Specific Aims

Understanding predictors and risk factors for vascular access complications is critical for focusing preventive and therapeutic efforts. Few serologic risk factors, however, have been identified, which has limited our ability to develop effective interventions.

Abnormalities in mineral homeostasis are common in patients with end-stage renal disease (ESRD) and are associated with numerous adverse outcomes, including vascular calcification and mortality.[1-5] Impaired excretion of phosphorus, decreased conversion of 25-hydroxy vitamin D to its active 1,25 D form, increased production of fibroblast growth factor 23 (FGF23) in the parenchyma and the resulting increase in parathyroid hormone (PTH) combine to form a complex interplay of factors that promote inflammation.

Vitamin D deficiency, specifically, may also have effects unrelated to mineral metabolism. Vitamin D deficiency has been associated with hypertension, insulin resistance, viral and bacterial infection risk, and multiple organ damage due to systemic inflammation.[6] Given the role of inflammation in vascular access complications, vitamin D may be of particular relevance for such events. Despite the importance of disordered mineral homeostasis for vascular outcomes in ESRD, few studies have investigated the role of these factors in vascular access outcomes.

The Choices for Health Outcomes in Caring for End-Stage Renal Disease (CHOICE) Study contains detailed data and extensive follow-up for a large cohort (n≥600) of incident hemodialysis patients, allowing us to directly address this critical issue.[7] The overall goal of our research program is to improve hemodialysis vascular access outcomes. The objectives of this application are to determine the association of markers of abnormal mineral metabolism with vascular access complications and failure. This application capitalizes on the unique resources and opportunities afforded by the data available in the CHOICE Study and the broad expertise of the investigators to achieve the following Specific Aims:

Specific Aim 1: Determine the association between markers of mineral metabolism and hemodialysis vascular access failure. *We hypothesize that lower levels of 25-hydroxyvitmain D (25 D) and higher levels of fibroblast growth factor 23 (FGF23), serum calcium, phosphorus, and parathyroid hormone (PTH) are associated with greater incidence of vascular access complications.* Outcomes of interest include access failure and loss of patency (i.e., need for revascularization).

Specific Aim 2: Determine whether the association between mineral metabolism markers and vascular accesses outcomes differ by type of permanent access. *We hypothesize that the association is present for both AV fistulae and AV grafts, but stronger in AV fistulae.*

This study will identify specific, potentially modifiable risk factors for vascular access complications and failure and elucidate the potential role of mineral homeostasis. It is anticipated that our findings will have a significant positive impact by focusing future interventions on pathways that can be most effectively modified in clinical practice.

Significance and Innovation

Preventing vascular access complications will require a greater understanding of the risk factors and pathophysiologic processes underlying such events. Few potentially modifiable risk factors have been identified, however. This has severely hampered our ability to develop effective interventions.

Declining kidney function leads to progressively deregulated mineral homeostasis, which is known to contribute to vascular calcification and to provide a pro-inflammatory milieu in patients with end-stage renal disease (ESRD).[2] These findings suggest an important role for mineral metabolism dysregulation in vascular access complications, as well. As these abnormalities are potentially modifiable with existing therapies, they provide an opportunity for significant clinical impact. Few studies, however, have investigated this potential association.

Morena et al analyzed 128 patients with an arteriovenous fistula (AVF). At baseline, stenoses were found in 50 of these fistulae.[8] None of the mineral metabolites were associated with the presence of stenosis. Over two years of follow-up, however, phosphorus, calcium x phosphorus product and PTH were higher in those 19 patients with subsequent fistula thrombosis than in those with no thrombosis. In a separate study of 128 patients undergoing permanent vascular access placement (112 AVF, 16 AV grafts), vitamin D deficiency, defined as plasma 25-hydroxycholecalciferol <20 ng/mL, was associated with a 2.3-fold greater (1.2-4.7) risk of vascular access failure after adjustment for relevant confounders.[9] [Figure] Other mineral metabolites (i.e., calcium, phosphorus, PTH) were not associated with vascular access outcomes. No studies, to our knowledge, have included analyses of the potential association of FGF23 with vascular access outcomes. Data from larger studies, including a greater number of AV graft, are needed to further elucidate the relationships between indicators of mineral metabolism and vascular access outcomes.

