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Association Between Post-Transplant Vitamin D, Metabolic Syndrome, and Post Transplant Diabetes Mellitus

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Abstract:	Background Associations between 25-hydroxyvitamin D (25(OH)D) deficiency and diabetes have been observed in the general population, but are less delineated in kidney transplant recipients (KTR), especially in the context of highly-prevalent metabolic syndrome (MetS) features in KTR. We hypothesized that vitamin D deficiency may present greater risk in KTR in those with greater burden of MetS features. Methods We retrospectively evaluated 1792 KTR with no treated diabetes at transplant between 1998 and 2018. Vitamin D was measured at ≥3 months post transplant. MetS features were defined by National Cholesterol Education Program, Adult Treatment Panel III (NCEP-ATP III) criteria. The primary outcome was treated post-transplant diabetes mellitus (PTDM) incidence. Results In 1792 non-diabetic KTR followed for 10956 patient-years, 237 patients developed PTDM. For KTR meeting NCEP-ATP-III criteria, with 4th-quartile 25(OH)D, there were 1.5 new diagnoses per 100 patient-years, versus 4.2 events per 100 patient-years in KTR with 1st-quartile 25(OH)D ($p<0.001$). In multivariate survival regression, vitamin D was, accounting for individual NCEP-ATP-III criteria, associated with PTDM (HR 0.93 per 10 nmol/mL 25(OH)D, $p=0.007$) independently of fasting blood sugar and HbA1c. In marginal effects analysis, MetS impact on PTDM increased as serum 25(OH)D

	levels decreased. Conclusions Our study suggests that decreased 25(OH)D is associated with increased PTDM, and this marginal impact worsens as KTR have an increased burden of MetS.
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Key point #3: as follow-up to "Key Points: Please state the 2-3 key points of the article. The responses included here will be included with your final published paper. The key points should be complete statements and not duplications of your keywords or index terms. At least two key points are	Higher vitamin D-25 levels blunt this association; in patients with high metabolic syndrome burden, vitamin D supplementation may be warranted.

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Association Between Post-Transplant Vitamin D, Metabolic Syndrome, and Post Transplant Diabetes Mellitus

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Abstract

Background

Associations between 25-hydroxyvitamin D (25(OH)D) deficiency and diabetes have been observed in the general population, but are less delineated in kidney transplant recipients (KTR), especially in the context of highly-prevalent metabolic syndrome (MetS) features in KTR. We hypothesized that vitamin D deficiency may present greater risk in KTR in those with greater burden of MetS features.

Methods

We retrospectively evaluated 1792 KTR with no treated diabetes at transplant between 1998 and 2018. Vitamin D was measured at ≥3 months post transplant. MetS features were defined by National Cholesterol Education Program, Adult Treatment Panel III (NCEP-ATP III) criteria. The primary outcome was treated post-transplant diabetes mellitus (PTDM) incidence.

Results

In 1792 non-diabetic KTR followed for 10956 patient-years, 237 patients developed PTDM. For KTR meeting NCEP-ATP-III criteria, with 4th-quartile 25(OH)D, there were 1.5 new diagnoses per 100 patient-years, versus 4.2 events per 100 patient-years in KTR with 1st-quartile 25(OH)D ($p<0.001$). In multivariate survival regression, vitamin D was, accounting for individual NCEP-ATP-III criteria, associated with PTDM (HR 0.93 per 10 nmol/mL 25(OH)D, $p=0.007$) independently of fasting blood sugar and HbA1c. In marginal effects analysis, MetS impact on PTDM increased as serum 25(OH)D levels decreased.

Conclusions

Our study suggests that decreased 25(OH)D is associated with increased PTDM, and this marginal impact worsens as KTR have an increased burden of MetS.

Introduction

Kidney transplantation is the gold standard treatment for end-stage kidney disease (ESKD), offering improved quality of life and increased survival rates for patients [1,2]. However, despite advancements in transplantation techniques and immunosuppressive therapies, post-transplant diabetes mellitus (PTDM) remains a significant cause of morbidity and mortality among kidney transplant recipients (KTR) [3].

Among the potential contributors to adverse outcomes in KTRs, 25-hydroxyvitamin D (25(OH)D) deficiency has emerged as an interesting, potentially modifiable candidate. 25(OH)D plays a pivotal role in various physiological processes beyond its classical role in calcium homeostasis, including immune modulation, endothelial function, and regulation of the renin-angiotensin-aldosterone system [4]. 25(OH)D deficiency is highly prevalent among KTRs, likely due to impaired 25(OH)D synthesis, increased catabolism, and limited exposure to sunlight [5].

The association between 25(OH)D deficiency, metabolic syndrome (MetS), and diabetes incidence has been extensively studied in the general population, highlighting its potential role in the development and progression of MetS and subsequent PTDM [6,7]. Data also exist on 25(OH)D deficiency and MetS components on PTDM and graft survival individually of each other [8,9]. However, limited data exist specifically addressing the impact of post-transplant vitamin D status and the co-occurrence of MetS features, and their influence on the incidence of post-transplant diabetes mellitus (PTDM) in KTRs.

The primary objective of this retrospective cohort study was hence to examine the associations between post-transplant 25(OH)D and co-occurence of features of MetS, and marginal impacts on a primary outcome of post-transplant diabetes mellitus (PTDM) incidence. This study was performed in a KTR population that was free of treated diabetes at the time of transplant and at the time of 25(OH)D measurement post-transplant, and with the prevalence of MetS features being determined at the time of 25(OH)D measurement.

Materials and Methods

2.1 Study sample and follow up

The study was conducted at St. Michael's Hospital (SMH), which is an urban university-affiliated medical-surgical center specializing in kidney transplantation. The hospital actively monitors a cohort of over 1700 kidney transplant recipients (KTR) and performs approximately 130 single-organ kidney transplants on adult patients each year. Follow-up visits are scheduled at regular intervals post-transplant, ranging from weekly to annually, during which trained staff record anthropometric measurements and resting blood pressure. Patients self-report their sex and ethnicity, and laboratory tests were conducted close to each clinic visit, with additional testing performed on a separate, more frequent schedule. The estimated glomerular filtration rate (eGFR) is calculated using the Modification of Diet in Renal Disease-7 equation [10].

