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Identifying the pattern of urine cast and crystals among diabetic and renal disease individuals: A prospective study

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Abstract

Urinalysis can reveal diseases that have gone unnoticed because they do not produce striking signs or symptoms. Examples include diabetes mellitus, various forms of glomerulonephritis, and chronic urinary tract infections. Microscopic identification reveals the current status of the kidney. The study aimed to determine the various types of urine cast, crystals and cells based on the disease condition which directly impact on the renal status. Sterile urine was processed at Roche U411 fully automated urine analyser for chemical constituents and microscopic identifications were done manually after centrifugation. Clinical chemistry analytes were measured using Beckman Coulter AU480 spectrophotometrically. Sodium and Potassium were analysed using direct ion selective electrode using ABL800 Rapid Diagnostics. Out of 50 samples, 16 were found as both CKD and diabetics whose protein and urine sugar is elevated. Based on the diseased condition the patterns were identified. This study proved that there is no direct correlation between the urine osmolality and the urine deposits (casts, crystals and cells) whereas particular types of casts, crystals and cells are present in specific diseased condition.

Keywords: Urine Cast, Crystals, diabetes, Renal Disease

Introduction

Osmolality is referred as the total number of particles which are present in the solution. The unit of osmolality is mmol/kg solvent (or) mOsmol/kg solvent. Osmolality were typically determined by combination and segregation of the molecules constantly present in the solvent. The urine osmolality is measured to assess the concentration of urine as specifically which differs from 50–1000 mmol/kg of water in healthy person which is related to the specific gravity in the range of 1.001–1.030. In some conditions the maximum concentration of urine can reach up to 1400 mOsmol/L after prolonged water loss. The changes in water-conserving mechanism will cause the high degree of changes in plasma osmolality whereas in urine osmolality it produces the same level of changes such as excessive water loss (or) water accumulation.[1-4]

Urine sodium is weakly correlated by measured urine osmolality and urine urea nitrogen and urine creatinine is firmly correlated with measured urine osmolality. There are ample of literature available in determination and calculation of urine osmolality using sodium (Na), potassium (K), ammonia(NH4), urine urea nitrogen, creatinine and protein.[5] Microscopic evidence of the deposit urine sample used to spot-out, the cast, crystals and cells.

The microscopic identification will reveal precise status of the excretory system

Nephron liberates the coagulated protein reflects results in the formation of the casts. They are cylindrical in shape and formed in the tubular portion of the kidneys along with it resembles the shape of the tubes. The most common type of cellular casts are red blood cell and white blood cell casts and others are granular casts, fatty casts and waxy casts which are also present in the urine sediment.

Urine is an eliminated waste product of body which contains dissolved solutes. The solid forms of these solutes are called as crystals. These crystals are classified and named based on their shape, colour and pH. Small, sand like crystals which don't have shape they are termed as amorphous crystals and some have the particular shape, such as needle-like crystals. Crystals are formed when urine cools after collection and they are not present inside the body. In healthy person, amorphous urates, uric acid crystals, calcium oxalates, amorphous phosphates are considered as normal constituents of the urine. The crystals are formed from the substances they treated as abnormal crystals which indicates the diseased condition or abnormal metabolic process. Calcium carbonate, cystine, tyrosine, leucine are some of the examples of abnormal crystals present in the urine.

If the crystals are from substances that are not normally in the urine, they are considered "abnormal." Abnormal crystals may indicate an abnormal metabolic process. Some of these include: Calcium carbonate, Cystine, Tyrosine, Leucine and so on.

