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Chemical Composition of Low-Molecular Weigth Organic Compounds (LMWOC) of Water Extracts from Cistanche Deserticola Stolones Depending on Treatment

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Abstract: *Ethnopharmacological relevance*: Cistanche deserticola (CD) known as "ginseng of the desert" is used in Chinese traditional medicine. Currently most Cistanche deserticola is gathered in Kazakhstan and exported to China and South Korea. Cistanche is considered to be stronger than ginseng. Few studies have investigated the chemical composition and biological properties of this plant. The present study was designed to explore biological activity, medicinal properties of Cistanche deserticola for potential production of herbal remedies, teas and energy drinks.

Key words: Cistanche % Bioactive substances % Stolone % Raw material treatment % Herbal tea % Energy drink

INTRODUCTION

The first published information about cistanche appeared 1,800 years ago in one of the oldest books on herbal medicine in China, known as Shennong Bencaojin (Dictionary of Chinese Materia Medica, 1977)[1]. Cistanche is used for disorders in male and female reproductive organs, diseases of the urinary system, musculoskeletal system, it also improves blood circulation. The dried succulent stems of Cistanche are one of the most widely used in traditional Chinese medicines, which earned the honor of being called the "ginseng of the desert". For the last 10 years there has been growing medical interests on herba Cistanche for its remarkable bioactivities including antioxidation, neuroprotection and antiaging [2]. In stolones of the desert plant Cistanche deserticola there is high concentration of biologically active compounds accumulated. In Kazakhstan Cistanche grows over a wide area and last 20 years is exported to China, however barely used in the country itself. There is vast variety of products that can be produced from Cistanche. Due to lack of processing technologies Kazakhstan does not receive profit from realization of this economically valuable plant of Kazakhstani flora. The aim of this work is the development of processing technologies of raw materials from Kazakhstani desert plant Cistanche deserticola, having high biological activity and creating herbal remedies for further mass production.

Aim of the Study: This work developed new drying and heat treatment techniques for conservation of large volumes of Cistanche stolones and technologies of extraction water soluble and low molecular weight organical components (LMWOC). Here, we have developed method of quality of raw material control by means of GCMC chromatography. In this work we introduced juice as a main ingredient of herbal teas and tinctures.

MATERIAL AND METHODS

Plant Material: Cistanche plant was collected from the Moinkum Desert, Kazakhstan. Work was carried out on the fields and in the laboratories. Field research consisted from trips to the desert Moinkum and collecting fresh crops of CD, by taking out stolones in saxaul (Haloxylon) forest.

Options of Extraction for GCMC Chromatography: For GCMS chromatography 5 options of extracts were prepared:

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- Fresh stolones pounded in mortar up to the homogeneous weight and centrifuged 10 minutes at 18 thousand rpm. Then to supernatant added methanol in the ratio 0.2: 0.8 (1st option).
- Fresh stolones cut into 4 parts (longitudinal and cross) for damage prevention and for better drying of a core of the plant. Extracted 8 g of stolones added to 500 ml of hot distilled water. Infused as the traditional way of brewing tea. Mixed methanol with the extract in the ratio 0.2: 0.8 (2nd option).
- Fresh stolones cut into 4 parts (longitudinal and cross) for damage prevention and the best drying of plant core. Extracted 8 g of stolones added to 500 ml of the hot distilled water. Boiled for 5 minutes. Mixed methanol with the extract in the ratio 0.3: 0.7 (3rd option).
- Dried up in the cut condition stolones crushed in a mill. 1 g of the received powder of Cistanche dissolved in 500 ml of the hot distilled water. Then infused as the traditional way of brewing tea. Mixed methanol with the extract in the ratio 0.3: 0.7 (4th option).
- Previously dried cut stolones crushed in the mill. 1g of the received Cistanche powder dissolved in 500 ml of hot distilled water and boiled for 10 minutes. Mixed methanol with the extract in the ratio 0.3: 0.7 (5th option).

Gas Chromatography: Qualitative complex of LMWOC in Cistanche stolones determined by gas chromatography with mass-selective detector (MSD) method.

Chromatography Regime Had the Following Parameters: Injector temperature-280 °C. The volume (drawings) in stick-1 mkl, Split = 6:1. MSD interface temperature-300°C. Gas carrier stream (helium)-1,5 ml/min., Mode: Constant flow. Column: DB-5MS, 0.25mm*30m*0.25mkm, Agilent company. Temperature mode of thermostat columns: 7 °C/minutes till 80°C; 320 °C (5 °C/minutes). Analysis Time 20-80 minutes.

Search of substances is carried out automatically by comparison of chromatogram integrated peak range in a sample with a library range.

Derivatization of LMWOC: Derivatization was applied to improve components detection and identification. For this purpose received extracts were hydrolyzed in KOH: took 1 ml of extract and added 0.1 ml 3M KOH in H2O. Hydrolyzed 1 hour at 80°C, solution cooled and added 4 ml of H_2O + 0.1ml 5M HCL and mixed. Added 5 ml of TBME, stirred up 15 min., centrifuged. the solution was evaporated and 100 mkl MSTFA (N-Methyl-N-(trimethylsilyl)trifluoroacetamide) was added to the dry deposit, maintained 30 min. at 60 °C. After cooling solution used for GCMS.

