Developing an LC-MS/MS method for measurement of tacrolimus and creatinine concentration from finger-prick blood collected using the Mitra device

RESEARCH REFERENCE NUMBERS

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Statistical Analysis Plan

Hypothesis:

The LC-MS/MS method for the quantitation of creatinine and tacrolimus in finger-prick samples collected from the Mitra device will produce results that are not statistically different from whole blood venous samples.

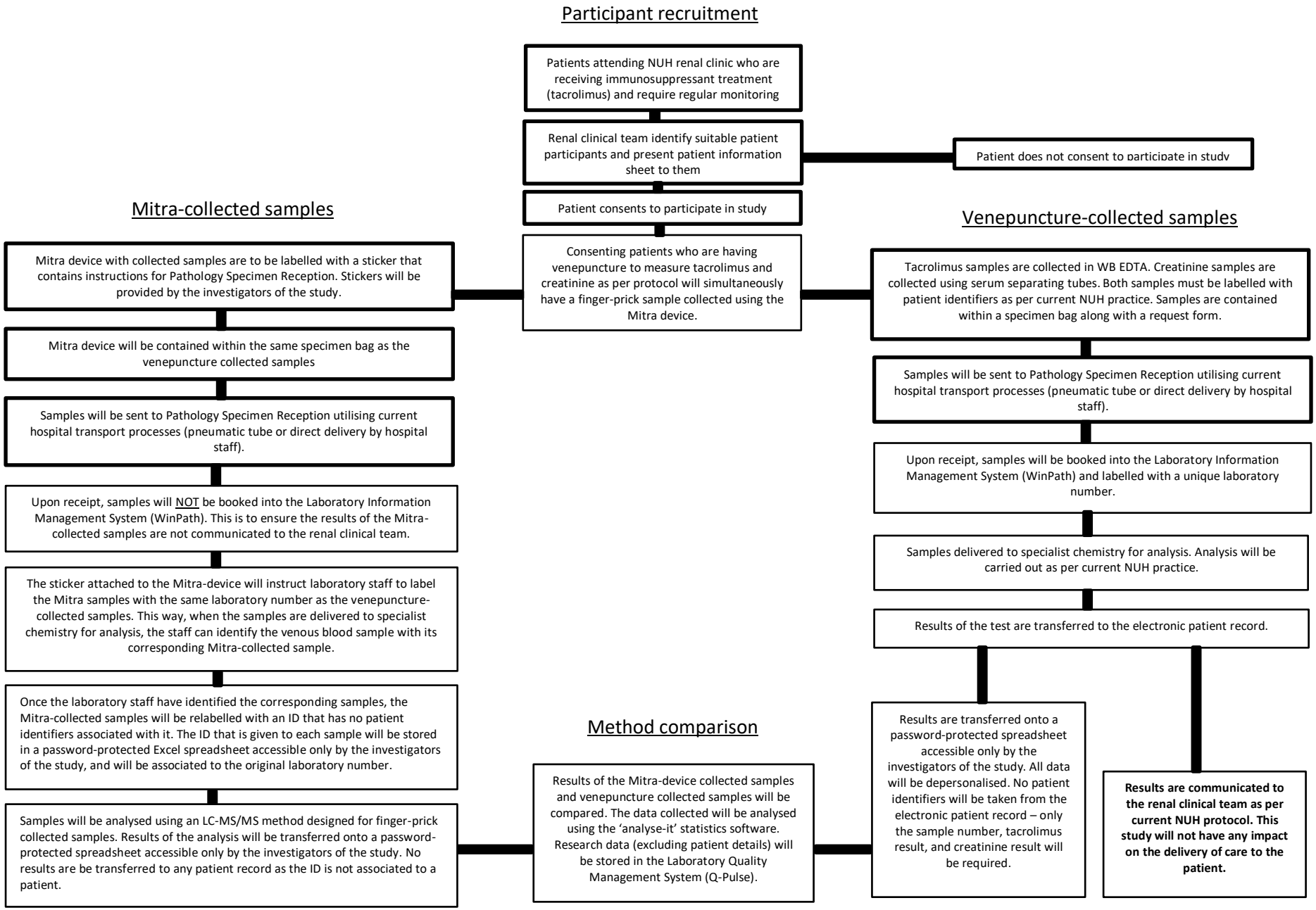
Methodology

Patients attending the Nottingham renal clinic for routine therapeutic drug monitoring for Tacrolimus will be consented to have a venous blood sample and a corresponding finger-prick sample (collected using the Mitra device) collected for tacrolimus and creatinine analysis (please see flowchart below).

50 samples will be collected from consented patients. The tacrolimus levels from the venous blood samples will be measured using the routine LC-MS/MS method and creatinine will be measured using the Abbott enzymatic creatinine method. The capillary-finger-prick samples collected using the Mitra device will be analysed using the optimised LC-MS/MS methods for creatinine and tacrolimus.

The results from the venous blood samples will be reported following routine laboratory processes (direct transfer of results from laboratory LIMS system to the electronic patient record). These results will then be transferred onto a password-protected spreadsheet accessible only by the investigators of the study. All data will be depersonalised. No patient identifiers will be taken from the electronic patient record – only the sample number, tacrolimus result, and creatinine result will be required. The results from the Mitra-collect finger-prick samples will be transferred to the same password-protected spreadsheet accessible only by the investigators of the study. These results will not be transferred to any patient record.