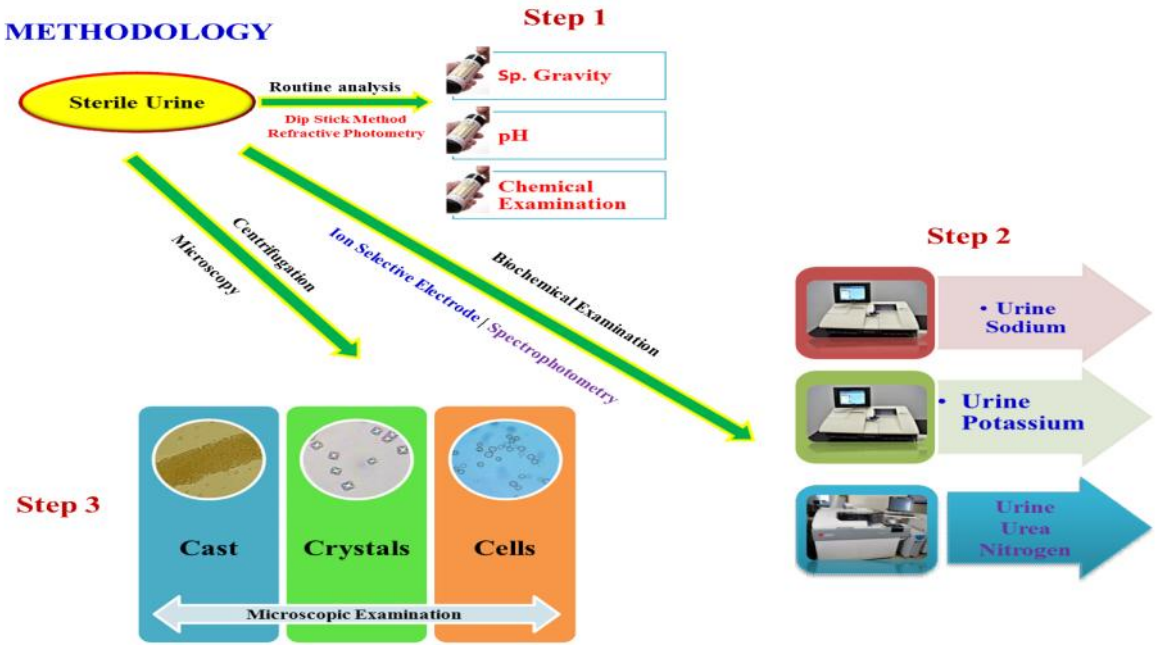
Cells: Usually epithelial cells, pus cells increase in the number in HPF is considered as the abnormal and the pattern decided the disease.

Hence study aimed to determine the various types of urine cast, crystals and cells based on the disease condition which directly impact on the renal status in diabetic and CKD.

Materials and Methods

Sample collection

Patients from Billroth Hospitals, Shenoy Nagar, Chennai-600030 are selected for the study and the proposal were approved by the Institutional Ethics committee (IEC No: 2019/Lab / 001). Urine samples were collected in the clean, dry and sterile container at ward with the assistance of the ward nurse or a patient attender. To avoid the contamination, the mid-stream portion of the urine was collected. After voiding the sample into the container, it was tightly closed with the lid. The patient ID, name, age, sex were written in the label and it was pasted on the container. The collected urine sample was placed in the box and maintained the cold chain during transport. The samples were received at the accession department and it was immediately sent to the respective department for further analysis.



Step 1

After receiving the sample, a small aliquot of sample was taken in a separate tube and it was used for routine analysis. The Chemstrip 10 UA test strip was dipped into the urine sample and while removing the strip it was wiped along the sides of the tube. The dipped test strip was placed in the tray of the instrument and strip was read using Roche Cobas U411 automated urine analyzer. The light passes through the dipped wet strip which gets refracted. The refracted emission will be captured and calculated as arithmetic value. The equipment functions by the methodology of refractive photometry. pH, Specific gravity , urine glucose and urine protein were used for the categorisation of individuals in diabetic and renal diseases.

Step 2

Other aliquot was taken for the biochemical examination to analyse urine sodium, Urine potassium and urine urea nitrogen. ABL 800 – Rapid Diagnosis were used for the analysis of (Sodium and potassium) which estimates by the principle of ion selective electrode and the urine urea nitrogen was analysed using in Beckman Coulter AU480 fully automated chemistry analyser, which reads by spectrophotometry at 340nm.

