to (2–24 mg) were transferred into three series of 10 mL volumetric flasks and the volumes were completed with acetonitrile. Twenty five µL aliquots of each solution were applied on thin layer chromatographic plates 20 × 20 cm using The micropipette. The spots were
spaced 2 cm apart from the bottom edge of the plates and the plates were put in chromatographic tanks which were previously saturated with the specified developing systems, for a distance of 16 cm. Plates were air dried and the spots were detected under UV lamp at 254 nm.

The drugs were scanned under the following parameters:

- Photo mode: reflection, lane: auto, scan mode: zigzag, swing width: 12 cm, beam width: 0.4 × 0.4 cm, wavelength: 235 nm.

The resulting outputs, chromatograms, and areas under peaks (AUP) were recorded and a calibration curve for each drug representing the relationship between AUP × 10^{-3} and its corresponding concentration in µg/25 µL (spot) was plotted. The following regression equations were computed to be used in calculations of concentrations.

\[
\begin{align*}
A &= 0.433C + 0.644 r^2 = 0.998 \text{ for almotriptan (1)} \\
A &= 0.182C - 0.1635 r^2 = 0.997 \text{ for eletriptan (2)} \\
A &= 0.516C - 0.0722 r^2 = 0.99 \text{ for rizatriptan (3)}
\end{align*}
\]

where A is the AUP × 10^{-3}, C is the concentration in µg/25 µL (spot), and r is the correlation coefficient.

**HPLC method**

Different aliquots of almotriptan and eletriptan stock standard solutions (0.25–5 mL) equivalent to (0.1–2 mg), and different aliquots of rizatriptan stock standard solution (0.25–4.5 mL) equivalent to (0.1–1.8 mg); were transferred into three series of 10 mL volumetric flasks and 1 mL aliquots of 100 µg/mL topiramate internal standard solution were added to each flask then volumes were completed using acetonitrile. Twenty µL aliquots of each solution were injected to HPLC system using auto sampler injector. The drugs were separated using Phosphate buffer pH3: methanol: acetonitrile (250:125:125 v/v) as a mobile phase at flow rate of 2 mL/min. and using a Hypersil C18, 5 mm column, (25 × 4 cm). The detection is done at 235 nm. The ratios between areas under peaks (AUP) of studied drugs and those of internal standard were recorded and a calibration curve for each drug representing the relationship between AUP ratio and its corresponding concentration in µg/mL was plotted. The following regression equations were computed to be used in calculations of concentrations.

\[
\begin{align*}
R &= 0.0487C + 0.309 r^2 = 0.9998 \text{ for almotriptan (4)} \\
R &= 0.1461C - 0.0763 r^2 = 0.9992 \text{ for eletriptan (5)} \\
R &= 0.0576C - 0.0372 r^2 = 0.9994 \text{ for rizatriptan (6)}
\end{align*}
\]

where R is the AUP ratio, C is the concentration in µg/mL, and r is the correlation coefficient.

**Stability indicating characteristics of the TLC densitometric method**

Twenty five micro liters of the laboratory prepared mixture solutions for each drug and its degradation products (prepared for stability indicating characteristics) were applied on TLC plates and the method was completed as mentioned under linearity starting from “and the plates were put in chromatographic tanks …. The concentrations of the intact drugs in the prepared mixtures were calculated using the regression equations (1–3).

**Stability indicating characteristics of the rF-HPLC method**

Twenty µL aliquots of each of the laboratory prepared mixture solutions for each drug were injected into HPLC system and the procedure was completed as mentioned [under General procedure and linearity, for HPLC method].

Concentrations of the intact drugs in the prepared mixtures were calculated using the regression equations 4–6.

**Application of proposed methods on Almotriptan, Eletriptan, and Rizatriptan in tablet form**

Different aliquots of the pharmaceutical dosage form solutions were transferred into three series of 10 mL volumetric flasks and the volumes were completed with acetonitrile. The procedures were completed as mentioned under General procedure and linearity. The same experiment was repeated applying the standard addition technique. The concentrations of the labeled and added standards of each of almotriptan, rizatriptan and eletriptan were calculated using the regression equations 1–6, for each of the studied drugs.
Results and Discussion

Densitometric method

The method was based on the different $R_f$ values of the drug of choice and its acid degradation product. Complete degradation was achieved by refluxing with hydrochloric acid for 5 hours. The $R_f$ values were found to be, 0.88, 0.294, and 0.353 for almotriptan, eletriptan and rizatriptan respectively, and zero for all acid degradation products. Different developing systems were used to obtain well defined spots of cited drugs and better separation from acid degradation product. Acetonitrile: methanol: dichloromethane: ammonia (50:30:15:5 v/v), ethyl acetate: methanol: ammonia (15:4:1 v/v) and, methanol: acetonitrile: ammonia (18:8:2 v/v) were used for almotriptan, eletriptan, and rizatriptan respectively. The use of ammonia prevents tailing and provides better separation.

