

COMPARATIVE STUDY ON THE METHODS FOR ASSESSMENT OF TOTAL URINARY PROTEIN

Thesis

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Master Degree of Clinical and Chemical Pathology

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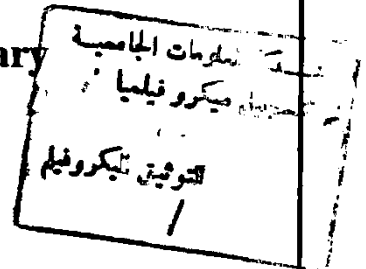
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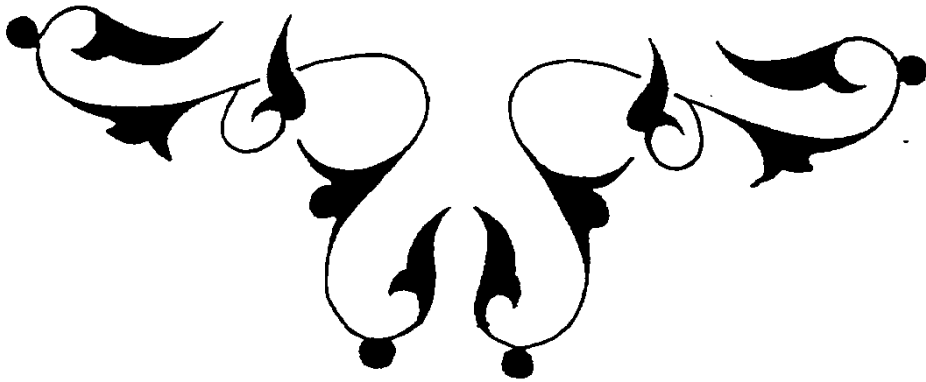
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INTRODUCTION

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The term proteinuria generally refers to increased rate of urinary protein excretion in excess of normal. Protein excretion is increased when the total daily excretion exceeds 150 mg/day or average rate of excretion exceeds 10-11 micrograms/minute (Maher, 1985).

The concentration of protein in a single voided sample of urine will vary inversely with urine flow. However, even at low flow rates, The concentration of protein in normal urine does not exceed 10 to 20 mg/100 ml. Thus, the occurrence of a higher concentration in any specimen of urine implies the existence of proteinuria (Manuel et al., 1970).

Proteinuria is a common laboratory abnormality among adult and children. It may take a physiological or pathological form. Physiological proteinuria occurs in the form of transitory elevation after intense exercise or work and exposure to cold (Kannel, 1984).

Pathological proteinuria may be due to renal or non renal disease. Proteinuria mainly albuminuria is an important parameter of renal function. So, albuminuria is mainly a manifestation of primary renal disease with such

conditions as pyelonephritis and acute glomerulonephritis. In those conditions the degree of proteinuria is slight usually accounting to less than 2 gm/day. In chronic glomerulonephritis, nephrotic syndrome including lipoid nephrosis and in some forms of hypertensive vascular disease protein loss may vary from a few grams to as much as 30 gm/day (Richmond et al., 1982).

Proteinuria is also encountered in non renal disease when they give rise to kidney lesion among those are lupus erythematosus, amyloidosis, toxemia of pregnancy, septicemia and certain forms of drug and chemical poisoning (Vacca et al., 1986).

AIM OF THE WORK

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Comparison of different methods used for assessment of total urinary protein to evaluate the most sensitive, accurate and the most applicable as a routine method in the clinical chemistry laboratories. The methods included in this study:

- The turbidimetric method by using trichloroacetic acid (manually prepared),
- Turbidimetric method using sulfosalicylic acid (manually prepared),
- Turbidimetric method using (Kit of stanbio laboratories*),
- Dye binding method of pyrogallol red molybdate by using kit of Biotrol** - and Biuret method using Biuret reagent (manually prepared).