Step 3

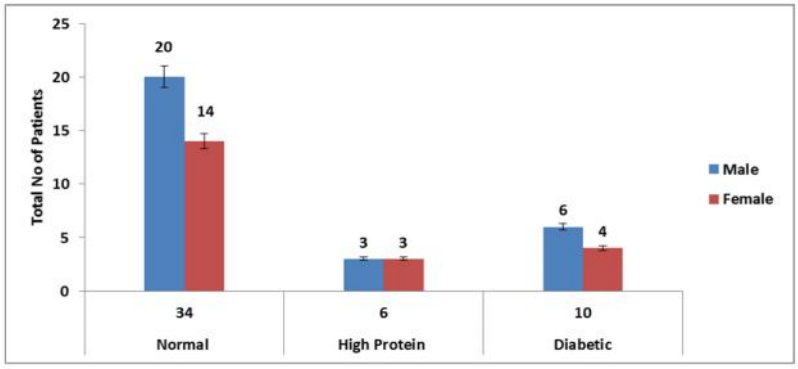
Another aliquot were centrifuged3000rpm for 10mins, the supernatant was discarded and the pellet was poured on the clean, greese-free slide. The fine pellet was covered with the coverslip without air bubbles and it was viewed under the light microscopy for the observation of cast, crystals and cells present in the urine. The microscopic examination of cells, crystals and other substances were recorded as the number observed per “low power field (LPF) and high power field (HPF)”. If epithelial cells, bacteria and crystals are observed, they were estimated as “few”, “moderate” or “plenty”.

Results

Categorization of patients based on the chemical examination of urine

Among 50 patients 34 were found as normal whereas others are considered as patients whose chemical concentration of urine abnormal. Remaining 16 patients were classified into two categories such as chronic renal disease and diabetic whose protein and urine sugar is high. Among those 16, 3 male and 3 female were found high protein and 6 male and 4 female have found high glucose **(Fig. 5.1) (Table 5.1).**

FIG. 1. CATEGORIZATION OF PATIENTS BASED ON THE CHEMICAL EXAMINATION OF URINE



Each bar represent the total no of cases taken for the study.
Since the study group confined to the single category and less no of samples.
5% Standard error mean (SEM) was employed to identify the difference

Table - 1

	Normal	High Protein	Diabetic
	34	6	10
Male	20	3	6
Female	14	3	4

Analysis of specific gravity and pH to identify the concentration of urine

Among 16 abnormal patients 1 male and 4 female has the specific gravity of 1.015 and 5 male and 4 female had the specific gravity of 1.020 and 2 male have specific gravity of 1.030 (Fig.5.2a) (Table 5.2a).

Among 16 abnormal patients 6 male and 6 female has the pH of 5.0 and 2 male and 2 female has the pH value of 6.0 (Fig.5.2b) (Table 5.2b).

FIG. 2. ANALYSIS OF SPECIFIC GRAVITY AND pH TO IDENTIFY THE CONCENTRATION OF URINE

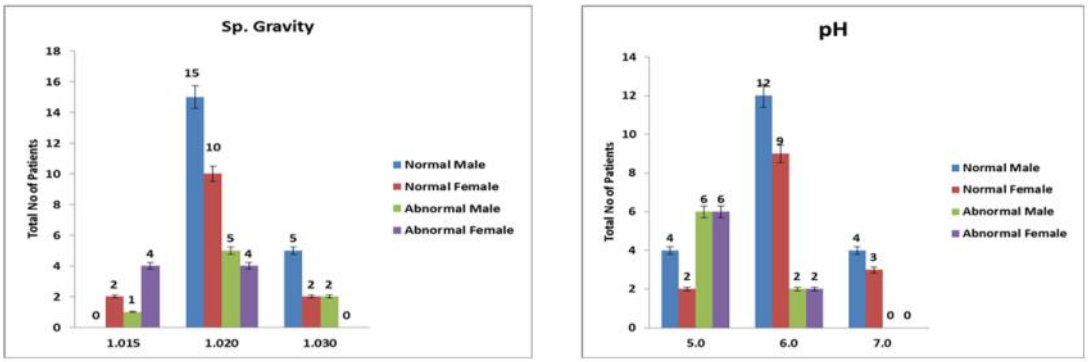


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Table - 2a

		Sp Gravity		
		1.015	1.020	1.030
Normal	Male	0	15	5
	Female	2	10	2
Abnormal	Male	1	5	2
	Female	4	4	0

Table - 2b

		pH		
		5.0	6.0	7.0
Normal	Male	4	12	4
	Female	2	9	3
Abnormal	Male	6	2	0
	Female	6	2	0

Estimation of urine osmolality using calculated method urine sodium, urine potassium and urine UUN

CKD and Diabetic patients are 16 in number, amongst 1 female has the low urine osmolality of below the 300 mOsm/kg, 9 male and 6 female has the normal urine osmolality of range is 300 – 900 mOsm/kg and none has found as high urine osmolality(Fig. 5.3) (Table 5.3).

