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Review

THE NUTRITIVE VALUE OF POULTRY DIETS CONTAINING SUNFLOWER MEAL SUPPLEMENTED BY ENZYMES

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The international limitations imposed on the utilization of meat and bone meals in animal diets, together with the increasing demand for soybean meal, create a necessity to search for other protein sources to economically balance compound feeds. In this regard it is important to note that sunflower is the best adapted high-protein crop available in some European regions and that is useful to use it in poultry farming as the replacement of other protein sources. Protein and many other nutrients are "imprisoned" to variable degrees, inside sunflower meal fibrous structures, and remain less available for digestion by the poultry's own proteases and other endogenous enzymes. Added exogenous enzymes (phytase, hemicellulase, cellulase, carbohydrase, protease, etc.) offer a number of creative possibilities for breakdown and "liberation" of these nutrients, their easier digestion and absorption, and thus development of new nutritional standards and new diets formulation. Supplementation of poultry diets containing sunflower meal by different enzymes increasingly contribute to sustainable poultry farming by enhancing production efficiency, increasing the effectiveness of nutrient utilization and upgrading in environmental protection.

KEY WORDS: sunflower meal, enzymes, poultry nutrition

INTRODUCTION

Sunflower meal (SFM), the important by-product obtained after the extraction of oil from sunflower seeds, is used as a protein source in animal nutrition. The amount and chemical composition of SFM depends on the variety of the seed, the processing method, efficiency of oil extraction and the degree of dehulling or decortication. In our country, the crude protein (CP) content of conventional SFM usually varies between 33% and 37%. The corresponding crude fiber (CF) contents are in the range between 18% and 23%. Thus, an inverse relation is seen between the CP and CF contents of SFM. These meals are mixtures of protein containing kernel and hulls in the approximate ratio of about 60:40% (1, 2). In other regions, CP content of SFM is very often lower than 28%, and CF content is higher than 30%. Due to high hull levels, these meals are mainly used

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for feeding cattle and sheep. But, using SFM in this way is wasting of valuable proteins. Therefore, some of the characteristics of SFM must be considered so that the maximum benefit may be gained from this feed ingredient (3-8).

An apparent disadvantage of SFM is that it contains a relatively high level of fiber compared to soybean meal (SBM). This characteristic of SFM may lead to bulky diets which may be a problem for young chicks, in particular, because their digestive system has a limited capacity. If SFM is incorporated at high inclusion rates, the nutrient and energy densities of the resulting diet may be significantly diluted and growth stagnant. The density of the diet is of prime concern in terms of nutrient intake and resultant growth rate (9, 10). Inclusion of high fiber ingredients is also limited because of the poor metabolizable energy contents. True metabolizable energy contents of sunflower meal is negativly correlated with CF and hull content (11). Separation of the hull, as the main source of fiber, from the kernel is the processing solution for improving the nutritional and commercial values of SP. The CF content should be decreased to a minimum by dehulling during processing of the sunflower seed for oil extraction or after oil extraction by different fractionation procedures of SFM based on diametrically opposed physical characteristics of the kernel and the hull. Several efficient fractionation procedures and complex systems for separating hulls from kernels and meals, rendering high yields of attractive protein fractions that contain 42-48 % of CP and 8-14% of CF have been developed at the Institute of Feed Technology – now renamed Research Centre Feed-to-Food at the University of Novi Sad, Serbia (2, 12, 13, 14). Technological solutions for the production of SFM with 44% of CP, have been introduced and implemented in several edible oil plants, based on some of these results. In the industrial conditions, from the initial SFM containing 37-38% of CP, 35-40% of sunflower meal with 44% of CP (protein fraction-through) and 65-60% sunflower meal with 33% CP (cellulosic fraction - overs) may be obtained (1, 2, 13). The separation of sunflower hulls from kernels with centrifugal separator is rather heavy due to the presence of hard conglomerates made of kernels and hulls adhered to them. The remarkable amount of these conglomerates flow over the sieve holes and directly reduced PF yield (13). The preliminary treating may be applied in order to crush the existing agglomerates and enable the subsequent separation of the kernels from the hulls adhered. It is very important that the hulls are not crushed into too small particles, so that it could be separated later by mechanical fractionation. Continuous investigations at our research Centre Feed to Food are focused on identifying solutions to increase yield and enhance the quality of decellulosed high protein SFM and we have made substantil progress in improving the technological process (14). However, hull removal has not been totally successful, probably because of the tight binding of the hull to the kernel, and it is useful to explore other options to upgrade SFM, so that the maximum benefit may be gained from this feed ingredient in monogastric animals diets (2, 9).