Linear relationships between AUP $\times 10^{-3}$ and concentration were obtained and the regression equations were computed (equations 1–3). The method was tested for selectivity by analyzing laboratory prepared mixtures containing different percentages of the drug with its degradation product. The method was found suitable for determination of the drug in the presence of 10% up to 90% of its degradation product. The mean percentage recoveries of the drugs in laboratory prepared mixtures were 99.98 ± 1.11, 100.69 ± 0.79, and 100.44 ± 0.82 for almotriptan, eletriptan, and rizatriptan respectively. Accuracy of the method was tested and the mean percentage accuracy was 99.50 ± 0.90, 100.59 ± 0.53, and 100.87 ± 0.59 for almotriptan, eletriptan and rizatriptan respectively. The method was applied on tablet form of each drug and the mean percentage recoveries were 100.64 ± 1.07, 100.57 ± 1.28, and 100.42 ± 0.50 for almotriptan, eletriptan and rizatriptan respectively. The validity of the methods was checked by applying the standard addition technique and the resulting percentage recoveries of the added authentic were 99.90 ± 0.83, 100.11 ± 1.28, and 100.02 ± 0.99 almotriptan, eletriptan and rizatriptan respectively. Regression data and validation parameters for the proposed TLC method for the determination of AM, EH and RB in presence of their degradation products are given in Table 1.

HPLC method

The method was based on the differences in retention time values of the drug of choice and its acid degradation product. Various mobile phase systems were attempted to be used for HPLC separation and solvent polarity optimization. Elution was carried out using phosphate buffer pH 3: methanol: acetonitrile (2: 1:1 v/v) at flow rate 2 mL/min and UV detection at 235 nm. Decreasing the pH to 2 gives bad resolution, while increasing the pH to 4 gives tailing of peaks. The retention times were found to be, 3.512 min, 3.109 min, and 6.361 min, 6.532 min and 6.057 min for almotriptan, eletriptan, and rizatriptan respectively. The method was applied on tablet form of each drug and the mean percentage recoveries were 100.64 ± 1.07, 100.57 ± 1.28, and 100.42 ± 0.50 for almotriptan, eletriptan and rizatriptan respectively. The method was applied on tablet form of each drug and the mean percentage recoveries were 100.64 ± 1.07, 100.57 ± 1.28, and 100.42 ± 0.50 for almotriptan, eletriptan and rizatriptan respectively. The validity of the methods was checked by applying the standard addition technique and the resulting percentage recoveries of the added authentic were 99.90 ± 0.83, 100.11 ± 1.28, and 100.02 ± 0.99 almotriptan, eletriptan and rizatriptan respectively. Regression data and validation parameters for the proposed TLC method for the determination of AM, EH and RB in presence of their degradation products are given in Table 1.

Table 1. Regression equation and validation parameters for the proposed TLC method for the determination of AM, EH and RB in presence of Their degradation products.

| Item                        | AM          | EH          | RB          |
|-----------------------------|-------------|-------------|-------------|
| $R_f$                       | 0.88        | 0.29        | 0.35        |
| Wavelength of detection     | 235 nm      | 235 nm      | 235 nm      |
| Linearity range             | 5–50 ± µg/spot | 5–60 ± µg/spot | 5–50 ± µg/spot |
| Regression equation & ($r^2$) | $Y = 0.433X + 0.644$ | $Y = 0.182X - 0.164$ | $Y = 0.516X - 0.072$ |
| $S_b$                       | 1.13 × 10^{-2} | 2.99 × 10^{-3} | 8.40 × 10^{-3} |
| $S_a$                       | 3.19 × 10^{-1} | 9.15 × 10^{-2} | 2.48 × 10^{-1} |
| LOD (µg)                    | 0.314       | 0.751       | 0.310       |
| LOQ (µg)                    | 0.940       | 2.504       | 1.034       |
| Confidence limit of the slope | 0.4328 ± 9.68 × 10^{-3} | 0.1821 ± 1.13 × 10^{-3} | 0.516 ± 2.75 × 10^{-3} |
| Confidence limit of the intercept | 0.6443 ± 2.73 × 10^{-1} | 0.1635 ± 3.45 × 10^{-2} | 0.0722 ± 8.11 × 10^{-2} |
| Accuracy                    | 99.98 ± 1.11 | 100.69 ± 0.79 | 100.44 ± 0.82 |
| Tablets ± SD               | 100.64 ± 1.07 | 100.57 ± 1.28 | 100.42 ± 0.49 |
| Added authentic ± SD       | 99.9 ± 0.83 | 100.11 ± 1.28 | 100.02 ± 0.99 |

**Note:** Y is the response, X is the concentration (µg/spot), a is the intercept, and b is the slope.
Figure 2. HPLC chromatogram of mixture of almotriptan, degradation product and topiramate as an internal standard.