FIG. 3. ESTIMATION OF URINE OSMOLALITY USING CALCULATED METHOD URINE SODIUM , URINE POTASSIUM AND URINE UREA NITROGEN

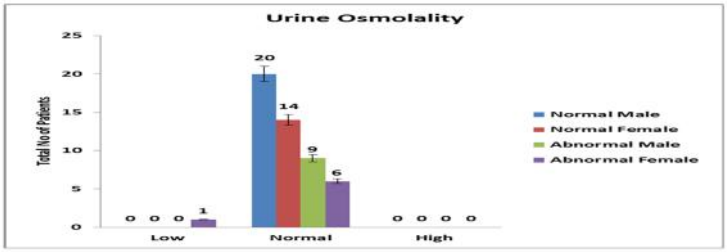

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Table - 3

	Urine Osmolality			
		Low	Normal	High
Normal	Male	0	20	0
	Female	0	14	0
Abnormal	Male	0	9	0
	Female	1	6	0

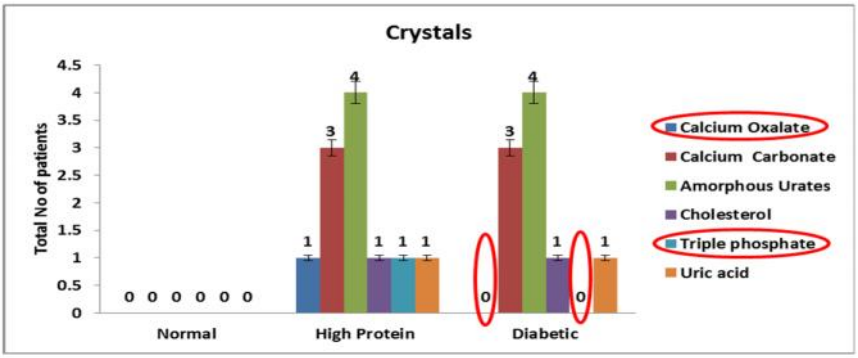
Urine Osmolality : (Urine Sodium x 2) + (Urine Potassium x 2) + 18 + (Urine Urea Nitrogen / 2.8)

Identification of different types of urine crystals in the patients of CKD and diabetes

Among 6 patients of high protein in their urine, 4 patients has the amorphous urate crystals,3 patients has the calcium carbonate crystals,1 patient has the calcium oxalate crystal, 1 patient has the cholesterol crystal, 1 patient has the triple phosphate crystal and 1 patient has the uric acid crystal.

Among 10 patients of high glucose in their urine, 4 patients has the amorphous urate crystals, 3 patients has the calcium carbonate crystals, 1 patient has the cholesterol crystal,1 patient has the uric acid crystal and none of the patient had the calcium oxalate and triple phosphate crystals (Fig. 5.4) (Table 5.4)

FIG. 4. IDENTIFICATION OF DIFFERENT TYPES URINE CRYSTALS IN THE PATIENTS OF CHRONIC KIDNEY DISEASE AND DIABETES



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5% Standard error mean (SEM) was employed to identify the difference

Table - 4

S.No	Type of Crystals	Normal	High Protein	Total High Protein	Diabetic	Total Diabetic
1	Calcium Oxalate	0	1	6	0	10
2	Calcium Carbonate	0	3	6	3	10
3	Amorphous Urates	0	4	6	4	10
4	Cholesterol	0	1	6	1	10
5	Triple phosphate	0	1	6	0	10
6	Uric acid	0	1	6	1	10

