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# Investigation into Chemical Composition of Powdered Lupine Seeds

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# Abstract

This work investigates into chemical composition of powdered seeds of non-conventional bean culture: lupine. It has been established experimentally that comminuted lupine seeds are loose fine powder of light brown color with attractive flat specific odor with flowery notes. Herbal flavor with typical traces of bean cultures. In chemical composition of lupine seeds proteins and fibers dominate, fat content is not higher than 10%. Total content of amino acids is 39.98 g/100 g of product. Fat acids of lipid complex of lupine seeds are characterized by dominating content (more than 50% of total) of oleic  $\omega$ -6 and linoleic  $\omega$ -3 acids. Lipids of lupine seeds together with triacylglycerol contain accompanying substances with dominating content of sterols, aliphatic alcohols, phospholipids and tocopherols. All these features make the lipid complex of lupine seeds a promising tool for adjustment of fatty acid composition of food products of functional and specialized purposes, BAA, as well as valuable stock for production of pharmaceutical substances and medications. This article describes chemical composition, calorific value as well as water-binding, fat-binding and emulsifying abilities of lupine seeds. Advantages and disadvantages of lupine in production of food products are determined. Food products with this protein enricher are reviewed. Possibility of lupine addition to confectionaries is considered instead of a classical component. However, there are some constraints in amount of added protein substance to food products since it contains alkaloids. According to international regulations the content of alkaloids in products can be 200 mg/kg (0.02%). Therefore, while developing food products with lupine it is required to perform experimental studies aimed at determination of optimum recipe.

Key words: lupine, enrichment, protein, food fibers

## INTRODUCTION

Plants are the richest source of proteins. High amount of crop, low expenses for processing as well as high biological value make it possible to replenish protein deficiency in human nutrition [1].

Food ration of an average Russian is short of 30% of protein. At present human demands in concentrated protein can be satisfied by plants. After removal of seed cover and certain processing according to patented techniques they acquire high nutrient value [2, 3].

Protein substances enrich organism with plastic properties required for building of structures subcellular inclusions (ribosomes, mitochondria, and so on) and maintain exchange between organism and environment. Proteins support numerous chemical transformations in organism, which provide its functionality as an integral whole. Proteins are continuously renewed in human organism irrespective of age. Exchange efficiency depends on composition of consumed food. According to recommendations of WHO and FAO, the optimum protein amount required for needs of adult human is 60-100 g per day [4-6].

Nowadays plant breeders and nutritional scientists pay high attention to bean cultures. In terms of chemical composition and nutrition value these cultures are close to animal protein: meat, fish and milk. In addition, they are inexpensive protein sources. One of them is lupine. Chemical composition and biological value of its seeds determine potentials of its use in food industry [1,7].

This plant can survive in severe environment, so it got its name from *Wolf* (from Latin *lupus*). There are about

200 kinds of this plants with different biological features. Four types of lupine are the most common in Russia: perennial, narrow leaved, yellow, and white. It is possible to highlight alkaloid-free (up to 0.025%), low alkaloid (0.025-0.1%) and alkaloid (more than 0.1%) varieties. All these types can be valuable raw material for production of new protein products in food industry. Experimental data obtained by Russian R&D companies demonstrated that the yield of white lupine exceeds those of other bean cultures, such as soy, pea, broad bean, by 1.3-2.4 times [7].

Presently food industry pays great attention to searching for new possibilities of Earth resources for food production. Plant products become more and more important in solution of protein deficiency, since their use is economically efficient and with lower labor consumption for their production [8]. In this regard studies of powdered lupine seeds in comparison with other protein enrichers is quite urgent.

Among plant raw material bean cultures are characterized by the highest protein value, including soy, pea, beans, lentil, lupine, chick pea, which contain high amounts of lysine and tryptophan, whereas wheat flour is short of these amino acids. Protein amount in seeds of most bean cultures often varies in the range from 25 to 30%. However, soy stands out sharply not only in terms of protein content but also of fat content. Thus, if in pea, bean, lentil and chickpea the protein content varies in the range of 20-24% and fat -1.5-4.5%, then in soy beans their contents are 35-40% and 17-20%, respectively. In terms of protein content lupines are close to soy. Lupine protein, as in soy, is well digested by human organism and

characterized by high biological activity. Lupine seeds contain up to 40% of protein, which includes all essential amino acids as well as sufficiently high content of lysine, threonine, and leucine. Table 1 summarizes biological value of plant raw materials, it can be seen that lupine is superior than all other materials except for soy.