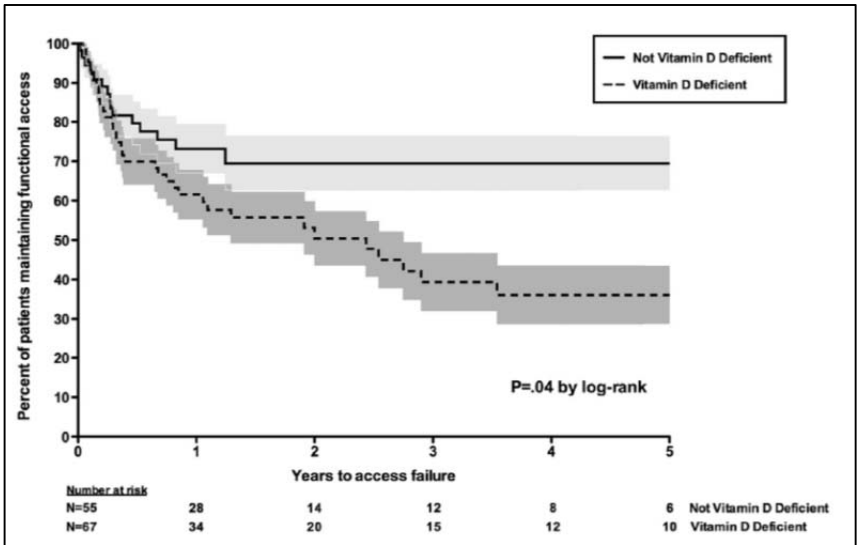

Fig 3. Kaplan-Meier curve shows access failure by vitamin D-deficient and nondeficient status.

The proposed project is innovative, in our opinion, because it will be the largest cohort of hemodialysis patients to be investigated for an association between with measured markers of mineral metabolism and vascular access outcomes. It will also be the first to include a complete panel of indicators, including FGF23, and will include a much greater number of AV grafts than previous studies.

Research Strategy

Study Population

We will address the Specific Aims utilizing data from the CHOICE Study. The CHOICE Study is a national, prospective cohort study of incident hemodialysis and peritoneal dialysis patients with the goal of investigating the associations of modality and dialysis dose with outcomes of dialysis care.[10] A total of 1,041 dialysis patients were recruited from 79 dialysis clinics associated with Dialysis Clinic, Incorporated (DCI; Nashville, TN), New Haven CAPD (New Haven, CT), and the Hospital of St. Raphael (New Haven, CT) from October 1995 through June 1998. Patients were enrolled a median of 45 days from initiation of chronic dialysis (98% within 4 months). The proposed analyses will be limited to patients who used hemodialysis as their initial renal replacement modality (n = 762), were enrolled at clinics associated with DCI (n = 735), and have vascular access information available (n = 616). Dr. Astor has extensive experience with the CHOICE Study, having published several manuscripts based on these data.[2,7,11-14]

Vascular Access Assessment

Access information was obtained through review of discharge summaries, dialysis flow sheets, and dialysis clinic progress notes.[7] The first and last date of use for each access was recorded. Permanent access failure was defined as the need for a surgical intervention to replace a poorly or nonfunctioning fistula or graft. Dr. Astor was integral to the original collection of vascular access data in the CHOICE Study and has used the data for numerous publications.[7,11-14]

Laboratory Assessments

Non-fasting, pre-dialysis blood specimens were collected from hemodialysis patients numerous times throughout the study. These specimens were centrifuged at 2,500 to 3,000 rpm for 15 minutes within 30 to 45 minutes of collection. Each blood collection was refrigerated and mailed overnight on ice to the DCI Central Laboratory for analysis or storage at -80°C.

C-terminal FGF23 (Immutopics, San Clemente, CA) and 25 D (Immunodiagnostic Systems, Scottsdate, AZ) were measured at a single timepoint in stored plasma samples drawn within 6 months of enrollment (median = 90 days). Serum PTH was measured using the Diasorin intact assay (Diasorin, Inc., Stillwater, MN). Serum albumin, hemoglobin, calcium, phosphorus and PTH were measured frequently throughout the study as part of clinical care. We will use the measurements closest in time to the date of the FGF23 and 25 D measurements.

Other Data Available

Additional data on CHOICE Study participants were collected through a combination of self-report and chart review. Available data includes demographics, medical history, presence and severity of comorbidity (assessed through the Index of Coexistent Disease [ICED]), body mass index (BMI),

Analyses

Analyses will include all CHOICE Study participants with available data who were using a permanent vascular access (AV fistula or AV graft) at the time of laboratory assays. The time from access creation to laboratory assay will be calculated and used in sensitivity analyses to exclude patients with a newly created (<30 days) access. Based on our prior work with the CHOICE Study, we expect 511 patients will have all laboratory data available.[Scialla, Banerjee]. We also expect approximately 70% of patients (n = 346) will be using a permanent access, with approximately 206 using an AV graft and 140 using an AV fistula. These 346 patients will comprise our study population.

Initial analyses will describe the distribution of each mineral metabolite in the study population. Calcium x phosphorus product will be calculated. These distributions will be checked for outliers, which will be excluded if needed. Values will be log-transformed if necessary to achieve approximate normality. Mineral metabolism markers will be analyzed as continuous variables, in quartiles, and in relevant categories (e.g., 25 D > or ≤ 20 ng/mL).

