SCIEX
Diagnostics

# Topaz™ System
# Analytical Performance

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# Topaz™ System Analytical Performance Data Sheet

**Illustration of analytical performance for Testosterone, Androstenedione, Cortisone, Cortisol, 11-Deoxycortisol, Corticosterone, 17-Hydroxyprogesterone, DHEA, and Progesterone.**

The SCIEX Topaz LC-MSMS System is intended to identify inorganic or organic compounds in human specimens. All laboratory-developed tests must be developed, verified, and validated in accordance with applicable laws and regulations prior to their use for clinical diagnostic purposes.

This document describes a test of the analytical performance of the SCIEX Topaz LC-MS/MS System to analyze the compounds Testosterone, Androstenedione, Cortisone, Cortisol, 11-Deoxycortisol, Corticosterone, 17-Hydroxyprogesterone, DHEA, and Progesterone in serum matrix.

The analytical performance data presented here is for illustrative purposes only to demonstrate the potential capabilities of the system. Performance in individual laboratories may differ due to a number of factors, including system configuration, laboratory methods, and operator technique. This document does not constitute a warranty of merchantability or fitness for any particular purpose, express or implied, including for the testing of the compounds analyzed in this experiment.

## Materials and Methods

The Topaz LC-MS/MS System was controlled and data processed using ClearCore™ MD Software (ver. 1.1). Commercially available serum calibrators and quality controls (UTAK P/N 51400, 51401, 51402, 51403 & 51404) containing the compounds of interest were processed using the following conditions:

**Sample Preparation Conditions**
Protein precipitation of 100 µL sample with ZnSO4 in methanol, centrifugation and direct injection.

**Liquid Chromatography Conditions**
Column: Phenomenex® Kinetix ® C8
Mobile Phase A: Water
Mobile Phase B: 95% Methanol
Flow Rate: 0.6 mL/min
Injection Volume: 50 µL
Gradient: Linear, 10-100% B over 6.5 min
Retention Time: Compound dependent: 3.0-4.5 min

**Mass Spectrometry Conditions**
Method Duration: 6.5 min
Polarity Positive: APCI
Transitions: Compound Dependent
Source Conditions: Flow Rate-Optimized

## Results

Analytical performance statistics including the concentration range evaluated, precision experiments (n=6 replicates), and functional sensitivity are shown in Table 1. Chromatogram of the compounds evaluated utilizing the described method is shown in Figure 1. Calibration curves over the defined concentration ranges for each compound are illustrated in Figure 2.

| Compound (units) | Range | %CV (at LLOQ) | Functional Sensitivity* |
|---|---|---|---|
| Testosterone (ng/dL) | 10-1000 | 13% | 4.12 |
| Androstenedione (ng/dL) | 5-500 | 14.8% | 1.20 |
| Cortisone (µg/dL) | 0.06-6 | 8.2% | 0.0033 |
| Cortisol (µg/dL) | 0.025-25 | 17.5% | 0.0064 |
| 11-Deoxycortisol (ng/dL) | 4-400 | 13.5% | 0.77 |
| Corticosterone (ng/mL) | 10-1000 | 6.7% | 16.85 |
| 17OH-Progesterone (ng/dL) | 10-1000 | 13.9% | 2.18 |
| DHEA (ng/mL) | 1-10 | 9.6% | 0.41 |
| Progesterone (ng/mL) | 0.25-25 | 10.3% | 0.20 |

**Table 1.** Measured range, percent coefficient of variation (%CV) at lower-limit of quantitation (LLOQ), and functional sensitivity for each compound evaluated.

**Functional Sensitivity is defined here as the lowest concentration achieving a CV of 20%.*

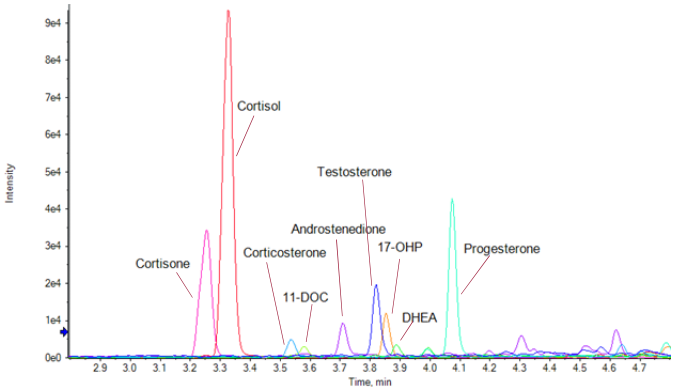


**Figure 1.** Chromatogram for all evaluated compounds using the Topaz System following the sample preparation and LC-MS/MS conditions.

