ANALISI E DISCUSSIONE DELLE RISPOSTE VEQ IMMUNOSOPPRESSORI CICLO 2018 – CDT CICLO 2018

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Laboratorio Generale
Settore Farmacotossicologia

Azienda Ospedaliero Universitaria Careggi

VEQ CICLO 2018 – RISULTATI 4 Chimica Clinica ciclo 2019 Etanolo, Ammonio, Farmaci, Farmaci immunosoppressori, Droghe abuso ciclo 2018

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CENTRO REGIONALE DI RIFERIMENTO

S.O.D. Sicurezza e Qualità in Laboratorio

Dal 2006 il centro è in possesso della certificazione secondo lo standard di riferimento UNI EN ISO 9001:2008 "Sistemi di gestione per la qualità - Requisiti" che nel 2018 ha aggiornato il suo sistema di qualità secondo la nuova versione della UNI EN ISO 9001:2015 "Sistemi di gestione per la qualità - Requisiti"

Visto l'importanza che i programmi V.E.Q. hanno acquisito in questi ultimi anni, il Centro ha visto la necessità di garantire la qualità dei programmi V.E.Q. gestiti, mediante l'Accreditamento secondo lo standard internazionale specifico di Riferimento UNI CEI EN ISO/IEC 17043 :2010 "Requisiti generali per le prove valutative interlaboratorio" con certificato di accreditamento PTP n° 0013 P rilasciato da ACCREDIA per 5 programmi, in data 26/11/2016.

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Attualmente i programmi ACCREDITATI secondo la UNI CEI EN ISO/IEC 17043 :2010 gestiti dal centro sono:

- Batteriologia
- Chimica clinica
- CDT
- Etanolo e Ammonio
- Farmacologia 1
- Farmacologia 2
- G6PDH
- HbA1c
- Immunosoppressori
- Marcatori Cardiaci
- Peptidi natriuretici
- Proteine specifiche
- Sangue occulto
- Coagulazione 1
- Parassitologia
- Ematologia
- Reticolociti
- Micobatteri

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IMMUNOSOPPRESSORI

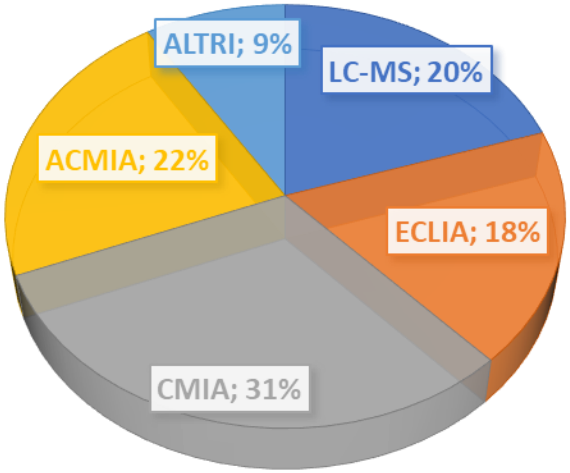
Campioni

Emolisati di origine umana liofilizzati per la detrminazione di Ciclosporina e Tacrolimus.

Il programma prevede l'invio di 8 campioni in 2 spedizioni.

AL PROGRAMMA 2018 HANNO PARTECIPATO
58 LABORATORI

CICLOSPORINA



Result Statistics	Unit	Number of Results
CMIA - Architect	µg/L	59
ECLIA - Roche	µg/L	41
EMIT	µg/L	7
CEDIA	µg/L	18
ADVIA	µg/L	6
ACMIA	µg/L	24
HPLC-MS	µg/L	123

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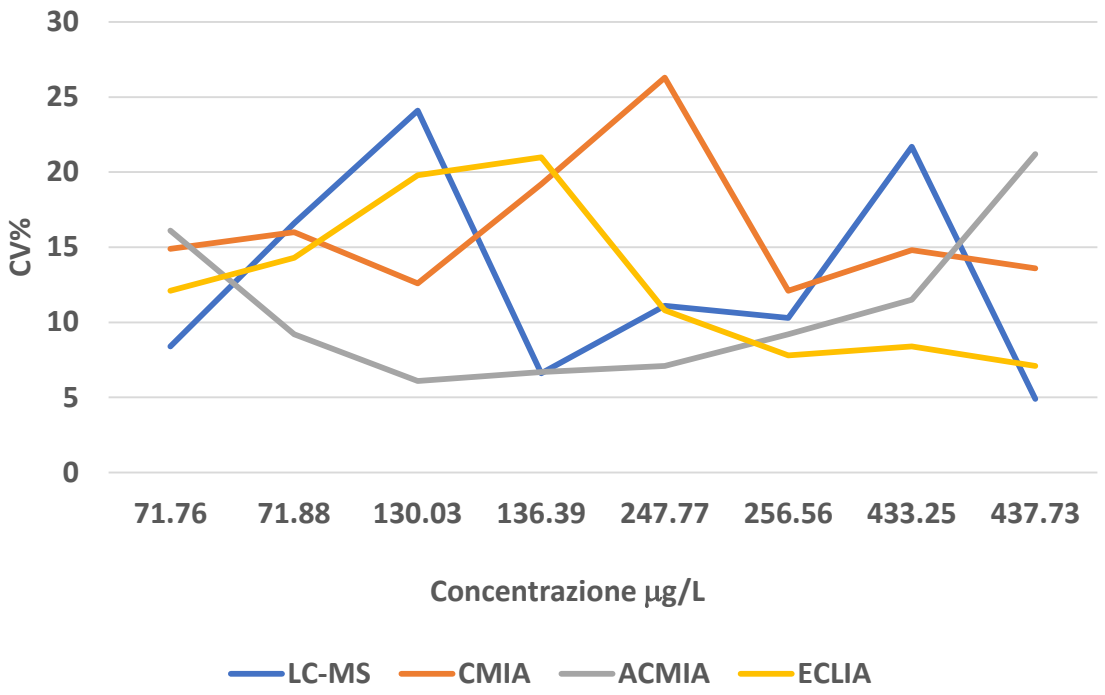
PROFILO IMPRECISIONE CICLOSPORINA



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Analita: *CICLOSPORINA* µg/L