The Used Equipment: Gas chromatograph: Agilent 6890N and mass-selective detector: Agilent MSD 5973. The used software: ChemStation© Agilent Technologies program.

Data Analysis: International libraries of chemical compounds were used for identification: Databases NISTO2.L, Wiley7n.1, PMW_Tox3.1. For rough preliminary analysis %-age of components correspondence was higher than 25, for more precise identification more than 70%. It is connected with the research purpose-production quality assessment by GCMS, instead of only chemical components identification of Cistanche.

RESULTS AND DISCUSSIONS

Cistanche Stolones Prossesing Technology Development: Previously applied in Kazakhstan CD stolones drying method required large amount of raw materials, therefore it was necessary to develop a less costly technology. Earlier CD stolones were dried up by the following methods:

- Spread in a thin layer on a flat surface and constantly turned in order to dry them evenly. Drying conducted not in a shadow, as usual, but in sunny open-air places. It was due to the spring period (April 10-May 10, depending on weather conditions of the year) and relatively cooler temperature for the desert.
- Treated with boiling water for 2-3 minutes for destruction of worm's larvae and conducted drying as in the first method.
- Plants cut lengthways (in Kazakhstan) or across (in China) and dried as in the first method. Required area for drying about 1600 tons of raw CD is equal to a field of small airfield.

These methods are characterized by large amount of waste and considerable shrinkage. So when drying whole stolones the outcome makes 1 to 8, i.e. 12%. It was necessary to create more modern and fast drying method, because during the process of drying the plant, with up to 1m length and thickness to 20cm, is still alive and is spending over nutrients. We created the following technology of raw materials processing:

Preliminary Raw Materials Processing: Quality of raw materials depends on preprocessing. It carries out before drying. For this purpose raw materials (stolones) were washed under running water to clean the remains of sand, clay, mud, spread out on a clean surface (large pieces of paper), viewed and removed the damaged part of the worm-eaten.

Technology of Cistanche Stolones Drying: Timely and properly collected raw drug plants should take the next important stage of preparation-drying. The compliance with the rules of drying is vital to preserve the medicinal properties of the plants. The purpose of drying is the rapid cessation in plants intracellular biochemical processes in which under the influence of cell enzymes there is a destruction of active substances. The fastest natural way of biochemical process termination is cell dehydration since the process can take place only in aquatic environment. In freshly harvested Cistanche plant materials the content of water is 56-64%. Removal of moisture up to 20% has reduced the rate of biochemical reactions and activity of enzymes. When level of water decreases by 10-14%, activity of enzymes ceases completely, i.e stops the intracellular processes, leading to the decomposition of active substances. Furthermore, reduction of the plant weight moisture leads to a delay and stops the development of different fungi and microorganisms, which also reduces the quality of raw materials.

Methods and conditions of various plants drying are numerous and depend on the type of the material and number of active substances, amount of moisture and etc. As the research results showed Cistanche deserticola mainly contains glycosides and sugar, in this regard the following parameters of raw materials drying were chosen:

- Shadow drying where stolones cut into small slices (1-1.5 x 5-7 cm) and dried naturally.
- Drying where stolones cut into small slices (1-1.5 x 5-7 cm) dried by hot air at 60 °C temperature (drying at this temperature goes quickly and digesting glycosides activity of the enzymes rapidly stops).

Basic principles of drying-timeliness of drying (not later than in 2 hours after collecting), raw materials preparation for drying, choosing temperature regime, purity of the drying room. For the given regime of drying the yield is 1:3, i.e. more than 30%.

Technology of Cistanche Stolones Freezing: According to long-term research we offer to store Cistanche stolones in the freezer. Before stolones were not normally frozen, but according to our observations the quality of raw CD does not deteriorate. Raw materials, after preparation, purification and washing, were placed in the refrigerator at the temperature-24°C. The storage period is 1-2 years without changes to a quality of the raw materials. After thawing, stolones were either cut into standard size pieces for subsequent drying, or juice was squeezed out or crude extracts were extracted.

Cistanche Stolones Storage: Well dried up medicinal raw materials have to contain hygroscopic moisture no more than 12-15%. It is necessary to store raw materials in packages; paper and cloth bags; boxes, the boxes covered with paper; in jars. When packing in packages, bags, jars and other containers inside labels were put with the title of raw materials type and the harvested time, the dried raw materials were stored in dry, cool and well aired rooms without access of direct sun light. Normally stolones storage period is 2-3 years.

Definition of Raw Materials Preparations Terms: Biological activity of freshly harvested stolones is high and local people use them for food. However harvesting is ongoing only for 10 days. Then worms and animals eat stolones. It is necessary to pick up optimal term of harvesting as on the 10^{th} day CD stolones tissue hardens and become not tasty. It is also required to standardize sizes of parts, stolones cleaning method and stolones cutting method. We offer to prepare stolones without visible signs of defeat by worms, to wash out surface of stolones with running water, to cut stolones on slices of 1-1.5 x 5-7 cm in size on industrial root cutter.

Chemical Complex of LMWOC: As well as the majority of desert plants Cistanche has a set of biologically active substances. The effect is similar to mezophytic ginseng and linked to a group of compounds. However, it is possible to tell with confidence that after identification and studying of individual components of Cistanche itself can act as a drug and not as a part of herbal complex. They are definitely natural and not artificial. Previously conducted research with sufficient reliability showed biological activity of pure powder, tinctures or extracts from Cistanche [3].