Table 1. Diological value of plant raw material		
Description of	Biological value, %	
vegetable stock		
Lupine	60	
Lentil	48	
Chickpea	51	
Pea	43	

Table 1. Biological value of plant raw material

One of features of lupine protein complex is existence of inhibitors of proteolytic enzymes: protease, invertase and others. However, in comparison with soy, pea and other bean cultures all kinds of lupine are characterized by their minimum content. Soy seeds contain in average 29-32 g of inactivated trypsin per 1 kg, and lupine seeds – 2-2.5 g, which characterizes them as more valuable raw material for food products. Lipids are also main components of lupine seeds, their fraction is from 5 to 12% of dry seeds.

In all lupine types the major portion in oil composition is occupied by linolenoic, linolic and oleic acids. Comparing on the basis of their content the oils of lupine, soy, pea it is possible to conclude that in terms of the most valuable unsaturated fat acids and their total amount the lupine oil is superior than that of pea, and the yellow lupine oil is similar to soy oil.

Lupine oils are rich in fat soluble vitamins and provitamins: tocopherols, sterols, and carotinoids. Lupine seeds contain also water soluble vitamins: thiamin, riboflavin, pyridoxine, biotin, folic acid, ascorbic acid. In terms of B vitamins they are comparable with seeds of other bean cultures (pea, soy) and significantly exceed wheat, rye, and other crops. In particular, lupine seeds are different in terms of  $\beta$ -carotene (0.30-0.49 mg %) and tocopherol (3.9-16.2 mg %) against 0.014-0.018 mg % and 1.1-5.5 mg % of crops. Therefore, lupine should be considered as food crop possessing some advantages in comparison with other bean plants, soy in particular. Thus, it is possible to recommend lupine for food products in order to improve biological value in all fields of food industry [8].

The studies were carried out with narrow leaved lupine, characterized by the earliest maturity and absence of toxic substances, which is advantageous for its application in food industry [9].

## METHODS

Protein weight fraction was determined using a DKL8 instrument for N2/protein determination (VELP SCIENTIFICA, Italy). Biological value of powdered sainfoin seeds was analyzed by experimental determination of amino acid composition using a KAPEL'-105M capillary electrophoresis system (Lumex, Russia [10].

Fiber weight fraction was determined using a FIBRETHERM FT12 instrument (Gerhardt, Germany) in accordance with Russian Standard GOST 10846-91. Fat weight fraction was determined using a SOXTHERM SOX414a facility for solid-liquid extraction, (Gerhardt, Germany) [11].

Carbohydrate weight fraction, including mono- and disaccharides, was determined by chromatography using high-pressure liquid chromatograph in acetonitrile–water mixture (77:23). Qualitative determination of diterpene glycosides was performed using densitometer (Germany) by calculations on the basis of spot color intensity.

Weight fraction of macro- and microelements (potassium, sodium, calcium, magnesium, iron, manganese, chromium, zinc, and copper) in dry stevia leaves and fat-free spices was determined by atomic absorption spectroscopy using AAS-1 instrument (Zeiss, Germany).

The experimental results were processed by means of Statistica 6.0, Microsoft Office Excel 2007 and Mathcad software packages.

All studies were performed on equipment of Research center of food and chemical technologies, Kuban state technological university.

# RESULTS

In terms of protein yield from hectare lupine exceeds crop and bean cultures. In addition, it can be cultivated on vast territory — from south boundary of forest-steppe zone to Leningrad oblast, as well as in Primorski and Khabarovsk krai, in Sakhalin and Kamchatka. Protein content in lupine seeds is from 30 to 55%, which is by nearly 4 times higher than in cereal grains [12]. Content of fractions is summarized in Table 2.

**Table 2. Content of fractions** 

Description	Content, %
Water soluble	23–44
Salt soluble	47–56
Alcohol soluble	1.9–11
Alkali soluble	3–9

It can be seen that lupine contains the lowest amount of water soluble protein in comparison with other bean cultures.

Chemical composition and calorific value of lupine are summarized in Table 3.

 Table 3. Chemical composition and calorific value of

 Junine

Tupine			
Description	Value		
Moisture content, wt %	8.3		
Raw protein content, dry weight, %	38.5		
Fat content, dry weight, %	9.4		
Fiber content, dry weight, %	22.2		
Calorific value, kcal	286.0		

In terms of synthesis and accumulation of protein lupine ranks No. 2 after soy, hence, it can be considered as a promising source for production of functional food. An important advantage of lupine protein in comparison with soy protein is that it actually does not contain protease inhibitors and does not provoke allergic reactions.

The properties of powdered lupine seeds are significantly determined by the state of its protein phase. Protein weight fraction in powdered lupine seeds is sufficiently high and equals to about 34–43%. Taking into account high value of powdered lupine seeds' protein components for human feeding, mainly of their amino acid composition, the data on amino acid content in powdered lupine seeds are summarized in Table 4.

