Efficient Detection Solution! Chromai Parallel LC Enables Simultaneous Determination of 11 Compounds in Cosmetics

I. Introduction

In cosmetics, magnesium ascorbyl phosphate, ascorbyl glucoside, kojic acid, niacinamide, 3-o-ethyl ascorbic acid, potassium 4-methoxysalicylate, ferulic acid, ellagic acid, 4-butylresorcinol, phenylethyl resorcinol, and tranexamic acid are commonly used as whitening and antioxidant ingredients, effectively providing antioxidant, whitening, and UV-protection effects.

However, long-term excessive use of cosmetics containing these ingredients may irritate the skin and cause allergic reactions.

This application refers to the Cosmetic Safety Technical Standard (2015 Edition) and employs the Leaps Duo Parallel Liquid Chromatography System to divide the 11 compounds into two groups for simultaneous analysis under different chromatographic conditions.

Group I includes magnesium ascorbyl phosphate, ascorbyl glucoside, kojic acid, niacinamide, 3-o-ethyl ascorbic acid, potassium 4-methoxysalicylate, ferulic acid, ellagic acid, 4-butylresorcinol, and phenylethyl resorcinol, while Group II consists of tranexamic acid.

This approach cuts the analysis time in half, significantly improving efficiency.

II. Experimental Conditions

Instruments and Equipment: Leaps Duo Parallel Liquid Chromatography System (P60 dual ternary gradient pump, A10D dual-valve autosampler, C10V2 column oven, D10 UV detector, D22 diode array detector).

Mixed Standard Working Solutions: Group I (10 compounds): magnesium ascorbyl

phosphate, ascorbyl glucoside, kojic acid, niacinamide, 3-o-ethyl ascorbic acid, potassium 4-methoxysalicylate, ferulic acid, ellagic acid, 4-butylresorcinol, and phenylethyl resorcinol, with concentrations of 1.00, 2.00, 5.00, 10.00, 25.00, and 50.00 μg/mL. Group II (tranexamic acid): concentrations of 5.00, 10.00, 20.00, 50.00, 100.00, and 200.00 μg/mL.

Group I: Accurately weigh 0.5 g of cosmetic sample (to 0.0001 g) into a 15 mL stoppered colorimetric tube. For mask-type samples, take the liquid portion; for foundation-type samples, mix thoroughly before sampling. Add methanol–water (1:1) to about 6–9 mL, vortex to mix. If dispersion is poor, add 3 mL dichloromethane, vortex until fully dispersed, then add methanol–water (1:1) for extraction. Ultrasonicate for 20 min, cool to room temperature, and transfer to a 10 mL volumetric flask. Make up to volume with methanol–water (1:1). Mix well, transfer to a centrifuge tube, centrifuge at 12000 r/min for 15 min. Filter the supernatant through a 0.22 μm membrane; the filtrate is used as the test solution.

1.1 Group II: Accurately weigh 0.5 g (to 0.0001 g) of cosmetic sample into a 15 mL stoppered colorimetric tube, add about 9 mL of 5% methanol solution, and vortex to mix thoroughly. Ultrasonicate for 10 min, cool to room temperature, transfer to a 10 mL volumetric flask, and make up to volume with 5% methanol. Mix well, transfer to a centrifuge tube, centrifuge at 12000 r/min for 15 min. Filter the supernatant through a 0.22 μm membrane; the filtrate is used as the test solution.

III. Chromatographic Conditions

1. Group I Chromatographic Conditions:

1.1 Lotus C18（4.6mm*250mm, 5 μm） Column: Lotus C18 (4.6 mm × 250 mm, 5 μm)

1.2 Column Temperature: 30 ℃

1.3 Injection Volume: 10 μL

1.4 Detection Wavelengths:230 nm for niacinamide; 250 nm for magnesium ascorbyl phosphate, ascorbyl glucoside, kojic acid, nicotinic acid, 3-o-ethyl ascorbic acid, potassium 4-methoxysalicylate, and ellagic acid; 280 nm for ferulic acid, 4-butylresorcinol, and phenylethyl resorcinol.