Representative picture of crystals in High Protein patients

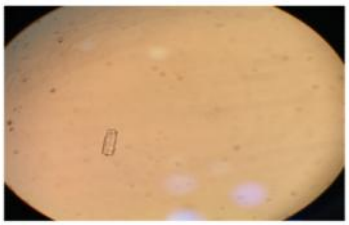


Plate 1.Triple phosphate

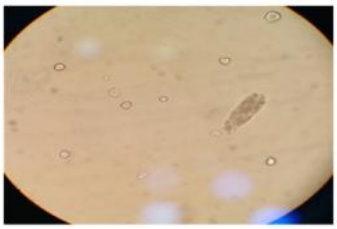


Plate 2.Calcium carbonate

Plate 3.Calcium oxalate

Representative picture of crystals in Diabetic patients

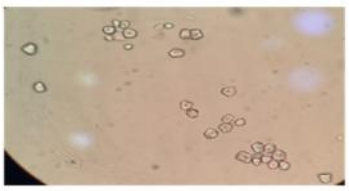


Plate 4.Calcium carbonate

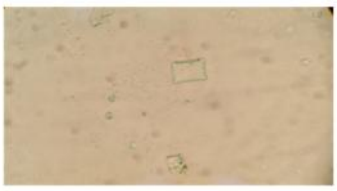


Plate 5.Uric acid

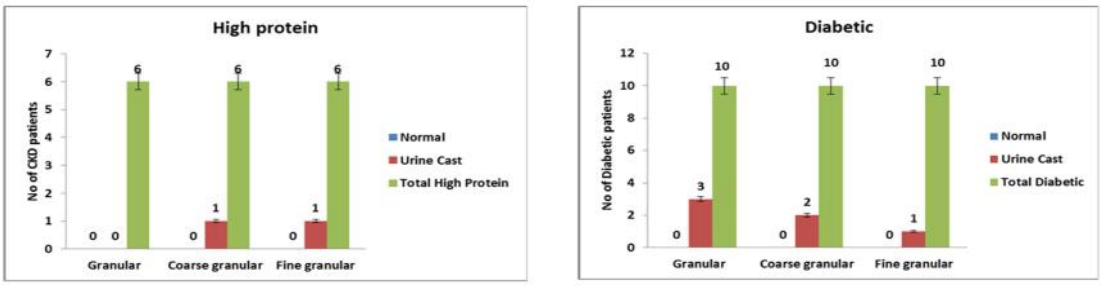
Plate 6.Amorphous urates

Identification of different types of urine casts in the patients of Chronic Kidney Disease and diabetes

The result shows that 6 patients of high protein have two types of granular casts. One found coarse granular casts and other has fine granular casts (Fig.5.5a) (Table 5.5).

While considering among diabetic cases whose glucose is high in urine. The result shows 3 patients have the granular casts, 2 patients have the coarse granular casts and 1 patient has the fine granular casts (Fig.5.5b) (Table 5.5).

FIG. 5. IDENTIFICATION OF DIFFERENT TYPES URINE CAST IN THE PATIENTS OF CHRONIC KIDNEY DISEASE AND DIABETES



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Since the study group confined to the single category and less no of samples.
5% Standard error mean (SEM) was employed to identify the difference

Table - 5

S.No	Type of Cast	Normal	Urine Cast	Total High Protein	Urine Cast	Total Diabetic
1	Granular	0	0	6	**3**	10
2	Coarse granular	0	1	6	**2**	10
3	Fine granular	0	1	6	**1**	10

Representative picture of Cast in High Protein patients

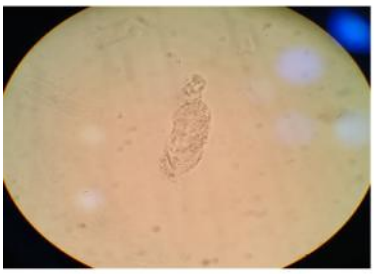

Plate 7.Fine granular

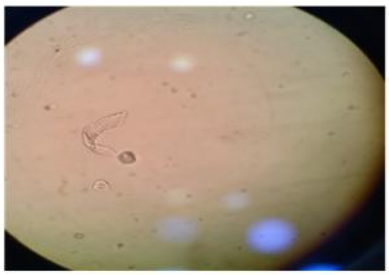

Plate 8.Fine granular

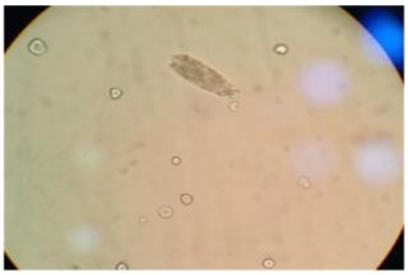

Plate 9.Coarse granular

Representative picture of Cast in Diabetic patients

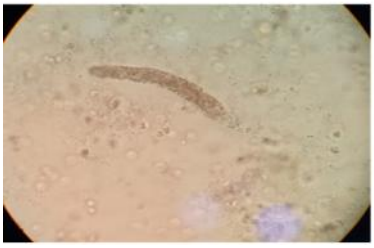

Plate 10.Fine granular

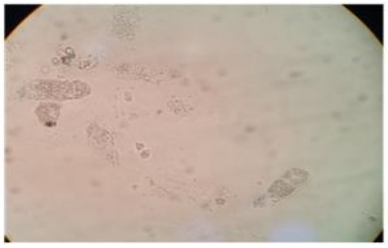

Plate 11.Coarse granular

Plate 12.Granular

Identification of different types of urine cells in the patients of Chronic Kidney Disease and diabetes

