Amino acid composition of the Chernobyl herb (Artemisia vulgaris L.) flora Russia and China

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Amino acid composition of mugwort herb (Artemisia vulgaris L.), grown in Russia and China

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SUMMARY

A method of thin layer chromatography (TLC) of amino acids of the herb wormwood (Artemisia vulgaris L.) of the flora of the Russian Federation and China has been developed. Using the developed technique, a comparative chromatographic analysis of the amino acid composition of extracts from wormwood herb was carried out: infusion, tincture with 70% alcohol; water-alcohol extracts (40%, 60%, 70%, 90%). Arginine, histidine, asparagine, proline, lysine, alanine, aspartic acid, methionine (PRC) were detected with the help of solutions of standard samples in a tincture of Chernobyl herb; arginine, histidine, asparagine, proline, lysine (RF). Arginine, histidine, asparagine, lysine, aspartic acid were found in herbal infusions (PRC and RF).

Key words: wormwood herb, Artemisia vulgaris L., amino acid composition, thin layer chromatography (TLC), flora of the Russian Federation, flora of the PRC.

RESUME

The Thin-Layer Chromatography (TLC) technique for amino acid analysis of Mugwort herb (Artemisia vulgaris L.) was developed. Then we conducted a comparative chromatographic analysis of amino acid composition of different sorts of extracts of the Mugwort herb (infusion, tincture at 70% alcohol, hydro-alcoholic extracts - 40%, 60%, 70%, 90%) with the developed method ... By using standard solutions of the amino acid samples we have discovered Arginine, Histidine, Asparagine, Proline, Lysine, Alanine, Aspartic acid, Methionine in Mugwort herb tincture (flora China), and Arginine, Aistidine, Asparagine, Proline, Lysine in Mugwort herb tincture (flora Russia). And we have discovered Arginine, Histidine, Asparagine, Lysine, Aspartic acid in Mugwort herb infusion (flora Russia and China).

Keywords: Artemisia vulgaris L.

Introduction

The flora of some regions of Russia and China in a number of positions coincides or is characterized by closely related plant species that are used in medicine in both countries. One of these plants is the common wormwood - Artemisia vulgaris L, the Asteraceae (Compositae) family - Asteraceae (Compositae).

The plant is official in Russia, but is not included in the State Pharmacopoeia of the PRC, despite its wide distribution in the territory of the PRC and its use in Chinese traditional medicine.

In the Russian Federation, the quality of dried wormwood herb is still standardized by FS 42-2094-83; there is no raw material regulation in China. In order to harmonize modern requirements for the quality of raw materials in the Russian Federation and China, it is urgent to develop a regulatory document that meets modern requirements for this kind of documentation in both countries.

In modern medical practice, natural medicines containing amino acids are becoming more and more popular, and there are more and more publications devoted to the study of the amino acid composition of various types of medicinal plant materials (MPRs) [3, 4, 5], including the TLC method [1, 2].

The aim of this study was a comparative study (by the method TLC) of the amino acid composition of the Chernobyl herb of the flora of the Russian Federation and China.

Materials and methods

The wormwood herb (Artemisia vulgaris L.) was harvested at the beginning of flowering in the Russian Federation (Moscow region) and in China (in Hebei province - Shijiazhuan city, Cangzhou city, Lantfang city and in Shandong province - Zibo and Qingdao) in 2009-2012. An infusion was obtained from the dried herb; tincture with 70% alcohol; water-alcohol extraction by extraction with 40%, 60%, 70%, 90% alcohol in a boiling water bath with a reflux condenser.

Thin layer chromatography was performed on DC-Alufolien Kieselgel 60 F254 plates, 0.2 mm thick (10×10 cm and 15×10 cm), Merck, Germany. Microsyringes, 10 µl, from Hamilton, Switzerland were used to apply the samples. In the course of the study, the optimal conditions for chromatography of the objects under study were worked out - the mobile phase, the time of saturation of the chamber, the time of activation of the chromatographic plates, the method of applying samples, etc. were selected.