 Table 4. Amino acid composition of powdered lupine

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Amino acid	Content, %	
Arginine	8.22	
Lysine	2.19	
Tyrosine	1.77	
Phenylalanine	1.56	
Histidine	1.05	
Leucine + isoleucine	4.80	
Methionine	2.67	
Valine	1.51	
Proline	1.75	
Threonine	1.78	
Serine	2.65	
Alanine	1.81	
Glycine	1.87	
Glutamine acid and glutamine	3.70	
Asparagine acid and asparagine	1.53	
Cystine	0.52	
Tryptophane	0.60	
Total:	39.98	

Lupine proteins contain essential amino acids. Human organism cannot synthesize them but they are important for correct organism functioning.

Arginine prevents hypertension process, promotes reinforcement of immune system. Sufficient amounts of arginine in human diet reduces probability of cancer tumors. Isoleucine and leucine play an important role in generation of hemoglobin, stabilize sugar content in blood and participate in numerous processes of life support. Glutamine has positive effect on digestive system, increases immune system, possesses neurally mediated properties [5].

Analysis of composition of lipid complex of powdered lupine seeds is of particular interest, since it is important for human organism. Composition of fat acids of lupine fat phase is summarized in Table 5.

# Table 5. Composition of fat acids of lupine fat phase

	Content	of fat acid, % of total		
Fat acid	Average	Rounding to 1 decimal according to Russian standard GOST 31663–2012		
C 16:0 palmitinic	8.17	8.2		
C 16:1 palmitoleic	0.95	1.0		
C 18:0 stearic	1.77	1.8		
C 18:1 oleic	57.01	57.0		
C 18:2 linolic	15.81	15.8		
C 18:3 linoleic	7.18	7.2		
C 20:0 arachic	1.13	1.1		
C 20:1 gondoinic	3.34	3.3		
C 22:0 behenic	3.53	3.5		
C 22:1 docosanoic	1.13	1.1		

As can be seen in Table 4, lipids in lupine seeds contain mainly linoleic ( $\omega$ -3) and oleic ( $\omega$ -9) acids. Oleic acid prevents dangerous diseases, such as thrombosis and atherosclerosis, as well as cancer process. Linoleic acid neutralizes saturated fats, thus preventing accumulation of cholesterol in vessels, governs functions of nervous system, improves utilization of fat soluble vitamins [11].

Fat phase of lupine seeds contains tocopherols, which regulate blood coagulation, improve blood circulation. Content of tocopherols in lupine is summarized in Table 6.

Table 6.	Content	of toco	pherols	in fat	phase
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Description	Value
α-tocopherol	2.35
β-tocopherol	not detected
γ–tocopherol	71.55
δ-tocopherol	not detected

# DISCUSSION

While investigating [1] into fraction composition of lupine seeds, shells, and kernels, it has been established that in comparison with other bean cultures lupine seeds contain the highest amount of protein, hence, it is more reasonable to use them for production of proteinous substances. Flour of lupine seeds is characterized by high emulsifying stability and lower critical concentration of gel formation in comparison with nut flour. This is related with the fact that shells of lupine seeds contain fiber, which is a good emulsifier and stabilizer. On the basis of performed studies is can be stated that the highest desirability function for mincemeat is achieved at 20% of lupine flour. The desirability function for dough recipe is based in 40% of lupine flour with regard to total flour weight.

A food encapsulant is known, containing jellified proteinous substances on the basis of lupine proteins embedded into continuous polysaccharide phase, where the mentioned jellified proteinous substances include micronutrient and where the encapsulant size is from 1  $\mu$ m to 5 mm.

The use of lupine seeds in production of items with complex raw stuff composition makes it possible to increase biological value of protein component of conventional plant raw materials and to decrease to some extent the existing protein deficiency.

The Chair of milk and food biotechnology, St. Petersburg State University of Refrigeration and Food Engineering, developed recipe and technology of fermented milk product using lupine seeds. A portion of cow milk was substituted for lupine milk in amount of 10-30%. With increase in the dosage souring time slightly decreased. This can be attributed to positive effect on acid formation exerted by food fibers in lupine milk. It was experimentally established that upon replacement of 30% of cow milk souring time of the mixture was reduced by 30-45 min [13].

The researchers [14] obtained concentrated product with high biological value on the basis of lupine seeds. In order to achieve this, lupine seeds are disintegrated and sieved on 100 µm screen. Then hydrolysis is carried out with preliminary prepared water-flour slurry (1:7). Total hydrolysis time is 3 h. After completion of the process the mixture is centrifuged, centrifugate is decanted and protein in it is deposited in isoelectric point (pH 3.0-3.2) by 35% solution of acetic acid. After protein deposition the mixture is centrifuged again. The protein paste is washed with water three times for complete removal of acetic acid, which is added upon protein deposition with subsequent centrifugation and drying. The obtained concentrated product of lupine seeds is characterized by improved functional properties, it can be applied in various fields of food industry, including meat industry, aiming in product enrichment with proper protein.