Results of the Mitra-device collected samples and venepuncture collected samples will be compared. The data collected will be analysed using the 'Analyse-It' statistics software and Bland-Altman and Passing-Bablok regression analyses will be completed to identify any statistical differences between the two LC-MS/MS methods. Research data (excluding patient details) will be stored in the Laboratory Quality Management System (Q-Pulse).


Participant recruitment
Patients attending NUH renal clinic who are receiving immunosuppressant treatment (tacrolimus) and require regular monitoring
Renal clinical team identify suitable patient participants and present patient information sheet to them
Patient does not consent to participate in study
Patient consents to participate in study
Consenting patients who are having venepuncture to measure tacrolimus and creatinine as per protocol will simultaneously have a finger-prick sample collected using the Mitra device.
Mitra-collected samples
Mitra device with collected samples are to be labelled with a sticker that contains instructions for Pathology Specimen Reception. Stickers will be provided by the investigators of the study.
Mitra device will be contained within the same specimen bag as the venepuncture collected samples
Samples will be sent to Pathology Specimen Reception utilising current hospital transport processes (pneumatic tube or direct delivery by hospital staff).
Upon receipt, samples will NOT be booked into the Laboratory Information Management System (WinPath). This is to ensure the results of the Mitra-collected samples are not communicated to the renal clinical team.
The sticker attached to the Mitra-device will instruct laboratory staff to label the Mitra samples with the same laboratory number as the venepuncture-collected samples. This way, when the samples are delivered to specialist chemistry for analysis, the staff can identify the venous blood sample with its corresponding Mitra-collected sample.
Once the laboratory staff have identified the corresponding samples, the Mitra-collected samples will be relabelled with an ID that has no patient identifiers associated with it. The ID that is given to each sample will be stored in a password-protected Excel spreadsheet accessible only by the investigators of the study, and will be associated to the original laboratory number.
Samples will be analysed using an LC-MS/MS method designed for finger-prick collected samples. Results of the analysis will be transferred onto a password-protected spreadsheet accessible only by the investigators of the study. No results are be transferred to any patient record as the ID is not associated to a patient.
Venepuncture-collected samples
Tacrolimus samples are collected in WB EDTA. Creatinine samples are collected using serum separating tubes. Both samples must be labelled with patient identifiers as per current NUH practice. Samples are contained within a specimen bag along with a request form.
Samples will be sent to Pathology Specimen Reception utilising current hospital transport processes (pneumatic tube or direct delivery by hospital staff).
Upon receipt, samples will be booked into the Laboratory Information Management System (WinPath) and labelled with a unique laboratory number.
Samples delivered to specialist chemistry for analysis. Analysis will be carried out as per current NUH practice.
Results of the test are transferred to the electronic patient record.
Method comparison
Results of the Mitra-device collected samples and venepuncture collected samples will be compared. The data collected will be analysed using the 'analyse-it' statistics software. Research data (excluding patient details) will be stored in the Laboratory Quality Management System (Q-Pulse).
Results are transferred onto a password-protected spreadsheet accessible only by the investigators of the study. All data will be depersonalised. No patient identifiers will be taken from the electronic patient record – only the sample number, tacrolimus result, and creatinine result will be required.
Results are communicated to the renal clinical team as per current NUH protocol. This study will not have any impact on the delivery of care to the patient.

Results

Please see Flowchart for details of participant flow.

Patients will be recruited and consented by the renal clinical team. Patient Information leaflets will be distributed to all volunteers and consent forms signed by the participants or parents/carers for paediatric patients. 50 patients will be included in the study and the results from the venous blood and capillary-finger prick samples compared.

The results from the venous blood samples will be processed and reported using the normal laboratory processes (electronic transfer of results from the laboratory LIMS system to the electronic patient record), ensuring there is no delay in receipt of routine results. As the results from the finger-prick blood samples collected using the Mitra device will not be reported there will be no change to tacrolimus dosing regimens based on these results. There are no anticipated adverse events expected.

Results of the Mitra-device collected samples and venepuncture collected samples will be compared. The data collected will be analysed using the 'Analyse-It' statistics software and Bland-Altman and Passing-Bablok regression analyses will be completed to identify any statistical differences between the two LC-MS/MS methods. Research data (excluding patient details) will be stored in the Laboratory Quality Management System (Q-Pulse).

Interpretation

Results of the Mitra-device collected samples and venepuncture collected samples will be compared. The data collected will be analysed using the 'Analyse-It' statistics software and Bland-Altman and Passing-Bablok regression analyses will be completed to identify any statistical differences between the two LC-MS/MS methods. Research data (excluding patient details) will be stored in the Laboratory Quality Management System (Q-Pulse).

The statistical analysis of the results will be compared to those previously reported by the Wythenshawe Laboratory (Marshall et Al. 2020) who have also introduced a finger-prick capillary collection method for tacrolimus and creatinine. The outcome of this study will be disseminated to the renal clinical team. If no statistically significant difference is identified between the methodologies, it is anticipated that the Mitra-devices could be utilised for remote blood collection and samples sent to the laboratory via postal services for tacrolimus and creatinine measurement. This would reduce the number of outpatient appointments required for patients requiring therapeutic drug monitoring.

References

Marshall, D. J., Kim, J. J., Brand, S., Bryne, C., & Keevil, B. G. (2020). Assessment of tacrolimus and creatinine concentration collected using Mitra microsampling devices. Annals of Clinical Biochemistry, 57(5), 389-396