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AN ENZYME–ALKALINE HYDROLYSIS OF FEATHER KERATIN FOR OBTAINING A PROTEIN CONCENTRATE FOR FODDER

Pencho Dalev

University of Sofia, Department of Biotechnology, Laboratory for New Protein Sources, 8 Dragan Tsankov, Sofia 1421, Bulgaria

Feathers are characterized by a keratin with a typically high content of small amino acid residues (glycyl, alanyl and seryl as well as cysteinyl and valyl ones). The complete solubilization of keratin structure is important for utilizing the great quantities of waste feather material as a source for protein fodder. Keratin, which amounts to ca. 90 % of the feather material is solubilized comparatively easily by reducing agents such as mercapto acetate, alkylation with iodoacetic acid (Harrap and Woods, 1964), copper sulphate, ammonia and sodium sulphite (Swan, 1961), sodium sulphite and sodium tetrathionate in the presence of 8 M urea (Bailey and Cole, 1961) and others. These approaches are however unsuitable for large-scale application. In that case the methodologies are based on enzyme hydrolysis and various pretreatment procedures such as ultrasound, ammonia etc. (Krilova and Popov, 1983; Weeks and Wildi, 1970; Gorjaev and Bikova, 1979). The main requirement is the employment of accessible and nontoxic reagents to obtain a product with high animal nutritional values. The known procedures employ costly equipment (ultra sound), high temperatures (140^{o}C) and lengtly processes (8 to 12 hrs). We here report results from studies of a biotechnological process for the hydrolysis of feathers which requires minimal time, low temperatures and accessible reagents and equipment.

MATERIALS AND METHODS

Reagents. Sodium hydroxide and hydrochloric acid, technical grade, containing heavy metals and toxic admitures within the limits of the requirements for fodder products, were used. The enzyme was alkaline protease B 72 from B. subtilis with an activity of 50000 U/mg produced at the Plant for Enzime Preparations. Botevgrad (Bulgaria).

Apparatus. The hydrolysis of the feather keratin was conducted in a 1 m^{3} reactor provided with a heating jacket, a disintegrator stirrer. The hydrolysis prodict was dried employing a „fluidized" drier.

Raw materials. Feathers obtained as a regular waste material from the poultry slaugter houses were used.

Procedure. The reactor was charged with 0,2 N aqueous sodium hydroxide. After heating to 90^{o}C this solution was added the feather material in portions and under stirring continuously. The temperature goes down to 70 - 80^{o}C and the alkaline pretreatment was carried out for 30 min. while activating periodically the desintegrating stirrer. At the end of the pretreatment stage the reaction mixture is a thick homogeneous mass. The pH of the mixture is then brought to 8,0 - 8,3 with 1:1 HCl and 0,5 g of enzyme per 100 g of feather material is added. The remperature is maintained at 55^{o}C and the mixture at the and of the process is a solution which after correcting to pH 7 is taken to dryness. The end prodict is a lyghtgray powder.

Analytical procedures. The content of protein, fat, ash, fiber, calcium and phosphorus were determined as described in the reference books on the analysis of foodstuffs (Lebedev and Usovich, 1969). The amino acid content was obtained by using an automatic amino analyzer type AAA - BIOTECHNA - 881 after hydrolysis with 6H HCl conducted for 24 hrs. at 110^{o}C. The gel filtration was carried out on a 20 cm x 80 cm Sephadex G-100 column with a phosphate buffer at pH 8,1.

RESULTS AND DISCUSSION

The combined action of three factors ensures the complete dissolution of keratin, namely alkaline pretreatment. mechanical disintegration in liquid medium and enzyme action. Separately, or when combined by two of them i.e. alkaline pretreatment enzyme desintegration they lead to a result that is unsatisfactory (Table 1).

Table 1. Treatment of feathers with enzyme 0,2 N NaON and with a mechanical desintegrator separately or in combination.

Mode of	Amount of solible protein as % of introduced substrate			
treatment	30 min	60 min	120 min	240 min
Enzyme-pH 8,55°C	4.8	12,5	17,6	21,4
NaON-0,2 N, 75°C	2,5	6,4	9,6	11,2
Disintegration	0,8	1,1	1,3	1,4
NaON-enzyme-desintegration	-	82,8	92,4	94,0

The extent of the hydrolysis of the keratin structure due to the action of the three factors can be seen from Molecular sieve chromatography that shows that from the combined process the major end product is divided between 50000 and 10000 D molecular mass. Its qualitative composition allows it to be described as a protein concentrate suitable as a fodder additive in combination with other protein concentrates. This is confirmed by some favourable results from preliminary experiments (78 % apparent digestibility) conducted with broiler chickens. The content of feather protein concentrate obtained by alkaline–enzyme–disintegration treatement, as percentage of dry mater is: Protein - 85,4; Fats - 1,22; Ash - 8,60; Fibers - 0,68; Calcium - 0,55; Phosphorus - 0,16. The amino acid content of feather protein concentrate as percentage per 100 g of protein (cist/e/in and tryptophan are omitted) is: Lysine - 2,17; Histidine - 0,58; Arginine - 6,98; Asparaginic acid - 7,79; Threonine - 3,49; Serine - 10,24; Glutaminic acid - 12,74; Alanine - 7,63; Valine - 8,02: Methionine - 1,20; Isoleucine - 4,90; Leucine - 8,11; Tyrosine - 2,85; Phenilalanine - 5,01; Proline - 8,90.

The feather protein concentrate significantly exceeds cereal fodder in Lysine and Methionine content but is inferior to fish flour and soya bean groats.

CONCLUSION

The considered procedure for obtaining protein concentrate from feather can be applied for the utilization of waste feathers from the poultry industry.

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