Relationship between phosphate reabsorption, age, tenofovir and bone mineral density

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Background

- The functional capacity of renal tubules to reabsorb phosphate declines with age (1).
- Renal tubular dysfunction is associated with combination antiretroviral therapy (cART), and the risk is increased when tenofovir (TDF) is co-prescribed with boosted protease inhibitors (PI) (2).
- Renal tubular dysfunction causes impaired reabsorption of phosphate in the proximal tubule, and in its most extreme form, Fanconi syndrome (3) which may be accompanied by osteomalacia and fractures.
- The extent to which age-related phosphate reabsorption is affected by TDF exposure and its effects on bone mineral density (BMD) in HIV-positive patients are unknown.

Aims

- **To investigate the effect of age on phosphate reabsorption in HIV-positive patients.**
- **To examine the effect of TDF exposure on age-related reductions in phosphate reabsorption.**
- **To investigate the factors associated with $TmPO_4$/GFR, a measure of phosphate reabsorption, and the relationship between $TmPO_4$/GFR, age, TDF exposure and BMD.**

Methods

- HIV-positive men attending the HIV outpatient clinic at Brighton, UK, were randomly selected to participate in a study investigating BMD and osteoporosis.
- The following data were collected at baseline:
  - Demographic and HIV-related details, including cART history and risk factors for low BMD.
  - Serum and urine creatinine (Cr), serum and urine phosphate (PO4), serum carboxy-terminal collagen crosslinks (CTX) to measure bone resorption and type 1 procollagen (P1NP) for bone formation were obtained from fasted, paired blood and urine samples.
  - Absolute BMD (g/cm^2) at the lumbar spine, the non-dominant total hip and the non-dominant femoral neck was measured using the GE Healthcare Lunar iDXA bone densitometer.
  - Phosphate wasting was assessed using maximum threshold for phosphate reabsorption ($TmPO_4$/GFR), based on a formula derived from the normogram by Walton and Bijvoet (4-6), where if $FePO_4$ = (urine PO^4/serum Cr)/(serum PO_4/urine Cr), then $TmPO_4$/GFR = (1-$FePO_4$)*serum PO_4 if (1-$FePO_4$) ≤ 0.86 and $TmPO_4$/GFR=0.3*(1-$FePO_4$)/[1-{0.8*(1-$FePO_4$)}]*serum PO_4 if (1-$FePO_4$) > 0.86.
- Correlation coefficients were used to evaluate relationships between variables and multivariate linear regression was used to investigate the relationship between $TmPO_4$/GFR and age, BMD and bone turnover, and all factors significant at the 10% level in univariable analysis were considered for entry into the model using a forward stepwise approach.

Results

- 411 men were included in the analyses (Table 1).
- 269 (65.5%) had $TmPO_4$/GFR <0.8 mmol/L, indicative of impaired phosphate reabsorption (6).

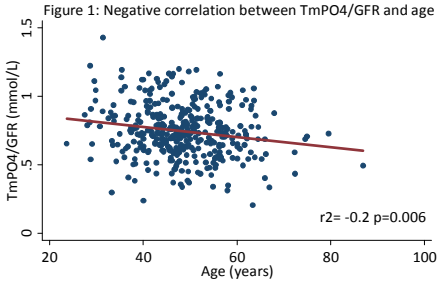
Table 1: Baseline demographics

	Total N=411		Total N=411
Age years, mean (SD)	47 (9.5)	On TDF at recruitment n (%)	347 (79)
White, n (%)	387 (94.2)	eGFR (CKD-Epi), mean (SD)	93.5 (17.1)
MSM, n (%)	382 (92.9)	$TmPO_4$/GFR, mmol/L, mean (SD)	0.8 (0.2)
Hepatitis B, n (%)	17 (4.0)	PTH, ng/L, mean (SD)	52.3 (21.4)
Hepatitis C, n (%)	58 (14.1)	ALP, IU/L, mean (SD)	82.0 (25.2)
Years since diagnosis, median(IQR)	9.3 (4.8, 15)	CTX, ng/ml, median (IQR)	2.1 (0.9, 5.3)
HIV clinical stage ≥3, n (%)	114 (29.4)	P1NP, ng/ml, median (IQR)	13.5 (5.6, 33.5)
CD4 nadir, median (IQR)	186 (95, 274)	BMD, g/cm^2, mean (SD)	
HIV RNA VL <40 copies/mL, n (%)	357 (86.9)	Lumbar spine	1.14 (0.16)
On cART at recruitment, n (%)	387 (88)	Non-dominant total hip	1.00 (0.14)
Years on cART, median (IQR)	9.5 (5.0, 15.5)	Non-dominant femoral neck	0.95 (0.14)

ALP: alkaline phosphatase; BMD: bone mineral density; cART: combination antiretroviral therapy; CKD-Epi: Chronic Kidney Disease Epidemiology Collaboration; CTX: carboxy-terminal collagen crosslinks; eGFR: estimated glomerular filtration rate; MSM: men who have sex with men; P1NP: type 1 procollagen; PI: protease inhibitor; PTH: parathyroid hormone; TDF: tenofovir; $TMPO_4$/GFR: renal threshold phosphate concentration; VL: viral load

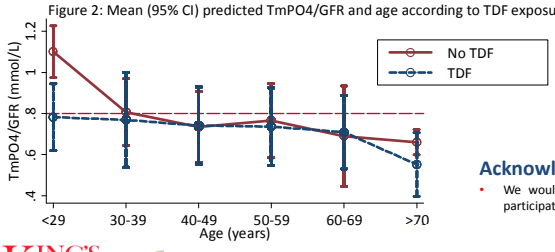
Effect of age on phosphate reabsorption

Figure 1: Negative correlation between TmPO4/GFR and age



- Figure 1 shows the relationship between $TmPO_4$ and age, which shows that there was a significant negative correlation (r^2=-0.2, p=0.006).
- Figure 2 shows the relationship between $TmPO_4$/GFR and age according to TDF exposure (TDF: n=347, cART without TDF: n=40, no cART: n=24).
- Among subjects aged 30 - 70 years on cART, $TmPO_4$/GFR did not differ among those exposed versus those not exposed to TDF.