In this study, we identified a cohort not treated for diabetes who were transplanted and followed at SMH between September 1, 1998 and December 31, 2018 who had a serum 25-hydroxyvitamin D (25(OH)D) level drawn at or beyond 3 months of transplant, extracting records from the SMH clinical patient data management system. No specific or centralized lab was used for 25(OH)D blood tests. No other exclusion criteria were applied. Clinical data relevant to the diagnosis of MetS were cross-sectionally obtained from time points closest to vitamin D-25 measurement. Followup for PTDM incidence and PTDM-free survival began at the time of vitamin D-25 measurement. Patients with formally diagnosed diabetes (diabetic nephropathy) or receiving treatment with oral hypoglycemics or insulin, at the time of transplant or at the time of vitamin D-25 measurement, were excluded from the study.

The study obtained approval from the institutional research ethics board (REB-10-204). As this study involved a retrospective review of clinic data pertaining to a prevalent KTR population, individual informed consent was not obtained. The study was conducted in accordance with the ethical principles outlined in the 2000 Declaration of Helsinki and the 2008 Declaration of Istanbul.

2.2 Patient Evaluation and Data Collection

Post-transplant diabetes mellitus (PTDM) diagnoses were regularly recorded through transcriptions from local and citywide hospital databases as part of routine patient care. In cases where additional information was needed, interviews with patients and their family physicians were conducted to supplement the data. The initiation or use of medications, including oral hypoglycemics and insulin was longitudinally referenced through chart review and/or patient data management systems at SMH, for determining PTDM-free survival.

In this study, patients were classified as having MetS on the basis of the NCEP-ATP-III definition [11]. According to the National Cholesterol Education Program Adult Treatment Panel III (NCEP ATP III), MetS is considered present when three or more of the following five criteria are met:

1. Waist circumference exceeding 40 inches (102 cm) for men or 35 inches (88 cm) for women.
2. Blood pressure higher than 130/85 mmHg.
3. Fasting triglyceride (TG) level exceeding 150 mg/dL (1.69 mmol/L) or TG-lowering medication use.
4. Fasting high-density lipoprotein (HDL) cholesterol level below 40 mg/dL for men or 50 mg/dL for women (1.03 and 1.29 mmol/L respectively) or lipid-lowering medication use.
5. Fasting blood glucose level over 100 mg/dL (5.6 mmol/L) or hypoglycemic medication use.

Fulfillment of these criteria were considered on the basis of patient status at the relevant blood draws and encounters nearest to the 25(OH)D bloodwork date. 25(OH)D in this study is analyzed as a continuous and categorized variable. When categorized, cutoffs for vitamin D subgroups were based on quartiles or median.

2.3 Statistical analysis

The statistical analyses were conducted using R, version 4.2.0. Reported data are presented as mean and SD for numeric variables or mean and percentage for categorical variables, unless otherwise specified. Statistical significance was determined using two-tailed hypothesis tests.

Missing values in the dataset were handled by exclusion from the relevant analyses without imputation. The appropriate statistical tests were used for comparisons, including t-test, ANOVA, Chi-square test, or Cox proportional hazards analysis, depending on the nature of the data, including tests for trends in proportions for data with ordinal independent variables. Survival curves are constructed based on Kaplan-Meier estimates with corresponding p-values based on log-rank test. To assess the normality and skewness of subgroups for relevant analyses, visual inspection with skewness testing were used (not shown). In analyses depending on categorization of 25(OH)D levels, quartiles or medians have been used as we have found poor agreement in the literature on definitive levels of normal/deficient vitamin D levels.

In marginal effects analysis using Cox regression models, selected covariates were chosen based on biological and clinical relevance, including sex, age, fasting blood sugar, HbA1c at the time of 25(OH)D measurement, in addition to 25(OH)D and MetS burden as measured by the number of NCEP-ATP-III criteria. Schoenfeld residuals plots and p-values were inspected to ensure the proportional hazards assumption in uni- or multivariate Cox models. Variable measurements were derived from recordings nearest to the time of vitamin D-25 bloodwork were used exclusively in all analyses, as event incidence and risk were measured from the time of vitamin D bloodwork post-transplant, and not from the time of transplant itself.

Results

3.1 Pre and post-transplant characteristics across MetS and vitamin D status

Data were available on 2136 KTR transplanted at St. Michael's Hospital between 1998 and 2018, with 344 KTR excluded from analysis on the basis of formally diagnosed or treated diabetes prior to or at the time of 25(OH)D measurement. A total of 1792 eligible KTR were hence enrolled in the study, providing a total of 10956 years of patient follow-up. Table 1 outlines baseline patient characteristics at 3 months post-transplant. Patients are grouped into four categories for comparisons: with and without MetS, and above or below median levels of serum vitamin D-25. 191 cases of new post-bloodwork PTDM are recorded over the followup interval, for an incident rate of 10.66% (191/1792), or 1.743 events per 100 patient-years. The vitamin D-25 median in all comers was 48.0 (IQR 38.0, 68.0, mean (SD) 54.3 (27.4)). No statistically significant difference was found between groups in ANOVA with age at vitamin D bloodwork (p=0.125). Average time to bloodwork from transplant was 2.81 years. At the time of vitamin D measurement, PTH levels were notably higher in patients with below-median vitamin D (above-median 25(OH)D PTH: 11.4±11.5 vs below-median 25(OH)D PTH: 17.9±20.5, p<0.001). Random blood glucose levels were also higher in KTR with below-median 25(OH)D regardless of MetS status (above-median 25(OH)D: 6.63±3.7 mmol/L vs below-median 25(OH)D: 7.19±3.6, p<0.001).

3.2 MetS status and vitamin D status and PTDM incidence

In KTR not meeting NCEP ATP III criteria for MetS, 4th quartile 25(OH)D (high vitamin D) patients had 1.8 new diagnoses of PTDM per 100 patient-years (years post-bloodwork free of event), while 1st quartile 25(OH)D patients had 3.4 events per 100 patient-years (p=0.011, all p-values reporting chi-squared test-for-trend in proportions). In KTR with MetS, for patients with 4th quartile 25(OH)D, there were 1.5 new diagnoses per 100 patient-years of follow-up, versus 4.2 new diagnoses per 100 patient-years in KTR with 1st quartile 25(OH)D (p<0.001).

Similarly, PTDM incidence rose with increased MetS burden, categorized by the number of features of MetS carried by each KTR 3-months post-transplant. KTR with 0 NCEP-ATP-III criteria had 1.00 new diagnoses per 100 patient-years while KTR with all (5) criteria had 7.4 new diagnoses per 100 patient-years ($p<0.001$).