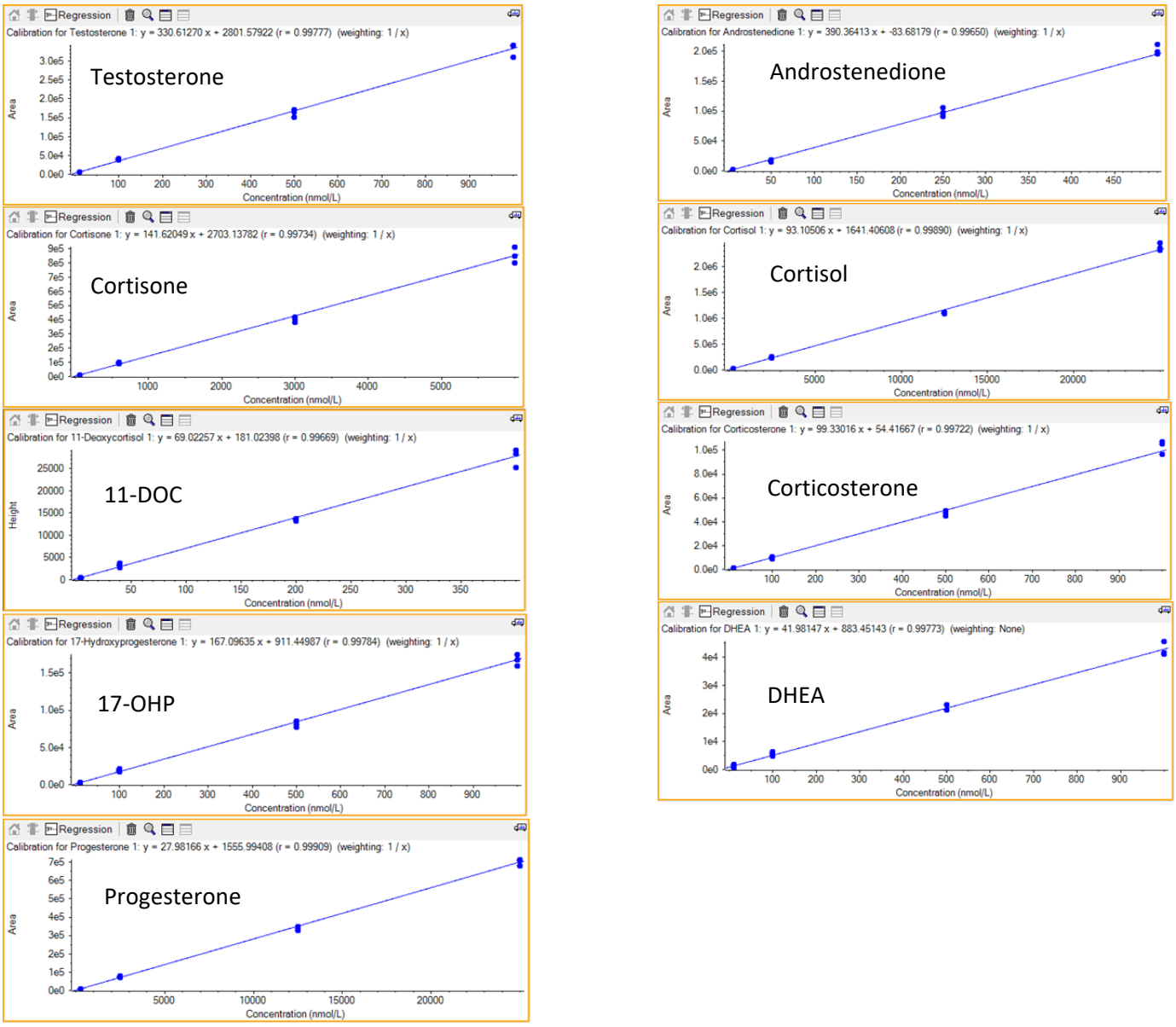


**Figure 2.** Calibration curves using ordinary least-squares regression and 1/x weighting for all the compounds evaluated over the defined concentration ranges in Table 1.