Based on the premise that SFM may contribute a significant portion of poultry diets and that it contain high levels of non-starch polysacharides (NSP) and phytates it would be useful to investigate the effect of enzyme supplementation on diets containing these ingredients. Dietary supplementation with enzyme preparations is not a new concept, but it becomes more fine-tuned with the production of specific enzyme preparations. The addition of exogenous enzymes (phytase, hemicellulase, cellulase, pectinase, carbohydra-

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se, protease, lipase, β-glucanase, etc.) offers a number of creative possibilities for breakdown and "liberation" of these nutrients, their easier digestion and absorption, and thus development of new nutritional standards and new diets formulation (15). The objective of the present study is to review some of research results and posible solutions obtained by using of enzymes as additives for upgrading the nutritive value of poultry diets containing sunflower meal.

Sunflower meal as the substrate for enzyme's actions

Enzymes are powerful but strictly specific catalysts that act on one or, at most, a limited group of compounds known as substrates. Careful focus should be directed to the physicochemical characteristics of target feed ingredients before applying enzymes to feed. Physicoohemical properties like major ingredients, target substrate and its amount, and physical structure have influenced the eficacy and onset of the enzymatic hydrolysis. By far, the varied enzyme efficacy due to ingredients cannot be simply explained. Amounts and characteristics of NSP, anti-nutritional factors, oligosaccharides and/or other components, physical structure of mainly starch and protein and the degree of feed processing have been compounded to represent the efficacy of supplemental enzymes (16). Thus, detailed information about chemical composition and nutritional properties of SFM is a prerequisite for successful use of enzymes as additives for upgrading the nutritive value of poultry diets containing this feedstuff. To describe the quality of SFM, chemical composition of two conventional and two high-protein SFM with 42 and 44% CP in comparation to SBM, as the »standard« that feed and animal producers want to achieve with other vegetable protein sources, is shown in Table 1.

Table 1. Chemical composition of soybean and sunflower meals*

Nutrient	Soybean meal	Sunflower meals			
Dry matter [%]	90	90	90	90	90
Crude protein [%]	44	34	37	42	44
Crude fibre [%]	5	23	18	16	12
Crude fat [%]	1.5	1.5	1.6	1.7	1.7
ME for poultry [MJ kg^{-1}]	9.25	5.44	6.70	7.20	7.95
Amino acids contents [%]					
Lysine	2.74	1.18	1.28	1.40	1.70
Methionine	0.60	0.72	0.79	0.92	1.10
Cystine	0.63	0.55	0.60	0.68	0.85
Threonine	1.72	1.21	1.32	1.44	1.70
Tryptophane	0.59	0.45	0.47	0.48	0.61
Arginine	3.28	2.68	2.97	3.27	4.10
Glycine	1.86	1.92	2.04	2.12	2.46
Serine	2.25	1.40	1.61	1.71	1.86
Histidine	1.17	0.82	0.89	0.96	1.10
Isoleucine	2.13	1.47	1.60	1.64	2.15
Leucine	3.40	2.12	2.33	2.64	2.90
Phenylalanine	2.22	1.50	1.64	1.95	2.10
Tyrosine	1.62	0.81	0.88	1.01	1.10
Valine	2.19	1.78	1.93	1.96	2.20

*Compiled from Tables (17, 18) and our own research (1); ME = Metabolizable energy;

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If properly processed, SFM with 44% of protein can be fully compared to SBM by its crude protein content (440 vs.440 g/kg). It contains considerably smaller amounts of lysine (17 vs. 27 g/kg), but significantly higher amounts of methionine (11 vs. 6) compared to SBM. The strong lysine deficiency in SFM has to be balanced by adding the lacking amount of lysine to obtain the utilisation of rich amino acid potential contained in sunflower protein (1, 13).