3.3 Features of MetS in association with PTDM incidence

In Figure 2, the 5 criteria of the NCEP ATP-III definition of MetS, as well as a sixth modifying criterion of sex were included in multiple Cox regression, as standalone criteria (top forest plot) and as continuous variables (bottom forest plot). Expressed as criteria, only waist-to-hip ratio (HR: 1.30, p=0.04) and fasting blood glucose (HR 4.36 per each mmol/L, $p<0.001$) were statistically significantly associated with PTDM-free survival. Expressed as continuous criteria, serum triglycerides were also statistically significant (HR: 1.25 per mmol/L, $p<0.001$). All criteria were also expressed as univariate Kaplan-Meier survival plots, with stratification by above/below median 25(OH)D status. In univariate plots, criteria using the waist-to-hip ratio, triglycerides, fasting glucose, and HDL were all statistically significant at $p<0.05$. 25(OH)D notably had variable effects on the hazard associated with meeting different MetS criteria in univariate KM plots. The predictive impact of each criterion was often differentially more marked in either the above-median group (e.g. HDL: Low 25(OH)D in high HDL vs low HDL p=0.75, vs. high 25(OH)D in high HDL vs low HDL p=0.004) or in the below-median group (e.g. triglycerides: Low 25(OH)D in high TG vs low TG p=0.003, vs. high 25(OH)D in high HDL vs low HDL p=0.035; p-values derived from pairwise tests from KM curves).

3.4 Kaplan-meier and multiple Cox regression for PTDM-free survival

Kaplan-Meier curves showing PTDM-free survival are shown for patients with above-median and below-median values of 25(OH)D with and without MetS (Figure 3). In KTR with MetS, there is a statistically significant difference in 5-year survival between 25(OH)D strata, with increased 25(OH)D being protective (p=0.021). In KTR without MetS, the same trend holds but falls below threshold for statistical significance (p=0.098).

3.5 Marginal effects of MetS criteria on association of Vitamin D and PTDM

In figure 4, a marginal effects plot is constructed to show the effects of increased MetS burden, represented by the number of NCEP ATP-III criteria, on the hazard ratios of low 25(OH)D for PTDM incidence. The multivariate Cox model represented also contains four other covariates selected on the basis of clinical relevance, including patient sex, age, and fasting glucose and HbA1c at the time of 25(OH)D bloodwork. Both 25(OH)D and MetS burden were statistically significantly associated with PTDM survival in the model (25(OH)D per 10 mmol/L HR: 0.93, p=0.007, NCEP-ATP-III criteria count HR: 1.31, p<0.001). In the plot, the effect of MetS burden appeared to be magnified at lower serum 25(OH)D levels.

Fasting glucose and HbA1c trended towards but did not achieve statistical significance for prognosticating PTDM-free survival, when included in this Cox model with 25(OH)D and MetS burden (p=0.653 and 0.300 respectively). In a separately conducted interaction terms analysis, the formal interaction term between 25(OH)D and metabolic syndrome burden was statistically significant (HR 1.08 [1.03-1.13], p=0.002, figure not shown).

Discussion

We had hypothesized that lower cross-sectional serum 25(OH)D measurements post-transplant would be associated with subsequent PTDM incidence, especially as the burden of metabolic syndrome increased in individual KTR. We found that low 25(OH)D levels appeared to indeed be associated with an increased relative hazard of MetS burden on PTDM survival, or vice versa.

Clinically, there is high prevalence of MetS the components thereof in pre- and post-transplant KTR [12]. Prior literature has linked MetS to PTDM, and PTDM and glucose intolerance to long-term adverse outcomes in KTR [13]; [14]; [15]. The impacts of MetS on renal injury are likely multifactorial, including insulin resistance and oxidative stress, heightened production of pro-inflammatory cytokines, augmented microvascular damage and subsequent renal ischemia, and more [16]. The interplay between MetS and CKD is complex and likely bidirectional [16]. The post-transplant milieu, especially with respect to the heavy immunosuppression and other medication burden, exposes patients to additional pathophysiology resulting in further graft injury. For example, corticosteroids increase resistance of the liver, muscles and peripheral tissues to insulin and decreases glucose uptake; calcineurin inhibition decreases intracellular nuclear transcription factors that promote insulin gene transcription [15].

However, prior to the International Consensus Guidelines in 2003 [12], there was no consistent definition for PTDM, and there has been little consistency between centers for diabetes screening pre-transplant as well. Numerous early studies on PTDM likely included individuals broadly without accounting for undetected or untreated diabetes and MetS before transplantation [12,14]. Since ICS 2003, significant advances in DM management in the general population have prompted the 3rd international PTDM Consensus Meeting in 2024 [17]. However KTR-specific evidence was recognized to be sparse, and as such the 2024 update includes opinion statements exclusively concerning optimal diagnostic methods and timing of assessments, and means of PTDM prevention. ICS 2003 and 2024 guidelines alike indicate that patients should be screened pre-transplant for MetS features as a known determinant of PTDM. The consideration of comorbid factors is an important step in the clinical assessment of patients pre-transplant, and is recommended to be used to individualize therapy and pre/post-transplant care, including potential vitamin D-25 supplementation.

The potential effects of vitamin D on insulin secretion and insulin resistance are pleiotropic and supported broadly in the laboratory and clinical literature [18]. Vitamin D receptors are expressed in pancreatic β-cells [19], and vitamin D responsive elements have been identified in human gene promoters encoding for insulin [20]. Clinically, in non-transplant populations, 25(OH)D levels are inversely correlated to DM prevalence [21], with 25(OH)D insufficiency being associated with insulin resistance and increased HbA1c [21]; [22].

In KTR specifically, the impact of MetS on PTDM has previously been investigated in smaller prospective and retrospective studies [8]. Separately, 25(OH)D deficiency has been identified as an independent risk factor of PTDM in kidney transplant [9]. The recent VITALE RCT [23] did not demonstrate improvement in non-skeletal complications of kidney transplant with vitamin D supplementation, including PTDM, nor improvement in HbA1c levels. However, MetS is a cluster of several modifiable factors outside of HbA1c; vitamin D deficiency may have marginally different impacts when comorbid with the syndrome, especially in diverse multi-ethnic cohorts [24]; [25]. Furthermore, the VITALE study considered PTDM incidence as part of a composite outcome, and was underpowered for assessment of individual components of the composite.