We will address Specific Aim 1 with survival analyses (i.e., time from assays to first access failure), consisting of unadjusted (Kaplan-Meier curves, log-rank tests) and adjusted analyses (Cox proportional hazards models). Initial analyses will examine each marker in separate models, first in unadjusted models and then adjusting for potential confounders in sequential models. Access failure incidence rates will be reported by quartile of each markers, and trends tested over the entire distribution.

We will first adjust for demographics (age, race, sex), then add access-realted variables (access type, time from first access use, time from starting dialysis, previous access failures), then add other clinical variables (BMI smoking status, comorbidities (ICED score, specific comorbidities). Models will appropriately account for potential clustering within dialysis centers with robust variance estimates. Models will initially include one marker of mineral metabolism at a time, with multiple markers being included in later models.

We will address Specific Aim 2 with similar methods, incorporating interaction terms for access type x marker values.

Statistical Power Considerations

Based on prior analyses from the CHOICE Study, we expect an overall access failure rate of approximately 80% per year. With our projected sample size of 346 patients, we expect to have 80% statistical power to detect an incidence rate ratio ≥1.34 between the upper and lower quartile of any marker. We will have even greater statistical power to detect trends across all quartiles or continuous associations. The statistical power to detect differences in these associations between access types will be more limited, but may provide insight on the relative importance of these markers for each type.

Expected Results

We expect to identify modifiable risk factors for vascular access failure and add to our limited understanding on the pathological processes underlying these events. These findings will be critical to designing interventional trials to decrease the enormous burden of vascular access complication in hemodialysis patients.

Alternative Approaches

We acknowledge that the studies proposed include analyses of numerous separate, but related, markers of mineral metabolism. These multiple comparisons are susceptible to random type 1 errors. We will remain cognizant of existing knowledge on the interactions between these markers and explore additional analyses to address findings supported by results pertaining to a single marker.

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Resources and Facilities

Office: The office of the PI is located on the fifth floor of the Medical Foundation Centennial Building (MFCB), 1685 Highland Avenue. The MFCB is attached to the UW Hospital and Clinics, UW Clinical Science Center, the Health Sciences Learning Center, and the William S. Middleton Memorial Veterans Hospital. The PI's office is locked and contains a desk, computer, and printer. Secretarial support is provided by the Department of Medicine. All Division of Nephrology faculty and staff are located in close proximity. Additional office space is available for research assistants and students, as well.

Computer: Offices of the PI and other personnel each have PCs (purchased 2014-5) that are equipped with all the necessary software for the proposed analyses. Software packages include, but are not limited to R, Stata, SAS, Microsoft Office (Word, Excel, etc), and Adobe illustrator CS5, Adobe Acrobat. Computer support is available through the Department of Medicine. Secure storage is available through the Department of Medicine shared servers,

Department of Medicine: The Department of Medicine in the University of Wisconsin School of Medicine and Public Health has approximately 350 faculty members in 13 clinical subspecialties, with approximately 100 research faculty who bring in $43.5 million dollars for research annually. The Department maintains several well-funded research programs, ranking it in the top 5% of internal medicine training programs in the U.S. and in the top 20 of academic departments of medicine with respect to National Institutes of Health funding. In addition, research opportunities exist with outside faculty on the University of Wisconsin - Madison campus. Department faculty collectively occupy approximately 50,000 assignable square feet coded as research space in 5 buildings across campus, including the Veterans Affairs Hospital and the University of Wisconsin Hospital. Department of Medicine faculty members direct seven National Research Service Awards T32 grants and two K12 Career Development grants from the National Institutes of Health. Department faculty have access to the following:

- Research support services – including assistance with pre- and post-award of any proposals written by trainees; assistance with manuscript writing; and a knowledge-rich, service-oriented "research guide" to facilitate access to institutional or human resources necessary to conduct research or engage in research-related training
- Department-supported biostatistician
- Computer network access and desktop support
- Access through institutional agreements to nearly all major biomedical and behavioral journals and 24-hour access to others.

University of Wisconsin-Madison Libraries: The University of Wisconsin-Madison has the 11th largest research library collection in North America. The campus library collections include more than 7.3 million printed volumes, 55,000 serial titles, 6.2 million microforms, 160 linear feet of manuscripts and over 7 million items in other formats, including government documents, maps, musical scores and more. There is also access to over 20,000 electronic journals, as well as e-books. The Health Sciences (Ebling) library occupies over 53,000 square feet and contains 404,848 print volumes, 5,200 online journals, 998 electronic books, and 75 electronic reference sources. The University of Wisconsin-Madison Library system is a member of: the Committee on Institutional Cooperation - comprised of Big Ten institutions plus the University of Chicago and the University of Illinois - Chicago, the Council of University of Wisconsin Libraries, and the Wisconsin E-Book Consortium. The University of Wisconsin-Madison Libraries supplement our on-site collections with the resources of the nationally recognized Center for Research Libraries.