Yurchenko et al. developed method of production of paste-like concentrated lupine [15], which could be applied as filler in milk, meat, baking, and confectionary industries. The method is comprised of preliminary selection of food lupine seeds in terms of quality, their mixing with water at 50-70°C by means of mechanic acoustic homogenizer with dispersing rotor in acoustic field with intensity of 100-500 W/kg. Then, the obtained mixture is pasteurized at 92-98°C in 3-5 min with subsequent cooling. This process sequence of concentrated product provides complete retention of valuable nutrient substances, essential amino acids, unsaturated fat acids, vitamins, mineral substances, food fibers. Protein fractions remain native and are well extracted from lupine seeds, which improves protein quality and increases its yield.

Prof. Zabodalova [16] obtained fermented milk and curd product with addition of dry proteinous substance. This researcher found that addition of proteinous concentrated product to fermented milk drink in amount of 2.0%, and to curd product in amount of 1.0% of weight of fermented mixture did not influence on souring and made it possible to obtain good curd. For fermented milk drink the curd is obtained without separation of whey, and for acid curd with well separated whey. Curd product is characterized by soft paste-like consistence. Addition of this proteinous filler in certain amount does not impair organoleptic performances of final product and enables enrichment of human diet with proteins, carbohydrates, vitamins, macro- and microelements.

The recipe of hard dough cookies is known [17], where powdered lupine seeds are used as bean semifinished product. The products on the basis of this powder are recommended for diabetics, since they promote decrease in sugar level in blood. In addition, lupine softly regulates blood pressure, influences positively on functioning of cardiovascular and nervous systems, prevents obesity, and decreases risk of cancer. Lupine seeds improve appetite and eliminate problems with digestion. Lupine can be used for infant food due to high content of nutritive substances beneficial for growing organism. Contrary to soy products, lupine does not provoke allergic reactions in organism. Lupine food fibers are characterized by enterosorbent properites: they reduce cholesterol content in blood and are required for prevention of certain diseases.

A method of production of toasts has been developed [18] involving use of lupine flour in amount of 15-17%, preliminary mixed with wheat flour. The considered flour is made of lupine seeds, treated in electrical field of super-high frequency in 130-140 s with subsequent cooling to ambient temperature, grinding, separation of seed shell in air flow at the rate of 7-8 m/s, comminuting and sieving. Herewith, the organoleptic properties of the toasts are improved, including color, taste and flavor, biological value increases, and dough fermentation time decreases.

A method of production of Mechta buns is known involving blended lupine hydrolyzate [19]. The hyrolyzate is prepared from mixture consisting of lupine flour and hen egg blend in the ratio of 1:3. Hydrolysis is performed in the presence of Neutrase ferment substance. This invention improves quality of buns due to increase in their biological value, optimum adjustment of chemical composition in terms proteins, carbohydrates, and mineral substances as well as adding of functional properties.

Kazanskaya developed high protein filler for bakery industry, which included natural plant component of bean species, namely: powdered lupine seeds.

Pruidze patented high protein filler, which included natural plant component: lupine seeds and food flavor made of spice mixture of powdered seeds of fennel and coriander; Golovchenko developed composition including pectin substances and plant protein from seeds of alkaloid-free white lupine comminuted to 0.25-5 mm, and plant component in the form of fruit pulp at the following ratio: comminuted lupine seeds – 10-40%, fruit pulp – 50-80%, food flavors –1-10%.

Non-milk cheese analog is known, where one and several certain and cleaned plant proteins are selected from reserve proteins, pea proteins, lupine proteins, bean proteins, chick pea proteins, and lentil proteins.

Makaron-Service company limited produces paste goods using powdered lupine as additional raw material [20] intended both for preventive measures and for satisfaction of human nutrient requirements, as well as to provide possibility of paste production on the basis of local agricultural products in the areas where wheat or rye cannot be cultivated.

## CONCLUSION

It has been established on the basis of organoleptic evaluation that comminuted lupine seeds are loose fine powder of light brown color with attractive flat specific odor with flowery notes. Flavor is herbal with typical traces of bean cultures.

The performed experiments demonstrated that powdered lupine seeds contained valuable food nutrients in protein and lipid complexes, which made it a promising ingredient for food technologies, including confectionary industry.

Therefore, the products on the basis of lupine seeds can be used as raw stuff for quality improvement of food products and as additives for development of recipes of new food products.

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