Main objective of our research was to determine the processing conditions on the quality of raw materials. As a marker of quality the component complex of LMWOC was used. Among which there are not yet identified components causing biological activity in Cistanche. The separation of the samples was carried out on the Agilent

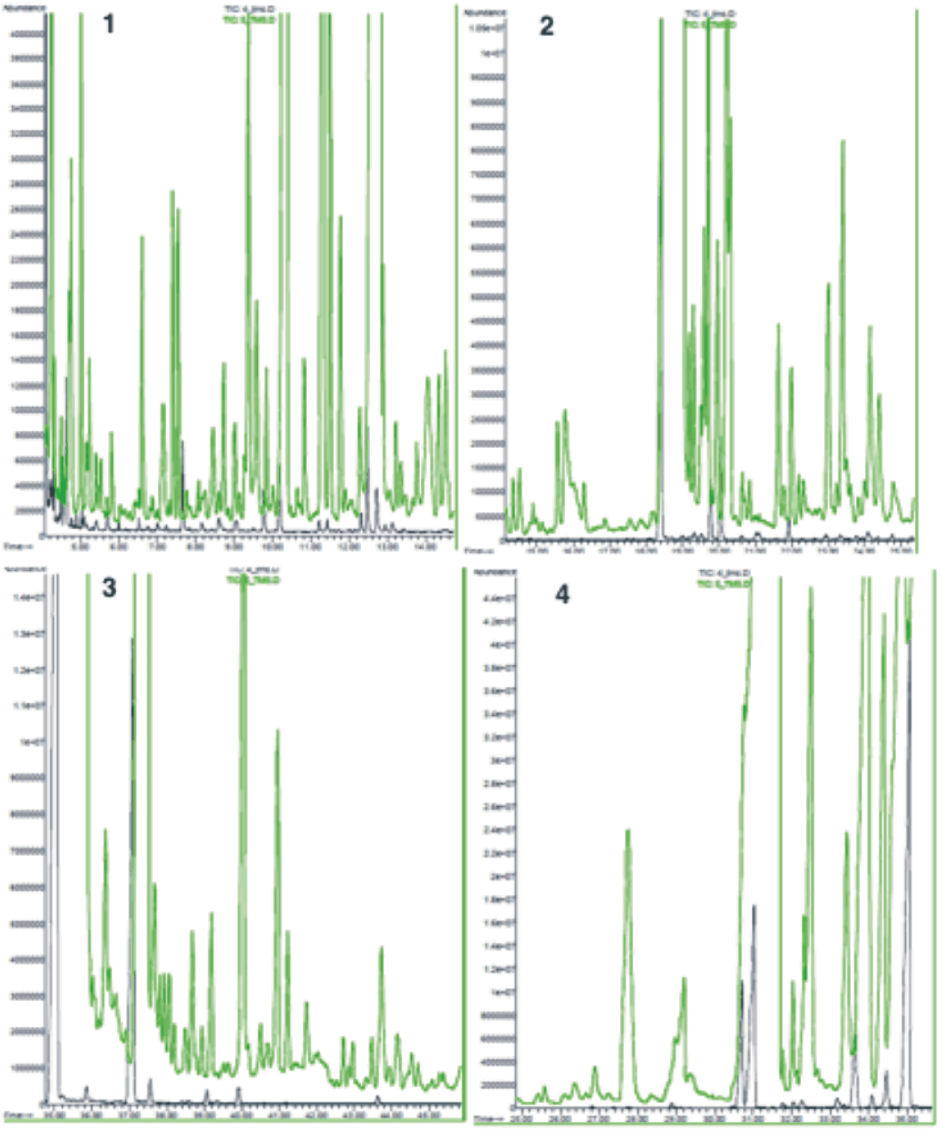

Fig. 1: Imposed chromatogram of the two options of crude Cistanche stolones: extracted by hot water (green color) and crude Cistanche stolones, treated by boiling water 5 min (separation time-90 minutes)

6890N chromatograph with the mass selective detector Agilent MSD 5973. The first studies showed that LMWOC of Cistanche is low volatile that caused identification of the minimum number of components. This was due to the fact that water extracts contained a large amount of nonpolar, non-volatile compounds.

After samples derivatization for chromatography separation improved significantly. In sample 4 on the chromatogram is possible to find 108 components, in sample 5-92 components, in sample 3-138 components, in sample 2-154 components, in sample 1-478 components. Between them there were identified: in the 5th sample-46, in the 4th sample-41, in the 2nd sample-89, in the 3rd sample-66, in the 1st sample-258 (Fig. 1). Identification in three world libraries of chemical compounds showed existence of large amount of carbohydrates and phenol connections.