1.5 Mobile Phase: A: 0.02 mol/L potassium dihydrogen phosphate solution; B: methanol

1.6 Gradient Table:

Time (min)	Pump (mL/min)	A%	B%
0.0	1.0	97	3
3	1.0	97	3
8	1.0	60	40
14	1.0	40	60
18	1.0	27	73
25	1.0	97	3
30	1.0	97	3

2. Group II Chromatographic Conditions:

2.1 Lotus C18（4.6mm*250mm，5 μm）Column: Lotus C18 (4.6 mm × 250 mm, 5 μm)

2.2 Column Temperature: 30 ℃

2.3 Injection Volume: 40 μL

2.4 Wavelength: 210 nm

2.5 Mobile Phase: B: Methanol, C: 0.05 mol/L potassium dihydrogen phosphate – 0.2% phosphoric acid solution

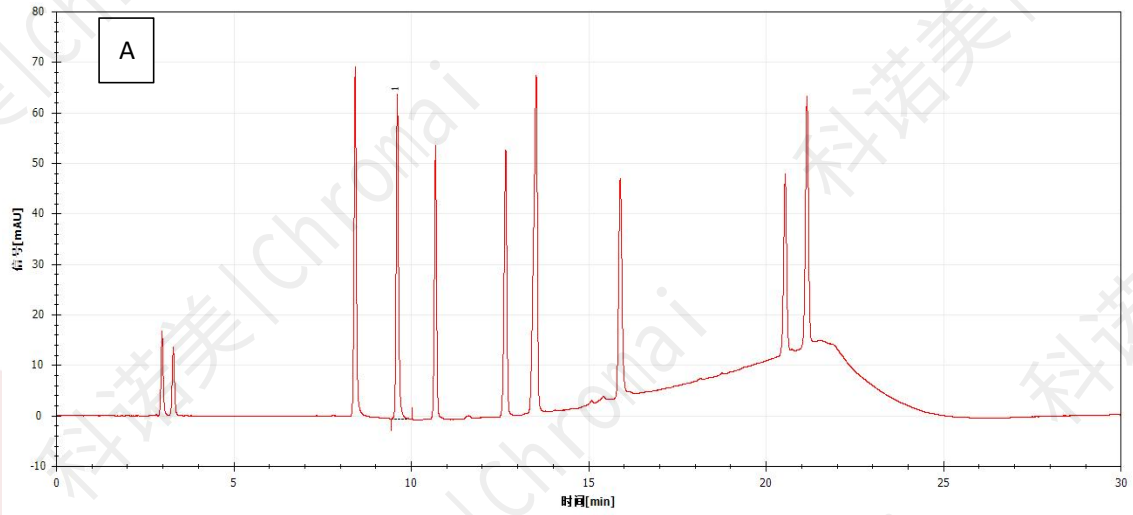
2.6 Isocratic Elution:

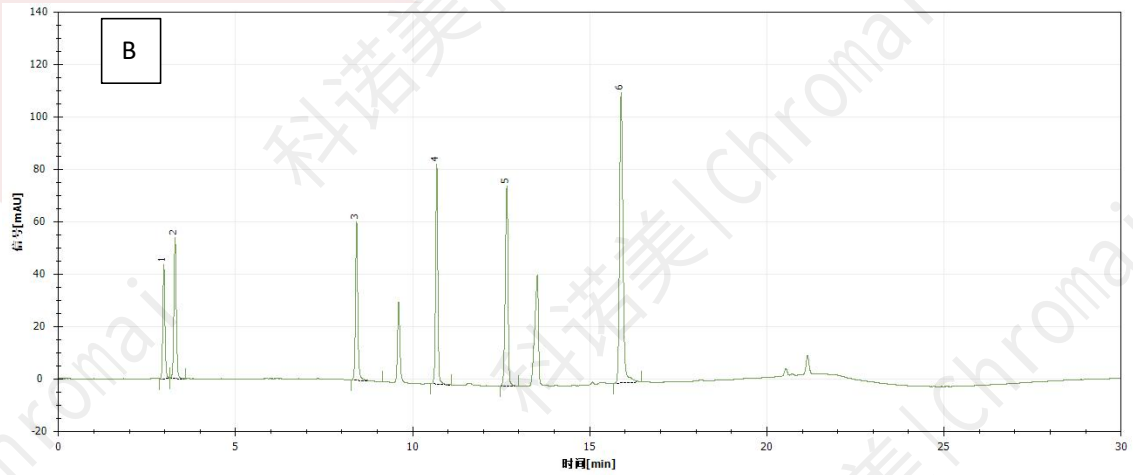
Time (min)	Pump (mL/min)	B%	C%
0.0	1.0	5	95

