ABSTRACT

Original Article

Urine Osmolality in Treatment-naïve HIV-positive Subjects in Southeast Nigeria

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Background and Objectives: Urine osmolality varies over a wide range of values in a healthy state. Dilute urine or concentrated urine may be observed in many environmental, physiologic, and disease conditions. Urine osmolality is not commonly evaluated in routine clinical practice and in human immunodeficiency virus (HIV) subjects. The factors that influence urine osmolality have not been completely identified. The aim of this study was to evaluate urine osmolality in treatment-naïve HIV subjects and to identify the factors that may influence dilute and concentrated urine in this group of patients. Methodology: This was a cross-sectional study of treatment-naive HIV subjects conducted in Federal Medical Centre (FMC), Owerri, Nigeria. Demographic and anthropometric data were obtained. Urine osmolality and other relevant investigations were conducted. Normal urine osmolality was defined as 24-h urine osmolality (24 HUOsm) 300-750 mOsm/kgH₂O, dilute urine as 24 HUOsm <300 mOsm/kgH₂O and concentrated urine as 24 HUOsm >750 mOsm/kgH₂O. The association between the variables and urine osmolality and the strength of variables to predict dilute urine and concentrated urine were determined. Results: The mean 24HUOsm was 564 ± 501 mOsm/kgH₂O and the mean spot urine osmolality (SUOsm) $464 \pm 271 \text{ mOsm/kgH}_{2}$ O. Normal urine osmolality was observed in 29.6%, dilute urine in 64.5%, and concentrated urine in 5.9% of the HIV subjects. There was a significant association between urine osmolality and body mass index (BMI), creatinine clearance, as well as serum cholesterol level. Only high-density lipoprotein cholesterol (HDL) predicted dilute urine, whereas BMI, spot urine protein, 24-h urine protein, spot urine creatinine, serum HDL, and CD4 cell count predicted concentrated urine. Conclusion: The prevalence of dilute urine was high among the treatment-naïve HIV subjects. Abnormalities of serum lipids, renal function, and weight were common in treatment-naïve HIV subjects who had dilute urine. There is a need for clinicians to routinely assess urine osmolality and further diagnose for dyslipidemia, renal function impairment, and abnormal weight in HIV subjects at the early stage of the infection.

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KEYWORDS: Dilute and concentrated urine, dyslipidemia, HIV, low CD4 cells count, Nigeria, underweight and obesity, urine osmolality

INTRODUCTION

T he ability of the kidneys to concentrate urine is usually determined by using the specific gravity of urine in clinical practice. Specific gravity is the ratio of weight of urine to an equal volume of water. However, urine osmolality has been shown to

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be more reliable than urine-specific gravity.^[1] Urine osmolality is not routinely evaluated in both the

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general population and HIV subjects in Sub-Saharan African countries.

It is known that urine osmolality varies over a wide range of values in a normal healthy state.^[2] Urine osmolality also shows individual variability, influenced by factors that include thirst threshold, tendency to concentrate urine, and the level of secretion of antidiuretic hormone (vasopressin).^[3-5]

In normal clinical states, urine osmolality is regulated by vasopressin and aquaporin receptors activities in the collecting ducts in response to plasma osmolality.^[6] Abnormalities of tubular function may impact on the osmolality of urine.^[2] In excreting soluble waste products, the kidney has the capacity to maximally concentrate solutes in urine many times as high as the plasma osmolality. By this, water conservation is achieved. Top on the list of the waste products is urea.^[3,7] Other determinants of the osmolality of urine include sodium, protein, glucose, and exogenous substances in the blood and water.^[8]

A study has shown that a low water intake, a low urine volume, or a high plasma copeptin level was associated with a poor kidney outcome.^[3,9] In addition, a study reported that urine osmolality at baseline was an important variable in monitoring renal patients.^[3,10] High urine osmolality contributes to kidney disease progression.^[3] Dilute urine is often associated with the inability of kidney to concentrate urine during early stages of chronic kidney disease.

The factors associated with dilute urine included female gender, age, race, body mass index (BMI), hypertension, water intake, and blood osmolality.^[11]

There is a paucity of studies on urine osmolality and the factors that influence dilute or concentrated urine in the general population and in HIV subjects emanating from Sub-Saharan African countries. This dearth of studies prompted us to carry out this study in treatment-naïve HIV subjects with a view to identifying those who might have dilute or concentrated urine and to further identify the factors that might impact negatively on urine osmolality. Identifying these factors and addressing them early would help stem down the morbidity and mortality of HIV infection.

