

Isolation, characterization and optimization of keratinolytic bacteria from chicken feather waste dumping site

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ABSTRACT

Chicken feathers are considered to be the waste products of poultry industry and can create a solid waste problem in the environment. Keratinases have the potential to degrade insoluble keratin predominantly present in feathers, hair, horns, and hooves. The aim of present study is to isolate, screen, and identify keratinolytic bacteria from feather waste dumping site and to optimize the culture conditions for maximum keratinase production and subsequent degradation of feathers. Fourteen bacteria were isolated from the feather waste dumping site at Virudhunagar, Tamil Nadu, India, and were screened for their keratinolytic property. Comparatively, three bacteria displayed better keratinolytic activity. Based on morphological and biochemical characteristics and 16S rRNA gene sequence analysis, they were identified as *Bacillus licheniformis*, *Glycomicrobacter arilantensis*, and *Serratia marcescens*, respectively. The effect of temperature, pH, feather concentration, and various substrates on the growth parameters of these bacteria was studied. The growth and protein production was high at 40°C for all bacteria. *B. licheniformis* and *S. marcescens* produced more protein at pH 8.5, whereas *G. arilantensis* produced more protein and grown well at pH 8. Hence, these bacteria were subjected to submerged fermentation using chicken feather powder (1%) as Carbon and Nitrogen sources. Among them, *G. arilantensis* is the superior in degrading feathers producing more protein (2.15 ± 0.04 mg/ml) and amino acids (0.498 ± 0.019 µg/ml). The microscopically observed masses of feather hydrolysates revealed that *G. arilantensis* degraded the chicken feathers more efficient comparatively other two bacteria.

1. INTRODUCTION

The poultry industries largely satisfy the ever increasing animal protein demand globally that has arisen due to population explosion [1]. People consume large quantity of chicken meat, consequently leads to the enormous generation of slaughterhouse waste [2]. Globally, 8.5 billion tons of poultry feather waste has been produced annually. In 2018, India produced 4.6 million tons of broiler chicken [3] and approximately 350 million tons of feather waste generated every year [4] and the feathers are indiscriminately dumped into the environment [5]. The accumulation of feathers in the environment leads to a variety of human ailments including cholera, fowl cholera, mycoplasmosis, and the outbreaks of H5N1 virus [6,7]. More than 90% of feathers are composed of protein and the major one is keratin. Extensive disulfide bonds and cross-linkages make keratin rigid and insoluble in water. Based on the sulfur content, keratins are classified as: hard keratins (feather, hair, hoof, and nail) and soft keratins (skin and callus) [8]. Sulfur content in keratin is high due to the presence of sulfur containing aminoacids such as cystine, cysteine, and methionine [9,10]. Keratin is remarkably resistant to degradation

by regular proteolytic enzymes such as pepsin, papain, and trypsin. Nevertheless, feathers are considered as a good source of protein and essential amino acids if systematically recycled [11].

Various strategies have been adapted to manage the slaughterhouse waste generated in India. However, feather waste still remains a challenge [12,13].

Conventional methods of feather degradation such as acid/alkali hydrolysis and steam pressure cooking destroy the heat sensitive essential amino acids such as lysine, methionine, and tryptophan and generate non-nutritive amino acids apart from consuming huge amount of energy [14-16]. Land dumping and incineration methods are likely to result in environmental destruction and generates highly toxic emissions [17]. The conventional method of processing feather waste is cost and labor intensive that may reduce the nutritive value. Protease is used for hydrolysis of the peptide bonds that link amino acids together in a polypeptide chain [18,19]. The enzyme-catalyzed reaction is cost effective, highly efficient, and easily selective which usually requires mild reaction conditions and less energy which consequently improves the value of feathers [20]. A group of secreted proteolytic enzymes called as keratinases have the ability to hydrolyze insoluble keratins more efficiently than other proteases, are produced by microorganisms and some insects [21]. Hence, the alternative method of using microorganisms and their enzymes in

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