The clinical relevance of our findings is twofold. Firstly, our results highlight the importance of assessing and addressing vitamin D status in KTR, particularly those with MetS. Vitamin D deficiency is prevalent in this population, and our findings suggest that correcting this deficiency may be particularly crucial especially in individuals with MetS to mitigate the risk of new-onset diabetes. Routine monitoring of serum 25(OH)D levels and targeted interventions, such as supplementation, may be warranted in these patients.

Secondly, our findings underscore the potential role of 25(OH)D as a potential modulating factor in the relationship between other MetS features and diabetes after kidney transplantation. Supporting this hypothesis, multivariate analysis with an interaction term for 25(OH)D by MetS criteria indicated positive interaction with a multiplicative effect of these variables (HR 1.08 [1.03-1.13], p=0.002). However, while the interaction term supports the hypothesis, the follow-up time was not sufficient for more dedicated interactions testing especially at higher metabolic syndrome burdens, and the analysis focuses on impacts for marginal effects. Furthermore, not all MetS features appear to represent the same risk for developing PTDM. Prior study in post-transplant major adverse cardiac events have demonstrated that different components of MetS have differential impacts post-transplant MACE [24].

This cohort study is limited by its retrospective nature, and should be considered hypothesis generating rather than capable of establishing causality. The analysis, despite coming from a multi-ethnic major urban hospital system, is still single-center. Vitamin D levels were limited to cross-sectional analysis at one point post-transplant, and routine post-transplant 25(OH)D testing had not begun until 2010-2011, with many patients not receiving testing until years post-transplant, with a mean time-to-bloodwork of 2.81 years. Patients at highest risk for PTDM, especially those transplanted between 1998-2010, may have developed DM prior to vitamin D measurement, and there is potential for informative censoring given the survival bias for patients having 25(OH)D levels drawn years post-transplant. The incident rate over post-bloodwork followup was 1.743 events per 100 patient-years, which may be lower than estimates in other cohorts, and represent this informative censoring. We could not assess the concurrent presence, or impact of vitamin D supplementation at the time of blood draw specifically, only baseline 25(OH)D levels. Pretransplant indicators of diabetes predisposition such as oral glucose tolerance or impaired fasting glucose were not available for our analysis. Lastly, intermethod variability is reported for 25(OH)D assays and may reduce the generalizability of the results [26] [27].

In this study we have demonstrated the differential impact of various MetS components with PTDM, although further mechanistic learnings are limited by the retrospective nature of this study. Our results indicate a compounding association between 25(OH)D deficiency and metabolic syndrome burden for PTDM risk. Identifying individuals with both specific, high-yield components of MetS and 25(OH)D deficiency as a high-risk subgroup may enable more targeted preventive strategies. Further dedicated, prospective research is needed to elucidate the underlying mechanisms.

References:

1. Wolfe RA, Ashby VB, Milford EL, Ojo AO, Ettenger RE, Agodoa LY, et al. Comparison of mortality in all patients on dialysis, patients on dialysis awaiting transplantation, and recipients of a first cadaveric transplant. N Engl J Med. 1999 Dec 2;341(23):1725–30.

2. Abecassis M, Bartlett ST, Collins AJ, Davis CL, Delmonico FL, Friedewald JJ, et al. Kidney transplantation as primary therapy for end-stage renal disease: a National Kidney Foundation/Kidney Disease Outcomes Quality Initiative (NKF/KDOQITM) conference. Clin J Am Soc Nephrol. 2008 Mar;3(2):471–80.

3. Markell M. Clinical impact of posttransplant diabetes mellitus. Transplant Proc. 2001 Aug;33(5A Suppl):19S – 22S.

4. Mathieu C, Gysemans C, Giulietti A, Bouillon R. Vitamin D and diabetes. Diabetologia. 2005 Jul;48(7):1247–57.

5. Cianciolo G, Galassi A, Capelli I, Angelini ML, La Manna G, Cozzolino M. Vitamin D in Kidney Transplant Recipients: Mechanisms and Therapy. Am J Nephrol. 2016 May 28;43(6):397–407.

6. Pereira-Santos M, Costa PRF, Assis AMO, Santos CAST, Santos DB. Obesity and vitamin D deficiency: a systematic review and meta-analysis. Obes Rev. 2015 Apr;16(4):341–9.

7. Melguizo-Rodríguez L, Costela-Ruiz VJ, García-Recio E, De Luna-Bertos E, Ruiz C, Illescas-Montes R. Role of Vitamin D in the Metabolic Syndrome. Nutrients [Internet]. 2021 Mar 3;13(3). Available from: http://dx.doi.org/10.3390/nu13030830

8. Porrini E, Delgado P, Bigo C, Alvarez A, Cobo M, Checa MD, et al. Impact of metabolic syndrome on graft function and survival after cadaveric renal transplantation. Am J Kidney Dis. 2006 Jul;48(1):134–42.

9. Le Fur A, Fournier MC, Gillaizeau F, Masson D, Giral M, Cariou B, et al. Vitamin D deficiency is an independent risk factor for PTDM after kidney transplantation. Transpl Int. 2016 Feb;29(2):207–15.

10. Pöge U, Gerhardt T, Palmedo H, Klehr HU, Sauerbruch T, Woitas RP. MDRD equations for estimation of GFR in renal transplant recipients. Am J Transplant. 2005 Jun;5(6):1306–11.

11. Huang PL. A comprehensive definition for metabolic syndrome. Dis Model Mech. 2009 May-Jun;2(5-6):231–7.

12. Davidson J, Wilkinson A, Dantal J, Dotta F, Haller H, Hernández D, et al. New-onset diabetes after transplantation: 2003 International consensus guidelines. Proceedings of an international expert panel meeting. Barcelona, Spain, 19 February 2003. Transplantation. 2003 May 27;75(10 Suppl):SS3–24.

13. Israni AK, Snyder JJ, Skeans MA, Kasiske BL, PORT Investigators. Clinical diagnosis of metabolic syndrome: predicting new-onset diabetes, coronary heart disease, and allograft failure late after kidney transplant. Transpl Int. 2012 Jul;25(7):748–57.

14. Shivaswamy V, Boerner B, Larsen J. Post-Transplant Diabetes Mellitus: Causes, Treatment, and Impact on Outcomes. Endocr Rev. 2016 Feb;37(1):37–61.

15. Cohen E, Korah M, Callender G, Belfort de Aguiar R, Haakinson D. Metabolic Disorders with Kidney Transplant. Clin J Am Soc Nephrol. 2020 May 7;15(5):732–42.

