Determination of Ephedrine HCl and Pseudoephedrine HCl in Xiao'er Kechuanling Oral Solution with Purospher® STAR RP-18e colµmn

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Ephedrine hydrochloride and pseudoephedrine hydrochloride can be used to reduce cold, allergic rhinitis, rhinitis and sinusitis caused by nasal congestion symptoms and control bronchial asthma. The same compounds are also found in the Chinese traditional medicine Xiao'er Kechuanling oral solution.

This application focuses on the determination of ephedrine HCl and pseudoephedrine HCl. Oral solution samples were distilled to extract the ephedrine HCl and pseudoephedrine HCl. Samples were then filtered through Millex® PTFE syringe filters, prior to HPLC-UV analysis using a Purospher® STAR RP-18 endcapped HPLC colµmn.

The limit of detection (LOD) and the limit of quantitation (LOQ) were 0.04 µg/kg and 0.12 µg/kg, respectively, for ephedrine, and 0.02 µg/kg and 0.06 µg/kg, respectively, for pseudoephedrine. The method can be used for determination of ephedrine HCl and pseudoephedrine HCl in Xiao'er Kechuanling oral solution.

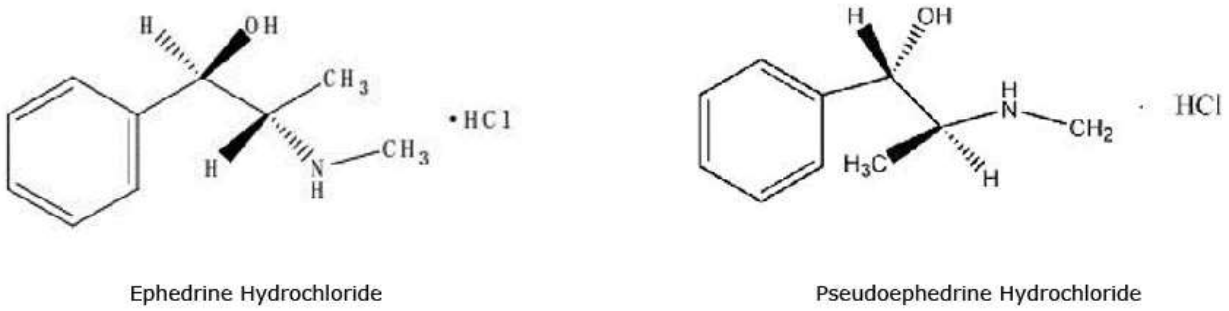

Figure 1. Chemical structures of compounds used in the study.

DETERMINATION OF EPHEDRINE HCL AND PSEUDOEPHEDRINE HCL IN XIAO'ER KECHUANLING ORAL SOLUTION

Purospher® STAR RP-18e

Experimental Conditions	
Colμmn Used:	Purospher® STAR RP-18e 250 x 4.6 mm, 5 μm (1.51456.0001)
Detection:	Waters e2695, 210 nm
Mobile phase A:	10 mM KH_2PO_4, adjust pH value to 2.3 with H_3PO_4
Mobile phase B:	Methanol
Gradient:	A : B = 89 : 11, v/v
Flow Rate:	1.0 mL/min
Injection Volμme:	20 μL
Temp:	30 °C
Standard	Dissolve appropriate amount of ephedrine HCl and pseudoephedrine HCl in mobile phase to obtain a solution containing 5 μg/mL of ephedrine HCl and 3 μg/mL of pseudoephedrine HCl
Sample:	Accurately weigh 5 mL of oral solution into a 500-mL round-bottomed flask, add into 120 mL of 10% sodiμm hydroxide, shake well, distill, collect ~95 mL distillate into a 100-mL volμmetric flask containing 5 mL of 10 mM of 10 mM KH_2PO_4 (adjust pH value to 2.3 with H_3PO_4). Make up to the tick mark with water. Filter with 0.45 μm filter membrane

Pressure drop:	2150 psi

Table 1. HPLC conditions.