	N.	Out	Media	C.V.	S.D.	Med.na
Tutti	55	1	433.25	14.19	61.5	424.00
Tuo Metodo	11	0	468.66	21.71	101.7	449.10

Campione	5 *(Scad. 09/07/2018)*
Tuo risultato	755.0

	Diff. S	Diff. %
Tutti	5.23	74.26
Tuo Metodo	2.82	61.10

Valutazione errore totale

1	2	3	4	5	6	7	8
O	O	O	O	X			

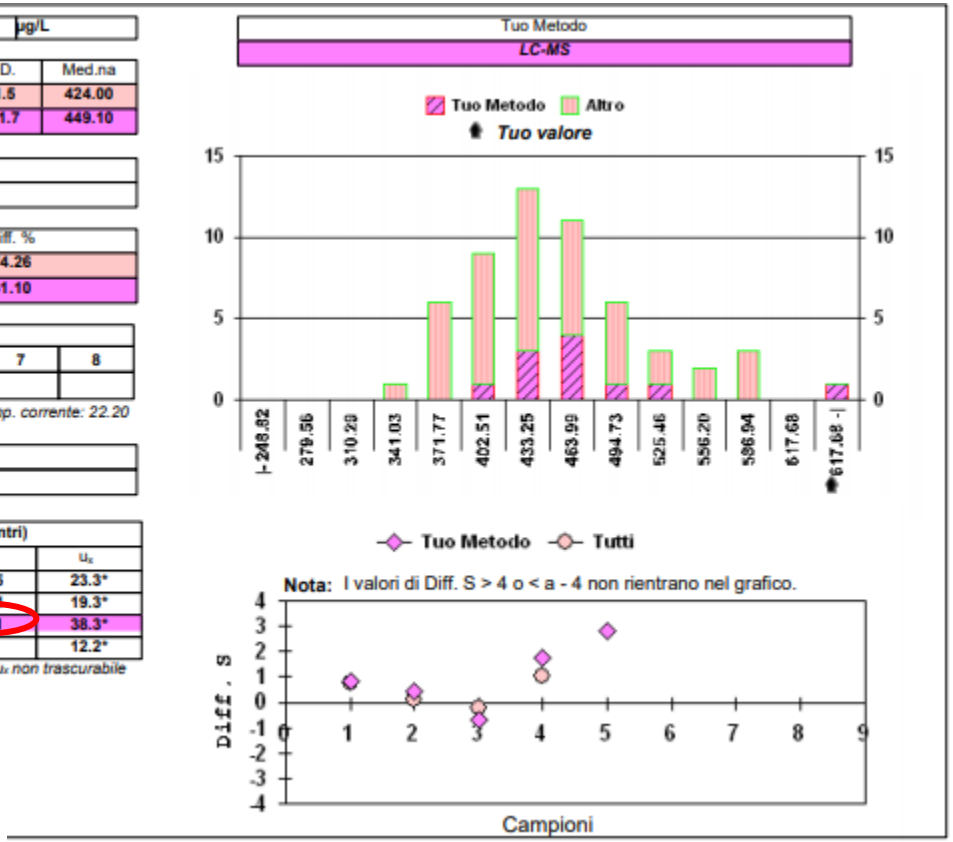
O = Interno X = Esterno rispetto ai L.A *L.A. camp. corrente: 22.20*

N. risultati numerici	55
N. risultati semiquantitativi/qualitativi	

Riepilogo x Metodo risultati numerici (> 7 Centri)

Metodo	N.	Out	M.	C.V.	u_x
CMIA	13	0	451.95	14.85	23.3*
ACMIA	12	0	464.07	11.51	19.3*
LC-MS	11	0	468.66	21.71	38.3*
ECLIA	10	0	365.27	8.44	12.2*

* u_x non trascurabile



numero risultati aberranti, evidenziati con le 2 seguenti iterazioni:

1) Eliminazione dei dati che non rientrano nel range "Mediana ± 80% valore Mediana";

2) Calcolo della media e S.D. dei dati rimanenti ed eliminazione dei dati che non rientrano nel range

"Media ± 3 S.D."

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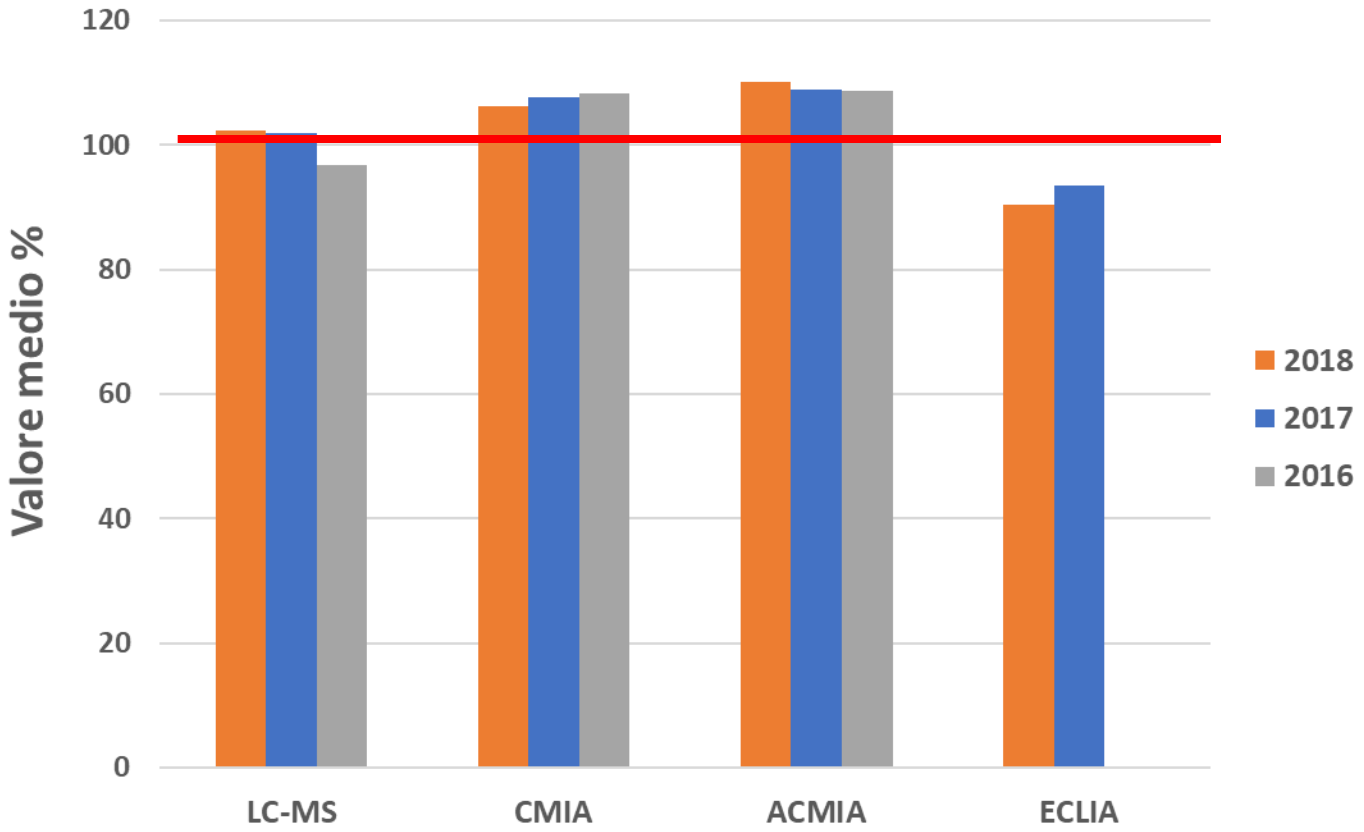

Il valor medio percentualizzato rispetto alla media di consenso, essendo necessariamente riferito alla media generale di tutti i metodi, rappresenta il comportamento dell'inaccuratezza di un gruppo.