After identification in the sample 4 were found: Butyric acid, Hexanoic acid, 5-Hexen-2-one, Ethanedioic acid, Glycine, Benzoic acid, Propanoic acid, 2-Butenedioic acid, 4-Dibenzofuranamine, 4,5-Difluoro-2-nitroaniline, 2-keto-gluconic acid, Malic acid, 2,3,4-Trihydroxybutyric acid, n-Hexadecanoic acid, Mefenamic acid, Ribonic acid, N,N-ethanamine, Dodecanoic acid, 1,3,5-Triazine, D-Erythro-Pentonic acid, 2-Pentenedioic acid, D-Glucose, Inositol, Benzamide, D-Gluconic acid, D-Fructose, 24-

Norchola-20,22-dien-14-ol, D-Galactose, Acrylic acid, D-Mannitol, beta.-L-Mannopyranose, Inositol, 1-Propene-1,2,3-tricarboxylic acid, Erythrose, Glucopyranose, Stearic acid, Isobonafousine, 8,2'-aminoanhydroadenosine alpha-D-Glucopyranoside, 2,3-Diacetyl-6-methyl-4,5-diphenyl-o-diacylbenzol, Guaicol-beta-d-glucopyranoside.

In sample 5 were found: 1,7-Octadiene, Propanoic acid, Hexanoic acid, 2-Ethyl-1,3-bis(trimethylsilyloxy)propane, oxalic acid, Butanoic acid, Glycine, Benzoic acid, Butanedioic acid, Pyridine, Diphenylamine, Pelargonic acid, Benzoic acid, 1H,3H-Naphthopyran, 1,2-Difluoro-3,3-dimethyl-1-amine, Methyl 2,3-Dihydro-1H-pyrrolo[1,2-a]indole-9-carbonic acid, Malic acid, 1,4-Bis(1,1-dimethyl**S**prop**S**yl)-2,5-dimeth**S**oxy**S**benzene, 4-Methoxy-1,2-diphenylbenzene, Threonic acid, 1,10-Phenanthroline, Acetamide, Triethylene glycol, D-Fructose, 24-Norchola-20,22-dien-14-ol, Mannose, Furocoumarine, D-Mannitol, Galactitol, L-Gluconic acid, Acetic acid, Mannose, Saccharic acid, Palmitic acid, meso-Inositol, Stearic acid, N,N'-propyldiyloxyethylen, alpha-D-Glucopyranoside, Maltose.

In sample 2 were found: 2-Fluorobenzoic acid, 2-Hydroxy-2-methyl-but-3-enyl 2-methyl-2(Z)-butenoate, 1-Naphthalenemethanol, 2-(5'-nitro-3'-thienyl)pyrimidine, Pentanoic acid, Acetic acid, 1-(3-Methylbutyl)-2,3,4,6-tetramethylbenzene, 4-Ketoglucose, Glycine, 4-Thiazolidinecarboxylic acid, Glycine, 1-Hydroxymethyl-8-methyldibenzothiophene, Phosphoric acid, Malonic acid, Fumaric acid, 4-Methyl-5-trifluoromethyl-4H-1,2,4-triazolin, Norphenazone, Malic acid, 2-Piperidinecarboxylic acid, Furosardonin A, 1-aza-1-(methoxyiminomethyl)-5-carbamoyl)-cyclonopropene, Succinic acid, 4-(4-Fluorophenyl)-1H-imidazole, Glucuronic acid, 2-Deoxy-D-galactose, Glutaconic acid, Catecholborane, Benzene, Mannose, Gluconic acid, alpha.-DL-Lyxopyranose, 6-Indolizinecarbonitrile, 1H-Indole-3-carboxaldehyde, 9H-Xanthen-9-one, Benzene, Saccharic acid, D-Fructose, Arabinose, Ribose, Glucose, Sorbose, Mannose, D-Xylopyranose, 24-Norchola-20,22-dien-14-ol, 1,2-diphenyl-4-pyrrol, L-Tyrosine,, Inositol, Gulonic acid, á-D-Glucose 1,6-bisphosphate tetra(cyclohexylammonium), Palmitelaidic acid, Palmitic acid, D-Gluconic acid, Xylonic acid, Guanosine, Inositol, margaric acid, Linoleic acid, Oleic acid, Stearic acid, Glucuronic acid, phthalic acid, Methyl â-D-galactoside, 16-Ketoestradiol, 4-Imidazolidinone, Docosanoic acid, Methyl á-D-glucoside, Quinoline, 8H-Dinaphtho[2,3-c:2',3'-h]phenol, Octyl glucoside, Maltose, Methyl á-D-glucoside, â-Sitosterol.

In sample 3 were found: S-Methyl methanethiosulfinate, Glycine, 2-Pentenal, lactic acid, Hexanoic acid, Acetic acid, N-Acetylelmerrillicine, propionic acid, Malonic acid, succinic acid, 4-(4'-Methoxyphenyl)-2-methyl-6-phenylpyridine, Fumaric acid, Butyric acid, 4,6,4'-trimethoxy-gris-3'-ene-3,2'-dione, Pyrimidinetetramine, Glutamine, L-Aspartic acid, Ethylamine, 1,2-bis(n-butylthio)-4,5-dimethoxybenzene, naphtho(1,8-bc)thiete 1-oxide, l-4,5-Dimethyl-3-(1-carboxyethyl)-.delta.(4)-thiazol, Arabinose, Mannose, D-Xylopyranose, benzofuran-2(3H),1'-[3]cyclohexene, D-Glycero-L-manno-Heptonic acid, Papaverine, Galactaric acid, Mucic acid, D-Fructose, alpha-l-idofuranuronic acid, 1-(2-Nitrobenzyl)isoquinoline, Sorbose, glucose, 24-Norchola-20,22-dien-14-ol, Xylose, 3-3-pyridinylferrocenophane, 1,2-diphenyl-4pyrrol, L-Tyrosine, D-Sorbitol, D-Mannitol, x,x,x-triisopropyl-o-xylene, Palmitic acid, Indole-3-acetic acid, Myo-Inositol, Stearic acid, Glycine, Sedoheptulose, 5-alpha-Androstane (steroids), Maltose, D-Turanose, 3-Morpholinoandrosta-3,5-dien, beta-d-glucopyranoside, Thymol-beta-d-glucopyranoside, Pregnan-17-ol, Talose, alpha-D-Glucopyranoside.