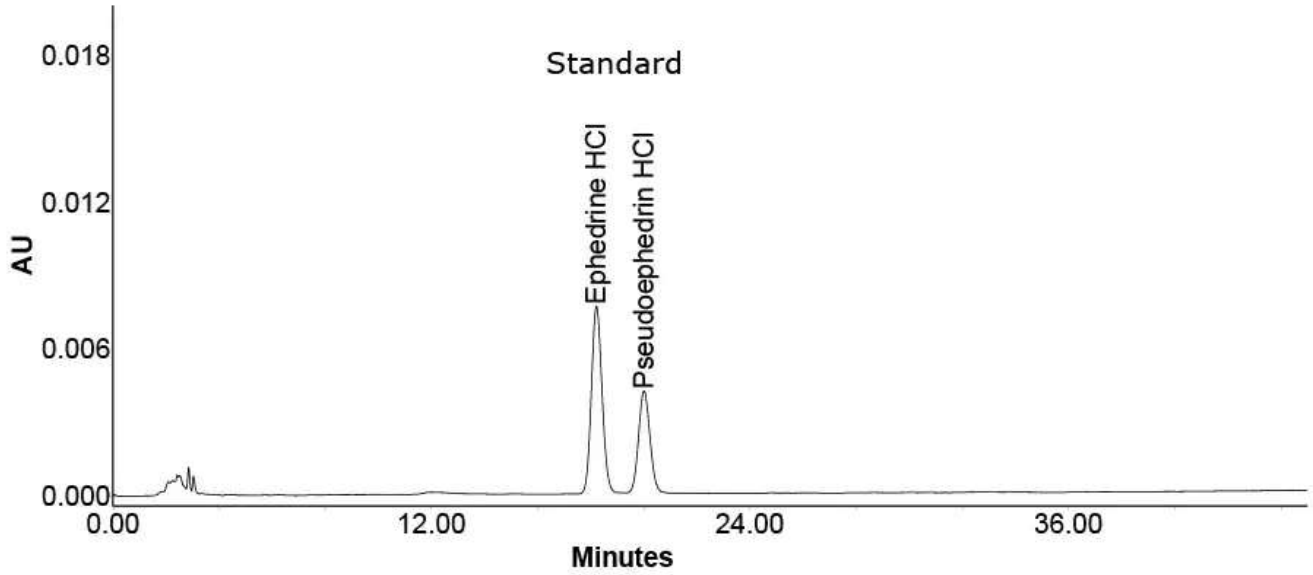

Figure 2. Chromatogram of ephedrine HCl and pseudoephedrine HCl standard solution.

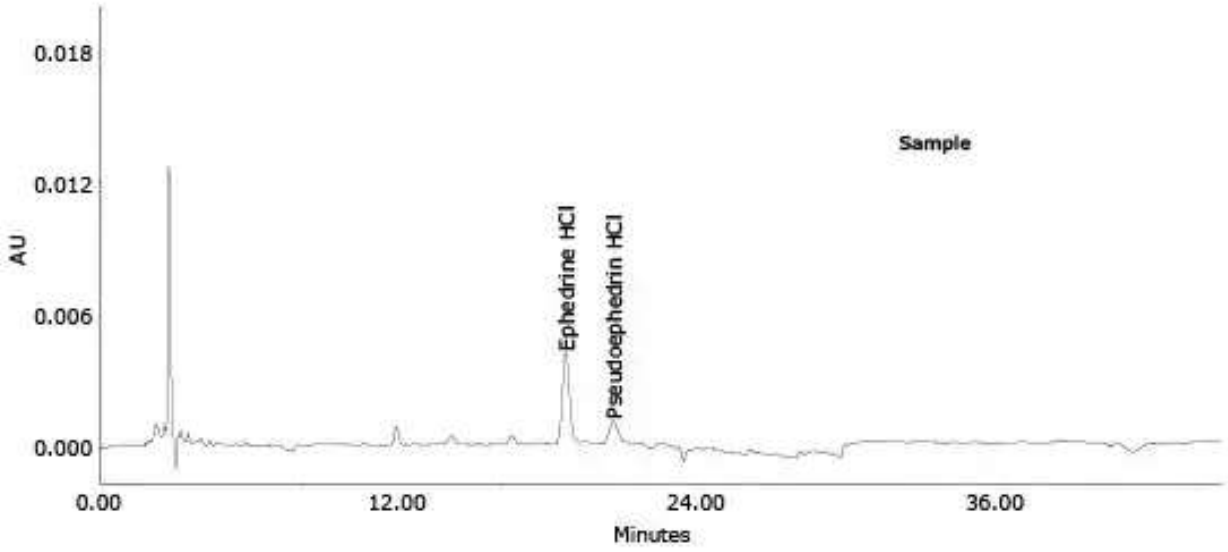

Figure 3. Chromatogram of in Xiao'er Kechuanling oral solution.