For the development of chromatograms, a 0.25% alcohol solution of ninhydrin was used. The detection solution was applied by spraying chromatograms from a spray bottle. The processing of the obtained chromatograms was carried out in visible light after the development of absorption zones.

Freshly prepared aqueous solutions of standard sample (CO) powders (10 mg / 10 ml) of the following AA were used as reference solutions: L-aspartic acid "pure" ("Reakhim", Russia); Asparagine ("Merck", Darmstadt, Germany); L-methionine "chda" ("Reakhim", Russia); L-proline "h", chromatographically homogeneous ("Reanal", Budapest, Hungary); L-lysine hydrochloride "pure", chromatographically homogeneous ("Reanal", Budapest, Hungary); L-Alanine "h" ("Reakhim", Russia); Alanine ("Reanal" Budapest, Hungary); L-histidine monohydrochloride (for biochemistry) (Merck, Darmstadt, Germany); L-Arginine (Merck, Darmstadt, Germany) [6].

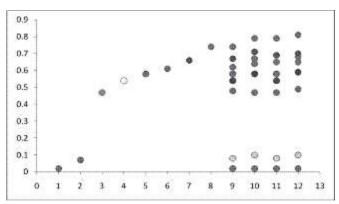
Research results and their discussion
When conducting research, the best separation of the absorption zones of AA of all

The studied extracts from the wormwood herb were observed during chromatography in the isopropanol - water (7: 3) system.

Methodology. The chromatographic chamber was pre-saturated with 5 ml of the mobile phase for 30 minutes, the plate was pre-washed and activated in an oven at 1100 C for 2–5 minutes. The starting line on the chromatographic plate is 1.5 cm from the bottom edge; sample volume - 5 µl; application - in the form of a strip 0.7 cm long and no more than 3 mm thick; the front of solvents was 7 cm. After passing the front and drying, the chromatogram was sprayed with 0.25% alcohol solution of ninhydrin, dried in air, and then placed in an oven for 2–5 minutes at a temperature of 110 ° C until adsorption zones appeared. Note: Processing of chromatograms should be carried out immediately after the development of adsorption zones, since the compounds formed during the interaction of ninhydrin with AA are rapidly destroyed.

The results are presented in table. 1-6, chromatogram scheme - on rice. one.

Chromatography of infusions of wormwood herb from the flora of the PRC and the Russian Federation revealed 8 zones of adsorption each (Fig. 1, Table 2).



Rice. 1. Chromatogram of the amino acid composition of the tincture (70% alcohol) and infusion of the herb of the chernorbylnik flora of the Russian Federation and China (system - isopropanol-water 7: 3); 1-8 - solutions of CO amino acids: 1 - arginine; 2 - histidine; 3 - asparagine; 4 - proline; 5 - lysine; 6 - alanine; 7 - aspartic acid; 8 - methionine; 9 - tincture of herbs from the flora of the PRC; 10 - water extract from the grass of the flora of the PRC; 11 - tincture

 $herbs\ of\ the\ flora\ of\ the\ Russian\ Federation;\ 12\ -\ water\ extract\ from\ the\ herb\ of\ the\ flora\ of\ the\ Russian\ Federation.$

Table 1

Characterization of the adsorption zones of solutions of standard samples (CO) of amino acids during TLC in the isopropanol - water (7: 3) system (average value of Rf of 3 definitions)

CO solution	Rf value	Zone color
Arginine	0.02	pink
Histidine	0.07	pink
Asparagine	0.47	brown
Proline	0.54	yellow
Lysine	0.58	pink
Alanin	0.61	pink
Aspartic acid	0.66	Violet
Methionine	0.74	pink

The Rf values and the colors of the AA adsorption zones of infusions practically coincide. As can be seen from the data in Fig. 1 and tab. 2, arginine, histidine, asparagine, lysine, and aspartic acid were presumably found on the chromatogram of infusions of wormwood herb from the flora of the People's Republic of China and the Russian Federation.

