Isolation, Identification, and Keratinolytic Activity of Several Feather-Degrading Bacterial Isolates

Taha I. Zaghloul,*,1 M. Al-Bahra,2 and H. Al-Azmeh2

1Department of Bioscience and Technology, Institute of Graduate Studies and Research, University of Alexandria, Alexandria, Egypt; and 2Department of Chemistry, Faculty of Science, Damascus University, Syria

ABSTRACT

Several feather-degrading bacterial isolates were isolated from Egyptian soil. These isolates were able to degrade chicken feather, when grown on basal medium containing 1% native feather as a source of energy, carbon, and nitrogen. Feather waste, generated in large quantities as a byproduct of commercial poultry processing, is nearly pure keratin, which is not easily degradable by common proteolytic enzymes. The isolates were identified according to the morphological characteristics, biochemical tests, and API 50 CHB Bacillus system. Proteolytic and keratinolytic activities of these isolates were monitored throughout the cultivation of the bacterial isolates on feather. Resulting soluble proteins, which were released as a result of the biodegradation of feather, were demonstrated by SDS-PAGE.

Index Entries: Bacillus; isolation; identification; keratinolytic activity.

INTRODUCTION

Environmental wastes are found in large quantities in many countries. Although some of them contain a considerable amount of protein and various carbon compounds, little attention is given to using them in a technological way. Recently, the authors have focused on the utilization of some environmental wastes, mainly feather waste. Feather waste, generated in large quantities as a byproduct of commercial poultry processing, is nearly pure keratin (1). Because of the high degree of disulfide bonds, hydrophobic interactions, and hydrogen bonds, keratin in its native state is not degradable by common proteolytic enzymes, such as pepsin, trypsin,
Feather waste is utilized on a limited basis as a dietary protein supplement for animal feedstuffs. Generally, the feather is steam-pressure-cooked or chemically treated before use (3). Biodegradation of feather by microorganisms represents an alternative method to improve the nutritional value of feather waste.

The present work reports the isolation of several feather-degrading bacterial isolates from soil. These isolates are able to degrade whole chicken feather when grown on basal medium containing 1% native feather. The isolates were classified as members of the genus *Bacillus*. Proteolytic and keratinolytic activities of these bacterial isolates were monitored throughout the biodegradation process.

**MATERIALS AND METHODS**

**Media**

Bacterial isolates were activated and grown on PY medium (4) (Bacto peptone, 10 g; Difco (East Molesley, Surrey, UK) yeast extract, 5 g; and NaCl, 5 g/L). PA medium is PY supplemented with 1.5% agar agar. Basal medium II (5) (NH₄Cl, 0.5 g; NaCl, 0.5 g; K₂HPO₄, 0.3 g; KH₂PO₄, 0.4 g; MgCl₂·6H₂O, 0.1 g; and yeast extract, 0.1 g/L), supplemented with 1% (w/v) whole chicken feather, was used to check the proteolytic and keratinolytic activity of the bacterial isolates. Feather plates were made as follows: 2 g of whole feather were treated with about 5 mL of NaOH (5 N), to the point that feather was converted to a soft paste. The volume was adjusted to 100 mL with basal medium II, after which the pH was adjusted to pH 7.0, and 1.5 g agar was added. The medium was autoclaved at 110°C for 10 min.

**Isolation of Microorganisms**

Several pieces of whole chicken feather were placed into wet soil in plastic containers. The containers were watered every 2 d for 2 wk, after which the partially degraded feather pieces were used to inoculate PY medium. The new culture was allowed to grow at 37°C for 24 h, with shaking. Suitable dilutions of this culture were plated on PA plates. Single colonies were screened for their ability to hydrolyze milk and keratin, by patching them into milk agar plates and feather plates, respectively. Alternatively, 100 mL of PY medium was inoculated with 1% soil suspension, and the culture was allowed to grow overnight, with shaking, at 37°C. Basal medium II, containing 1% whole feather, was inoculated with 1 mL of the above culture, and the new culture was allowed to grow at 37°C for 2, 4, and 6 d. A sample was taken every 2 d, diluted, and plated on PA plates. Single colonies were checked on milk plates and feather plates, as described above.