16. Prasad GVR. Metabolic syndrome and chronic kidney disease: Current status and future directions. World J Nephrol. 2014 Nov 6;3(4):210–9.

17. Sharif A, Chakkera H, de Vries APJ, Eller K, Guthoff M, Haller MC, et al. International consensus on post-transplantation diabetes mellitus. Nephrol Dial Transplant. 2024 Feb 28;39(3):531–49.

18. Pittas AG, Lau J, Hu FB, Dawson-Hughes B. The role of vitamin D and calcium in type 2 diabetes. A systematic review and meta-analysis. J Clin Endocrinol Metab. 2007 Jun;92(6):2017–29.

19. Johnson JA, Grande JP, Roche PC, Kumar R. Immunohistochemical localization of the 1,25(OH)2D3 receptor and calbindin D28k in human and rat pancreas. Am J Physiol. 1994 Sep;267(3 Pt 1):E356–60.

20. Maestro B, Dávila N, Carranza MC, Calle C. Identification of a Vitamin D response element in the human insulin receptor gene promoter. J Steroid Biochem Mol Biol. 2003 Feb;84(2-3):223–30.

21. Scragg R, Sowers M, Bell C, Third National Health and Nutrition Examination Survey. Serum 25-hydroxyvitamin D, diabetes, and ethnicity in the Third National Health and Nutrition Examination Survey. Diabetes Care. 2004 Dec;27(12):2813–8.

22. Hyppönen E, Power C. Vitamin D status and glucose homeostasis in the 1958 British birth cohort: the role of obesity. Diabetes Care. 2006 Oct;29(10):2244–6.

23. Courbebaisse M, Bourmaud A, Souberbielle JC, Sberro-Soussan R, Moal V, Le Meur Y, et al. Nonskeletal and skeletal effects of high doses versus low doses of vitamin D in renal transplant recipients: Results of the VITALE (VITamin D supplementation in renAL transplant recipients) study, a randomized clinical trial. Am J Transplant. 2023 Mar;23(3):366–76.

24. Prasad GVR, Huang M, Silver SA, Al-Lawati AI, Rapi L, Nash MM, et al. Metabolic syndrome definitions and components in predicting major adverse cardiovascular events after kidney transplantation. Transpl Int. 2015 Jan;28(1):79–88.

25. Prasad GVR, Bhamidi V. Managing cardiovascular disease risk in South Asian kidney transplant recipients. World J Transplant. 2021 Jun 18;11(6):147–60.

26. Courbebaisse M, Souberbielle JC, Thervet E. Potential nonclassical effects of vitamin D in transplant recipients. Transplantation. 2010 Jan 27;89(2):131–7.

27. Binkley N, Krueger D, Cowgill CS, Plum L, Lake E, Hansen KE, et al. Assay variation confounds the diagnosis of hypovitaminosis D: a call for standardization. J Clin Endocrinol Metab. 2004 Jul;89(7):3152–7.

Tables:

Table 1. Baseline recipient characteristics and summary statistics. Criteria for metabolic syndrome are based on NCEP-ATP III criteria. “Above median” vitamin D-25 level is >48 nmol/mL. Mean and (standard deviation) are provided for continuous variables. Binary variables are indicated as count and (percentage).

	No metabolic syndrome		Metabolic syndrome			
Vitamin D status	Below median	Above median	Below median	Above median	p	missing
n	286	346	594	566		
Patient demographics						
Age at transplant (years)	50.12 (14.22)	47.89 (13.22)	50.54 (13.71)	51.03 (13.73)	**0.007**	0
Age at bloodwork (years)	52.98 (13.78)	52.71 (12.97)	52.50 (13.32)	54.25 (13.17)	0.125	0
Mean years follow-up	5.69 (4.38)	8.58 (5.14)	4.58 (3.81)	6.43 (4.72)	**<0.001**	0
Ethnicity					**0.001**	0
Black/Afro-Caribbean	17 (5.9)	12 (3.5)	43 (7.2)	26 (4.6)		
East Asian	36 (12.6)	29 (8.4)	72 (12.1)	49 (8.7)		
Other/unknown	157 (54.9)	205 (59.2)	329 (55.4)	349 (61.7)		
South Asian	23 (8.0)	17 (4.9)	42 (7.1)	16 (2.8)		
White	53 (18.5)	83 (24.0)	108 (18.2)	126 (22.3)		
Patient characteristics						
Systolic BP (mmHg)	128.58 (15.09)	127.65 (15.12)	128.25 (15.98)	128.62 (15.48)	0.848	11.5
Diastolic BP (mmHg)	80.35 (10.13)	81.19 (9.61)	80.83 (10.44)	80.33 (9.74)	0.625	11.5
BMI (kg/m^2)	27.46 (12.87)	27.12 (12.05)	26.44 (10.50)	27.62 (22.21)	0.664	14.0
Waist-to-hip ratio	0.96 (0.07)	0.99 (0.75)	0.99 (0.57)	0.95 (0.08)	0.396	19.0
Smoking status (%)	87 (34.5)	127 (41.8)	205 (37.5)	203 (40.5)	0.255	10.5
Male sex (%)	167 (58.4)	203 (58.8)	365 (61.6)	344 (60.8)	0.757	0.1
Live donor (%)	176 (61.5)	151 (43.6)	363 (61.1)	278 (49.1)	**<0.001**	0.0
Pre-transplant MACE (%)	47 (16.4)	47 (13.6)	87 (14.6)	96 (17.0)	0.491	0.0
Acute rejection	17 (5.9)	9 (2.6)	45 (7.6)	27 (4.8)	**0.010**	0