Chromatography of tinctures on 70% alcohol of wormwood herb of the common flora of the PRC and RF revealed 8 zones of adsorption each (Table 3).

As can be seen from the data in the table. 3, on the chromatogram of the tincture (70% alcohol) of the herb of wormwood of the common flora of the PRC, arginine, histidine, asparagine, proline, lysine, alanine, aspartic acid, methionine were presumably found; flora of the Russian Federation - arginine, histidine, asparagine, proline, lysine.

Chromatography of aqueous-alcoholic extracts with 40% alcohol and 60% alcohol from the wormwood herb of the common flora of the PRC and RF revealed 9 adsorption zones each (Table 4).

table 2
Characteristics of the most clearly defined zones of adsorption during TLC infusions
herbs wormwood of the common flora of the PRC and the Russian Federation in the isopropanol - water system
(7: 3) (average Rf value from 3 definitions)

Настой травы полыни Значение Rf		ой травы полыни		Значение Rf	Цвет зоны
		Раствор СО			
КНР	РФ	Цвет зоны	STATISTICS OF PERSONS		
0,02	0,02	розовый	Аргинин	0,02	розовый
0,1	0,1	розовый	Гистидин	0,07	розовый
0,47	0,49	бурый	Аспарагин	0,47	бурый
0,58	0,59	розовый	Лизин	0,58	розовый
0,64	0,65	фиолетовый	Аспарагиновая кислота	0,66	фиолетовый
0,67	0,68	розовый			
0,71	0,7	фиолетовый			
0,79	0,81	розовый	,		

Table 3
Characterization of the most clearly defined adsorption zones during TLC tinctures on 70% alcohol herbs of wormwood of the common flora of China and the Russian Federation in the system

isopropanol - water (7: 3) (average Rf value from 3 determinations)

Настойка травы полыни на 70 % спирте Значение Rf			D. D.	100	
		TT	Раствор СО	Значение Rf	Цвет зоны
КНР	РФ	Цвет зоны			
0,02	0,02	розовый	Аргинин	0,02	розовый
0,08	0,08	розовый	Гистидин	0,07	розовый
0,48	0,47	бурый	Аспарагин	0,47	бурый
0,54	0,54	желтый	Пролин	0,54	желтый
0,58	0,58	розовый	Лизин	0,58	розовый
0,62	0,65	розовый	Аланин	0,61	розовый
0,67	0,69	фиолетовый	Аспарагиновая кислота	0,66	фиолетовый
0,74	0,79	розовый	Метионин	0,74	розовый

As can be seen from the data in the table. 4, we obtained similar chromatograms of aqueous-alcoholic extracts of 40% and 60% alcohol for wormwood herb from the Russian Federation and China. Arginine, histidine, asparagine, methionine were presumably found on the chromatogram of the aqueous-alcoholic extraction of 40% and 60% alcohol from the herb of wormwood of the common flora of the PRC; flora of the Russian Federation - arginine, histidine, aspartic acid.

Chromatography of aqueous-alcoholic extracts with 70% alcohol from the wormwood herb of the common flora of the PRC and RF revealed 10 and 8 zones of adsorption, respectively (Table 5).

As can be seen from the data in the table. 5, on the chromatogram of an aqueous-alcoholic extraction of 70% alcohol from the herb of wormwood of the common flora of the PRC, arginine, histidine, asparagine, proline, lysine, alanine, aspartic acid, methionine were presumably found; flora of the Russian Federation - arginine, histidine, asparagine, proline, lysine; aspartic acid, methionine.

Chromatography of aqueous-alcoholic extracts with 90% alcohol from the wormwood herb of the common flora of the PRC and RF revealed 9 adsorption zones each (Table 6). The Rf values and colors of the AA adsorption zones of the extracts coincide.

As can be seen from the data in the table. 6, on the chromatogram of aqueous-alcoholic extracts with 90% alcohol from the herb of wormwood of the common flora of the PRC and the Russian Federation, arginine, histidine, asparagine, and methionine were presumably found.