MATERIALS AND METHODS

This was a cross-sectional study, comprising of 375 treatment-naïve HIV-positive subjects, recruited from an HIV clinic of Federal Medical Centre, Owerri, Nigeria. The study was conducted from April to August 2011. Inclusion criterion was treatment-naïve HIV-seropositive status in subjects who were within the age range of

16–65 years. The subjects who had known pituitary, adrenal, renal diseases, or terminal illness, those who were pregnant, and subjects not willing to give informed consent were excluded from the study. The study was approved by the Ethics Research Committee of the hospital.

With the help of a questionnaire, anthropometric and demographic data were obtained. Our laboratory technicians evaluated the questionnaire and obtained the relevant data. Because the study was hospital based, it was not pretested, as data collection was not difficult. In both English and our native language, the aim of the study was explained to the subjects. The place of domicile and origin, age, and gender of the subjects were obtained. Weight and height were measured and BMI was calculated as the ratio of weight/height² (kg/ m²). Blood pressure measurements were also taken.^[12]

Clear instructions were given to all the subjects on how to collect a 24-h urine sample. At 8.00 a.m. on the designated day, the subject voided urine and discarded. Thereafter, on the next day, the subject collected all his urine into a container until 08.00 a.m. A day-time (08.00 a.m. the second day) random spot urine sample and blood samples were collected at the end of the 24-h urine sample collection.^[13,14] From the random spot urine samples collected, spot urine protein (SUP) level, spot urine creatinine (SUCr) level, and spot urine osmolality (SUOsm) were measured. In addition, from the 24-h urine samples collected, 24-h urine protein (24HUP) level, 24-h urine creatinine (24HUCr) level, and 24-h urine osmolality (24HUOsm) were measured. Hemoglobin (Hb) and serum creatinine (SCr) levels were measured from the blood samples collected. Other tests performed on the collected blood samples were HIV screening test, fasting blood glucose, and fasting serum lipid profile (FSLP) [total cholesterol, triglyceride (TG), high-density lipoprotein (HDL) cholesterol. and low-density lipoprotein (LDL) cholesterol]. Osmolality was determined by the freezing point depression method using the Precision Osmette 5002 osmometer, creatinine by modified Jeff's method, and protein by a photometric method. Creatinine clearance (ClCr) and SUCr/osmolality (SUCOR) were also determined.

The potential predictors of 24HUOsm evaluated were BMI, CD4 cell count, SCr, SUOsm, SUP, SUCr, 24HUP, 24HUCr, SUCOR, serum cholesterol, serum TG, serum HDL, serum LDL, and ClCr.

Statistical analysis

The data were analyzed using SPSS version 17.0 (SPSS Int. Chicago, II, USA). The distribution and

characterization of clinical and laboratory features among the HIV-positive subjects with different levels of 24HUOsm were analyzed using cross tabulation, whereas statistical significance of association of these variables with 24HUOsm changes was determined using student *t*-test. Multivariate linear regression analyses were used to determine the strength of variables to predict dilute urine and concentrated urine. All tests were two-tailed. $P \le 0.05$ was taken as statistically significant.

Definition of terms

- Normal urine osmolality: 24HUOsm 300–750 mOsm/kgH₂O
- Dilute urine: 24HUOsm <300 mOsm/kgH₂O
- Concentrated urine: 24HUOsm >750 mOsm/kgH₂O.