Figure 3. HPLC chromatogram of mixture of eletriptan, degradation product and topiramate as an internal standard.

Figure 4. HPLC chromatogram of mixture of rizatriptan, degradation product and topiramate as an internal standard.
Table 2. Regression equation and validation parameters for the proposed HPLC method for the determination of AM, EH, and RB in presence of Their degradation products.

| Item                          | AM            | EH            | RB            |
|-------------------------------|---------------|---------------|---------------|
| Retention time                | 3.512 min     | 3.109 min     | 3.845         |
| Wavelength of detection       | 235 nm        | 235 nm        | 235 nm        |
| Linearity range               | 10–200 µg/mL  | 10–200 µg/mL  | 10–180 µg/mL  |
| Regression equation & \( r^2 \) | \( Y = 0.0487X + 0.309 \) \( r^2 = 0.9998 \) | \( Y = 0.1461X - 0.076 \) \( r^2 = 0.9995 \) | \( Y = 0.0576X - 0.037 \) \( r^2 = 0.9994 \) |
| \( S_b \)                     | 2.14 × 10^{-4} | 2.54 × 10^{-3} | 5.21 × 10^{-4} |
| \( S_a \)                     | 2.34 × 10^{-2} | 2.77 × 10^{-1} | 5.47 × 10^{-2} |
| LOD (µg)                      | 2.66 µg       | 1.19 µg       | 1.33 µg       |
| LOQ (µg)                      | 8.85 µg       | 3.96 µg       | 4.44 µg       |
| Confidence limit of the slope | 0.0487 ± 6.76 × 10^{-5} | 0.1461 ± 2.35 × 10^{-3} | 0.576 ± 3.85 × 10^{-4} |
| Confidence limit of the intercept | 0.309 ± 7.4 × 10^{-3} | 0.0763 ± 2.57 × 10^{-1} | 0.0372 ± 4.04 × 10^{-2} |
| Accuracy                      | 100.62 ± 0.91 | 100.62 ± 0.42 | 99.92 ± 1.22  |
| Tablets ± SD                  | 100.64 ± 1.07 | 100.28 ± 0.83 | 100.51 ± 0.99 |
| Added authentic ± SD          | 99.9 ± 0.83   | 100.02 ± 0.99 | 99.59 ± 1.076 |

Note: \( Y \) is the response, \( X \) is the concentration (µg/mL), \( a \) is the intercept, and \( b \) is the slope.

Table 3. Statistical comparison between results of analysis of the studied drugs applying the proposed methods and reference methods.

