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BRIEF COMMUNICATION

Discrepancy between self-perceived mycophenolic acid-associated diarrhea and stool water content after kidney transplantation

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Abstract

Background: Diarrhea is a well-known side effect of mycophenolic acid (MPA) use in kidney transplant recipients (KTRs). It is unknown whether self-reported diarrhea using the Modified Transplant Symptom Occurrence and Symptom Distress Scale (MTSOSD-59R) corresponds to stool water content and how both relate to MPA usage.

Methods: MTSOSD-59R questionnaires filled out by 700 KTRs from the TransplantLines Biobank and Cohort Study (NCT03272841) were analyzed and compared with stool water content. Stool samples (N = 345) were freeze-dried, and a water content ≥80% was considered diarrhea.

Results: Self-perceived diarrhea was reported by 46%, while stool water content ≥80% was present in 23% of KTRs. MPA use was not associated with self-perceived diarrhea (odds ratio(OR) 1.32; 95% confidence interval(CI), 0.87–1.99, p = .2), while it was associated with stool water content ≥80% (OR 2.88; 95%CI, 1.41–5.89, p = .004), independent of potential confounders. Adjustment for prior MPA discontinuation because of severe diarrhea, uncovered an association between MPA use and self-perceived diarrhea (OR 1.80; 95%CI, 1.13–2.89, p = .01).

Conclusions: These results suggest that reporting bias could add to the discrepancy between both methods for diarrhea assessment. We recommend use of objective biomarkers or more extensive questionnaires which assess information on stool frequency and stool consistency, to investigate post-transplantation diarrhea.

KEYWORDS
diarrhea, kidney transplantation, MTSOSD-59R, mycophenolic acid, side effects, stool water content

Diarrhea is a common side effect of immunosuppressive therapy, affecting up to 50% of patients following kidney transplantation.[1] Use of mycophenolic acid (MPA) has been identified as the main culprit.[2,3] In a previous survey, it was demonstrated that clinicians underestimate the prevalence of post-transplantation diarrhea.[4] However, the clinical consequences of this diarrhea are serious,

Study registry: clinicaltrials.gov; identifier: NCT03272841.

since it is associated with increased numbers of hospital admission, graft failure, higher health-care costs and even an increased risk of premature mortality.[5-7] In 2008, the 59-item Modified Transplant Symptom Occurrence and Symptom Distress Scale (MTSOSD-59R), which is used to measure patients subjective appraisal of side effects from immunosuppressive therapy, was validated in kidney transplant recipients (KTRs).[8] Symptom experience in KTRs has not been extensively studied, and it is currently unknown whether self-reported diarrhea using the MTSOSD-59R corresponds to objective markers of diarrhea and how both relate to MPA use. We aimed to investigate this by objectively assessing diarrhea using stool water measurements.

As part of the TransplantLines Biobank and Cohort Study (clinicaltrials.gov identifier: NCT03272841),[9] 700 stable outpatient KTRs who received a donor kidney ≥1 year ago, filled out the MTSOSD-59R questionnaire prior to a study visit between June 2015 and August 2019. This questionnaire consists of 59 items assessing the occurrence and distress of side effects caused by immunosuppressive therapy, including diarrhea,[8] which is assessed by the following question: "Did you experience diarrhea during the past four weeks?". Answers were scored on a 5-point Likert scale (0 = never; 1 = sometimes; 2 = regularly; 3 = almost always; 4 = always). Additionally, participants were asked to collect a stool sample using a FecesCatcher (TAG Hemi VOF, Zeijen, The Netherlands) the day prior to the study visit and to store the sample on ice. Upon arrival at the University Medical Center Groningen (UMCG) stool samples were immediately stored at −80°C. Stool samples were available from 345 participants, which were freeze-dried for 48 hours under 0.5 bar at −50°C to assess the stool water content. Stool water content ≥80% was considered as diarrhea.[10,11] The study protocol was approved by the International Review Board of the UMCG (IRB identifier: 014-077), adheres to the UMCG Biobank Regulation, and is in accordance with the Declaration of Helsinki and the Declaration of Istanbul.[9] All participants signed a written informed consent.