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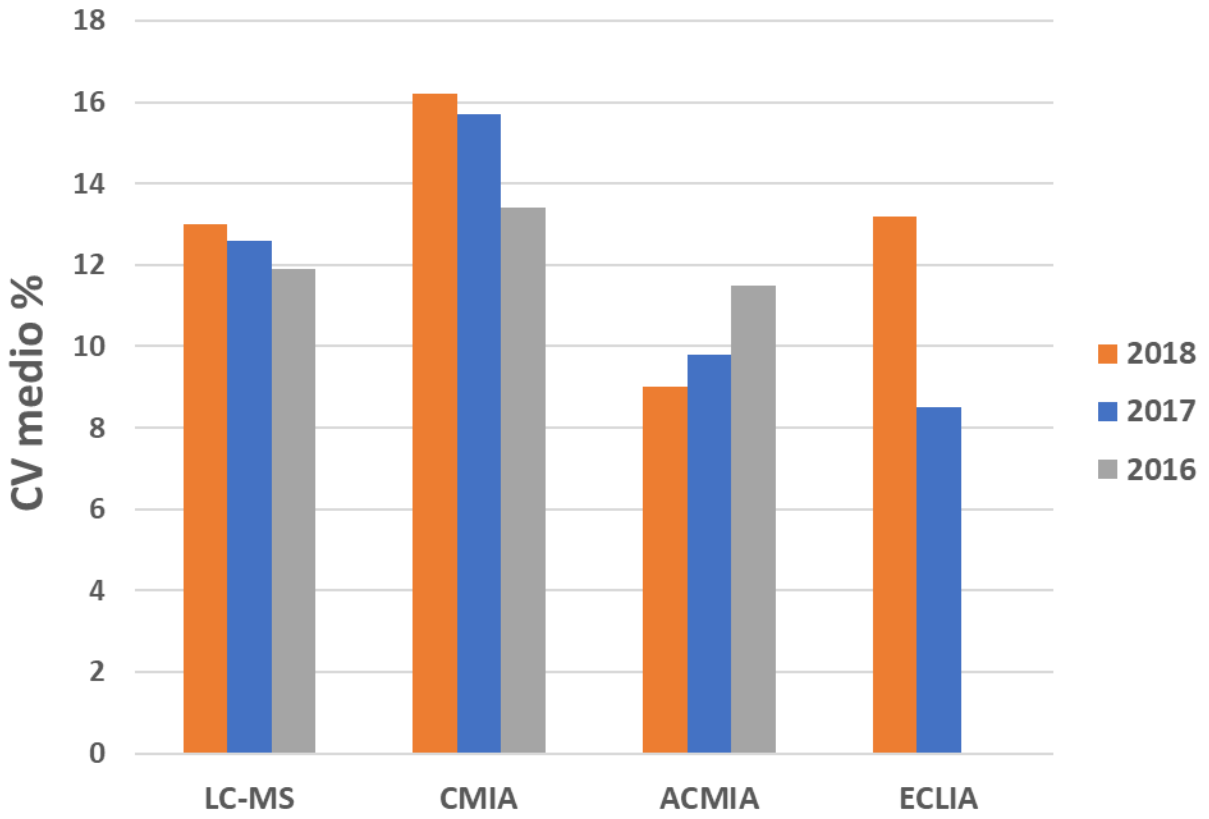
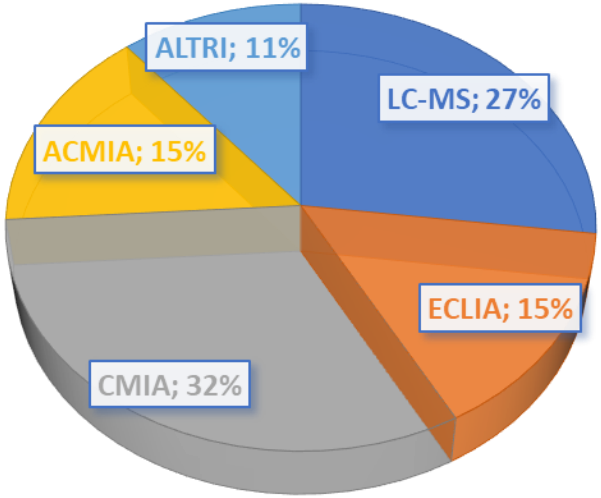


TACROLIMUS



Result Statistics	Unit	Number of Results
CMIA - Architect	µg/L	84
ECLIA - Roche	µg/L	31
EMIT	µg/L	7
QMS	µg/L	8
HPLC-MS	µg/L	135
ACMIA	µg/L	21

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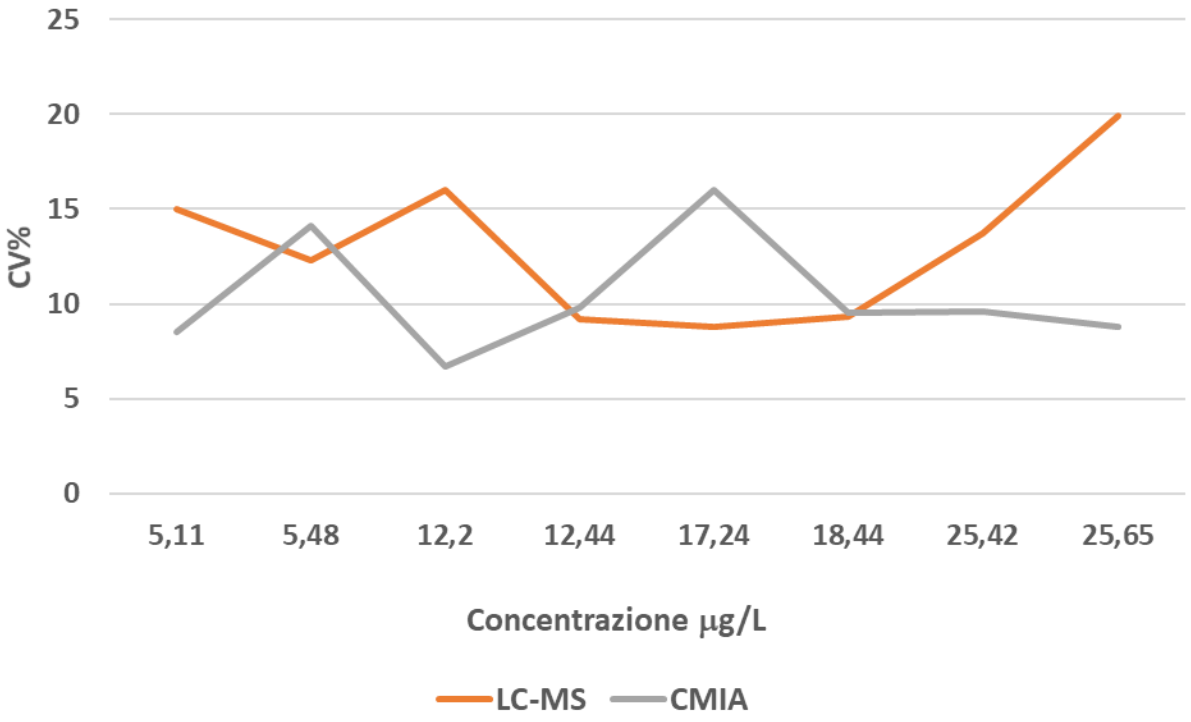
PROFILO IMPRECISIONE TACROLIMUS



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Analita: TACROLIMUS	N.	Out	Media	C.V.	S.D.	Med.na
						µg/L
Tutti	41	1	12.20	13.95	1.7	12.05
Tuo Metodo	11	0	12.14	16.05	1.9	11.70

Campione	5 (Scad. 09/07/2018)
Tuo risultato	17.8

	Diff. S	Diff. %
Tutti	3.29	45.90
Tuo Metodo	2.98	46.62

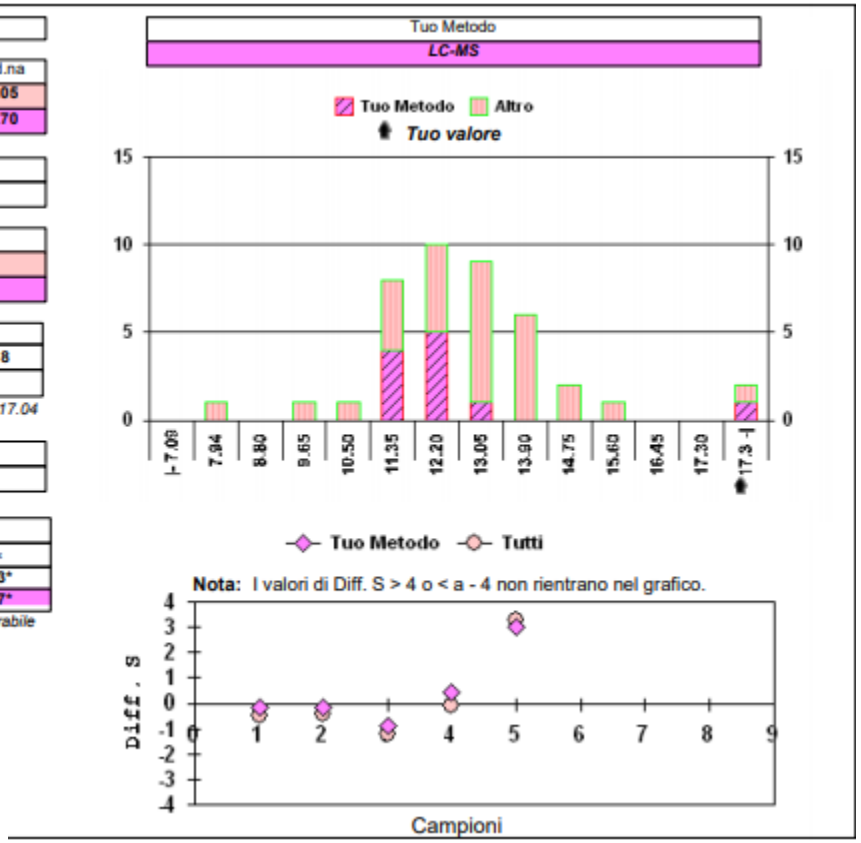
Valutazione errore totale							
1	2	3	4	5	6	7	8
O	O	O	O	X			