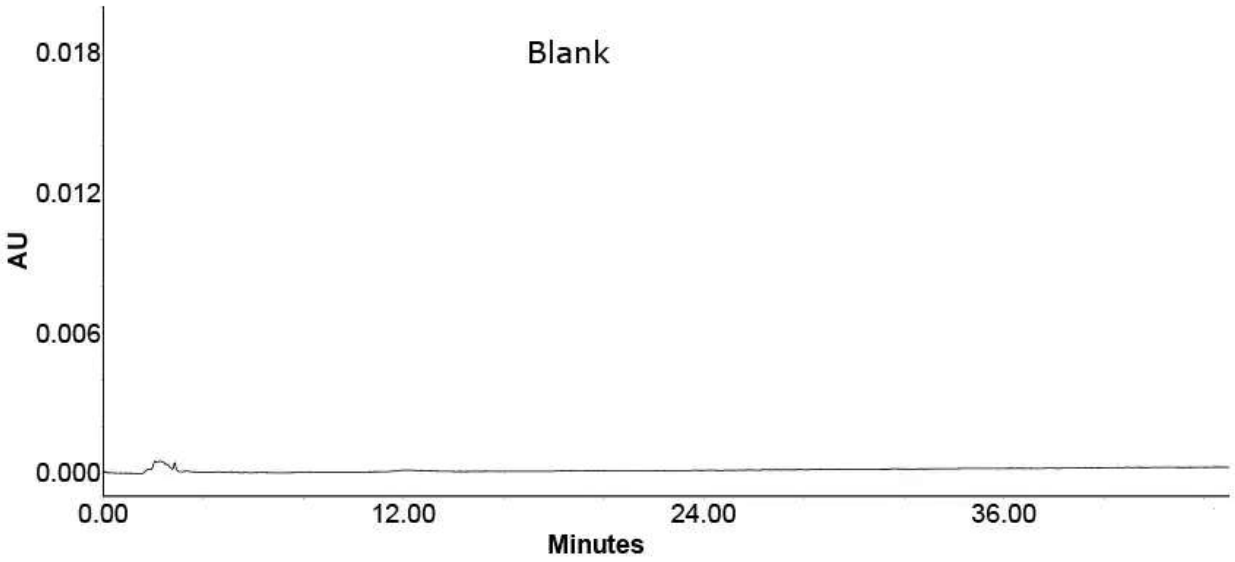

Figure 4. Chromatogram of solvent blank.

No	Compound	Retention Time (min)	Resolution	Theoretical Plates	Tailing Factor
1	ephedrine HCl	18.2	--	9698	1.1
2	pseudoephedrine HCl	20.0	2.4	10292	1.1

Table 2. Specificity data of ephedrine HCl and pseudoephedrine HCl.

SPECIFICITY AND REPEATABILITY – EPHEDRINE AND PSEUDOEPHEDRINE

Specificity: Inject reference solution and determine the retention time of desired analyte in presence of other components like impurities.

No	Compound	Retention Time (min)	Area (%)	Tailing Factor	RRT
1	ephedrine HCl	18.2	56.4	1.1	1.0
2	pseudoephedrine HCl	20.0	32.8	1.1	1.1

Table 3. Specificity data of ephedrine and pseudoephedrine HCl.

Standard repeatability (5 µg/mL) - ephedrine HCl	
STD 1	212571
STD 2	227584
STD 3	215716
STD 4	212961
STD 5	251431
Mean	1547859
Standard Deviation	16480.3
RSD (%)	1.1

Table 4. Standard repeatability data of ephedrine HCl.

Standard Repeatability (5 µg/mL) - Pseudoephedrine HCl	
STD 1	123655
STD 2	123017
STD 3	126350
STD 4	122884

STD 5	133801
Mean	1327102
Standard Deviation	4611.6
RSD (%)	0.3

Table 5. Standard repeatability data of pseudoephedrine HCl.

CALIBRATION DATA - EPHEDRINE

Linearity (area mAU*min), ephedrine HCl

Conc. (µg/mL)	Mean Area
0.10	3380
0.20	8013
0.40	16834
0.5	20141
1.0	35864
2.0	79213
4.0	166302
5.0	212571
6.0	272766

Linearity (area mAU*min), ephedrine HCl

Table 6. Linear range of ephedrine HCl.

LOD and LOQ, ephedrine HCl

Conc. (µg/mL)	Mean Area (mAU*min)
0.10	3380
0.20	8013
0.40	16834
0.50	20141
STEYEX	508.2
Slope	42344.5
LOD (ppm)	0.04
LOQ (ppm)	0.12

Table 7. LOD and LOQ of ephedrine HCl.

