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DETERMINATION OF PROTEIN IN MUNICIPAL SLUDGE OF KARACHI UNIVERSITY CAMPUS

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ABSTRACT

The protein content of 20 primary sludge samples of domestic origin was estimated by Kjeldahl and Lowry's methods as described in Standard Methods for the Examination of Water and Wastewater. The results obtained by the two methods were comparable as shown by their statistical analysis.

Key-words: Protein estimation, municipal sludge, University campus, Kjeldahl and Lowry's methods

INTRODUCTION

Municipal sewage sludge has long been considered a valuable manure but in the recent years its possible use as animal feed or feed supplement has been considered because of the presence of an appreciable amount of protein which could be used to replace conventional sources of protein in animal feeding stuffs (Chishti *et al.,* 1992: Berzedits *et al.,* 1981; Christiansen and Mirchell, 1978).

During past few decades a great deal of work has been done on the development of economically useful products and by-products from sewage sludge (Leary, 1954; Forester, 1973, Sridhar and Pillai, 1973). The presence of protein in sewage sludge and its extraction has been reported by many workers (Christiansen and Mirchell, 1978). On dry weight basis sewage sludge contains considerable amount of protein which can be separated and used in animal feed without any adverse affect (Chishti *et al.,* 1992; Sridhar and Pillai, 1973). In addition, to protein and other valuable materials sludge has also been investigated by some workers for its metal composition and its possible effects on plants and animal tissues (Furr *et al.,* 1976; Hansen, 1976; Hinesly, 1976). Recovered solids could be rendered into animal feed supplement or sprayed as fertilizer (Gupta and Kim, 1995; Rooney, 1992; Cai *et al.,* 1992; Smith, 1991; Totzke, 1991; Hopwood, 1977; Woodard *et al.,* 1977; Westerman *et al.,* 1989).

Many techniques, such as chromatography and dialysis for the extraction and purification of proteins have been developed for the studies of plant and animal cells and tissues. These techniques although work for many industrial and domestic wastes but are time consuming and are not generally applicable to routine protein determination with the facilities available at the treatment plants. Knowledge of the amount of proteins in wastewater, sludge and similar substances would be useful not only for determining the pollution-effects on the environment but also for determining the possible usage of the resulting materials in agriculture including animal nutrition. The aim of this work was to develop a suitable and economical method for the recovery of protein from primary sludge for its possible reuse.

MATERIALS AND METHODS

The methods used for the estimation of sludge protein were Kjeldahl and Lowry method as described in the Standard Methods for the Examination of Water and Wastewater (APHA, 1992) and (Lowry *et.al.,* 1965).

Kjeldahl's Method

A 10 ml of domestic sludge sample was taken in 300 ml Kjeldahl flask and it was heated in the presence of sulphuric acid, potassium sulphate and mercuric sulphate till the solution became colorless or pale yellow. After digestion the residue was mixed with 10 ml sodium hydroxide-sodium thiosulphate (500 g; 25g/l) solution and ammonia was distilled over and nesslerized. The absorbance was determined photometrically at 425 nm and the concentration was computed using standard curve.

Lowry's method

Reagents

i. Solution A
20 g of sodium carbonate was mixed with 4gm sodium hydroxide and diluted to one litre with distilled water.
ii. Solution .B
100 mg of sodium tartarate was mixed with 50 mg copper sulphate in 10 ml distilled water.
iii. Solution C
1.0 ml of Folin ciocalten phenol reagent was mixed with one ml distilled water.
iv. Solution D
1.0 ml of Sol. B was mixed with 40 ml of Sol. A.

One ml of sample was taken in a test tube to which 5 ml of solution D was added. Thoroughly mixed and allowed to stand for 15 minutes at room temperature. 0.5 ml of solution C was then added to this tube and mixed immediately. Absorbance was taken at 650 nm on a spectrophotometer (SP-6-400 Pye Unicamp).

Table 1 Comparison of Kjeldahl's method and Lowry's method for protein estimation in sludge.

Sample No.	**Kjeldahl protein (N x 6.25) gm/l**	**Lowry's protein (gm/1)**
1	8.2	8.1
2	9.0	8.6
3	9.2	9.2
4	10.5	9.5
5	11.1	11.0
6	11.8	11.5
7	13.1	12.5
8	10.9	11.2
9	10.0	10.2
10	10.9	11.0
11	10.9	11.0
12	11.6	11.5
13	11.5	11.5
14	12.7	12.5
15	12.7	12.5
16	11.0	10.8
17	11.5	11.8
18	11.0	11.5
19	10.5	10.8
20	10.8	10.4

(t-computed <t 0.1, 0.05)

RESULTS AND DISCUSSION

An accurate determination of the concentration of protein in sludge is difficult because no chemical reaction is known to be specific for sludge proteins. This leaves with the choice of attempting to extract the protein in a purified form and then analyzing it with different methods.

The chemical structure of protein suggested two methods for determining their concentration in sludge. A quantitative test for the amount of organic Kjeldahl nitrogen (OKN) in a sample which is an indication of the amount of protein present and colourimetric reaction for a specific functional group or amino acid associated with protein molecules (Mattock , 1978).

Typical results of estimation of protein in sewage sludge by the two methods viz. Kjeldahl's method (OKN x 6.25) and Lowry's method seem to be quite comparable for protein estimation (Table 1). The t test and F test for the two methods indicated that there is no significant difference between the two methods.

The Kjeldahl's method has an advantage over Lowry's method. It can be used for protein estimation in both liquid and solid samples whereas Lowry's method suitable only for liquid samples. It is a sensitive and acceptable method for protein estimation using Folin Ciocalteu phenol reagent. In sludge sample of ---- protein content varied from 8.2 to 13.1g/L by OKN method and from 8.1 to 12.5 g/L by Lowry's method.

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