O = Interno X = Esterno rispetto ai L.A. L.A. camp. corrente: 17.04

N. risultati numerici	41
N. risultati semiquantitativi/qualitativi	

Riepilogo x Metodo risultati numerici (> 7 Centri)					
Metodo	N.	Out	M.	C.V.	u_x
CMIA	13	0	12.80	6.71	0.3*
LC-MS	11	0	12.14	16.05	0.7*

* u_x non trascurabile



Diff% (ET) >L.A.

crocetta rossa, indica che il risultato non è rientrato nei L.A.

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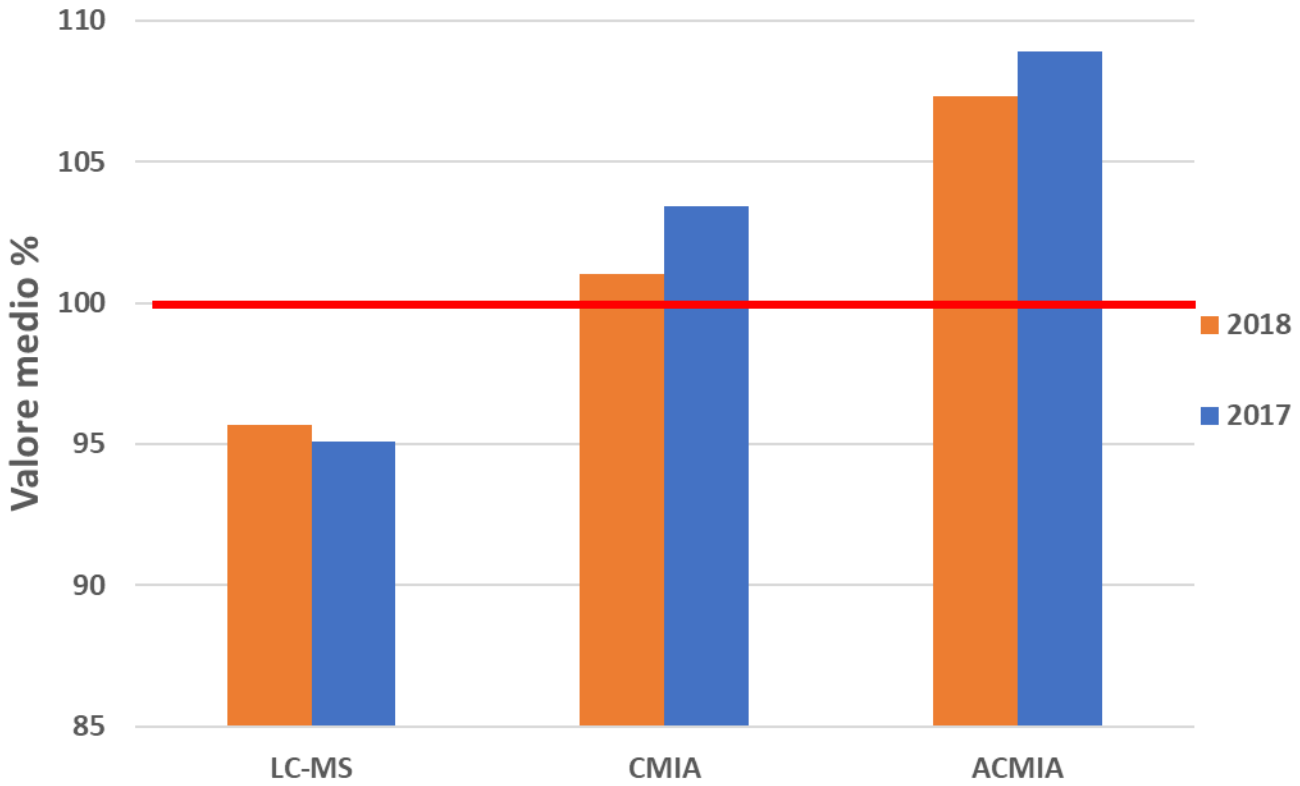

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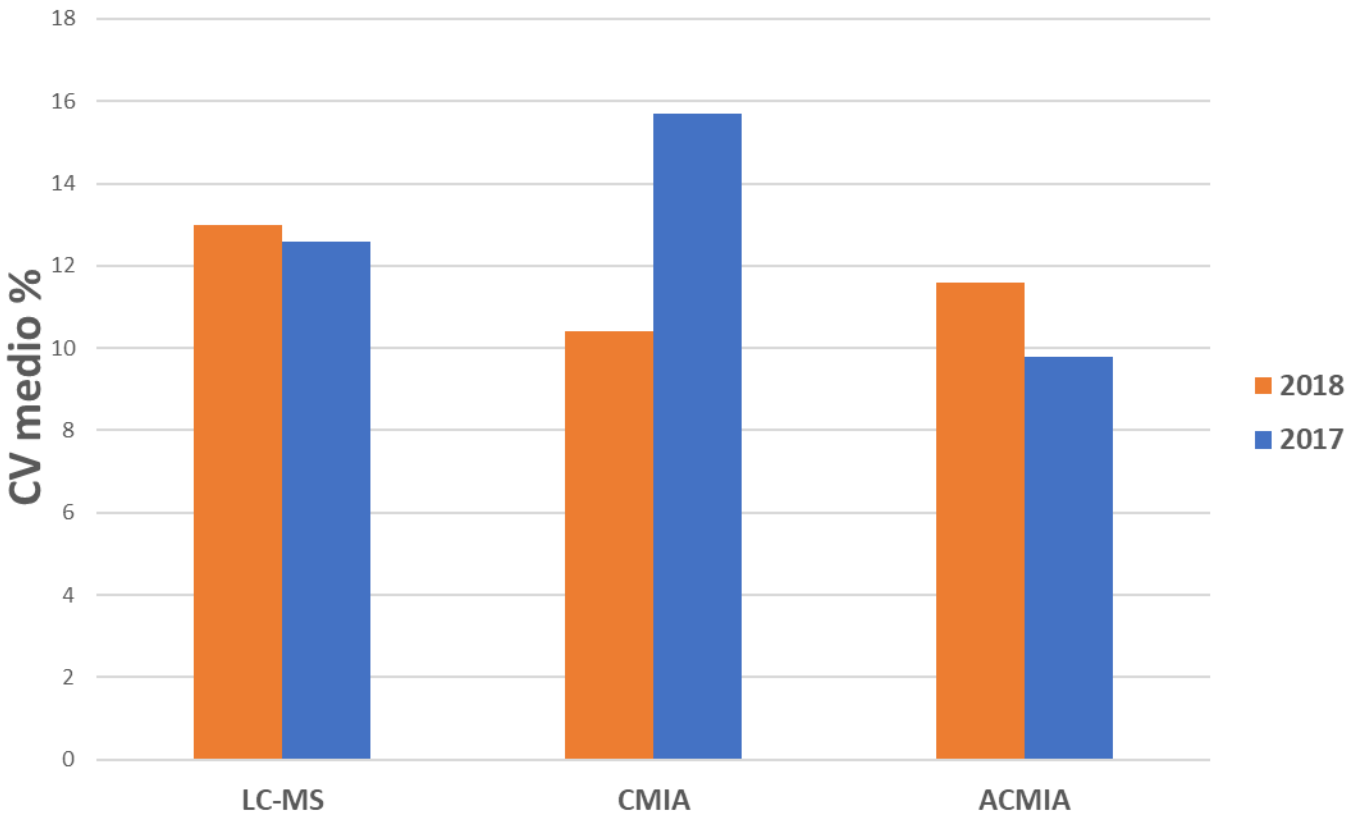