## Conclusions

Based on the above performance testing, the following results were obtained:

**Assay Linearity:** All compounds exhibited a linear dynamic range of at least 3 orders of magnitude.

**Reproducibility:** At each LLOQ, the precision (%CV) was <18% for all compounds evaluated as determined by n=6 replicates. CV data is based on calculated concentration without internal standard.

**Sensitivity:** Functional sensitivity, as defined by a CV of <20%, was <5 ng/dL for all compounds with the exception of Cortisol (6.4 ng/dL) and Corticosterone (16.85 ng/dL).

The Topaz System exhibited capability to deliver sensitive and reproducible analytical performance for Testosterone, Androstenedione, Cortisone, Cortisol, 11-Deoxycortisol, Corticosterone, 17-Hydroxyprogesterone, DHEA, and Progesterone in serum matrix.

# Topaz™ System Analytical Performance Data Sheet

## Illustration of Analytical Performance for Methamphetamine, Morphine, Benzoylecgonine, Methadone, Phencyclidine, Propoxyphene, and Methaqualone in Urine

The SCIEX Topaz LC-MS/MS System is intended to identify inorganic or organic compounds in human specimens. All laboratory-developed tests must be developed, verified, and validated in accordance with applicable laws and regulations prior to their use for clinical diagnostic purposes.

This document describes a test of the analytical performance of the SCIEX Topaz LC-MS/MS System to analyze the compounds Methamphetamine, Morphine, Benzoylecgonine, Methadone, Phencyclidine, Propoxyphene, and Methaqualone in urine matrix.

The analytical performance data presented here is for illustrative purposes only to demonstrate the potential capabilities of the system. Performance in individual laboratories may differ due to a number of factors, including system configuration, laboratory methods, and operator technique. This document does not constitute a warranty of merchantability or fitness for any particular purpose, express or implied, including for the testing of the compounds analyzed in this experiment.

## Materials and Methods

The Topaz LC-MS/MS system was controlled and data processed using ClearCore™ MD Software (ver. 1.1). Commercially available urine calibrators and quality controls (Bio-Rad P/N 438) containing all compounds of interest were processed using the following approach:

**Sample Preparation Conditions**
Dilution with water/Acetonitrile solution using 200µL sample, followed by direct injection

**Liquid Chromatography Conditions**
Column: Phenomenex® Kinetex® Biphenyl
Mobile Phase A: Water/0.1% Formic Acid
Mobile Phase B: Acetonitrile/0.1% Formic Acid
Flow Rate: 0.7mL/min
Injection Volume: 5µL
Gradient: Gradient 10-85%B over 7 min
Retention Time: Compound dependent 2.9-4.8 min

**Mass Spectrometry Conditions**
Method Duration: 7 min
Polarity: Positive Electrospray
Transitions: Compound Dependent
Source Conditions: Flow Rate-Optimized

## Results

Analytical performance statistics including the concentration range evaluated, precision experiments (n=6 replicates), and functional sensitivity are shown in Table 1. Chromatogram of the compounds evaluated utilizing the described method is shown in Figure 1. Calibration curves over the defined concentration ranges for each compound are illustrated in Figure 2.

| Compound (units) | Range | %CV (at LLOQ) | Functional Sensitivity* |
|---|---|---|---|
| Methamphetamine (ng/mL) | 0.75-750 | 15.3% | 1.99 |
| Morphine (ng/mL) | 1.5-1500 | 14.2% | 6.27 |
| Benzoylecgonine (ng/mL) | 0.225-225 | 16.3% | 85.14 |
| PCP (ng/mL) | 0.19-19 | 12.2% | 3.43 |
| Methadone (ng/mL) | 0.225-225 | 15.4% | 0.58 |
| Propoxyphene (ng/mL) | 2.25-225 | 13.9% | 7.81 |
| Methaqualone (ng/mL) | 0.225-225 | 12.3% | 0.11 |

**Table 1.** Measured range, percent coefficient of variation (%CV) at lower-limit of quantitation (LLOQ), and functional sensitivity for each compound evaluated.