* Stanbio Inc 230 East Houston street, san Antonio Texas 78202

** Biotrol (Rossatta Co.), 132 Tahrir St. Dokki, Cairo, Egypt, Tel 717457 - 701992.

REVIEW OF LITERATURE

I- PROTEINURIA

[A] NORMAL PROTEINS IN URINE:

These are derived from plasma and urinary tract. The basement membranes of the glomeruli behave as ultrafilters for plasma proteins. Filtration of protein is dependent on its molecular size and its concentration. Normally high molecular weight proteins (>100.000 dalton) do not appear in glomerular filtrate while, plasma proteins with molecular weight less than 50.000 to 60.000 pass through the glomerular membrane and are reabsorbed by the proximal tubular cells. Proteins with M.W 15.000-40.000 are filtered more readily but in lesser quantities because of their low plasma concentration (Tietz, 1987).

The normal amount of proteins excreted by healthy individuals is up to 150 mg/24 hour or 10 mg/dl, depending on urine volume. This amount consists of about one third of albumin and the remaining plasma proteins include many small globulin like retinol binding, B₂ microglobulin, immunoglobulin light chains and lysozyme are excreted in small amounts. Tamm Horsfall glycoprotein constitutes about one third or more of the total normal protein excretion (Rock et al., 1987).

Proteins which are derived from the urinary tract include:

1) Tamm-Horsfall mucoprotein:

This is the most abundant one which constitute about one third of the normal total daily protein excretion. It is synthesized by the epithelial cells in the distal tubule, it possess antiviral activity and is involved in cast formation.

2) Urokinase (Plasminogen activator):

This is an antifibrinolytic enzyme that is secreted by the tubular cells into the lumenal fluid.

3) Secretory IgA:

This type of immunoglobulin is synthesized by the renal tubular epithelial cells and it is responsible for the local defence mechanism of the renal tubules (Tietz, 1987).

Table (1): Normal urinary protein level:

	Normal range of concentration in mg/100 ml	Approximate diurnal output mg/day
Total protein	10-12	100-150 mg/day
Mucoprotein	3-5	50-70
Albumin	2.5 - 4.5	40-50
Globulin	1.5-2.5	6-10

(Schosinsky et al., 1974)

[B] TYPES OF PROTEINURIA:

1) Intermittent proteinuria:

This type is benign in nature in which urine specimen has an abnormal quantity of protein. It can be further described as:

a) Benign transient Proteinuria:

It describes the isolated finding of urine containing abnormal amount of protein that disappears within few days. This condition is most often seen in children or young adults. It is found by chance during routine chemical examinations, there is no evidence of progression to renal disease and it is considered to be idiopathic (Rennke and Venkatachalam, 1977).

b) Functional proteinuria:

It is characterized by excretion of urinary protein in the absence of renal disease. Functional proteinuria may accompany a febrile illness, strenuous exercise, cold or emotional stress, congestive heart failure, hypertension, seizures, abdominal operation and therapy with sympathomimetic drugs. It is usually of glomerular origin and renal vasoconstriction has been implicated as the primary mechanism. This type of proteinuria usually resolves

with appropriate treatment or rest within 2-3 days (Abuelo, 1983).

c) Postural or orthostatic proteinuria:

It occurs in individuals only while they are in the upright or lordotic position. The first morning voided specimen is invariably normal in protein content in these individuals. The mechanism by which postural proteinuria occurs is renal vasoconstriction as well as venous congestion which is produced by the lordotic or passively erect posture (Thompson and Wootton, 1970).

Transient postural proteinuria is considered to be benign. But individuals with this type of proteinuria should be checked periodically at six months intervals. If proteinuria is chronic or not entirely related to posture it will suggest underlying renal disease. The evaluation for orthostatic proteinuria involves a minimum of three urine samples collected up on first arising in the morning and three samples obtained in the afternoon or evening for three days. Trace or negative results with urine dipsticks for morning (supine) urine samples together with one positive reading or greater in the afternoon (upright) samples indicate an orthostatic pattern (Rytand and spreiter, 1981).