12	1.0	5	95

一、实验结果 V. Test result

1. 标准溶液谱图 Standard Solution Chromatograms





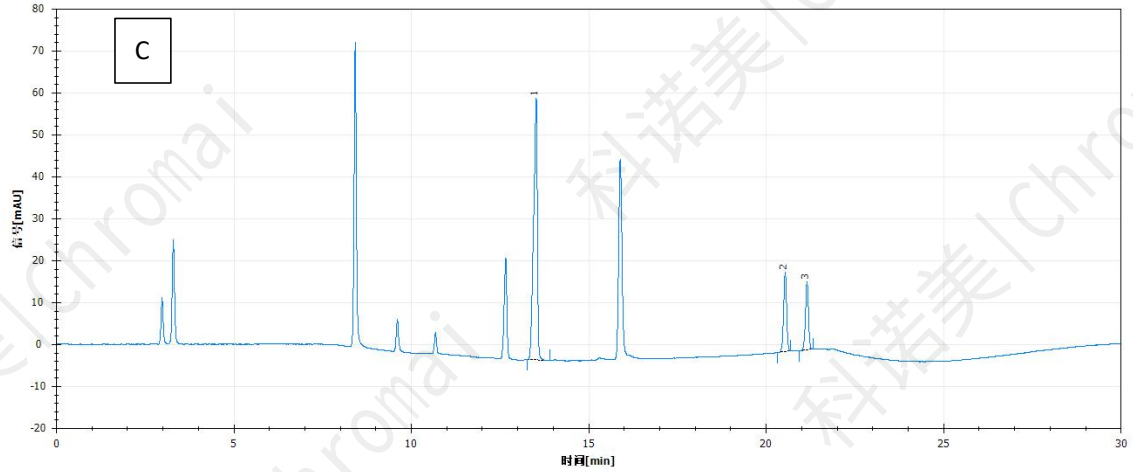


Figure 1 Chromatogram of Standard Solutions for Group I (Ten Raw Materials)

(A: 230 nm, 1. Nicotinamide; B: 250 nm, 1. Magnesium Ascorbyl Phosphate, 2. Ascorbyl Glucoside, 3. Kojic Acid, 4. 3-O-Ethyl Ascorbic Acid, 5. Potassium 4-Methoxysalicylate, 6. Ellagic Acid; C: 280 nm, 1. Ferulic Acid, 2. 4-Butylresorcinol, 3. Phenylethyl Resorcinol)

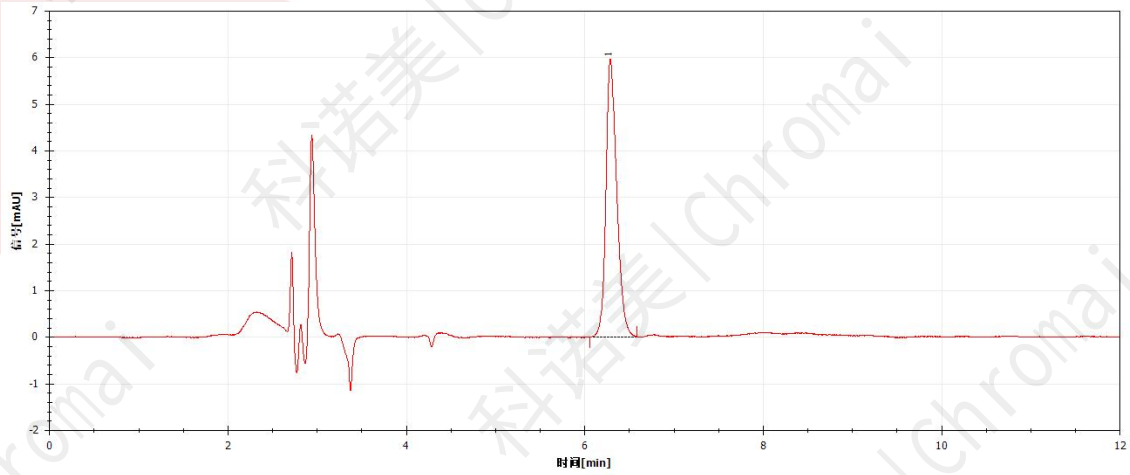


Figure 2 Chromatogram of Tranexamic Acid Standard Solution for Group II

2. Calibration Curves and Detection Limits

Standard working solutions were injected sequentially from low to high concentrations. Calibration curves were plotted with concentration as the x-axis and peak area as the y-axis. Within the linear range, the correlation coefficients (R^2) of all compounds were greater than 0.9997.