Delayed graft function	11 (3.8)	9 (2.6)	26 (6.1)	18 (3.2)	**0.030**	0
3-month post-transplant bloodwork						0
HDL (mmol/L)	1.31 (0.48)	1.33 (0.44)	1.33 (0.46)	1.42 (1.03)	0.087	
LDL (mmol/L)	2.64 (1.10)	3.12 (9.75)	2.51 (0.93)	2.67 (1.01)	0.297	8.4
Non-HDL cholesterol (mmol/L)	3.51 (1.44)	3.37 (1.05)	3.44 (2.01)	3.45 (1.47)	0.764	14.3
Triglycerides (mmol/L)	1.95 (1.38)	1.83 (1.02)	1.89 (1.04)	1.80 (1.07)	0.288	8.4
24-hour urine protein	0.86 (2.26)	0.50 (0.85)	0.82 (1.47)	0.60 (1.17)	0.046	5.8
Fasting blood gluc. (mmol/L)	6.20 (2.25)	5.80 (1.79)	6.38 (4.92)	6.15 (5.17)	0.255	56.8
Random blood gluc. (mmol/L)	6.63 (4.09)	6.01 (2.03)	6.57 (2.67)	6.30 (3.91)	**0.039**	0.9
Apolipoprotein A1	1.49 (0.32)	1.50 (0.27)	1.50 (0.32)	1.55 (0.28)	**0.050**	0.4
Apolipoprotein B	0.83 (0.24)	0.84 (0.23)	0.84 (0.25)	0.83 (0.23)	0.946	22.9
Apo-B/Apo-A1 ratio	0.58 (0.20)	0.58 (0.19)	0.58 (0.24)	0.55 (0.18)	0.166	22.5
Serum creatinine (umol/L)	133.79 (92.79)	128.14 (74.87)	130.57 (64.94)	129.98 (69.35)	0.810	23.2
GFR (MDRD-7)	60.78 (77.30)	53.89 (18.04)	54.54 (21.28)	67.18 (261.81)	0.457	0
C-reactive protein (mg/L)	8.64 (25.19)	6.49 (14.80)	6.70 (20.18)	6.49 (15.57)	0.401	0
Albumin to creatinine ratio (mg/mmol)	16.69 (59.11)	9.82 (32.30)	10.41 (26.96)	9.37 (23.55)	**0.023**	0.4
Uric acid (mmol/L)	392.55 (115.41)	393.11 (106.43)	388.95 (111.74)	388.99 (100.50)	0.929	2.5
PTH (ng/mL)	19.09 (22.14)	11.23 (11.97)	17.85 (20.25)	11.84 (12.00)	**<0.001**	14.2
Vitamin D-25 (nmol/mL)	32.26 (10.01)	76.73 (23.06)	31.67 (10.05)	72.91 (20.96)	**<0.001**	0.3
Prescriptions at 3 months						0
Tacrolimus (vs. cyclosporine)	168 (75.0)	196 (75.7)	345 (81.8)	332 (81.0)	0.079	
Statin (%)	137 (52.9)	147 (50.2)	253 (46.5)	276 (55.5)	**0.030**	11.1
ACE-inhibitor (%)	49 (18.9)	72 (24.6)	62 (11.4)	100 (20.1)	**<0.001**	11.1
ARB (%)	49 (18.9)	71 (24.2)	92 (16.9)	97 (19.5)	0.087	11.1
Oral hypoglycemic (%)	38 (14.7)	29 (9.9)	63 (11.6)	56 (11.3)	0.356	11.1
Prednisone (%)	197 (87.9)	220 (84.9)	376 (89.1)	366 (89.3)	0.332	26.6

Figures:

Figure 1. PTDM incidence proportions across quartiles of vitamin D and number of criteria for metabolic syndrome met. Criteria for metabolic syndrome are based on NCEP-ATP III criteria. Hypothesis testing conducted via chi-square test for trends in incidence proportions. Years of follow-up, case counts, and event counts per 100 patient-years are included in annotations above individual bars. [2 column]