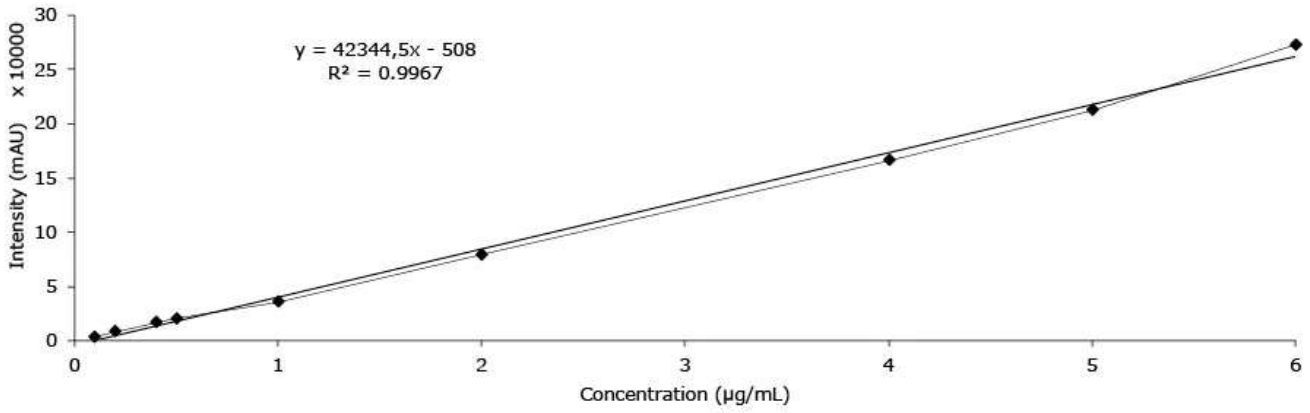

Figure 5. Calibration curve of ephedrine HCl.

CALIBRATION DATA - PSEUDOEPHEDRIN

Linearity (area mAU*min), pseudoephedrine HCl

Conc. (µg/mL)	Mean Area
0.06	1976
0.12	4684
0.24	8952
0.30	11653
0.60	19605
1.2	45571
2.4	98038
3.0	123655
4.0	179102

Table 8. Linear range of pseudoephedrine HCl.

4.2 LOD and LOQ - pseudoephedrine HCl

Conc. (µg/mL)	Mean Area (mAU*min)
0.06	1976
0.12	4684
0.24	8952
0.30	11653
STEYEX	255.2
Slope	39369.9
LOD (ppm)	0.02
LOQ (ppm)	0.06

Table 9. LOD and LOQ of pseudoephedrin HCl.

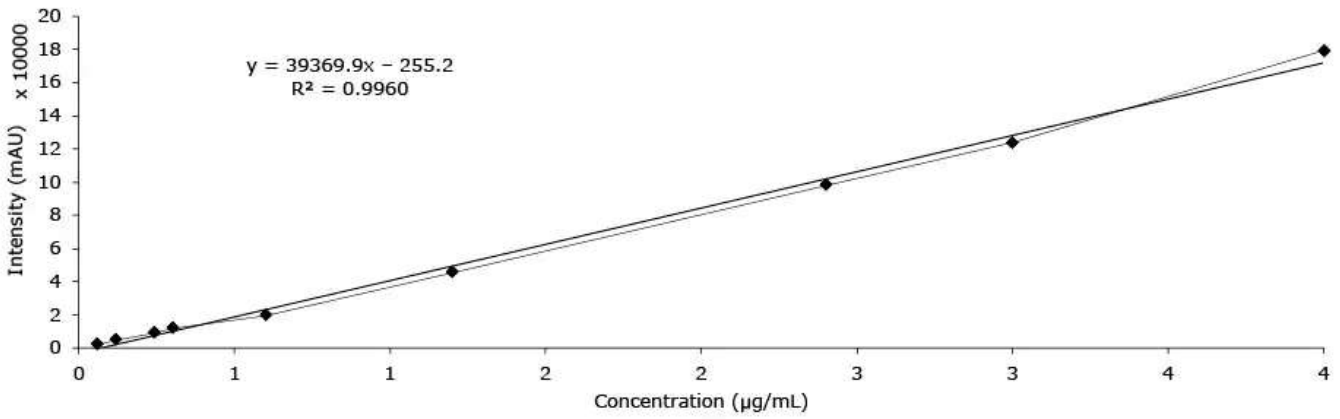

Figure 6. Calibration curve of pseudoephedrine HCl.

CONCLUSION

The method can be used for ephedrine HCl and pseudoephedrine HCl in Xiao'er Kechuanling oral solution.

Materials

Product No.	Description	SDS	Pricing
1.51456	**Purospher® STAR RP-18 endcapped (5 µm) Hibar® RT 250-4.6** suitable for HPLC		Expand
1.04877	**Potassium dihydrogen phosphate** for analysis (<= 0.005% Na) EMSURE® ACS,ISO,Reag. Ph Eur	↓	Expand
1.00573	**ortho-Phosphoric acid 85%** for analysis EMSURE® ACS,ISO,Reag. Ph Eur	↓	Expand
1.00030	**Acetonitrile** gradient grade for liquid chromatography LiChrosolv® Reag. Ph Eur	↓	Expand
1.06007	**Methanol** gradient grade for liquid chromatography LiChrosolv® Reag. Ph Eur	↓	Expand
1.15333	**Water** for chromatography (LC-MS Grade) LiChrosolv®	↓	Expand
SLCR033	**MILLEX®-LCR Syringe Filter** 0.45 µm Hydrophilic PTFE, 33 mm, Nonsterile		Expand