RESULTS

938

The mean 24HUOsm was $564 \pm 501 \text{ mOsm/kgH}_2\text{O}$ and the mean SUOsm $464 \pm 271 \text{ mOsm/kgH}_2\text{O}$. The mean values of other variables are shown in Table 1. Dilute

Table 1: Characteristics of variables in HIV subjects					
Variables (mean±SD)	HIV subjects				
Body Mass Index (kg/m ²)	26.2±5.4				
Hemoglobin (g/dl)	11.2±1.8				
CD4 cells	416±209				
SUOsm (mOsm/kgH,O)	464±271				
Spot urine Protein (mg/dl)	11.8 <mark>9±19.13</mark>				
Spot urine creatinine (mg/dl)	137.21±98.47				
24-h urine protein (g)	0.187±0.290				
24-h urine creatinine (mg)	1507±781				
24HUOsm (mOsm)	564±501				
SUCOR (mg/dl/mOsm/kgH ₂ O)	0.422±0.486				
Cholesterol (mmol/l)	4.26±0.90				
Triglyceride (mmol/l)	1.23±0.37				
HDL (mmol/l)	1.18±0.39				
LDL (mmol/l)	2.05±0.58				
Creatinine clearance (mls/min)	91.42±22.98				
SD=standard deviation, SUOsm=spot urine osmolality,					

24UOsm=24-h urine osmolality, SUCOR=spot urine creatinine/ osmolality ratio, HDL=high density lipoprotein cholesterol, LDL=low density lipoprotein cholesterol, urine was present in 242 (64.5%) of the HIV subjects, whereas concentrated urine was observed in 22 (5.9%).

There was a significant association between BMI and urine osmolality, P = 0.016. Out of the 242 subjects who had dilute urine (24HUOsm < 300 Osm/kgH₂O), 98 (40.5%) had BMI 18.5–24.9 kg/m², 86 (35.5%) BMI 25.0–29.9 kg/m², 12 (5.0%) BMI < 18.5 kg/m², 86 (35.5%) BMI 25–29.9 kg/m², whereas 46 (19.0%) had BMI \geq 30 kg/m². This showed that the prevalence of dilute urine (<300 mOsm/kgH₂H) was high among the subjects whose weight were normal and declined with underweight but increased with obese subjects [Table 2].

[4CD] cells count did not show any significant association with 24HUOsm, P = 0.132 [Table 2].

There was a significant association between ClCr and 24HUOsm, P = 0.021. In total, 242 subjects had dilute urine (24HUOsm < 300). Out of this number, 124 (51.5%) had ClCr ≥ 90 mls/min, 88 (36.5%) ClCr 50–89 mls/min, whereas 29 (12.0%) had ClCr 30–59 mls/min. This showed that the prevalence of dilute urine declined as ClCr declined. In total, 22 subjects had concentrated urine (24HUOsm > 750 mOsm/kgH₂O). Out of this number, 14 (63.6%) had ClCr ≥ 90 mls/min, 6 (27.3%) had ClCr 60–89 mls/min, This demonstrated that the prevalence of concentrated urine declined as ClCr 30–59 mls/min. This demonstrated that the prevalence of concentrated urine declined as ClCr declined [Table 2].

Hb has no significant association with 24HUOsm, P = 0.196S [Table 2].

The association between serum cholesterol and 24HUOsm was significant, P = 0.008. In total, 242 subjects had dilute urine. Of these, 220 (90.9%) had a desirable serum cholesterol level, 18 (7.4%) had a borderline serum cholesterol level, whereas 4 (1.7%) had a high serum cholesterol level. This showed that the prevalence of dilute urine decreased as the serum cholesterol level increased. In total, 22 subjects had concentrated urine. Out of this number, 18 (81.3%)

Table 2: Distribution a	ind characterization	of selected	l risk fac	tors at different level	ls of 24-h urin	e osmolality	y in
treatment-naïve HIV-positive subjects (<i>n</i> =375)							

Variables	24-h urine osm	24-h urine osmolality Levels (mOsm/kgH,O) (no/%)				Р
	<300	300-750	>750			
BMI (kg/m ²) <18.5	12 (3.2%)	10 (2.7%)	2((0.5%)	15.584	0.009	0.016
18.5-24.9	98 (26.1%)	28 (7.4%)	2 (0.5%)			
25.0-29.9	86 (22.9%)	47 (12.5%)	12 (3.2%)			
≥30	46 (12.2%)	27 (7.2%)	6 (1.6%)			
CD4 (cells/ml) <200	29 (7.7%)	17 (4.5%)	0 (0.0%)	4.053	0.036	0.132
≥200	213 (56.8%)	94 (25.1%)	22 (5.9%)			
Hb (g/dl) ≥12.0	72 (19.1%)	44 (11.7%)	6 (1.6%)	8.619	0.210	0.195
10.0-11.9	114 (30.3%)	44 (11.7%)	12 (2.3%)			