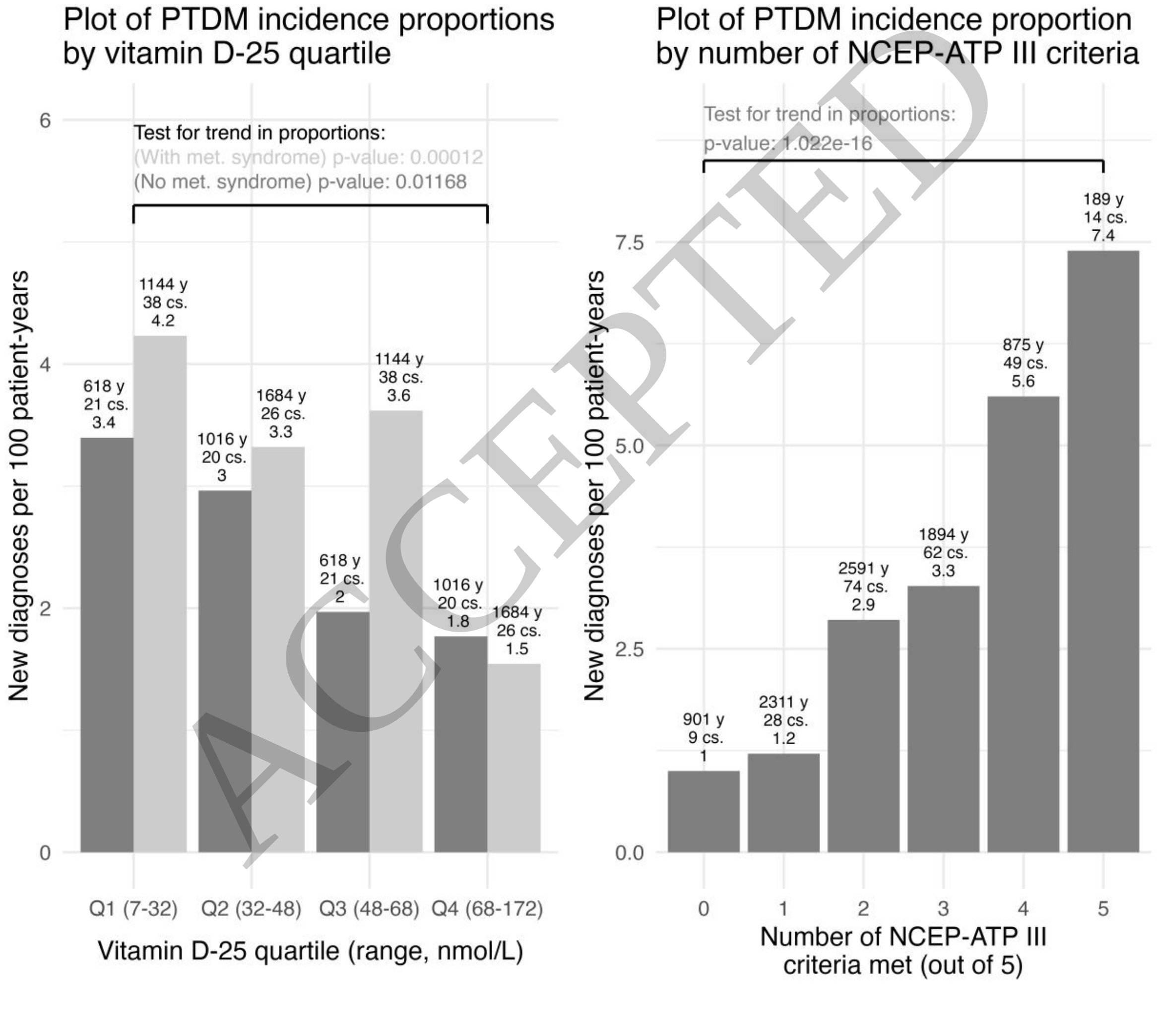


Figure 2. Features of the NCEP-ATP-III definition of metabolic syndrome and association in multivariate logistic regression with PTDM survival. Forest plots demonstrating hazard ratios of various components of the NCEP-ATP-III definition of metabolic syndrome in multivariable regression. Samples where all columns of data (metabolic syndrome criteria) were not fully accounted for were excluded from the analysis. The upper forest plot uses binary variables based on the NCEP-ATP-III criteria (e.g. blood pressure criterion is met, 0=no, 1=yes). The lower forest plot uses continuous versions of the relevant variables from the definition. Below, Kaplan-Meier survival curves demonstrate univariate impacts of each of the criteria on PTDM-free survival. Solid line = below median 25(OH)D, dashed line = above median 25(OH)D; Black line = does not meet MetS criteria, grey line = does meet MetS criteria. Note the y-axis range of the KM plot for fasting glucose is wider than other plots, and spans from 0.5 to 1.0. [2 column]

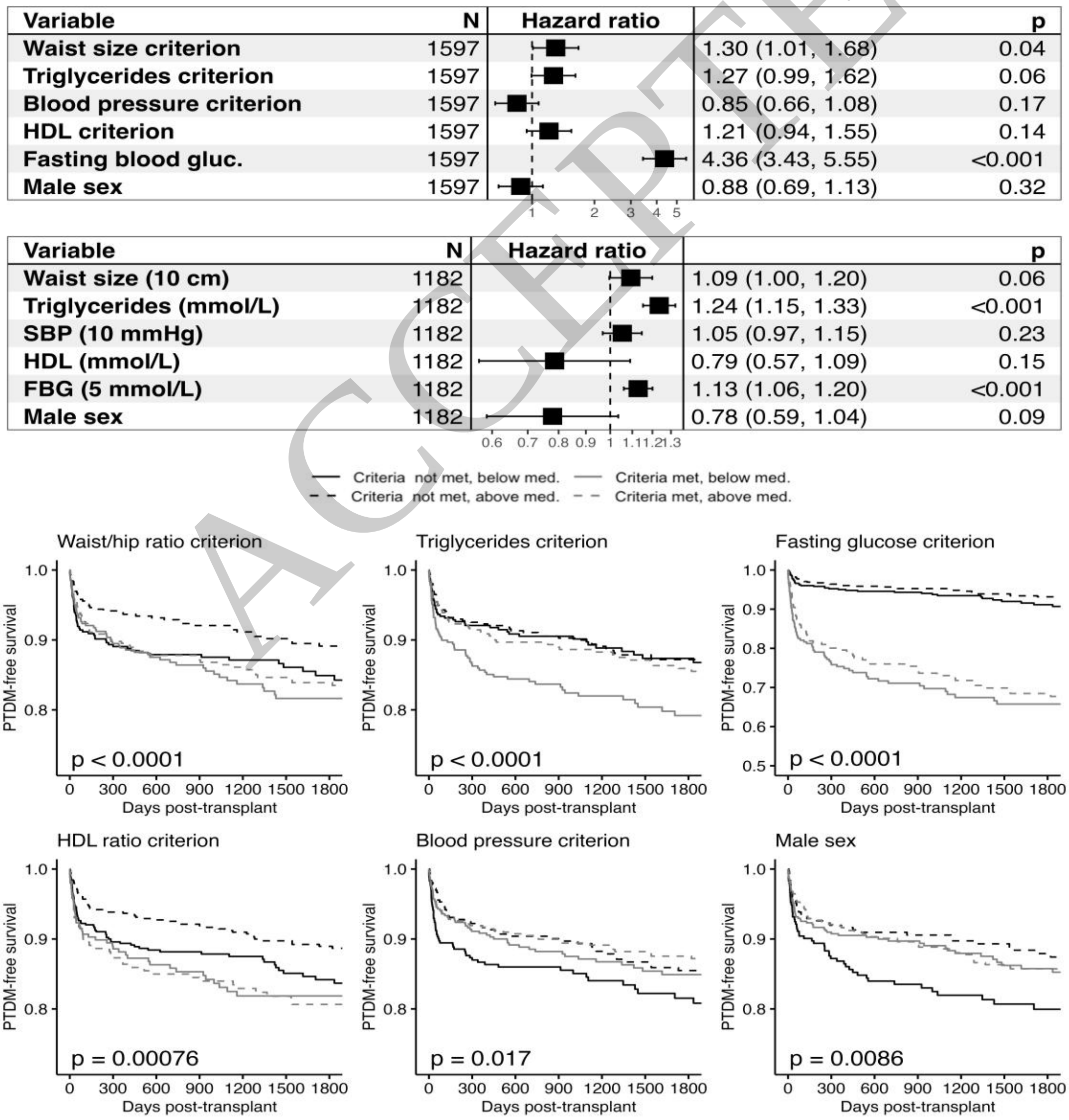


Figure 3. Kaplan-Meier survival curves for PTDM incidence across vitamin D median levels. Criteria for metabolic syndrome are based on NCEP-ATP III criteria.Vitamin D-25 median in this cohort is 48 nmol/mL. Risk tables and cumulative events tables are included below for each KM plot respectively. [2 column]