IMPORTANZA TDM IMMUNOSOPPRESSORI

- ✓Alterazioni nell'assorbimento
- ✓Stretto intervallo terapeutico (livelli terapeutici vicini a livelli tossici)
- ✓Tossicità a concentrazioni elevate
- ✓Interazioni tra farmaci in politerapia (induzione o inibizione enzimatica)
- ✓Patologie del paziente che alterano la farmacocinetica del farmaco (renali, epatiche)
- ✓Scarsa aderenza alla terapia.
- ✓Disfunzione genetica

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CICLOSPORINA

Time Post Transplant (Months)	Cyclosporine Trough Concentration (ng/mL) Tandem Mass Spectrometry Assay	Cyclosporine C_2 Concentration (ng/mL) Tandem Mass Spectrometry Assay
ADULT Kidney and Kidney Pancreas Transplant Recipients (Oct 2014)		
Less than 1	300 to 350	1300
1 to 2	250 to 300	1100
3 to 6	150 to 250	800 to 900
7 to 12	125 to 200	700
Greater than 12	75 to 125	450-600
PEDIATRIC Kidney Transplant* Recipients (Oct 2014)		
Less than 1	200 to 250	Not used
1 to 2	150 to 200	Not used
2 to 3	100 to 150	Not used
Greater than 3	80 to 100	Not used
**as per Dr. Matsell November 1 2012*		
ADULT Liver Transplant Recipients (Dec 2014)		
0 to 3	250 to 275	750 **
3 to 6	200 to 250	600 **
6 to 9	150 to 200	450 **
9 to 12	125 to 150	450 **
Greater than 12	100 to 125	450 **
** **Cyclosporine C_2** is not routinely used in liver transplant recipients		
ADULT Lung Transplant Recipients (Nov 2014)		
Less than 1	275 to 300	Not used
1 to 3	250 to 275	Not used
3 to 6	200 to 250	Not used
6 to 12	150 to 200	Not used
Greater than 12	125 to 150	Not used
Greater than 12 month with decrease in renal function	Switch to tacrolimus if possible	

Time Post Transplant (Months)	Cyclosporine Trough Concentration (ng/mL) Tandem Mass Spectrometry Assay	Cyclosporine C_2 Concentration (ng/mL) Tandem Mass Spectrometry Assay
ADULT Heart Transplant * (Nov 2014)**		
When eGFR is greater than 45 mL/min/1.73 m^2		
Less than 1 month	Not used	1200 to 1400
2 to 3 months	Not used	1000 to 1200
4 to 5 months	Not used	800 to 1100
6 to 12 months	Not used	700 to 1000
12 to 24 months	Not used	600 to 800
Greater than 24 months	Not used	400 to 600
When eGFR is less than 45 mL/min/1.73 m^2		
Less than 1 month	Not used	1000 to 1200
2 to 3 months	Not used	800 to 1100
4 to 5 months	Not used	700 to 900
6 to 12 months	Not used	600 to 800
12 to 24 months	Not used	400 to 600
Greater than 24 months	Not used	300 to 400
Patients Transplanted Greater Than 15 Years Ago		
0 to 3 months	300 to 350	Not used
3 to 6 months	200 to 300	Not used
6 to 12 months	150 to 250	Not used
Greater than 12 months	100 to 150	Not used
*** MOTOWN study used immunoassay not tandem mass spectrometry assay to analyze cyclosporine concentrations		

BC TRANSPLANT Provincial Health Services Authority

Gennaio 2019

TACROLIMUS

Time Post-Transplant (Months)	Tacrolimus Trough Blood Concentration (ng/mL) 12 hours Post-Dose Tandem Mass Spectrometry Assay
ADULT Kidney and Kidney/Pancreas Transplant Recipients (Oct 2014)	
Less than 1	8 to 12
1 to 3	6 to 9
Greater than 3	4 to 8
PEDIATRIC Kidney Transplant Recipients (Oct 2014)	
Month 1	10 to 12
Month 2 and 3	8 to 10
Month 4, 5 and 6	6 to 8
After Month 6	4 to 6
ADULT Liver Transplant Recipients* (Dec 2014) Individual situations may vary. Please contact Vancouver General Hospital Transplant Clinic if you have any questions.	
Less than 1	6 to 9*
1 to 3	4 to 8*
Greater than 3	4 to 6*
Greater than 12	3 to 5*
*** for patients with renal dysfunction aim for the lower therapeutic target**	
ADULT Lung Transplant Recipients (Nov 2014)	
0 to 3	10 to 12
4 to 12	8 to 10
Greater than 12	6 to 8
Greater than 12 months with eGFR < 50 mL/min/1.73 m^2	4 to 6
ADULT Heart Transplant Recipients (Nov 2014)	
Less than 3	9 to 12
3 to 6	8 to 9
6 to 12	6 to 8
Greater than 12	4 to 8

B) Drugs that **INCREASE** CSA/TAC/ Sirolimus levels			
Antimicrobial: ▪ erythromycin, clarithromycin (Biaxin®) ▪ azole antifungals (fluconazole, itraconazole, voriconazole)	↓ CSA/TAC/sirolimus metabolism, ↑ rate of absorption, ↓volume of distribution • delayed / major ↓CSA/TAC/sirolimus	↑ CSA/TAC/sirolimus levels, ↑ risk of toxicity	Monitor CSA/TAC/sirolimus levels following addition, dose change or discontinuation. Monitor serum creatinine
Antidepressar fluoxetine, flu greater than s venlafaxine, r paroxetine			
Cardiovascula ▪ diltiazem, ▪ amiodaro			

Drug A) Drugs that **DECREASE** CSA/TAC/ Sirolimus levels	Possible Mechanism / Onset and severity	Adverse Effects	Management
Anticonvulsants: ▪ phenytoin ▪ carbamazepine ▪ phenobarbital, primidone	Enzyme induction ↑ CSA/TAC/sirolimus metabolism • delayed / major • delayed/ moderate • delayed / major	↓ effectiveness of CSA/TAC/sirolimus which may lead to rejection	↑ CSA/TAC/sirolimus dose by 30% and monitor levels following addition, dose change or discontinuation.
Antimicrobial: ▪ rifampin ▪ caspofungin **(tacrolimus ONLY)**	Induction of hepatic enzymes • delayed / major Mechanism is unknown • delayed/ moderate	Same as above	Monitor CSA/TAC/sirolimus levels following addition, dose change or discontinuation. Monitor tacrolimus level closely when caspofungin is initiated or dose changes and when caspofungin discontinued.