Mean age of the study population was 56 ± 13 years, and 60% were male (Table 1). KTRs were included at a median time after transplantation of 3.9 [1.0–10.6] years, and mean estimated glomerular filtration rate (eGFR) was 51.1 ± 17.5 ml/min/1.73 m^2. MPA was used by a majority of KTRs (75.4%). Self-perceived diarrhea (MTSOSD score 1–4) was reported by 46% of KTRs, while diarrhea assessed objectively by stool water content ≥80% was present in 23% of patients. There was a weak correlation between results of the MTSOSD-59R and the stool water content (Spearman's rho = 0.24, $p < .001$). Interestingly, in a crude logistic regression analysis, MPA use was not associated with self-perceived diarrhea symptoms (MTSOSD score 1–4) (Odds ratio (OR) 0.98; 95% confidence interval (CI), 0.69–1.39, $p = .9$), while MPA use was significantly associated with diarrhea using stool water content (OR 2.15; 95% CI, 1.16– 3.99,

TABLE 1 Characteristics of 700 kidney transplant recipients from the TransplantLines Study

	Total Population	MPA non-users	MPA users	*p*-value
Number of participants, n (%)	700 (100)	172 (25)	528 (75)	n/a
Demographics				
Age, y	56 ± 13	57 ± 14	55 ± 13	.2
Male sex, n (%)	421 (60)	91 (53)	330 (63)	.03
Height, cm	173.5 ± 9.9	171.8 ± 10.0	174.1 ± 9.9	.008
BSA, m^2	1.96 ± 0.22	1.90 ± 0.22	1.98 ± 0.21	<.001
Diabetes, n (%)	188 (27)	50 (29)	138 (26)	.5
Time after transplantation, y	3.9 [1.0–10.6]	10.2 [2.5–18.8]	2.2 [1.0–8.0]	<.001
Renal function parameters				
eGFR, ml/min/1.73 m^2	51.1 ± 17.5	44.9 ± 17.0	53.1 ± 17.2	<.001
Serum creatinine, µmol/L	123.0 [102.0 –154.0]	133.0 [111.0 –176.0]	122.0 [100.3–151.0]	<.001
Immunosuppressive therapy				
Prednisolone, n (%)	683 (97.6)	166 (96.5)	517 (97.9)	0.3
Tacrolimus, n (%)	467 (66.7)	92 (53.5)	375 (71.0)	<0.001
Cyclosporine, n (%)	101 (14.4)	40 (23.3)	61 (11.6)	<0.001
mTOR inhibitors, n (%)	27 (3.9)	21 (12.2)	6 (1.1)	<0.001
Azathioprine, n (%)	75 (10.7)	75 (43.6)	0 (0)	<0.001
Diarrhea				
Self-perceived diarrhea, n (%)	323 (46)	80 (47)	243 (46)	0.9
Stool water content, %	74.9 ± 6.5	73.6 ± 6.4	75.4 ± 6.4	0.02
Stool water content ≥80%, n (%)	80 (23)	15 (15)	65 (27)	0.01

Note: Data are presented as mean ±SD, median with interquartile ranges (IQR) or number with percentages (%). Abbreviations: BSA, body surface area; eGFR, estimated glomerular filtration rate; mTOR inhibitors, mammalian target of rapamycin inhibitors.

p = .02). These results remained materially unchanged independent of adjustment for potential confounders including, age, sex, body surface area (BSA), eGFR, time after transplantation, diabetes, and use of other immunosuppressive medication (ie, tacrolimus, cyclosporine and mTOR inhibitors) (Table 2). Since the discrepancy between both methods for assessment of diarrhea was suggestive of reporting bias in the MTSOSD-59R, we hypothesized that KTRs in which MPA treatment was discontinued more than 4 weeks prior to the study visit because of severe diarrhea, potentially still reported this side effect as present in the MTSOSD-59R. To investigate this, we thoroughly reviewed all electronic patient records to obtain information about MPA discontinuation and the cause for this. Indeed, MPA was discontinued in 111 KTRs in the past, for which severe diarrhea was the most common reason (n = 46, 41%), Figure 1. Other common reasons for discontinuation were viral infections (ie, CMV, EBV, BK, and Hepatitis) and pulmonary pathology, including bronchiectasis and recurrent (upper) airway infections (Figure 1). The median time interval between MPA discontinuation and the study visit was 3.1 [0.9–7.3] years in the total subset of 111 KTRs in which MPA therapy was discontinued and 2.1 [0.8–7.4] years in the subset of 46 KTRs in which MPA therapy was discontinued because of severe diarrhea. In addition, in 28 KTRs an MPA dose reduction was performed at 1.0 [0.8–3.0] years prior to the study visit to ameliorate diarrhea symptoms. When we adjusted for MPA cessation because of severe diarrhea in a logistic regression analysis, a significant association between current MPA use and self-perceived diarrhea symptoms was uncovered (OR 1.80; 95% CI, 1.13–2.89, p = .01, Table 2). Additional adjustment for MPA dose reduction did not materially change the association (OR 1.77; 95% CI, 1.10–2.83, p = .02, Table 2).