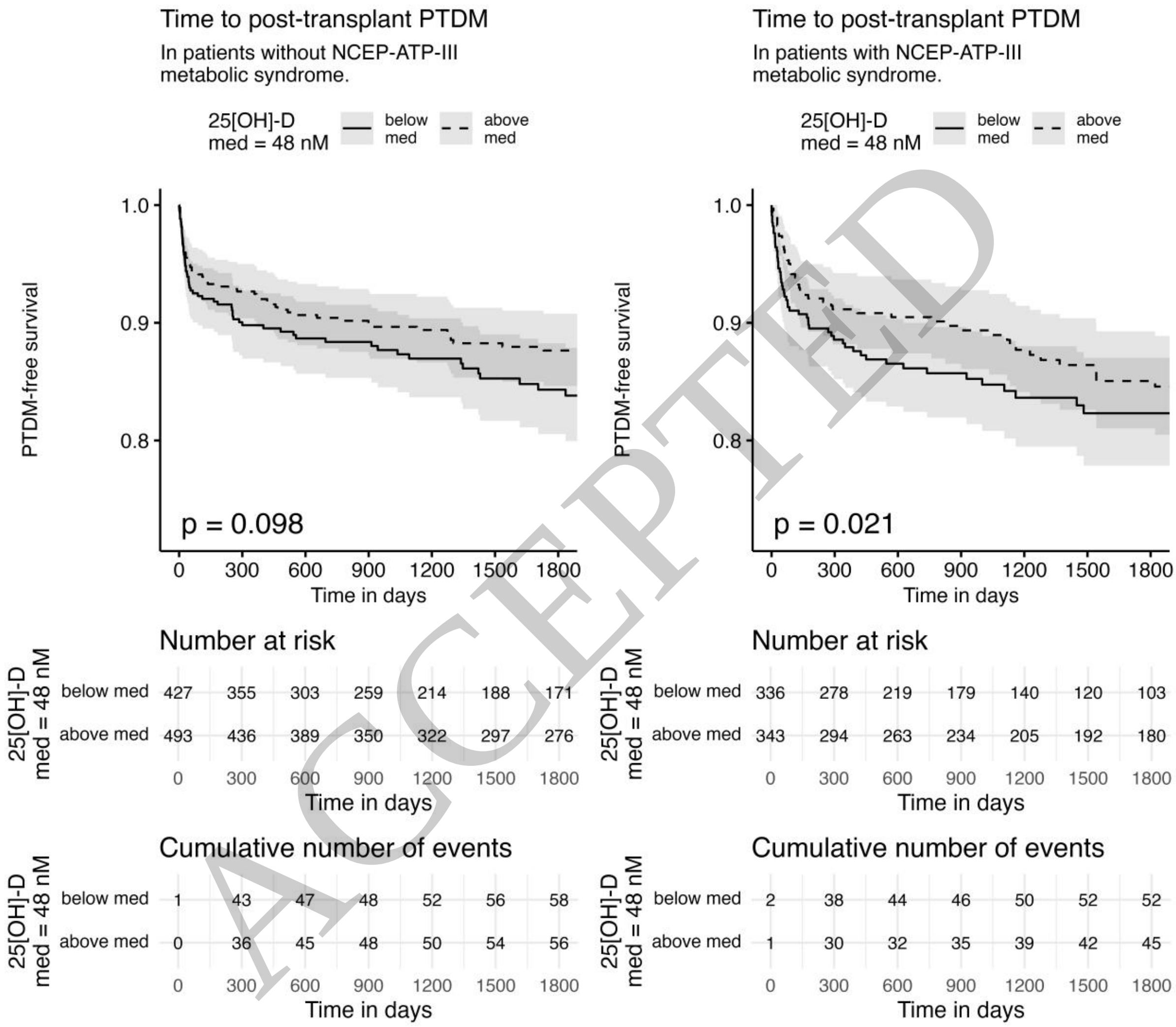
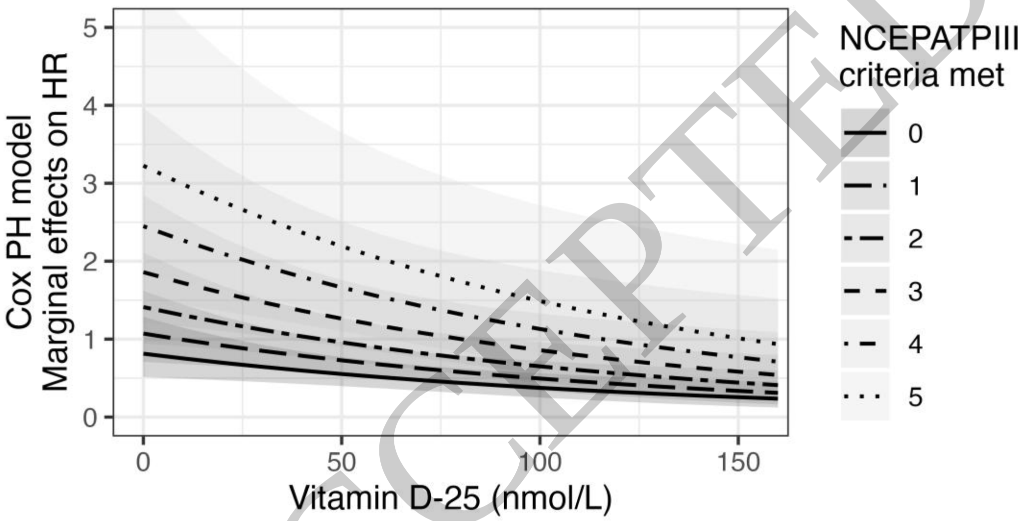


Figure 4. Multiple Cox regression and marginal effects plot for PTDM incidence across vitamin D level and number of metabolic syndrome criteria met. A plot demonstrating the marginal effects on estimated hazard ratios from a Cox survival regression of the effects of Vitamin D-25 on downstream PTDM survival at different levels of the metabolic syndrome, based on criteria of the metabolic syndrome met at the time of bloodwork. No interaction terms are used in this regression model. A table containing hazard ratio estimates with confidence intervals and p-values is included for the depicted model. [2 column]



	HR [95% conf. int.]	p-value
Vitamin D-25 (10 nmol/L)	0.93 [0.88, 0.98]	0.007
NCEPATPIII criteria met	1.31 [1.16, 1.48]	<0.001
Male sex	0.90 [0.68, 1.19]	0.455
Age (10 years)	1.22 [1.09, 1.37]	0.001
Fasting sugar (mmol/L)	1.01 [0.98, 1.03]	0.653
HbA1c (%)	0.57 [0.19, 1.66]	0.300

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Disclosure Updated Date: November 25, 2024

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Employer: St. Michael's Hospital; Research Funding: Paladin Canada; and Honoraria: Paladin Canada;.

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Name: G.V. Ramesh Prasad
Manuscript ID: K360-2024-000650R1
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Name: Lindita Rapi
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K. Vansjalia has nothing to disclose.

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Name: Karan M Vansjalia
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