Table 2: Contd						
Variables	24-h urine osn	24-h urine osmolality Levels (mOsm/kgH,O) (no/%)				Р
	<300	300-750	>750			
<7.0	2 (0.5%)	4 (1.1%)	0 (0.0%)			
ClCr (mls/min) ≥90 mls/min	124 (33.1%)	61 (16.3%)	14 (3.7%)	11.522	0.006	0.021
60-89	88 (23.5%)	49 (13.1%)	6 (1.6%)			
30-59	29 (7.7%)	2 (0.5%)	2 (0.5%)			
24HUP <0.300 g	209 (55.7%)	92 (24.5%)	20 (5.3%)	6.943	0.295	0.316
≥0.300 g	32 (8.5%)	20 (5.3%)	2 (0.5%)			
FSLP (mmol/l)						
Chol T Des (<5.2	220 (58.5%)	96 (25.5%)	18 (4.8%)	13.655	0.025	0.008
BorderL (5.2-6.2)	18 (4.8%)	16 (4.3%)	2 (0.5%)			
High (>6.2)	2 (0.5%)	4 (1.1%)	0 (0.0%)			
LDL Des (<2.6)	201 (53.6%)	92 (24.5%)	16 (4.3%)	1.591	0.488	0.451
BorderL (2.6-4.1)	40 (10.7%)	20 (5.3%)	6 (1.6%)			
HDL Low (<1)	77 (20.5%)	46 (12.2%)	8 (2.1%)	2.912	0.237	0.233
High (≥ 1)	165 (43.9%)	66 (17.6%)	14 (3.7%)			
TG Des<1.7)	216 (57.4%)	104 (27.7%)	18 (4.8%)	23.390	0.93	0.001
BorderL (1.7-2.2)	22 (5.9%)	6 (1.5%)	1 (0.3%)			
High (>2.2)	4 (1.1%)	2 (0.5%)	2 (0.5%)			

Anyabolu, et al.: Urine osmolality in treatment-naïve HIV-positive subjects

LHR=Likelihood ratio; BMI=Body mass index; Waist Circ=Waist circumference; ClCr=Creatinine clearance, 24HUP=24-h urine protein; FSLP=Fasting serum lipid profile; CholT=Total cholesterol, Des=Desirable; BorderL=Borderline; LDL=Low density lipoprotein cholesterol; HDL=High density lipoprotein cholesterol; TG=Triglyceride

Table 3: Correlation of 24HUOsm with selected variables							
in treatment-naïve HIV-positive subjects (<i>n</i> =375)							
Variables	Correlation coefficient (r)	Р					
Body mass index	0.193	< 0.001					
CD4 cells count	0.073	0.160					
Spot urine protein	0.067	0.196					
Spot urine creatinine	0.178	0.001					
24-h urine protein	-0.036	0.484					
24-h urine creatinine	0.128	0.013					
24-h urine volume	-0.052	0.313					
SUCOR	0.009	0.865					
Serum creatinine	0.048	0.356					
Serum cholesterol (total)	0.039	0.453					
Serum triglyceride	0.075	0.145					
Serum HDL	-0.151	0.003					
Serum LDL	0.034	0.511					
Creatinine clearance	0.082	0.112					
SUCOR=Spot urine creatinine osmolality ratio: HDL=High							

SUCOR=Spot urine creatinine osmolality ratio; HDL=High density lipoprotein cholesterol; LDL=Low density lipoprotein cholesterol

Table 4: Multivariate linear regression of variables
with 24HUOsm <300mOsm/kgH ₂ O in treatment-naïve
HIV-positive subjects (n=242)

	1	3		/
Variables	Beta	t	Р	95% CI
Body mass index	0.092	1.002	0.317	-1.272-3.806
Spot urine creatinine	0.098	0.956	0.340	-0.084 - 0.241
24-h urine creatinine	0.019	0.224	0.824	-14.037-17.609
HDL	0.219	2.988	0.003	0.338-1.648
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CI=Confidence Interval; HDL=High density lipoprotein cholesterol

had a desirable serum cholesterol level, 2 (9.1%) had a borderline serum cholesterol level, and 2 (9.1%) had a high serum cholesterol level. This also showed that the prevalence of concentrated urine declined as serum cholesterol level increased [Table 2].