A large number of investigations have found a direct dependence between the energy value of SFM and the contents of dietary fiber (11, 19). As a result of these investigations, various formulas have been set, on which the base energy value of SFM can be predicted trough nutrient content of individual fractions of fibers. Janssen and Care (20) also pointed that the content of dietary fiber might be a good assumption of feed's nutritive value due to the impact of sunflower shell and core content of cell walls, and the strong negative correlation between crude fiber content and digestibility of CP and fat. The values of the metabolic energy of SFM were ranked from 4.94 to 9.39 MJ kg^{-10}, where the lower values are associated with higher levels of hemicellulose, and crude fiber (9). The low level of metabolic energy for poultry can be overcome by adding fat to the diets with SFM (9).

Unlike most other oilseed meals, SFM is not known to have harmful anti-nutritional factors. Namely, SFM contains the polyphenolic compounds, chlorogenic and a cafeic acid, but the concentrations of these antinutritional factors is not toxic in poultry diets (21, 22). However, phytic acid is considered as anti-nutritional factor in poultry because it binds phosphorous and other important nutrients and decreases their availability. Phosphorous is an essential nutrient in poultry diets, and its efficient use is essential for economic poultry production. Unavailable phosphorous is simply excreted and the result is a serious phosphorous pollution problem. In order to become available to broiler chicks, phosphorous from vegetable sources must be hydrolyzed with phytase as a catalyst, to inositols and inorganic phosphates which are readily absorbed in the digestive tract (23). By releasing phosphorous from phytate molecule, phosphorous supplementation in diets may be considerably reduced or even cancelled, thus leading to the reduction of phosphorus excretion, with beneficial effects on the environmental issues (24). Furthermore, phytate has also the potential to form indigestible complexes with cations (Mg, Ca, Fe) and bind with protein (25).

SFMs are used in animal nutrition mainly as protein sources, but they also contain significant amount of dietary fibre (DF), which is defined as the sum of lignin and polysaccharides that are not digested by endogeneous secretion of the digestive tract of non-ruminant animal species. In this nutrition context, the term DF includes any polysacharide reaching the hindgut and so includes resistant starch and NSP (26). Polysaccharides are macromolecular polymers of simple sugars or monosacharides linked together by glycosidic bonds. NSPs have glycosidic bonds other than the bonds of starch which in some case cause their resistance to starch degrading enzymes (27). The NSPs found in feedstuffs are primarily components of plant cell walls and they represent a group of heterogeneous compounds differing considerably in chemical composition and physical properties (28).

The types and levels of carbohydrates in SFM depend to a great extent on the technology of seed processing and the degree of dehulling or decortication and, hence, the data available in the literature differ greatly. According to the literature data, SFM with 33% crude protein contains 1-4% starch, 26-41% DF including 9-11% arabinoxylans, 18-23% cellulose, 9-10% lignins, 2-5% pectins (2, 28, 29). In order to develop techniques to counteract the antinutritive effects of soluble NSP, and understanding their chemistry, physical properties and behavior on ingestion by monogastric is crucial. Further work is required to characterise the type, levels and nutritive activity of the NSP and other DF component found SFM.

It is convenient to classify five major classes of fibers, according to their chemical structure and to their properties: four classes of water-insoluble polymers (lignins, cellulose, hemicelluloses, pectic substances) and one class of various water-soluble non-starch polysacharides and oligosaccharides (water soluble pectins, β-glucans, arabinoxilans) (27). Solubility of fiber components is linked to their effects in the digestive tract of animals. NSPs are generally defined as water-soluble or insoluble. Plants generally contain a mixture of both soluble and insoluble NSPs in a ratio that varies according to the type and stage of maturity. The types and levels of carbohydrate polymers and monomers in sunflower seed are shown in Table 2. (28).