Conclusions:

1. Developed a TLC method for the analysis of the sum of amino acids (AA), contained in the herb wormwood. Using the developed technique, a comparative chromatographic analysis of the AA-composition of various alcoholic extracts from the herb wormwood of the common flora of the PRC and the Russian Federation was carried out.

Table 4

Characteristics of the most clearly defined zones of adsorption during TLC of aqueousalcoholic extracts of 40% and 60% alcohol from wormwood herb of the common flora of the PRC and RF in the isopropanol - water (7: 3) system (average Rf value from 3

definitions)

Водно-	пиртовые изв и 60 % с	лечения 40 % :пиртом	спиртом
из травы полыни обыкно- венной флоры КНР		из травы полыни обыкно- венной флоры РФ	
значение Rf	цвет зоны	значение Rf	цвет зоны
0,02	Розовый	0,02	Розовый
0,08	Розовый	0,1	Розовый
0,41	Розовый		
0,47	Бурый	0,47	Розовый
0,55	Фиолетовый		
		0,58	Бурый
0,64	Розовый	0,64	Фиолетовый
		0,71	Розовый
0,75	Розовый	0,77	Розовый
0,82	Розовый	0,82	Розовый
0,87	Желтый	0,87	Желтый

Table 5 Characteristics of the most clearly defined zones of adsorption during TLC of aqueous-alcoholic extracts with 70% alcohol from grass wormwood

China and the Russian Federation in the isopropanol - water system (7: 3) (average value of Rf from 3 definitions)

	a c i i i i	.01.0)		
Водно-с	пиртовые изв	лечения 70 %	спиртом	
из травы полыни обыкно- венной флоры КНР		из травы полыни обыкно- венной флоры РФ		
значение Rf	цвет зоны	значение Rf	цвет зоны	
0,02	Розовый	0,02	Розовый	
0,08	Розовый	0,08	Розовый	
0,48	Бурый	0,47	Бурый	
0,54	Желтый	0,54	Желтый	
0,58	Розовый	0,58	Розовый	
0,62	Серый			
0,67	Фиолетовый	0,65	Фиолетовый	
0,74	Розовый	0,74	Розовый	
0,81	Желтый	1770000000		
17.		0,84	Желтый	
0,87	зеленый			

2. It was found that infusions and water-alcohol extracts (90% ethanol) from the herb of wormwood of the common flora of the PRC and the Russian Federation are similar, and the rest of the extractions differ insignificantly in the AA composition. Using solutions of standard samples, it is highly likely that we

most of the AAs present on the chromatograms of the studied objects were identified.

3. In the herb of wormwood of the common flora of the PRC, the presence of arginine is shown, histidine, asparagine, proline, lysine, alanine, aspartic acid, methionine; flora of the Russian Federation - arginine, histidine, asparagine, proline, lysine, aspartic acid, methionine (it was not possible to detect alanine, in contrast to the herb wormwood from the PRC).

Table 6

Characteristics of the most clearly defined zones of adsorption during TLC of aqueousalcoholic extracts with 90% alcohol from the herb of wormwood of the common flora of China and the Russian Federation in the isopropanol - water (7: 3) system (average Rf value out of 3 definitions)

Водно-с	пиртовые изв	лечения 90 %	спиртом	
из травы полыни обыкно- венной флоры КНР		из травы полыни обыкновенной флоры РФ		
значение Rf	цвет зоны	значение Rf	цвет зоны	
0,02	Розовый	0,02	Розовый	
0,08	Розовый	0,08	Розовый	
0,41	Розовый	0,41	Розовый	
0,47	Бурый	0,47	Бурый	
0,55	Фиолетовый	0,55	Фиолетовый	
0,64	Розовый	0,64	Розовый	
0,75	Розовый	0,75	Розовый	
0,82	Розовый	0,82	Розовый	
0,87	Желтый	0,87	Желтый	

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