No significant association was found between 24HUOsm and serum LDL level (P = 0.451) on one hand, and between 24HUOsm and serum HDL level (P = 0.233) on the other [Table 2].

There was a significant association between serum TG level and 24HUOsm, P = 0.001. In total, 242 subjects had dilute urine. Out of this number, 216 (89.3%) had a desirable serum TG level, 22 (9.1%) had borderline serum TG level, whereas 4 (1.7%) had a high serum TG level. This demonstrated that the prevalence of dilute urine declined as serum TG level increased. Twenty-two subjects had concentrated urine. Out of this number, 18 (81.8%) had a desirable serum TG level, whereas 2 (8.1%) had a high serum TG level, indicating that the prevalence of concentrated urine declined as the level of serum TG increased [Table 2].

There was no significant association between 24HUP and 24HUOsm, P = 0.316 [Table 2].

Only serum HDL predicted dilute urine, whereas BMI, SUP, 24HUP, SUCr, serum HDL, and CD4 cells count predicted concentrated urine [Tables 3-5].

Table 5: Multivariate linear regression of variables with 24HOsm >750mOsm/kgH ₂ O in treatment-naïve								
$\frac{\text{HIV-positive Subjects } (n=22)}{\text{Variables}}$								
Body mass index	0.132	11617278.0	< 0.001	1.364-1.364				
Waist	0.576	44309220.3	< 0.001	2.773-2.773				
circumference								
Spot urine protein	0.620	95528492.3	< 0.001	6.707-6.707				
24-h urine protein	-0.056	84434467.8	< 0.001	5,607-5.607				
Spot urine creatinine	-0.837	-102385156.7	< 0.001	-0.497-0.497				
HDL	0.253	27950347.4	< 0.001	1.941-1.941				
CD4 cells count	-0.208	-37442703.3	< 0.001	-0.110-0.110				
Underweight=Body mass index <18.5. CI=Confidence interval;								

HDL=High density lipoprotein cholesterol

DISCUSSION

The prevalence of dilute urine, 64.5%, found in this study was high compared to 8.1% documented by Yeh *et al.*^[11] The difference, perhaps, could be explained, in part, by the study design. Our study subjects were treatment-naïve HIV-positive in an HIV center, whereas theirs study subjects were non-HIV in a general population. In addition, our study was conducted in Nigeria, whereas theirs in USA. A study, however, noted that high baseline urine osmolality could contribute to kidney disease progression.^[3] A very high prevalence of dilute urine apparently supports the fact that our study subjects were unlikely to progress fast if they were chronic kidney disease patients.

In this study, the prevalence of concentrated urine (5.9%) we observed was similar to 3.1% reported in another study,^[11] despite the differences in climatic background, race, levels of industrialization, and HIV status of the study population.

Our study showed that BMI has a significant association with dilute urine, similar to the finding documented in one study.^[11] Our study demonstrated that the prevalence of dilute urine was high among the subjects whose weights were normal, declined with underweight but increased with obese subjects. Urine osmolality, in healthy and some disease states, may be influenced by differences in the excretion of solutes such as sodium, which is found to vary according to the BMI.^[15] This implies that dilute urine is likely to occur more readily in subjects who are underweight and less in obese subjects. However, our observation showed the opposite. Perhaps, among treatment-naïve HIV subjects, those who were obese were more disposed to over-hydration, compared to those who were underweight who might have under-hydration, accounting for the observed difference in the prevalence of dilute urine.

We found that ClCr was a significantly associated with dilute urine. It was also observed that the prevalence

of dilute urine declined as ClCr declined. Some studies demonstrated that urine osmolality correlated positively with ClCr (r = 0.60, P < 0.01).^[3] They further showed that increased hydration consequently led to dilute urine that lowered vasopressin secretion that significantly lowered ClCr.^[3,16-18] Our finding is in keeping with the observation that concentrated urine was associated with declining kidney function,^[3] implying that the prevalence of dilute urine declined as ClCr declined, as found in our study.