Figure 2: Mean (95% CI) predicted TmPO4/GFR and age according to TDF exposure



Factors associated with $TmPO_4$/GFR

- In univariate analyses, there was no association with ethnicity, hepatitis B, diabetes, hypertension or CTX.
- P1NP was borderline significant (Table 2) but this small effect was no longer present after adjustment for potential confounders.

Table 2: Age and 25(OH) vitamin D are associated with $TmPO_4$/GFR

	Crude β (95%CI)	P-value	Adjusted β (95% CI)*	P-value
Age, years, per 10 year increase	**-0.05 (-0.07, -0.03)**	**<0.001**	**-0.03 (-0.05, -0.01)**	**0.003**
Prior AIDS defining condition	-0.05 (-0.1, -0.01)	0.01	-0.02 (-0.06, 0.03)	0.4
Nadir CD4 count, per 10 cells/μl increase	0.003 (0.001, 0.004)	<0.001	0.001 (-0.0004, 0.003)	0.2
Current log HIV RNA VL	0.06 (0.03, 0.09)	0.000	0.02 (-0.01, 0.05)	0.2
On TDF at recruitment	-0.05 (-0.1, -0.01)	0.01	-0.04 (-0.09, 0.004)	0.07
On PI at recruitment	-0.05 (-0.09, -0.01)	0.01	-0.04 (-0.08, 0.002)	0.07
25(OH) vitamin D	**0.001 (0.0002, 0.002)**	**0.02**	**0.001 (0.0001, 0.002)**	**0.03**
P1NP	0.0003 (0.000, 0.001)	0.047	0.0003 (-0.0001, 0.001)	0.1

25(OH) vitamin D: total plasma 25-0H vitamin D (nmol/L); AIDS: acquired immune deficiency disease; CD4: CD4 positive cell count per 10 cells/μl increase; PI: protease inhibitor; PTH: parathyroid hormone (ng/L); TDF: tenofovir; $TMPO_4$/GFR: renal threshold phosphate concentration (mmol/L)
*Also adjusted for hepatitis C status (hepatitis C antibody positive), smoking, body mass index and P1NP

Relationship between $TmPO_4$/GFR, age and BMD

- We hypothesized that lower $TmPO_4$/GFR would be associated with lower BMD (i.e., a positive correlation).
- We observed no correlation between $TmPO_4$/GFR and BMD spine or BMD total hip in the overall cohort (Fig 3a/b); in those > 50 years, a weak negative correlation was observed between $TmPO_4$/GFR and BMD spine (Fig 3a).
- After adjustment for potential confounders, the negative association between BMD spine and TmPO4/GFR remained significant in patients > 50 years age (Table 3).

Figure 3a: Relationship between TmPO4/GFR and BMD spine

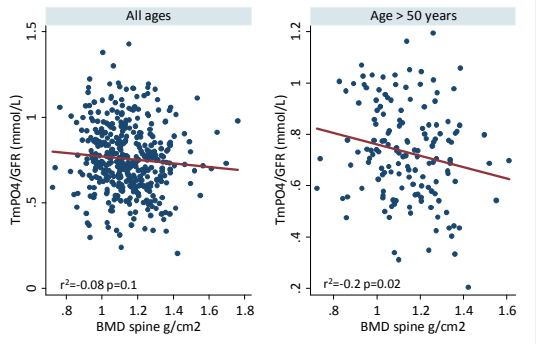


Figure 3b: Relationship between TmPO4/GFR and BMD hip

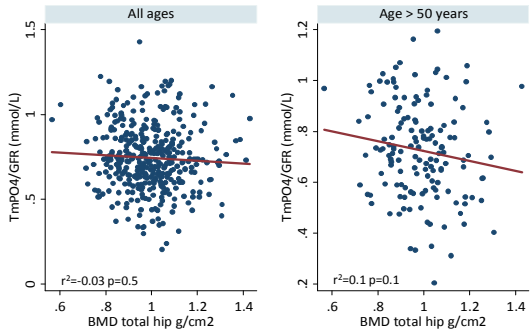


Table 3: Multivariable regression of the association between BMD spine (g/cm^2) and $TmPO_4$/GFR in those >50 years

	Age > 50 years N=148			
	Crude β (95%CI)	**P-value**	**Adjusted β (95% CI)***	**P-value**
$TmPO_4$/GFR	**-0.2 (-0.3, -0.03)**	**0.02**	**-0.2 (-0.4, -0.08)**	**0.003**
Years since diagnosis	**-0.005 (-0.009, -0.002)**	**0.003**	**-0.005 (-0.008, -0.001)**	**0.01**
BMI	**0.01 (0.006, 0.02)**	**<0.001**	**0.1 (0.005, 0.02)**	**<0.001**

BMI: body mass index; $TMPO_4$/GFR: renal threshold phosphate concentration (mmol/L)
*Also adjusted for current PI use, estimated GFR (eGFR) using CKD-Epi and ethnicity

Conclusions

- **In HIV-positive men, reduced phosphate reabsorption was present in two-thirds of patients.**
- **$TmPO_4$/GFR declined with age but was not significantly associated with TDF exposure, increased bone resorption/formation, or reduced BMD.**
- **Our results suggest that in patients stable on antiretroviral therapy, $TmPO_4$/GFR is of limited use in identifying patients at increased risk of bone loss.**

Acknowledgements

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