In sample 1 were found: 2-methyl-1,3-butanediol, Cyclohexene, Lactic acid, milk acid, Butanoic acid, Butyric acid, Propanoic acid, Acetic acid, N-methylaminopropionic acid, t-butylpentamethyldisiloxane, 1,2-Ethanediamine, 1H-Indole-3-acetonitrile, Pyridine, Phosphoric acid, 2-Benzylidenehydrazono-3-methyl-2,3-dihydrobenzothil, L-Alanine, Propanedioic acid, Malonic acid, 2,3,1-Benzodiazaborine, Glycine, 1-(5',6',7'-trimethoxyisoquinolin-8'-yl)ethanone, 3,8-Dioxa-2,9-disiladec-5-yne, Arabino-Hexos-2-ulose, Benzeneacetic acid, Phenylacetic acid, 1(2H)-Phenanthrenone, 12-Methoxy-19-norpodocarpa-8,11,13-tetraen-3-one, Hexanoic acid, caproic acid, 3-Pyridinecarboxylic acid, 2-Desoxy-pentos-3-ulose, l-Threonine, 2-Butenoic acid, Crotonic acid, Butanedioic acid, Succinic acid, 2-Butenedioic acid, Fumaric acid, Tetrahydrobenzene, 2H-Cyclopentacyclooctene, Methoxybismethane, L-threonine, fatty acid, Acrylic acid, 1H-Indole, 4-Nitrodibenzothiophene, Methanamine (gas), Benzacridine, 7-phenyl-2-azafluoren-9-one, 12,13-dihydroxytotara-8,11,13-trien-7-one, 3-Benzoaziridine, Malic acid, 4'-Methyl-3-(2-thienyl)acrylophenone, 4'-Methyl-3-(2-thienyl)acrylophenone, Butane, L-Proline, L-Aspartic acid, (3,6-D2)Tricyclo[6.2.2.0(2,7)]dodeca-2(7),9-diene, Retinoic acid (vitamin A acid), 1,2,3,4-Tetrahydro-6-methoxy-2,4-diphenylquinoline, 2,3,4-Trihydroxybutyric acid, 1(2H)-Phthalazinone, 4-Hydroxyphenylethanol, Tyrosol, Pentanedioic acid, Glutaric acid, (3,6-D2)Tricyclo[6.2.2.0(2,7)]dodeca-2(7),9-diene, thieno[2',3':4,5]thieno[2,3-c]quinolone, 3-Phenpropenoic acid, hydrocinnamic acid, Hexanedioic acid, Adipic acid,