**Functional Sensitivity is defined here as the lowest concentration achieving a CV of 20%.*

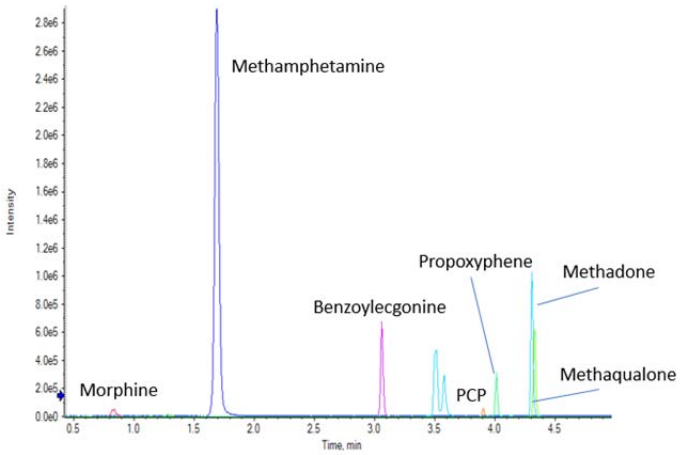


**Figure 1.** Chromatogram for all evaluated compounds using the Topaz System following the sample preparation and LC-MS/MS conditions.

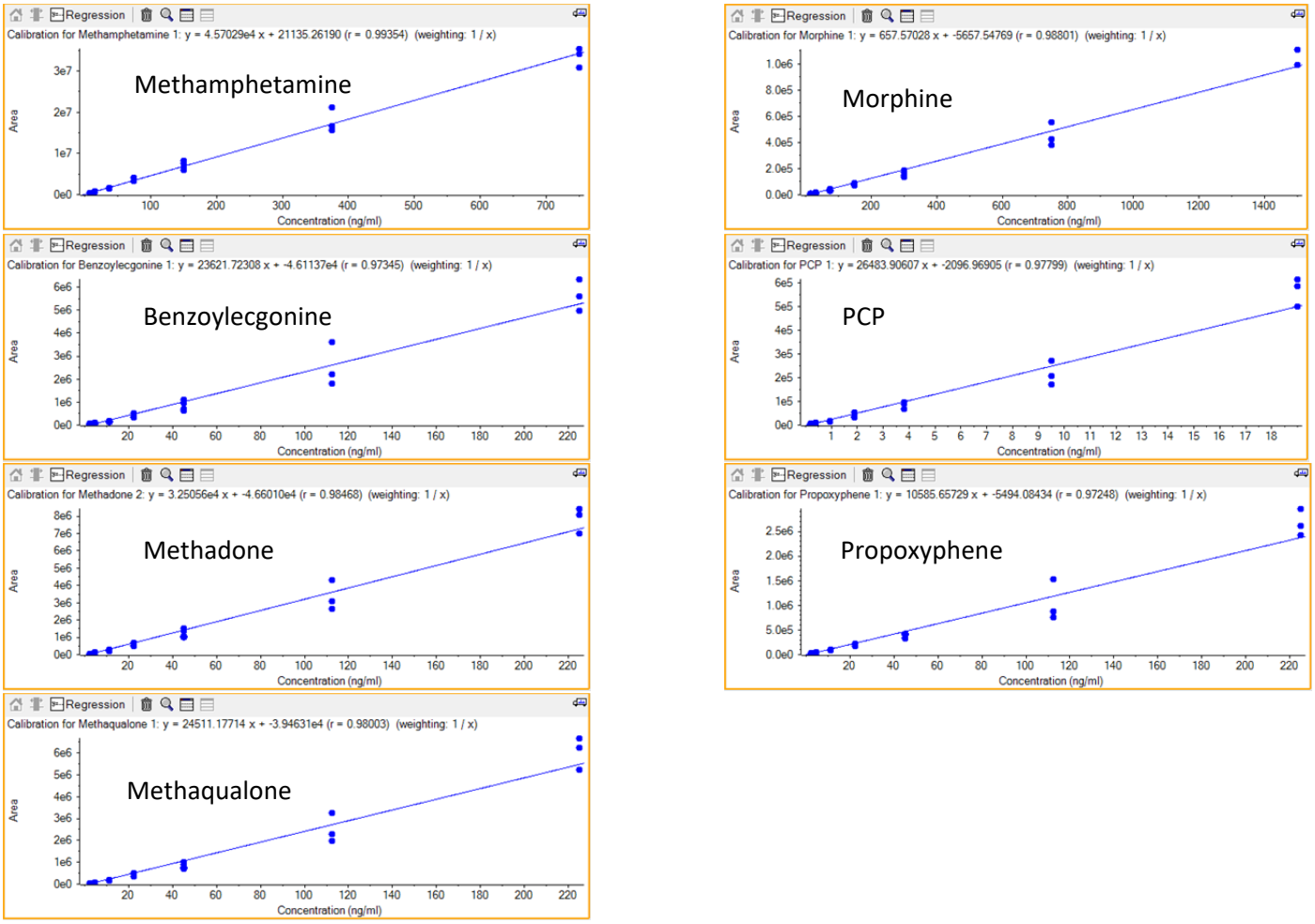


**Figure 2.** Calibration curves using ordinary least-squares regression and 1/x weighting for all the compounds evaluated over the defined concentration ranges in Table 1.