Table 1 Results of Calibration Curves and Detection Limits

Group	Peak	Compound	Concentration range (μg/mL)	Regression equation	Correlation coefficient	Detection limit (μg/g)
I	1	Magnesium Ascorbyl Phosphate	1.00~50.00	Y=18.709X-4.0885	r=0.9998	0.80
	2	Ascorbyl Glucoside	1.00~50.00	Y=25.472X-3.9978	r=0.9999	2.00
	3	Kojic Acid	1.00~50.00	Y=29.147X-9.1987	r=0.9999	2.20
	4	Nicotinamide	1.00~50.00	Y=31.721X-7.0329	r=0.9999	2.00
	5	3-O-Ethyl Ascorbic Acid	1.00~50.00	Y=37.621X-7.6843	r=0.9999	2.80
	6	Potassium 4-Methoxysalicylate	1.00~50.00	Y=43.264X-10.984	r=0.9999	0.80
	7	Ferulic Acid	1.00~50.00	Y=46.209X+3.9078	r=0.9999	1.00
	8	Ellagic Acid	1.00~25.00	Y=78.067X-37.932	r=0.9996	5.40
	9	4-Butylresorcinol	1.00~50.00	Y=11.897X-3.3951	r=0.9999	5.40
	10	Phenylethyl Resorcinol	1.00~50.00	Y=10.030X-2.2452	r=0.9999	9.20
II	1	Tranexamic Acid	5.00~200.00	Y=0.9654X+0.0848	r=0.9997	22.20

3. Repeatability

The same concentration of standard solutions containing the ten Group I compounds (such as magnesium ascorbyl phosphate) and tranexamic acid were injected six times. The repeatability results are shown in Table 2. The RSD% of retention time was less than 0.15%, and the RSD% of peak area was less than 1.5%.

Table 2 Repeatability Results

Group	Compound	Retention time RSD%	Peak area RSD%
I	Magnesium Ascorbyl Phosphate	0.13	1.09

	Ascorbyl Glucoside	0.11	0.71
	Kojic Acid	0.13	0.97
	Nicotinamide	0.13	0.70
	3-O-Ethyl Ascorbic Acid	0.08	0.89
	Potassium 4-Methoxysalicylate	0.14	0.80
	Ferulic Acid	0.10	0.88
	Ellagic Acid	0.14	1.00
	4-Butylresorcinol	0.07	0.94
	Phenylethyl Resorcinol	0.06	0.91
II	Tranexamic Acid	0.12	0.15

4. Spike Recovery

Different concentrations of the target compounds were spiked into real cosmetic samples. The calculated spike recovery results are shown in Table 3. The recoveries ranged from 82.8% to 104.0%.

Table 3 Spike Recovery Results

Group	Compound	Spike concentration（μg/mL)	Spike recovery（%)
I	Magnesium Ascorbyl Phosphate	2.50	92.00
	Ascorbyl Glucoside	2.50	96.80
	Kojic Acid	2.50	98.80
	Nicotinamide	2.50	96.40
	3-O-Ethyl Ascorbic Acid	2.50	95.20
	Potassium 4-Methoxysalicylate	2.50	104.00
	Ferulic Acid	2.50	82.80
	Ellagic Acid	1.00	85.00
	4-Butylresorcinol	2.50	95.20

	Phenylethyl Resorcinol	2.50	92.00
II	Tranexamic Acid	30.00	98.37

Conclusion

In this application, a rapid method for the determination of eleven compounds, including magnesium ascorbyl phosphate, in cosmetics was established using the Chromai Leaps Duo parallel liquid chromatography system.

All compounds showed correlation coefficients greater than 0.9997; retention time RSDs below 0.15%; and peak area RSDs below 1.5%. The spike recoveries in real samples ranged from 82.8% to 104.0%.

This method offers a high degree of automation, excellent stability, high sensitivity, and simple operation. By using a dual-injection system, analytical efficiency was greatly improved, cutting the detection time in half. The method is suitable for the rapid determination of eleven cosmetic compounds including magnesium ascorbyl phosphate.