L-phenylalanine, Xylonic acid, Lyxonic acid, D-Glucitol, Sorbitol, 1,2,4,8-Tetramethylbicyclo[6.3.0]undeca-2,4-diene, L-Altrose, D-Arabinose, 5,10-Dihydro-10-(1'-methyl-1'-nitroethyl)indenol, 6,7-Bis-2,3-naphthalenedicarboxyl, 1H-Indole, Mannose, Galactitol, Xylitol, 1-Propene-1,2,3-tricarboxylic acid, trans-Aconitic acid, D-Xylose, l-Glutamine, Kaurane-16,18-diol, 1-Methyl-4-ethyl 2-phenylsuccinate, D-Fructose, 7-Isopropenyl-1,4a-dimethyl-4,4a,5,6,7,8-hexahydroxil, Arabinose, Stigmast-22-en-3-ol, Glucose, beta.-D-Galactofuranose, 24-Norchola-20,22-dien-14-ol, Glucose-1,2,4-Trisnaphthalene, 4-(N-(3,4-dichlorophenyl)amino)-5,6-dimethyl-7H-phenol, 2-(p-Biphenylyl)-4-mentyl-3-phenylpyridine, 3.alpha.-Phenylcholest-5-en-3-beta.-ol, Glucosamine, galaktaric acid, Gulonic acid, 3-Bromo-5-ethoxy-4-hydroxybenzaldehyde, 7.alpha.-(1,2-epoxy-1-methylethyl)-2.alpha.-hydroxybenzaldehyde, di(methyl) 1-ethyl-2,9-dihydro-1H-carbzole, Galactaric acid, Mucic acid, Galactonic acid, D-Galacturonic acid, 2,4,6(1H,3H,5H)-Pyrimidinetrione, 1H-Indole-3-acetic acid, Gluconic acid, Inositol, Arabino-hexaric acid, Linoleic acid, 11-cis-Octadecenoic acid, vaccenic acid, L-Tryptophan, á-D-Glucopyranose, 2-Keto-d-gluconic acid, (Z)-1,3,4,5-Tetrahydro-3,3,3',4'-tetramethyl-5-ox, 3,9-diacetoxy-1,2-dimethoxy-7-formyl-6a,7-dehydropropene, Sedoheptulose, 2-Oxo-5-benzoyl-4-phenyl-6-(4-tolyl)-1,2-dihydropropene, 1-iodo-2-methoxy-5-methylbenzene, Phosphine, 1H-Indole-2,3-dione, 2-O-Glycerol-á-D-galactopyranoside, 2,5-di-(E)-(2-carbomethoxystyryl)thiophene, 2H-Furol, Sorbitol, 2-(1-aza-4-oxa-cyclohexyl)-benzoxazole, 2-(2-Bromo-4-methylphenoxy)-N'-{[1-(4-nitrophenyl, 1,1,4a-Trimethyl-5,6-dimethylenedecahydronaphthalene, Uridine, D-Glucuronic acid, sedoheptulose, Testosterone phenylpropionate, methyl dihydropimarate, 2-Deoxy-galactopyranose, Estra-1,3,5(10)-trien-17-ol, 3-methoxy-, acetate, Azulene, Naphthalene, 2H-Cyclopentacyclooctene, 3,9-diacetoxy-1,2-dimethoxy-7-formyl-6a,7-dehydropropene, 4,4'-bis(3,4-dimethoxystyryl)-2,2'-bipyridine, glutamic acid, Naphthalene, 5H-Dibenzo[a,d]cycloheptene, 5-methylene, Glucopyranose, á-D-Glucopyranoside, 2-[2'-Phenoxathiinyl]cinchoninic acid, Vitamin D3 dimethylphosphate, trans-Cembranolide, 1H-Indole, Galactose, Adenosine, Pregn-4-ene-3,20-dione, 13-glucopyranosyloxydocosanoic acid, Trifluorosilyl-octamethylcyclotetrasilazane, Lactose, Maltose, 2-(2-Bromo-4-methylphenoxy)-N'-{[1-(4-nitrophenyl, 2-O-glyceryl-á-D-glucopyranoside, Melibiose, 3-Bromo-5-ethoxy-4-hydroxybenzaldehyde, Octamethylcyclotetrasilazane, 5-(p-Chlorophenyl)-3-(6-methyl-3-pyridyl)-1-(p-tolyl)-2-pyrazoline, beta-l-Galactopyranoside, 13-glucopyranosyloxydocosanoic acid, Cliogoinol, D-Glucose, D-Turanose, 2-bianthrone, 2-Dimethyl(pentafluorophenyl)silyloxyadamantane, 4-cyano-7-mesityl-5H-thieno[2,3-c]thiopyran, beta-D-galactopyranuroni, trans-3'-Dimethylamino-4-(methylthio)chalcone, Thymolphthalein, D-Glucose, 6,7-dihydroxycoumarin-.beta.-d-glucopyranoside, 6,7-bis(trimethylsilyl)-4-methoxy-1-azabiphenylene, 5.beta.-Androst-7-en-6-one, 2,5-Bis(5-formyl-4-propyl-2-pyrrolyl)thiophene, Benzenesulfonic acid, 2-Pyridone, Bisacodyl-M, beta-Sitosterol, 2-Pyridinamine, 1,4,6-Trimethyl-1,2,3,4-tetrahydroquinoxaline-2,3-dione, Turanose, 2-phenyl-2-p-anisyl-3,3-bis(trifluoromethyl)oxira, Patchouli alcohol, Benzoic acid, Phenethylamine, 1,1,4a-Trimethyl-5,6-dimethylenedecahydronaphthalene, Flavone acetic acid, (4-Hydroxy-3-methoxyphenyl)ethylene glycol, Guaicol-.beta.-d-glucopyranoside, threo-2,5-Hexodiulose, Methyl-19-norisoanticopalate (deuterate), 1,1,4a-Trimethyl-5,6-dimethyl decahydronaphthalene, Scopolin, (E)-N-Phthaloyl-2,3-dehydrophenylalanine, 10H-Phenoxazine, alpha.-D-Glucopyranoside, 2-Dimethyl(pentafluorophenyl)adamantane, Glucose, 4-Piperidineacetic acid, Ethyl 2-[4-chlorophenyl]-7,8-benzocinchoninate, Ethyl 2-[4-chlorophenyl]-7,8-benzocinchoninate, 11.beta.-Hydroxyapoaromadendrone.