Pus cells: This result shows the normal individual of 34 both male and female , 24 individual have the range of 1-2 pus cells and 10 persons have the range of 2-4 pus cells in their urine which is considered as normal while compare with biological references intervals.

Among 6 CKD patients, 1 has the range of 2-4 pus cells and 5 patients have the range of 8-10 pus cells in their urine. Where as in the 10 diabetic patients, there are 3 patients have 2-4 pus cells, 5 have 8-10 pus cells and 2 patients have plenty of pus cells in their urine which is considered as highly abnormal of infected (Fig.5.6a) (Table 5.6a).

FIG. 6a. IDENTIFICATION OF DIFFERENT TYPES URINE CELLS IN THE PATIENTS OF CHRONIC KIDNEY DISEASE AND DIABETES

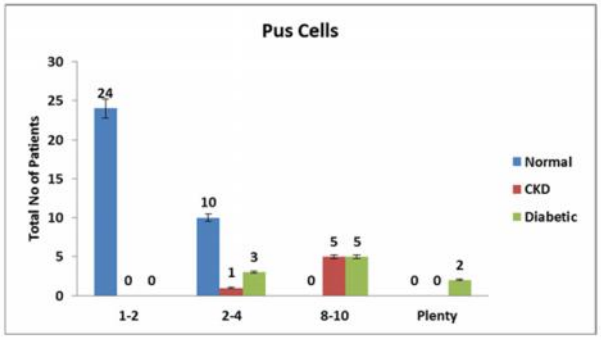

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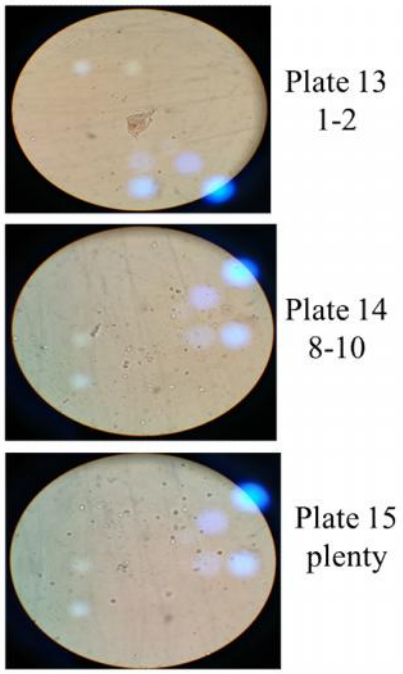
Table – 6a

Pus	1-2	2-4	8-10	Plenty
Normal	24	10	0	0
CKD	0	1	5	0
Diabetic	0	3	5	2



Representative picture of the respective groups
Normal | High Protein | Diabetic

Epithelial cells: The results for 34 normal individual, 8 persons has the range of 1-2 epithelial cells and 26 persons has the range of 2-4 epithelial cells in their urine which are normal.

For 6 CKD patients, 1 has the range of 2-4 epithelial cells, 2 patients slides found of 8-10 epithelial cells, 3 patients has the plenty of epithelial cells in their urine. While compare with the 10 diabetic patients, 1 patient has the range of 2-4 epithelial cells, 1 has 8-10 epithelial cells and 8 patients has the plenty of epithelial cells in their urine (Fig.5.6b) (Table 5.6b).