Table 2. Carbohydrates in sunflower seed (% dry matter) (28)

Carbohydrate	Soluble	Insoluble	Total
Starch			1.4
Total NSP	4.5	23.1	27.6
Cellulose		8.7	8.7
Rhamnose	0.2	0.3	0.5
Fucose	0.1	0.1	0.2
Arabinose	0.6	3.0	3.6
Xylose		5.3	5.3
Mannose	0.1	1.1	1.2
Galaktose	0.3	0.9	1.2
Glucose		0.4	0.4
Uronic acids	3.2	3.4	6.6

The NSP content of feedstuffs influences various aspects of animal performance. The high NSP content in SFM limit their energy value and even more their protein value. Their nutritional effects in monogastric animals are diverse and, in some cases, extreme. It is, however, generally conceded that the major detrimental effects of NSP are associated with the viscous nature of these polysaccharides, their physiological and morphological effects on the digestive tract, and the interaction with the microflora of the gut.

Soluble fibers increase intestinal transit time, delay gastric emptying, delay glucose absorption, increase pancreatic secretion, and slow down absorption, whereas insoluble fibers decrease transit time, enhance water holding capacity and assist faecal bulking in non-ruminant animals (3, 28). These include the effects on voluntary feed intake, supply of available energy to the animal, including the digestibility and utilization of nutrients other than NSPs and gut and animal health. These effects can be attributed to the effects of NSPs on gut microorganisms, viscosity and water-holding capacity of the digesta (27).

Some investigations have suggested that the negative effect of NSP can be overcome by dietary modifications, including supplementation of diets with suitable exogenous enzyme preparations (30).

Enzymes and sunflower meal in poultry nutrition

Regardless of the goal of animal production, enzymes will aid performance through removal of antinutritive factors (ANF), breaking open cell walls, releasing phytate phosphorus, improving protein digestibility and provision of substrates for beneficial microflora. Exogenous enzymes supplemented to feed are theoretically capable for improving digestibility of feed by hydrolyzing the substrates that hinder digestion and specific antinutritional factors. However, in reality the practices do not exert the theoretically expected benefits.

Type and age of animal, quality of feed ingredients, including presence of target substrate and optimization of multi-enzyme combination are factors that are to be taken into account upon using feed enzymes. Therefore, it needs a very careful approach to decide the type of enzymes and their additional levels. (31).

Enzymes make it possible to upgrade the nutritional value of a feedstuff and can be added to feeds as "multi-enzyme" products that contain a variety of different activities or "specific-enzyme" products which are responsible for single-type of enzymatic activity based on a particular dietary substrate, such as NSP (15). Not only the total fiber content, but also the physical and chemical structure of fibrous polysaccharides and their anatomical arrangement within each specific ingredient, affect the accessibility of enzymes for digestion of nutrients (27). Undoubtedly, a total depolymerisation of the NSP require extremely complex enzyme activities. There are various types of fiber-degrading or NSP-breaking enzymes.

The efficacy of feed enzymes depends on their substrate specificity, activity and stability. Therefore, it is often very difficult to select potentially useful enzymes available in the market. Selection of effective enzyme products for use in sunflower-containing diets requires detailed knowledge of the substrates and their breakdown patterns in the gastrointestinal tract of the target species (31). Many commercially available enzyme formulations differ in their composition with respect to the number of individual enzymes and their activities. Selected microbial enzymes must degrade NSPs to an extent that can lower the viscosity in the intestine and improve feed utilization. Extensive research has revealed that enzyme usage increases the efficiency of utilization of the feed. It is now well documented that enzymes supplementation breaks NSP polymeric chains into smalller pieces, reduces the gut viscosity, and hence improves the nutritive value of fibrous feedstuffs.

Poultry do not produce enzymes for the hydrolysis of NSP present in the cell wall of SFM and they remain unhydrolized. This results in low feed efficiency. Most of the research work on NSP enzyme application in poultry feeds has focused on cereal grains, such as wheat, barley, rye and triticale, or, alternatively, on diets based on corn and SBM, with very little targeted to alternative protein meals like sunflower meal. Inconsistent results have been reported by several authors regarding the use SFM with enzyme supplementation in poultry diets (16).