| Statistical item | Drug studied | Reference method | HPLC proposed method | TLC proposed method |
|------------------|--------------|------------------|----------------------|---------------------|
| n                | Almotriptan  | 5                | 6                    | 6                   |
| Mean             | 100.31       | 100.62           | 100.49               |
| SD               | 1.03         | 0.91             | 0.89                 |
| variance         | 1.06         | 0.83             | 0.81                 |
| SE               | 0.46         | 0.37             | 0.37                 |
| LOD              | 1.19         | 1.19             | 0.94                 |
| LOQ              | 3.96         | 3.96             | 0.32                 |
| Student t test   | 0.32 (1.833)* | 0.86 (1.833)*     | 1.32 (5.19)*         |
| F ratio          | 1.28 (5.19)* | 1.32 (5.19)*     | 1.32 (5.19)*         |
| n                | Eletriptan   | 5                | 6                    | 6                   |
| Mean             | 99.68        | 100.62           | 100.19               |
| SD               | 0.95         | 0.42             | 1.26                 |
| variance         | 0.90         | 0.18             | 1.58                 |
| SE               | 0.65         | 0.17             | 0.51                 |
| LOD (µg)         |              | 0.75             | 0.75                 |
| LOQ (µg)         |              | 2.51             | 2.51                 |
| Student t test   | 0.93 (1.833)* | 0.38 (1.833)*     | 1.26 (5.19)*         |
| F ratio          | 1.24 (5.19)* | 1.26 (5.19)*     | 1.26 (5.19)*         |
| n                | Rizatriptan  | 5                | 6                    | 6                   |
| Mean             | 100.66       | 99.92            | 100.87               |
| SD               | 0.74         | 1.23             | 0.59                 |
| variance         | 0.55         | 1.49             | 0.34                 |
| SE               | 0.33         | 0.50             | 0.24                 |
| LOD (µg)         |              | 0.31             | 0.31                 |
| LOQ (µg)         |              | 1.04             | 1.04                 |
| Student t test   | 0.74 (1.833)* | 0.33 (1.833)*     | 1.59 (5.19)*         |
| F ratio          | 2.75 (6.62)* | 1.59 (5.19)*     | 1.59 (5.19)*         |
Linear relationships between AUP ratios and concentration were obtained and the regression equations were computed (equations 4–6). The method was tested for selectivity by analyzing laboratory prepared mixtures containing different percentages of the drug with its degradation product. The method was found suitable for determination of the drug in the presence of 10% up to 90% of its degradation product. The mean percentage recoveries of the drugs in laboratory prepared mixtures were 99.44 ± 0.76, 100.05 ± 1.02, and 100.45 ± 0.97 for almotriptan, eletriptan and rizatriptan respectively. Accuracy of the method was tested and the mean percentage accuracy was 100.62 ± 0.91, 100.62 ± 0.42, and 99.92 ± 1.22 for almotriptan, eletriptan and rizatriptan respectively. The method was applied on tablet form of each drug and the mean percentage recoveries were 100.32 ± 1.31, 100.03 ± 0.60, and 100.03 ± 0.85 for almotriptan, eletriptan and rizatriptan respectively. The validity of the methods was checked by applying the standard addition technique and the resulting percentage recoveries of the added authentic were 99.73 ± 1.36, 100.02 ± 1.00, and 99.59 ± 1.08 for almotriptan, eletriptan and rizatriptan respectively. Regression data and validation parameters for the proposed HPLC method for the determination of AM, EH and RB in presence of Their degradation products are given in Table 2.

Statistical comparison between the results obtained by the suggested densitometric, and RP—HPLC methods with the reference methods of analysis of the cited drugs was carried out and no significant difference, between them was indicated, as shown in Table 1.

Statistical comparison between the result of analysis of the proposed TLC and HPLC methods and the reference methods shows no significant differences. As shown in Table 3.

Author Contributions
Conceived and designed the experiments: NGMA, HN. Analysed the data: RIEB, NGMA. Wrote the first draft of the manuscript: HN. Contributed to the writing of the manuscript: RIEB. Agree with manuscript results and conclusions: RIEB, NGMA. Jointly developed the structure and arguments for the paper: RIEB, NGMA. Made critical revisions and approved final version: RIEB. All authors reviewed and approved of the final manuscript.

References
1. Sean C Sweetman, Martindale, The complete drug reference, Pharmaceutical Press, London, Chicago, 34th edition 2005.
2. The Merck Index, Merck and Co., INK, White house station, NJ USA. 14th edition, 2006.
3. Phani-Kumar A, Ganesh VRL, Subba-Rao DV, et al. J Pharm Biomed Anal. 2008;46(4):792–8.
4. Manufacture procedure obtained from European Egyptian Pharm. Ind. Co. Cairo, Egypt by personal communication.
5. Sagirli O, Onal A, Sensory D. Chromatographia. 2008;68(3–4):269–73.
6. Cooper JDH, Muirhead DC, Taylor JE. J Pharma Biomed Anal. 1999;21(4):787–96.
7. Manufacture procedure obtained from Pfizer, Co. Cairo, Egypt by personal communication.
8. Manufacture procedure obtained from Epico. Co. Cairo by personal communication.
9. Antonucci V, Wright L, Toma P. J Liq Chromatogr Relat Technol. 1998;21(11):1649–70.
10. Jocic B, Zecevic M, Zivanovic L, Licanski A. Anal Lett. 2007;40(10–12):2301–16.
11. Qin YP, Zou YG, Liang MZ, Yu Q, Yaowu J. Fenxi Zazhi. 2003;26(1):7–9.
12. Chen J, Jiang XG, Jiang WM, Mei N, Gao XL, Zhang QZ. Chromatogr B: Anal Technol Biomed Life Sci. 2004;805(1):169–73.
13. Qin Y, Miao H, Lin M, et al. Chromatogr B Anal Technol Biomed Life Sci. Dec 2006;844(2):268–77.
14. Vishwanathan K, Bartlett MG. J Rapid common mass spectrum. 2000;14(3):168–172.
15. Mahuzier PE, Clark BJ, Crompton AJ, Altira KD. J Sep Sci. 2001;24(9):784–8.
16. HPLC determination of topiramate in the finished product (Topiramate tablets). The method was achieved with private communication with Cairo, Co. Cairo, Egypt.
17. British Pharmacopoeia, Her Majesty stationary office, London, 1965 (2007).
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