FIG. 6b. IDENTIFICATION OF DIFFERENT TYPES URINE CELLS IN THE PATIENTS OF CHRONIC KIDNEY DISEASE AND DIABETES

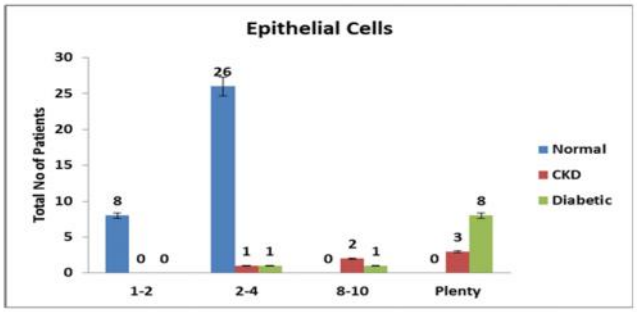


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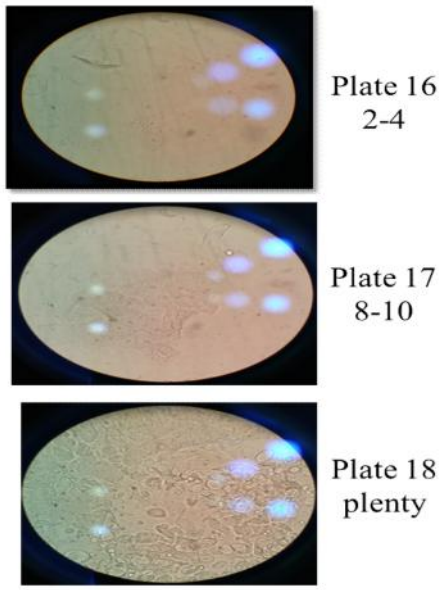
Table –6b

Epithelial Cells	1-2	2-4	8-10	Plenty
Normal	8	26	0	0
CKD	0	1	2	3
Diabetic	0	1	1	8



Representative picture of the respective groups
Normal | High Protein | Diabetic

RBC's: Results shows the counts of RBC's for the normal 34 persons, among that 22 has very occasional RBC's, 8 persons has 1-2 RBC's/ high power field and 4 persons has 2-4 RBC's in their urine. While comparing with 6 CKD patients, 2 patients has the range of 1-2 RBC's and 4 patients has the range of 2-4 RBC's in their urine. Whereas 10 diabetic patients, 5 patients has the range of 1-2 RBC's and 5 patients has the range of 2-4 RBC's in their urine (Fig.5.6c) (Table 5.6c).

FIG. 6c. IDENTIFICATION OF DIFFERENT TYPES URINE CELLS IN THE PATIENTS OF CHRONIC KIDNEY DISEASE AND DIABETES

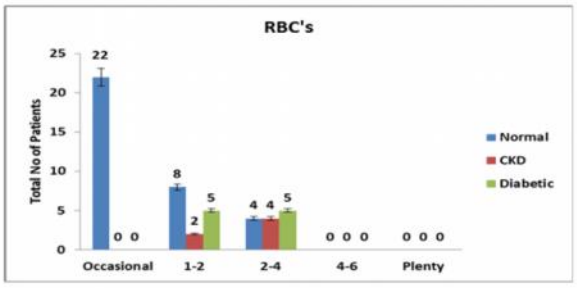


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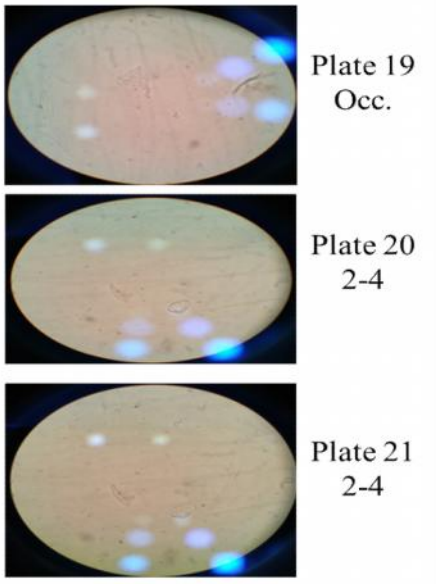
Table – 6c

RBC	Occasional	1-2	2-4	4-6	Plenty
Normal	22	8	4	0	0
CKD	0	2	4	0	0
Diabetic	0	5	5	0	0