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Clinical Chemistry 59:11
1630–1637 (2013)

Drug Monitoring and Toxicology

Standardization of LC-MS for Therapeutic Drug Monitoring of Tacrolimus

Thomas M. Annesley,[1*] Denise A. McKeown,[2] David W. Holt,[2] Christopher Mussell,[3] Elodie Champarnaud,[3] Leonie Harter,[4] Lisa J. Calton,[4] and Donald S. Mason[5]

Analytical Services International

assay to assess whether interlaboratory imprecision could be improved through standardization of LC-MS analysis. Further, we compared the results obtained us-

described (*4*). Briefly, 10 patient pools (target 0–25 ng/mL) and 10 tacrolimus-supplemented samples (target 2–25 ng/mL) were prepared in duplicate. The tacrolimus-supplemented samples were prepared in EDTA anticoagulated human whole blood from Biological Specialty. Tacrolimus hydrate (99.7% purity) was purchased from Enzo Life Sciences. Pooled patient samples were from kidney transplant patients receiving tacrolimus. The samples were pooled from the volumes remaining after routine therapeutic drug monitoring and would otherwise have been discarded. The resulting 40 samples were randomized and distributed blindly to the participating centers. Four of the patient pools were prepared in sufficient volume (approximately 20 mL) to allow for value assignment by an exact-matching isotope dilution mass spectrometry (EM-IDMS) method (the reference measurement procedure) at LGC.

Il solo criterio di inclusione nello studio è quella di avere la stessa piattaforma strumentale e lo stesso kit commerciale

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SAMPLE ANALYSIS

Samples were analyzed initially at ASI by an LC-MS method used for value assignment of samples distributed by the International Proficiency Testing Scheme for tacrolimus. This method has previously been shown to be in good agreement with the EM-IDMS method used at LGC *(4)*.

Participating laboratories analyzed the samples in the test panel as unknowns, following the directions for use that accompany the test kits, and as previously described *(5)*.

DATA ANALYSIS

Data from the participating laboratories were used to estimate interlaboratory imprecision. The data were also compared to the ASI method as an initial measure of accuracy. Accuracy was further evaluated for a subset of the patient pools by comparing the mean concentrations from the 7 laboratories to the EM-IDMS method.

Table 1. Test method assay imprecision with tacrolimus proficiency panel.

	Abbott Architect, 17 sites[a]			MassTrak LC-MS Assay, 7 sites				
Panel	Tacrolimus[b,c]	Mean[c]	CV, %	Tacrolimus[b,c]	Mean[c]	CV, %	Minimum[c]	Maximum[c]
Supplemented samples								
	2.0	1.9	9.3	2.0	2.0	12.2	1.6	2.3
	4.0	4.1	6.0	4.0	4.0	7.4	3.5	4.3
	6.0	6.3	5.1	6.0	5.9	6.6	5.3	6.3
	8.0	8.3	6.3	8.0	7.5	7.8	6.4	8.1
	10.0	10.4	6.5	10.0	9.8	6.1	9.1	10.8
	15.0	15.8	5.8	15.0	15.1	3.7	14.4	16.0
	20.0	20.9	6.9	20.0	19.9	6.1	18.4	21.7
	25.0	26.0	6.7	25.0	25.2	6.5	24.0	28.8
Patient pools								
	2.9	2.9	7.9	2.6	2.8	4.6	2.5	2.9
	4.5	4.8	5.8	4.5	4.6	5.0	4.4	5.1
	6.4	6.7	6.2	6.6	6.9	5.3	6.5	7.7
	8.5	9.1	6.1	7.8	8.0	4.4	7.4	8.5
	10.4	10.8	3.9	10.0	10.3	4.1	9.8	11.0
	12.2	12.8	4.9	12.1	12.4	5.1	11.6	13.4
	13.1	13.5	9.5	14.2	15.1	4.9	14.2	16.3
	16.1	16.8	5.8	17.1	17.5	2.2	16.9	17.9
	20.2	21.5	5.9	20.7	20.7	5.4	19.4	22.7
	26.5	27.1	6.9	24.2	25.6	2.0	25.1	26.7
Mean CV, %			6.4			[illegible]		

[a]The data for the Architect assay are from Levine et al. *(4)*.
[b]The concentration for the supplemented samples is the amount of tacrolimus added to blank whole blood. The concentration for each pool from patients receiving tacrolimus was quantified by the validated liquid chromatography–tandem mass spectrometry method at ASI. Samples were tested in duplicate by each method.
[c]Concentrations expressed as nanograms per milliliter.

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Table 2. Comparisons of the MassTrak LC-MS method to ASI LC-MS and EM-IDMS methods.

Panel	Ma
P-02	
P-04	
P-06	
P-09	

[a]Measured values are expr
[b]Concentrations are expres
[c]Between MassTrak (MT) L
[d]EM-IDMS values are expre
procedure, including the ur
[e]Between MassTrak (MT) L

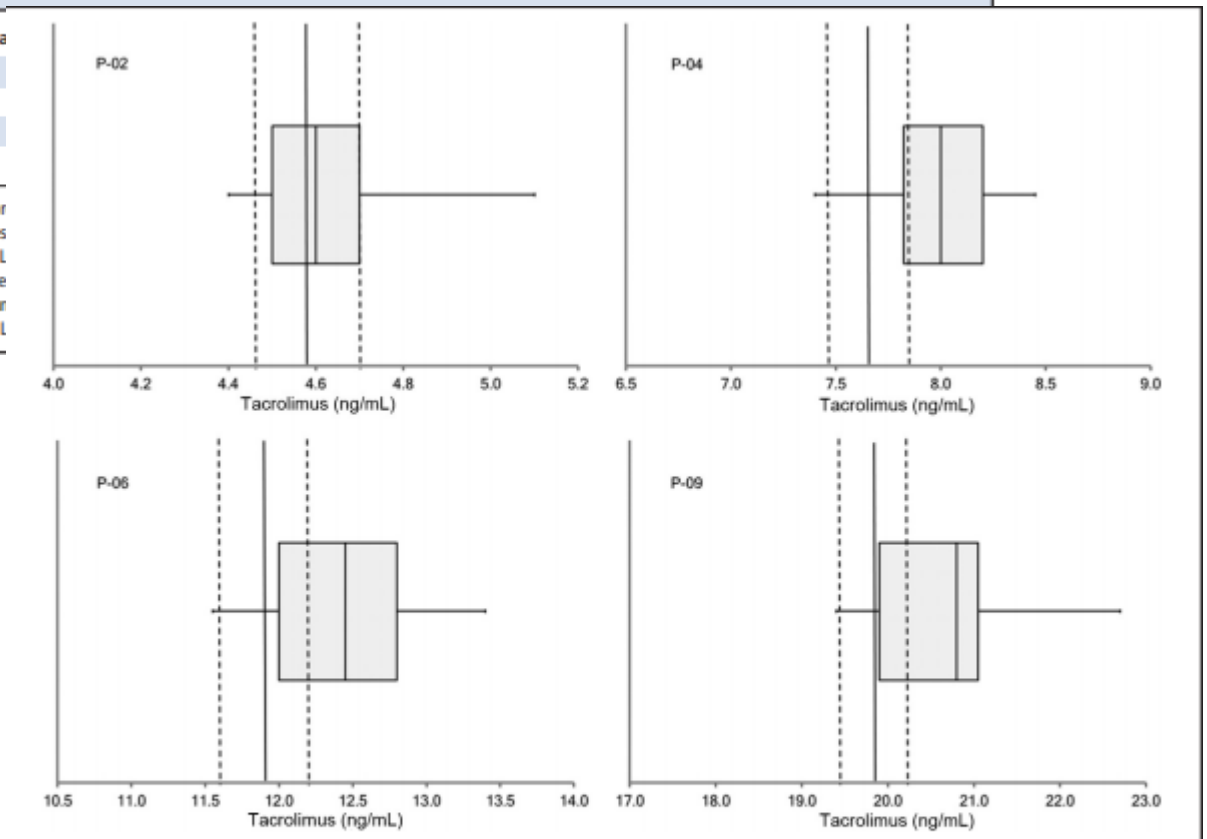

Fig. 1. Box-and-whisker plots for patient pools P-02, P-04, P-06 and P-09.

The left boundary of the box represents the first quartile of results, the right boundary represents the third quartile of results, and the line intersecting the box represents the median value of results from the 7 laboratories. The whiskers represent the lowest and highest values returned by any laboratory. For comparison, the reference values (solid line) and associated measurement uncertainties (dashed lines) for the EM-IDMS method are overlaid.