The most various and rich on a chemical composition were spectrum of compounds in extract from a crude Cistanche stolones (sample 1). In comparison with samples of crude stolone extracts treated with temperature and boiling water [2, 3], in extract of crude stolones without treatment reveal large number of components absent at heated-up samples: 2-methyl-1,3-butanediol, Cyclohexene, Lactic acid, N-methylaminopropionic acid, 1,2-Ethanediamine, 2-Benzylidenehydrazono-3-methyl-2,3-dihydrobenzothil, L-Alanine, 2,3,1-Benzodiazaborine, 1-ethanone, 3,8-Dioxa-2,9-disiladec-5-yne, Arabino-Hexos-2-ulose, Benzeneacetic acid, Phenylacetic acid, 1(2H)-Phenanthrenone, 3-Pyridinecarboxylic acid, 2-Desoxy-pentos-3-ulose, l-Threonine, Crotonic acid, Tetrahydrobenzene, 2H-Cyclopentacyclooctene, Methoxybismethane, L-threonine, fatty acid, Acrylic acid, 1H-Indole, 4-Nitrodibenzothiophene, Methanamine (gas), Benzacridine, 7-phenyl-2-azafluoren-9-one, 12,13-dihydroxytotara-8,11,13-trien-7-one, 3-Benzoaziridine, 4'-Methyl-3-(2-thienyl)acrylophenone, Butane, L-Proline, L-aspartic acid, Tricyclododeca-2,9-diene, Retinoic acid (vitamin A acid), 1,2,3,4-Tetrahydro-6-methoxy-2,4-diphenylquinoline, 2,3,4-Trihydroxybutyric acid, 1(2H)-Phthalazinone, 4-Hydroxyphenylethanol, Tyrosol, 3-Phenpropenoic acid, hydrocinnamic acid, Adipic acid,

L-phenylalanine, Xylonic acid, Lyxonic acid, 1,2,4,8-Tetramethylbicycloundeca-2,4-diene, 5,10-Dihydro-10-indenol, 6,7-Bis-2,3-naphthalenedicarboxyl, trans-Aconitic acid, l-Glutamine, Kaurane-16,18-diol, 1-Methyl-4-ethyl 2-phenylsuccinate, 7-Isopropenyl-1,4a-dimethyl-4,4a,5,6,7,8-hexahydroxil, Stigmast-22-en-3-ol, 1,2,4-Trisnaphthalene, 5,6-dimethyl-7H-phenol, 2,4-mentyl-3-phenylpyridine, 3.alpha.-Phenylcholest-5-en-3-beta.-ol, Glucosamine, 7alpha-2.alpha.-hydroxybenzaldehyde, 1-ethyl-2,9-dihydro-1H-carbzole, 2,4,6(1H,3H,5H)-Pyrimidinetrione, 1H-Indole-3-acetic acid, Arabino-hexaric acid, vaccenic acid, L-Tryptophan, Sedoheptulose, 1-iodo-2-methoxy-5-methylbenzene, Phosphine, 2,5-di-(E)-(2-carbomethoxystyryl)thiophene, 2H-Furol, Sorbitol, 2-(1-aza-4-oxa-cyclohexyl)-benzoxazole, 2-(2-Bromo-4-methylphenoxy)-N'-{[1-(4-nitrophenyl, 1,1,4a-Trimethyl-5,6-dimethylenedecahydronaphthalene, Uridine, D-Testosterone phenylpropionate, methyl dihydropimarate, 2-Deoxy-galactopyranose, Estra-1,3,5(10)-trien-17-ol, 3-methoxy-acetate, Azulene, 2H-Cyclopentacyclooctene, 3,9-diacetoxy-1,2-dimethoxy-7-formyl-dehydropropene, 4,4'-bis(3,4-dimethoxystyryl)-2,2'-bipyridine, glutamic acid, 5H-Dibenzocycloheptene, 5-methylene, 2-cinchoninic acid, Vitamin D3 dimethylphosphate, trans-Cembranolide, Adenosine, Pregn-4-ene-3,20-dione, 13-glucopyranosyloxydocosanoic acid, Trifluorosilyl-octamethylcyclotetrasilazane, Lactose, 2-O-glyceryl-á-D-glucopyranoside, Melibiose, 3-Bromo-5-ethoxy-4-hydroxybenzaldehyde, octamethylcyclotetrasilazane, 5-(p-Chlorophenyl)-3-(6-methyl-3-pyridyl)-1-(p-tolyl)-2-pyrazoline, beta-l-Galactopyranoside, 13-glucopyranosyloxydocosanoic acid, Cliogoinol, 2-bianthrone, 4-cyano-7-mesityl-5H-thieno[2,3-c]thiopyran, beta-D-galactopyranuroni, trans-3'-Dimethylamino-4-(methylthio)chalcone, Thymolphthalein, 6,7-dihydroxycoumarin-.beta.-d-glucopyranoside, 6,7-bis(trimethylsilyl)-4-methoxy-1-azabiphenylene, 5.beta.-Androst-7-en-6-one, 2,5-Bis(5-formyl-4-propyl-2-pyrrolyl)thiophene, Benzenesulfonic acid, 2-Pyridone, Bisacodyl-M, 2-Pyridinamine, 1,4,6-Trimethyl-1,2,3,4-tetrahydroquinoxaline-2,3-dione, Turanose, 2-phenyl-2-p-anisyl-3,3-bis(trifluoromethyl)oxira, Patchouli alcohol, Phenethylamine, 1,1,4a-Trimethyl-5,6-dimethylenedecahydronaphthalene, Flavone acetic acid, ethylene glycol, Guaicol-beta-d-glucopyranoside, threo-2,5-Hexodiulose, Methyl-19-norisoanticopalate (deuterate), 1,1,4a-Trimethyl-5,6-dimethyl decahydronaphthalene, Scopolin, (E)-N-Phthaloyl-2,3-dehydrophenylalanine, 10H-Phenoxazine, 2-Dimethyl(pentafluorophenyl)adamantane, Ethyl 2-[4-chlorophenyl]-7,8-benzocinchoninate, 11.beta.-Hydroxyapoaromadendrone.