Representative picture of the respective groups
Normal | High Protein | Diabetic

Discussion

Hippocrates reflected “One can obtain considerable information concerning the general trends by examining the urine,” and properly described urine as a filtrate of blood, one of the four humors of the body next to yellow bile, black bile, and phlegm. Recently, in the early-mid 1800s, an English physician named Richard Bright pioneered the field of kidney research and became known as the “father of nephrology.” Measurement of urine osmolality (Uosm), the gold standard in the estimate of urine rapt ability, is a valuable contrivance for the assessment of renal function in such distinct clinical conditions as acute kidney injury (AKI) and chronic kidney disease (CKD). Although a correlation does exist between Urine deposits and Uosm, at least under normal physiological conditions [6,7] the assumption that Urine deposits accurately reflects Uosm, which underlies any clinical decision based on Urine deposits, has not been formally tested and, in fact, has been recently challenged. In the present study we examined the relation between Urine concentration and the cast, crystals and the cells.

Today urinalysis continues to be a powerful tool in obtaining crucial information for diagnostic purposes in medicine. Body fluid homeostasis is forbidden by the kidney. The determination of serum and urine osmolality is technically effort less practice, and if accurately functional, is of considerable use in the clinical management of water and electrolyte disturbances. In normal subjects with low urine solute concentration the correlation between urine SG and osmolality is good. This correlation becomes deprived at elevated urine solute concentration for the reason that a greater proportion of the solute is urea in addition to non ionized substances.[8]

Marked changes in urine osmolality may occur without being reflected in the specific gravity either in amount or direction of change. Therefore, specific gravity readings around the magic "1.010" mark may be highly misleading. Specific gravity readings of a duplicate urine sample were shown to vary upto 0.005 units for a given interpreter and upto 0.012 units between interpreters. It is evident that if accurate measurement of renal concentrating ability is desired, osmolality is superior to specific gravity measurements. Concept, physiology, and clinical application of osmolal although specific gravity (SG) alone has been used to classify urine concentration [9], use of urine osmolality and urine SG provide a more accurate definition [10]. Hyposthenuria describes urine with a SG less than 1.008 and an osmolality below 269 mOsm/kg. Isosthenuric urine is similar to plasma in concentration with an SG of 1.008–1.012 and osmolality between 260–300 mOsm/kg. Hypersthenuric urine has a SG greater than 1.012 with an osmolality greater than 300 mOsm/kg. Normal adults have hypersthenuric urine. Normal foals have hyposthenuric urine [10].

The reasons why the relationship between Urine deposits and Uosm is less consistent than might be expected are unclear. In the present study, the effect of the possible presence of glucose and/or protein in urine was corrected by applying appropriate equations.[11] However, the association between Urine deposits and Uosm remained loose even after samples containing these solutes were excluded ($p < 0.05$). It should be noted that a myriad of other solutes, commonly encountered in the urine of patients with renal disorders, such as drugs and iodinated radiocontrast agents, could increase urine density, chief to overestimation of the renal concentrating ability.[12-14] Even “physiologic” solutes, such as sodium, potassium and urea, can emerge in generally varying proportions in the urine of both healthy and diseased subjects, each of them exerting a diverse influence on urine density.[15,16] The impulsiveness of these effects helps to explicate the erratic connection amid Urine deposits and Uosm.[17]

The study reveals the pattern of urine deposits such as Granular cast was highly present in diabetic patients and both the granules of fine and coarse present in both type of patients such as CKD and diabetics. In case of crystal the study shows the amorphous urates, calcium carbonate, cholesterol and uric acid were found in both the cases. In addition CKD samples found triple phosphate and calcium oxalate. While observing the cells the study reveals most of the diabetic cases have high and plenty of pus cells and epithelial cells whereas CKD patients alone have the RBCs.

This patterns shows that the renal diseases patients are very prone to the excess release of the protein and its metabolites which results in the formation of Cast, crystals and cells. But in case of diabetic cases its purely depend on the localized infection which acquired from the less immunity and Immunocompromised by administration of insulin and medicines.

Conclusion

In the present study, we hypothesized that there will as definite correlation with the urine osmolality and the urine microscopic examination patterns. Hence the study reveals a new concept that the Urine osmolality variation is not correlated with any of the specific diseases. The variation in the osmolality is purely depending on the current clinical condition of the particular patient. In another way, the patterns of the casts, crystals and cells are directly proportional to the concentration of the urine. This study proves the pattern of the urine deposits in the high protein patients and the diabetics. Hope this finding helps to pave a pathway for the further identification of patterns in other diseases.

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