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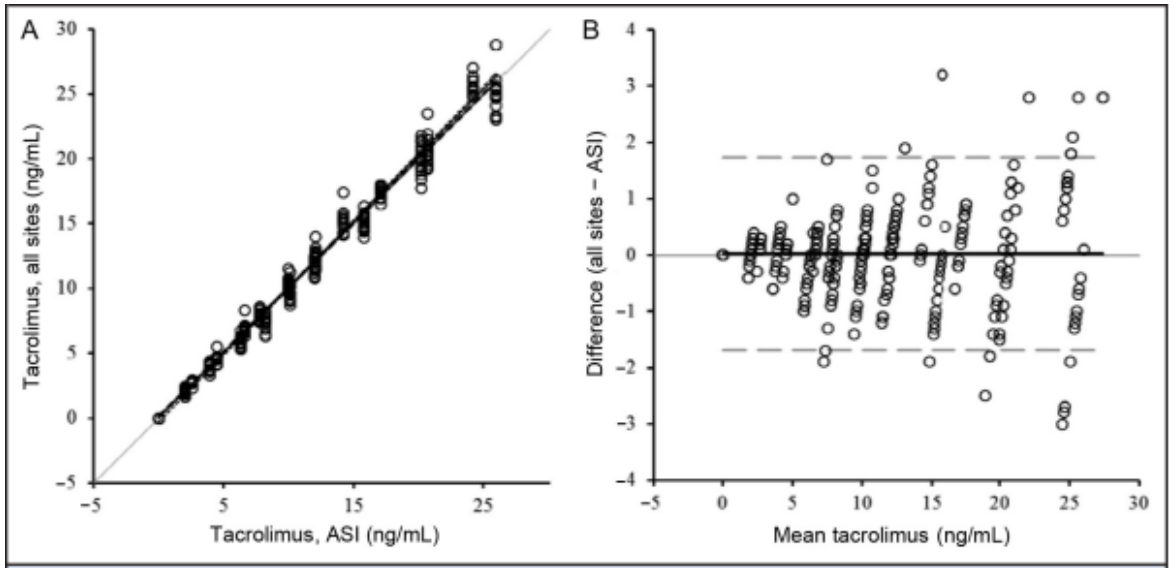

Fig. 2. Passing–Bablok regression (A) and Bland–Altman bias estimation (B) plots comparing the combined data for the 7 sites using the MassTrak assay to data obtained with the assay in use at ASI.

For the regression plot (A), the gray line is the line of identity, the black line the Passing–Bablok fit ($y = 1.02x - 0.02$, $r =$ 0.99), and the dashed lines the 95% CI bands. For the Bland–Altman plot (B), the gray line represents zero bias, the black line the bias (0.03 ng/mL), and the dashed lines the 95% limits of agreement (−1.69 to 1.74 ng/mL).

The results of our study demonstrate that the standardization of key analytical variables (calibration materials, sample pretreatment protocols, and chromatography) in the LC-MS analysis of tacrolimus yields highly reproducible tacrolimus measurements across laboratories. This standardization was

been ill-defined to date (*2*). We believe this to be the first study to demonstrate standardization of an LC-MS assay for tacrolimus across multiple laboratories, and it could provide a model for future studies that aim to quantify the impact of individual parameters that contribute to the variability in LC-MS assays. Of course, standardization is not a substitute for good laboratory practices such as internal assay validation and system performance checks, which are also key elements for generating accurate and precise results.

Assuring the Proper Analytical Performance of Measurement Procedures for Immunosuppressive Drug Concentrations in Clinical Practice: Recommendations of the International Association of Therapeutic Drug Monitoring and Clinical Toxicology Immunosuppressive Drug Scie

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Elaine M. Billaud, PharmD, PhD,§
*Mercè Brunet, PhD,** Paweł K.*
Loralie J. Langman, PhD,§§ Pierre
*Eberhard Wieland, MD,† and Pierre Wallemacq, PhD****

Whereas, as stated above, intralaboratory precision enhancement is in the hands of the individual laboratory, interlaboratory bias can only be addressed by an initiative focusing on the establishment of higher-order reference materials and/or higher-order reference procedures to ensure worldwide measurement traceability.[45–47] Such initiatives

Ther Drug Monit • Volume 38, Number 2, April 2016

drugs, including ISDs. Currently, only one ISD reference material—a commercially available whole-blood standard for tacrolimus (ERM-DA110a)—is listed in the JCTLM database. Some candidate reference methods have been published[59]; however, none has been reviewed and listed by the JCTLM. Consequently no reference measurement service is available to ensure the comparability of routine platforms through the establishment of a traceability chain.

the Web site of the Joint Committee for Traceability in Laboratory Medicine (www.bipm.org/jctlm, JCTLM).

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E’ STATO FATTO UN QUESTIONARIO CON 128 DOMANDE DIVISE IN 8 SESSIONI PER CERCARE DI IDENTIFICARE LE CAUSE DELLA GRANDE VARIABILITÀ INTERLABORATORIO NEI PROGRAMMI VEQ.

76 LABORATORI DI 14 PAESI HANNO PARTECIPATO

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- ✓ **TEMPI TRA RACCOLTA DEL CAMPIONE E ANALISI VARIANO DA 1H A 1 SETTIMANA**
- ✓ **CONTROLLO DELLA TEMPERATURA NEL TRASPORTO E STOCCAGGIO.**
- ✓ **PROCEDURE DI ESTRAZIONE MANUALE POCO CONTROLLATE E STANDARDIZZATE**
- ✓ **NON UTILIZZO NEI METODI LC-MS DI STANDARD INTERNI DE**
- ✓ **UT SP**
- ✓ **FC**

Taken together, 3 main reasons for variability were suggested from the survey results; a lack of standardization of analytical methods and sample testing practices; a lack of the use of appropriate reference materials, calibrators, and control samples; and inconsistency of regulatory requirements and the level of compliance to internationally accepted laboratory practice guidelines in different parts of the world.

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✓ DESIGN DEL METODO

Meaningful method design must meet clinical needs. The analysis frequency, time to report results, laboratory workflow issues, desired analytical range, and minimal requirements for assay precision (ie, derived from biological variation and the impact on clinical decision-making) are the cornerstones for measurement method selection. For ISDs,

✓ VALIDAZIONE DEL METODO O VERIFICA DEL METODO

✓ GESTIONE DEL CICLO DI VITA DEL METODO

The long-term consistency of the results generated with a method is of high importance in transplantation medicine. ISDs are used in life-long treatment of most transplant patients; individualized target ranges are usually established post-transplantation with a certain method (eg, LC-MS/MS at the transplantation center). Usually patients stay attached with their transplantation center for several years with visiting time intervals from some weeks to several months. Hence, the ISD-TDM platform must be stable over such times; any long-term inconsistency of results may negatively impact dosing decisions and the patient outcome. Therefore, a method life-cycle management should be established to guarantee that analytical performance documented during method validation is continuously reproduced.

Stable analytical performance over time is based on a robust assay performed by well-trained personnel on well-maintained instruments. Assay bias and assay imprecision are kept low by monitoring the assay while it is being performed and by following good laboratory practices. Measures to avoid calibration bias include participation in an external quality assurance program, the use of external commercial calibrators, QC materials (preferably from different manufacturers), and the availability of certified reference materials and reference methods for all ISDs.