This study showed that both serum cholesterol and serum TG levels have a significant association with dilute urine. From the literature search, we could not find any study that assessed the influence of dilute urine on these lipids. Both high serum cholesterol and high serum TG levels, in isolation or in combination with low serum HDL, as components of dyslipidemia, are known risk factors of both micro- and microvasculopathy that might be largely systemic but could be regional.^[19,20] Ishemic renal states resulting from vasculopthy usually affect tubular functions and generate dilute urine. This tends to suggest that our study subjects might have some degree of renal compromise associated with dilute urine and dyslipidemia.

Conversely, ClCr, serum cholesterol, and serum TG levels have a significant association with concentrated urine, as shown in our study. This showed that abnormalities of serum lipids were influenced by high urine osmolality. Studies were sparse, from the literature search, that evaluated the impact of serum lipids on urine osmolality. One study conducted in a general out-patient clinic population did not find any correlation between 24HUOSm and ClCr. in contrast to our finding.^[21] Similar to our finding, a study has reported a high correlation between urine osmolality and ClCr.^[3] Glomerular diseases with little or no involvement of the interstitial compartments might be associated with dyslipidemia, proteinuria, and passage of reasonable amounts of solutes in urine. This could, perhaps, in part, account for the high osmolar urine associated with high serum cholesterol and high serum TG levels, found in our study subjects. This observation tends to suggest that these treatment-naïve HIV subjects might have some degree of proteinuric, glomerular diseases, and some level of renal function impairment.

Our study noted that only serum HDL was the predictor of dilute urine. The literature search did not reveal any study on serum HDL as a potential predictor of dilute urine. However, low serum HDL has been documented as an independent risk factor for the development of microvascular disease affecting the kidney.^[19,20] Renal microvasculopathy might involve the interstitial and tubular compartments that might lead to the loss of renal concentrating ability and subsequently to the production of dilute urine.

BMI, SUP, 24HUP, SUCr, serum HDL, and CD4 cell count were the predictors of concentrated urine. However, co-linearity variance was skewed, as the subjects on which the attributes studied were small in number.

The presence of protein in urine influences its osmolality. As a result, the higher the level of protein in urine, the higher is the osmolality of that urine sample. This could explain the direct positive correlation between 24HUP and 24HUOsm found in this study, in treatment-naïve HIV subjects.

Urine creatinine is also used for the evaluation of the concentration of urine. The positive correlation between 24HUOsm and urine creatinine in our study is in agreement with a study that found that both could be used interchangeably to evaluate urine concentration and dilution with some measure of agreement.^[11] However, we did not evaluate the validity of using 24HUOsm and 24HUCr for estimating dilute and concentrated urine.

In this study, we observed a positive direct correlation between CD4 cells count and high urine osmolality. Low CD4 cell count and underweight are usually found in wasting disease, another name by which HIV infection is known. This condition is frequently associated with a salt-losing syndrome.^[22] This might explain the direct positive correlation between CD4 cells count and concentrated urine found in our study subjects.

CONCLUSION

The prevalence of dilute urine was high among treatment-naïve HIV subjects. Abnormalities of serum lipids, renal function, and weight were common in treatment-naïve HIV subjects who had dilute urine. There is a need for clinicians to routinely assess urine osmolality and further diagnose for dyslipidemia, renal function impairment, and abnormal weight in HIV subjects at the early stage of the infection.

Limitations

Serum copeptin would have given a better objective assessment of vasopressin in relation to urine osmolality in this study, if copeptin was measured. Plasma osmolality and urine sodium level were not measured; they would have contributed in categorizing dilute urine. A larger study population would have averted the skewed co-linearity variance that voided the potential predictors of concentrated urine in this study.

What is already known about this topic

- Urine osmolality is not commonly evaluated in routine clinical practice
- Many environmental, physiologic, and disease conditions affect urine osmolality
- Some associated factors of dilute urine and concentrated urine have been identified; concentrated urine is associated with the progression of chronic kidney disease.

What this study adds to knowledge

- The prevalence of dilute urine was high, whereas that of concentrated urine was low in treatment-naïve HIV-positive subjects in Nigeria
- Dyslipidemia, renal function impairment, and abnormal weight were common in treatment-naïve HIV subjects who have dilute urine
- There is a need for clinicians to routinely assess urine osmolality and further diagnose for dyslipidemia, renal function impairment, and abnormal weight in HIV subjects at the early stage of the infection.

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Conflicts of interest

There are no conflicts of interest.

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942