It has been recognized that the disruption of the cell wall matrix of SFM by exogenous microbial enzymes can lead to easy access of the endogenous proteolytic enzymes to digest the entrapped proteins (32). Results obtained by Raza et al. (33) showed significant differences ($p<0.05$) among different experimental diets (varying in the level of SFM and CF) for weight gain and feed conversion. The highest weight gain was observed in chicks fed on the diet containing 10% SFM (6% CF in diet) with enzyme Grindazym GP 5000, (produced by DuPont^{TM} Danisco® Company and containing xylanase, P-glucanase and pectinase), while lowest gain was observed in chicks fed on the same diet without enzyme supplementation. Francesch et al. (34) reported the results of dietary supplementation of an enzyme preparation (Grindazym GP 5000) containing xylanase, P-glucanase and pectinase activity included for four months in a barley : SFM-based (60 : 20%) layer diet. There was no significant effect of enzyme supplementation on the rate of lay, daily food intake or body weight gain. However, a significantly positive effect was observed during the first four weeks on egg weight and egg size, and there was also a reduction in the percentage of dirty eggs. High doses of enzyme also improved excreta quality by reducing its water content. Sorensen (35) has reported that supplementation of SFM-based diets with the same enzyme increased the nutrient utilisation of this product, both in layers and broilers. Improvement in the performance of broilers with the addition of multienzyme in high sunflower diets were also noticed by Meussen, (36), Raj et al. (37), and Kocher et al., (32). Mushtaq et al. (38) reported that enzyme supplementation had a pronounced effect in low nutrient concentration and high SFM diets for broilers

According to the results of Tavernari (39) the enzyme complex consisting of cellulase, β-glucanase, xylanase and phytase had a significant effect on the weight gain only during the starter phase with the diets contained 20% SFM, which is possibly explained by the immature digestive system of broilers at this age. Oliveira et al. (40) evaluated two sunflower meal inclusion levels (0 and 15%) with or without an enzyme complex (cellulase, protease and amylase) in the diet of 21 to 42-day-old broilers, and did not find any significant interaction between SFM and the enzyme complex. These authors concluded that the dietary inclusion of 15% SFM improves live performance, but does not affect carcass yield. El Sherif (41) found that SFM concentrated with energy and supplemented with lysine and methionine was effectively utilized in grower and finisher broiler diets in place of SBM without adverse effect on the production, and there were no beneficial effects of enzyme supplementation with this SFM. The results of Kocher et al. (32) clearly indicate that comercial enzyme products have some effects in diets containing high concentrations of SFM. However, these effects could not be seen after detailed analyses of feed and digesta, and did not result in a significant improvement in the growth performance of broilers. Meng and Slominski (42) and Tabook et al. (43) have indicated that the addition of commercially available multiactivity enzyme products did not result in an improved broiler performance in diets, especially having increased concentration of SFM. These controversial results reported regarding the nutritive value of SFM were attributed to the differences in the variety, method of processing, age of birds and feed formulation techniques employed in these studies (9).

It is clear from the literature that phytate levels in poultry feed are variable, and have a negative effect on the overall efficiency of nutrient utilization by decreasing mineral and protein solubility and digestibility, and increasing endogenous secretions (23). First research works regarding the usage of phytase in animal feed were conducted some thirty years ago, but without wider application in field conditions. As a result of the growing environmental concerns and more stringent environmental regulations, however, research related to the production and application of phytase in animal feed has been intensified in the recent years. The use of phytase has become standard practice to reduce phosphorus levels in the environment and to compensate for the drastic increase of the cost of inorganic phosphates.

In order to be made available to broiler chicks, phosphorous from vegetable sources must be hydrolyzed with phytase as a catalyst, to inositol and inorganic phosphates, which are readily absorbed in the digestive tract (44). Results of numerous experiments have shown that degradation of phytate by phytase has a twofold positive effect – release of phosphorous and release of minerals, proteins and digestive enzymes. By releasing phosphorous from phytate molecule, phosphorous supplementation in diets may be considerably reduced or even cancelled, thus leading to the reduction of phosphorus excretion (45), with beneficial effects on the environment. Phytase increases the digestibility of phytate from around 25% to 50-70% in poultry, and its use has been on the increase since banning the use of animal protein sources, such as meat and bone meal, in the EU. It is also understood that phytase can improve the digestibility of other nutrients as well as energy (46, 47).

All obtained results about the use of phytase in poultry feeding indicate that with the addition of various phytase preparations in differently formulated diets, this enzyme improves availability of phytic phosphorous and other nutrients (44, 48, 49, 50) and reduces phosphorous excretion (45,51).