A rigorous internal quality assurance program that includes both system suitability testing (control of temperature in different compartments of the instrument, signal accuracy, signal stability signal intensity, signal recording, retention times, etc.), and revalidation of critical analytical parameters for existing methods is strongly recommended. Laboratories should have established protocols for these procedures that should be conducted on a regular basis to ensure continuous fitness with regard to analytical specifications and clinical requirements. For instance, the intro-

Continuous education and training of TDM laboratory personnel is an integral part of ensuring a high level of analytical quality. Therefore, establishing programs to maintain an adequate educational and training level of the personnel involved in analysis and reporting or interpreting the results is strongly recommended. Finally, it should be reminded that the clinical effectiveness of TDM (of ISDs particularly) largely depends also on the respect of the sampling hours and of any preanalytical recommendations as correct sampling from catheter sytsems.[150,151] Continuous education should therefore ideally include the nursing staff and health care professionals.

DE GRUYTER

Diagnosis 2019; 6(1): 57–68

Review

Adrian Klak, Steven Pauwels and Pieter Vermeersch*

Preanalytical considerations in therapeutic drug monitoring of immunosuppressants with dried blood spots

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Keywords: dried blood spot; dried blood spot testing; drying time; filter paper; hematocrit; immunosuppressants; microsampling; preanalytical phase; stability; therapeutic drug monitoring.

Abstract

Dried blood spot (DBS) sampling consists of the deposition of a drop of capillary blood (preferably from the finger in adults) on filter paper by either the patient or a medical professional. The spot is then left to dry (drying times typically range from a couple of hours to overnight drying) and is shipped to the laboratory. After arrival at the laboratory, a sam is then followed by a of the sample [3]. D

Multiple studies have been published regarding the TDM of immunosuppressive agents in DBSs (most notably tacrolimus, sirolimus and everolimus), all of which concluded that DBS sampling is a valid alternative for venous sampling. However, most studies have been performed in controlled environments and were focused on analytical performance, often using venous sampling i.e. the application of a drop of ethylenediamine tetraacetic acid (EDTA) blood by a laboratory technician] instead of true capillary sampling. The impact of preanalytical factors specific to DBSs may be underestimated. In as many as 40% of published assays using DBSs for various analytes, not

FATTORI PREANALITICI

- ✓ TIPO DI CARTA ASSORBENTE
- ✓ OMOGENEITA' DELLO SPOT
- ✓ VOLUME DI SANGUE
- ✓ EMATOCRITO
- ✓ TEMPO DI ESSICCAMENTO
- ✓ STABILITA' DELL'ANALITA
- ✓ POSIZIONE DI «PUNCH»

Twenty-one methods and clinical validations were included [8, 10, 16–34]. In addition, three studies evaluating a single DBS-specific parameter in the context of immunosuppressants [15, 35] and three studies using TDM of tacrolimus in DBSs as a validated method for pharmacokinetic studies were included [9, 36, 37]. Two studies

Table 1: Influence of blood spot volume in method and clinical validations.

Study	Card type	Range, µL	Crit.	Drug	Effect of variation in blood spot volume
Koster et al. [32]	Whatman nr. 10535097	30–90	15%	TaC SiR EvE	No significant effect on analyte concentration
Sadilkova et al. [28]	Whatman 903	25–100	n/a	TaC SiR	No significant effect on analyte concentration
Den Burger et al. [25]	Whatman 903	20–100	15%	TaC SiR EvE	No significant effect on analyte concentration 20 µL spot and HC: approx. –17.5% bias 20 µL spot and LC: approx. –19% bias
Li et al. [30]	Whatman 903	15–50	15%	TaC	No significant effect on analyte concentration
Martial et al. [22]	Whatman 903	20–60	15%	TaC	No significant effect on analyte concentration
Knapen et al. [20]	Whatman 903	20–50	15%	EvE	At 20% Hct: difference of slope of calibration curve >15%
Koster et al. [21]	Whatman FTA DMPK-C	30–70	15%	TaC SiR EvE	No significant effect on analyte concentration

TaC, tacrolimus; SiR, sirolimus; EvE, everolimus; n/a, not available; crit., acceptability criterion; HC, high concentration; LC, low concentration.

Table 2: Influence of Hct in method and clinical validations.

Study	Card type	Hct range or mean, %	Crit	Drug	Hct effect on bias
Koster et al. [32]	Whatman nr. 10535097	20–50%	15%	TaC	No significant effect after correction for target Hct
				SiR	Hct 20% and HC: –20% bias
				EvE	Hct 20% and HC: –28% bias
Koster et al. [32]	Whatman nr. 10535097	Mean:	15%	TaC	No significant effect (no correction)
		Outpatient: 38.4%		SiR	
		Inpatient: 29.5%		EvE	
Sadilkova et al. [28]	Whatman 903	20–45%	n/a	TaC	No significant effect (no correction)
				SiR	
Den Burger et al. [25]	Whatman 903	22–41%	15%	TaC	No significant effect after correction for target Hct
				SiR	
				EvE	
Li et al. [30]	Whatman 903	23.2–48.6%	15%	TaC	No significant effect (no correction)
van Boekel et al. [9]	Whatman nr. 10535097	39%	–	TaC	No significant effect on correlation with venous blood (primary outcome)
Koster et al. [21]	Whatman FTA DMPK-C	23–53%	15%	Tac	No significant effect
				EvE	
				SiR	Hct 28% and LC: –15.1% bias
Knapen et al. [20]	Whatman 903	20–50%	15%	EvE	Hct 20% and HC: –18 to –21.9% bias
Martial et al. [22]	Whatman 903	15–50%	15%	TaC	Hct 15% and MC: –24% bias
					Hct 20% and MC: –16% bias

TaC, tacrolimus; SiR, sirolimus; EvE, everolimus; n/a, not available; Hct, hematocrit; crit., acceptability criterion; HC, high concentration; MC, medium concentration; LC, low concentration.

Tacrolimus was not significantly influenced by blood spot volume, Hct effects, blood spot inhomogeneity and drying times. Furthermore, tacrolimus was found to be sufficiently stable at low and high temperatures, as no significant degradation was noted after 5 days at 60°C. This lack of interference makes tacrolimus a viable candidate for DBS sampling, even in a home-based setting.

Sirolimus and everolimus were more prone to interference from the studied preanalytical factors. They exhibited significant bias in blood spots with low volume, although the clinical impact is minimal, as these spots would have been rejected due to insufficient filling. High temperatures significantly impacted the stability of these compounds at 60°C and 70°C, temperatures which can be reached in postal boxes during summer months.

Furthermore, sirolimus and everolimus were more susceptible to Hct-related effects than tacrolimus, as they exhibited variations in recovery (up to –26%) which were dependent on Hct, concentration and drying time. A significant Hct-dependent bias on concentration was also found at low Hct and high concentration. Although the

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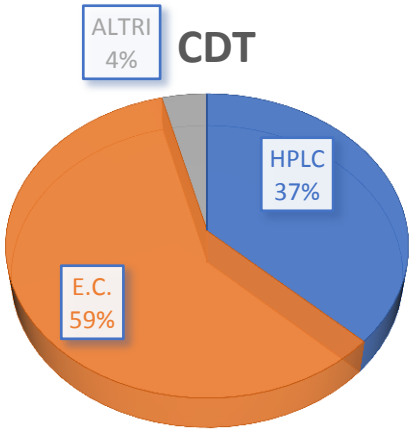
CDT

Campioni

Sieri di origine umana, liofili per la determinazione della CDT.
Il programma prevede 6 campioni inviati in 2 spedizioni.

AL PROGRAMMA 2018 HANNO PARTECIPATO 125 LABORATORI



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Campione	Metodo	Media	CV %
1	EC HPLC	4.0 4.2	12.5 12.8
2	EC HPLC	1.3 1.8	7.9 14.7
3	EC HPLC	1.4 1.9	12.4 18.9
4	EC HPLC	0.7 1.2	13.2 21.6
5	EC HPLC	1.9 2.4	10.1 15.1
6	EC HPLC	1.2 1.8	13.8 21.4

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IFCC approved HPLC reference measurement procedure for the alcohol consumption biomarker carbohydrate-deficient transferrin (CDT): Its validation and use☆

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