Next, we investigated the association between MPA trough levels and self-perceived diarrhea. In total, MPA trough levels were available in 360 KTRs (68% of all MPA users) of our cohort. In a crude logistic regression analysis, MPA trough levels were not significantly associated with self-perceived diarrhea (MTSOSD score 1–4) (Odds Ratio (OR) 0.97; 95% Confidence Interval (CI) 0.88–1.07, p = .6, Table S1). This result remained materially unchanged after adjustment for potential confounders (OR 0.94; 95% CI 0.84–1.06, p = .3, Table S1). Thereafter, we investigated the association between MPA trough levels and the stool water content. In total, MPA trough levels were available in 153 KTRs (44%) of the subset of 345 KTRs of which a stool sample was available. In a crude logistic regression analysis, MPA trough levels were not significantly associated with stool water content ≥80% (OR 1.00; 95% CI 0.88–1.14, p = .9, Table S1). Also this result remained materially unchanged after adjustment for potential confounders (OR 1.00; 95% CI 0.87–1.16, p = .9, Table S1).

Our results support the suggested hypothesis that KTRs still report side effects of immunosuppressive therapy outside the recall period of four weeks for which the questionnaire was developed. This hypothesis is, however, based on the assumption that diarrhea symptoms in the 46 patients in which MPA was discontinued because of severe diarrhea did resolve, which we could not verify. The finding that MPA trough levels are not associated with diarrhea is in agreement with the current opinion that neither systemic levels of MPA, nor its metabolites, are associated with diarrhea, but that MPA metabolites exert local toxicity within the gastrointestinal tract. It has previously been shown that neither MPA dose, nor MPA-area under the curve (AUC), nor the two hour AUCs of acyl and phenolic glucuronide metabolites of MPA, were associated with diarrhea in KTRs.[12] MPA-glucuronide undergoes enterohepatic recirculation via biliary excretion and by intestinal deconjugation trough bacteria with specific β-glucoronidase (GUS) activity.[13] In a recent study, it was discovered that treatment with vancomycin, an antibiotic which eliminates gut bacteria with GUS activity, prevented MPA induced gastrointestinal toxicity in mice.[14] Moreover, it has been demonstrated that co-treatment with ciclosporine reduces the incidence of diarrhea, potentially via the inhibition of transporters that facilitate the biliary excretion of MPA metabolites.[15] These findings support the hypothesis that local intestinal exposure to MPA

TABLE 2 Association of MPA use with self-perceived diarrhea and stool water content in kidney transplant recipients

Number of participants	Self-perceived diarrhea *N* = 700		Stool water content ≥80% *N* = 345	
MPA use	OR (95%CI)	*p*-value	OR (95%CI)	*p*-value
Crude	0.98 (0.69; 1.39)	.9	2.15 (1.16; 3.99)	.02
Model 1	0.97 (0.68; 1.38)	.9	2.20 (1.17; 4.13)	.02
Model 2	1.18 (0.80; 1.74)	.4	2.58 (1.29; 5.16)	.007
Model 3	1.32 (0.87; 1.99)	.2	2.88 (1.41; 5.89)	.004
Model 4	1.80 (1.13; 2.89)	.01	-	n/a
Model 5	1.77 (1.10; 2.83)	.02	-	n/a

Note: Model 1: MPA use adjusted for age, sex, BSA.
Model 2: model 1 + adjustment for eGFR, time since transplantation, diabetes.
Model 3: model 2 + adjustment for tacrolimus, cyclosporine, mTOR inhibitors.
Model 4: model 3 + adjustment for MPA cessation because of diarrhea.
Model 5: model 4 + adjustment for MPA dose reduction because of diarrhea.
Abbreviations: MPA, mycophenolic acid; OR, odds ratio.