Supplementation of poultry diets with exogenous enzymes as additives for upgrading the nutritive value of poultry diets containing SFM has been investigated in our Institute. The higher body weight by 8.36% and better feed conversion ratio by 2.84 % was observed in chicks fed on a diet containing 15% SFM (with 44% CP) supplemented with enzymes (protease, hemicellulase, pectinase, β-glucanase) with respect to the trial diets without enzyme supplementation (29). With the diet containing 10% SFM with 33% CF supplemented by enzymes (cellulase, protease, lipase, α-amylase, β-glucanase) higher body weight by 10.23% and better feed conversion ratio by 8.15 % were obtained in comparasion with the diets without enzymes (52).

In one of our latest investigation (53), experimental diets were formulated on the basis of corn, high-quality decellulosed SFM containing 44% of protein (20% in diets) and SBM. Five broiler diets were tested. The first was a commercial diet based on corn and SBM and in the others 20% of SBM was replaced with SFM containing 44% of crude protein. The treatments were as follows: A – SBM without supplement; B – SFM without supplement; C – SFM supplemented with l-lysine HCl; D – SFM supplemented with l-lysine HCl and enzyme complex containing protease, hemicellulase, pectinase and β-glukanase; E – SFM supplemented with l-lysine HCl and phytase and 30% lower phosphorous content. Both enzymatic supplements positively influenced live weight and feed conversion ratio (FCR) in treatments D and E. There were no significant differences between these two treatments, but significant differences ($P<0.01$) were found for live weight and FCR between treatments B, D and E. Significantly positive ($P<0.01$) effect of added enzymes was found in treatments D and E in comparison with treatment A. It was also found that the treatment E with reduced phosphorous level and added l-lysine HCl and phytase decreased the level of phosphorous in broiler feces up to 20%. The decrease of available phosphorous level in diet E did not have any adverse effect on broiler feeding performances. From this experiment, it can be concluded that the supplementation of poultry diets with 20% SFM containing 44% of crude protein by l-lysine HCl and enzyme complex (protease, hemicellulase, pectinase and β-glukanase) or phytase increasingly contribute to the sustainable poultry farming by enhancing the production efficiency, increasing the effectiveness of nutrient utilization and upgrading environmental protection.

CONCLUSION

In spite of some conflicting results, in most studies SFM has been found to be a promising source of protein for poultry. Most of our research work suggested that the negative effect of SFM can be overcome by processing technologies and dietary modifications including supplementation of diets with suitable exogenous enzyme preparations. In this context, research efforts have been directed to identify novel, alternative and economically viable SFM + enzyme combinations for a succesful replacement of other protein sources in poultry diets.

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Review

ХРАНЉИВА ВРЕДНОСТ ОБРОКА ЗА ЖИВИНУ КОЈИ САДРЖЕ СУНЦОКРЕТОВУ САЧМУ ДОПУЊЕНУ ЕНЗИМИМА

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Међународна ограничења наметнута за коришћењу месно-коштаног брашна у оброцима животиња, заједно са повећањем потражње за сојином сачмом, стварају потребу за проналажењем других извора протеина да би се смеше за животиње избалансирале на економичнан начин. У том смислу важно је напоменути да је сунцокрет најбоље прилагођен високо-протеински усев доступан у појединим европским регионима и да га је корисно употребљавати у живинарству као замену за друге изворе протеина. Протеини и многе друге хранљиве материје су "затворени" до променљивог степена, унутар влакнасте структуре сунцокретове сачме што смањује њихову доступност за варење сопственим протеазама и додатим ендогеним ензимима. Додати ендогени ензими *(*фитазе, хемицелулазе, целулазе, карбохидразе, протеазе...) нуде низ креативних могућности за разлагање и "ослобађање" тих хранљивих материја, њихово лакше варење и апсорпцију, а тиме и за развој нових прехрамбених стандарда и нове формулације оброка. Допуњавање оброка живине који садрже сунцокретову сачму различитим ензимима све више доприноси одрживом узгоју живине, унапређењем ефикасности производње, повећањем ефикасности коришћења хранљивих материја и унапређењем у заштити животне средине.

Кључне речи: сунцокретова сачма, ензими, исхрана живине

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