Thus, drying and heat treatment lead to loss from chromatogram spectrum organic acids (Ethanedioic acid, Fumaric acid, 2-keto-gluconic acid, Mefenamic acid, Ribonic acid, Dodecanoic acid, Isosaccharinic acid, 3-Methylglutaconic acid, 2-Keto-l-gluconic acid, D-Gluconic acid), sugars (D-Fructose, Mannose, Maltose, Glucofuranose, beta.-D-Glucopyranose, D-Altro-2-Heptulose, Bete-D-glucopyranaside, Thymol-beta-d-glucopyranoside) and some other components. According to the received results and chemical composition analysis of each sample, it is recommended for production of herbal teas samples 4 and 5 and for receiving the most active extract sample 1.

Extracts from CD have a long tradition in China in the treatment of disorders in male and female reproductive organs, diseases of the urinary system, musculoskeletal system. Hence, there were made attempts to reveal substances or compounds responsible for biological activity of CD.

Investigations of Kobayashi H and others showed that Cistanche, as desert plant, has different organic compounds: alkaloids, iridoids, kankanoside, îrobanchin, 6-methyl indole, 3-methyl-3ethylhexane, 2,6-bis(1,1-dimethylethyl)-4-methyl phenol, heptadecane, 2-methyl-5-propyl nonane, nonadecane, eicosane, henicosane. Water-soluble faction includes N,N-dimethyl glycine methyl ester, betaine, sitosterol, daucosterol, triacontanol, acteoside, 8-epiloganic acid, stearic acid, 2-nonacosanone, bis-2-ethyl-hexyl-phthalate. Following compounds have pharmacological actions as stimulation sexual and anticancer actions: 2'-Acetylacteoside; Acteoside; Bicyclo [2,2,2]oct-5-en-2-ol; 2,6-Bis(1,1-dimethylethyl)-4-methyl phenol;Bis-2-ethyl-hexyl-phthalate Cistachlorin;Cistanin; Cistanoside B; Cistanoside C; Cistanoside D; Cistanoside E; Cistanoside G; Cistanoside H; Daucosterol; 4,6-Dimethyl dodecane; *N*,*N*-Dimethyl glycine methyl ester;3,6-Dimethyl-undecane; (2,5-Dioxo-4-imidazolidinyl)carbamic acid; Echinacoside;Eicosane; 8-Epiloganic acid; Geniposidic acid; Laxative; Heneicosane, Heneicosanic acid; Heptadecane; Leonuride; Liriodendrin; 2-Methyl-5-propyl nonane; 3-Methyl-3-ethylhexane; 6-Methyl indole; 2-Nonacosanone; *n*-Nonadecane ; Phenylalanine; â-Sitosterol; Stearic acid; Succinic acid; n-Triacontanol; Tubuloside B [4-10]. The main idea of our work was to use greater biological activity of Cistanche rather than ginseng. We expect to produce herbal teas and energy drinks based on Cistanche deserticola. For this purpose it is necessary to develop technology of raw materials processing from stolones harvested in Moinkun desert, storage and selection of the content of herbal teas and

energy drinks. In the beginning the study of 5 samples on the Agilent 6890N chromatograph with the mass selective detector Agilent MSD 5973 revealed only minimum number of components. This was due to the fact that water extracts contained large number of nonpolar, nonvolatile compounds. Derivatization improved quality of chromatographic separation. In sample 4 on chromatogram can be seen 108 components, in sample 5-92 components, in sample 3-138 components, in sample 2-154 components, in sample 1-478 components. With high correspondance we identified the following number of components: in sample 5-46, in sample 4-41, in sample 2-89, in sample 3-66, in sample 1-258 LMWOC. There are large amounts of carbohydrates and phenolic compounds among them.

CONCLUSION

In Moinkum desert annually around 200 tonnes of dried stolones of CD is produced. With previous method of drying in order to produce such amount 1600 tonnnes of raw material was required. To process such large amount within 10 days of vegetation of CD not laboratory, but large scale technology is required. We changed of raw material processing technology by placing at the begining the slowdown of stolones metabolism. We have done it by cutting stolones in pieces and treating it with hot air. This led to drying of the surface driness and further drying within few days. To maintain the quality of products in tight deadlines in processing we applied freezing of the stolones. This does not lead to quality depreciation. However, allows in the future production of juice and dry powder. We have first introduced a new product-Cistanche juice. According to our data the juice is not only rich for sugars, phenol compounds, but also proteins and polysaccharides. Later during drying, especially as a whole, breaks up.

This work developed new drying and heat treatment techniques for conservation of Cistanche stolones and technologies of extraction of water soluble LMWOC. Here, we have developed method of quality of raw material control by means of GCMC chromatography. Research of chemical composition of dry and crude Cistanche stolones, treated with high temperature and boiling water and water-soluble extract for receiving herbal teas and energy drinks from the Kazakhstani plant of Cistanche deserticola found that water-soluble extract is the most suitable for production of products with biologically active substances. Drying and heat treatment significantly reduces the quality of products, resulting in a depletion of LMWOC content.

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