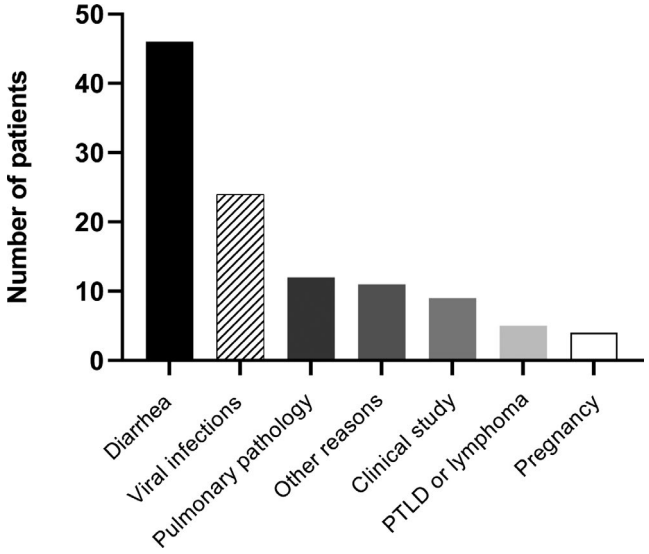

FIGURE 1 Reasons for discontinuation of MPA therapy among kidney transplant recipients from the TransplantLines Study. Abbreviations: PTLD, post-transplant lymphoproliferative disease

metabolites, instead of higher plasma levels, lead to intestinal toxicity and diarrhea.

Some limitations of this report should be acknowledged. The weak correlation between the MTSOSD-59R questionnaire and stool water content might be explained by differences in timing of both measurements. Stool water measurements only provide information about a single stool collection and thus whether diarrhea is present at time of sample collection, while the MTSOSD-59R questionnaire intends to cover a recall period of four weeks. However, if patients indeed experience chronic diarrhea due to MPA use, one might expect that the stool sample would also be indicative of diarrhea. Another limitation is that the MTSOSD-59R does not contain questions that generate information on stool frequency and stool consistency. This may also have led to the lack of agreement between self-perceived diarrhea and stool water content. Furthermore, the presence of diarrhea can also have other underlying causes that are not included in this report.

Lastly, among the KTRs who collected a stool sample the prevalence of diarrhea was 23%, which might actually be an underestimation. Patients with severe diarrhea might experience difficulties with sample collection and might therefore be less willing to collect a stool sample. Regardless, objective stool water measurements are easily performed and less prone to bias and may therefore be an interesting additional and potentially even more suitable tool for the investigation of post-transplantation diarrhea.

In conclusion, these results suggest that reporting bias could add to the discrepancy between both methods for assessment of diarrhea. Given the clinical relevance of post-transplant diarrhea and the increased use of the MTSOSD-59R in clinical research, we would like to recommend other researchers in the field to use objective biomarkers, such as the stool water content, or more extensive questionnaires which also assess information on stool frequency and stool consistency, to assess and investigate the occurrence of post-transplantation diarrhea.

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CONFLICT OF INTEREST

The authors of this manuscript have no conflicts of interest to disclose.

AUTHORS CONTRIBUTIONS

RMD and SJLB involved in research design. RMD involved in performance of the research and drafting article. RMD, JCS, and AP involved in data analysis/interpretation. JCS, AP, CA, HJMH, and SJLB involved in critical revision of article. CA, HJMH, and SJLB involved in supervision/mentorship. Each author contributed important intellectual content during the manuscript drafting and agrees with submission of the manuscript.

DATA AVAILABILITY STATEMENT

All data presented in this study can be made available by the data manager of the Transplantlines study, by mailing to datarequest.transplantlines@umcg.nl.

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SUPPORTING INFORMATION

Additional supporting information may be found online in the Supporting Information section.

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