## Conclusions

Based on the above performance testing, the following results were obtained:

**Assay Linearity:** All compounds exhibited a linear dynamic range of at least 3 orders of magnitude.

**Reproducibility:** At each LLOQ, the precision (%CV) was <17% for all compounds evaluated as determined by n=6 replicates. CV data for all compounds evaluated is based on calculated concentration without internal standard.

**Sensitivity:** Functional sensitivity, as defined by a CV of <20%, was <8 ng/mL for all compounds with the exception of Benzoylecgonine (85.1ng/mL)

The Topaz System exhibited capability to deliver sensitive and reproducible analytical performance for Methamphetamine, Morphine, Benzoylecgonine, Methadone, Phencyclidine, Propoxyphene, and Methaqualone in urine matrix.

# Topaz™ System Analytical Performance Data Sheet

**Illustration of Analytical Performance for Methamphetamine, Morphine, Benzoylecgonine, Methadone, Phencyclidine, Amphetamine, and Oxazepam in Oral Fluid**

The SCIEX Topaz LC-MS/MS System is intended to identify inorganic or organic compounds in human specimens. All laboratory-developed tests must be developed, verified, and validated in accordance with applicable laws and regulations prior to their use for clinical diagnostic purposes.

This document describes a test of the analytical performance of the SCIEX Topaz LC-MS/MS System to analyze the compounds Methamphetamine, Morphine, Benzoylecgonine, Methadone, Phencyclidine, Amphetamine, and Oxazepam in oral fluid matrix.

The analytical performance data presented here is for illustrative purposes only to demonstrate the potential capabilities of the system. Performance in individual laboratories may differ due to a number of factors, including system configuration, laboratory methods, and operator technique. This document does not constitute a warranty of merchantability or fitness for any particular purpose, express or implied, including for the testing of the compounds analyzed in this experiment.

## Materials and Methods

The Topaz LC-MS/MS System was controlled and data processed using ClearCore™ MD Software (ver. 1.1). Commercially available urine calibrators and quality controls (Bio-Rad P/N 438) containing all compounds of interest were processed using the following conditions:

**Sample Preparation Conditions**
Dilution with Water/Acetonitrile solution using 200µL sample, followed by direct injection

**Liquid Chromatography Conditions**
Column: Phenomenex® Kinetex® Biphenyl
Mobile Phase A: Water/0.1% Formic Acid
Mobile Phase B: Acetonitrile/0.1% Formic Acid
Flow Rate: 0.7mL/min
Injection Volume: 5µL
Gradient: Gradient 10-85%B over 7 min
Retention Time: Compound dependent: 2.9-4.8 min

**Mass Spectrometry Conditions**
Method Duration: 7 min
Polarity: Positive electrospray
Transitions: Compound dependent
Source Conditions: Flow rate-optimized

## Results

Analytical performance statistics including the concentration range evaluated, precision experiments (n=6 replicates), and functional sensitivity are shown in Table 1. Chromatogram of the compounds evaluated utilizing the described method is shown in Figure 1. Calibration curves over the defined concentration ranges for each compound are illustrated in Figure 2.

| Compound (units) | Range | %CV (at LLOQ) | Functional Sensitivity* |
|---|---|---|---|
| Methamphetamine (ng/mL) | 0.75-750 | 9.5% | 0.704 |
| Morphine (ng/mL) | 1.5-1500 | 14.0% | 0.757 |
| Benzoylecgonine (ng/mL) | 0.225-225 | 16.4% | 0.425 |
| PCP (ng/mL) | 0.19-19 | 19.7% | 0.224 |
| Methadone (ng/mL) | 0.225-225 | 7.0% | 0.083 |
| Amphetamine (ng/mL) | 2.25-225 | 11.2% | 0.520 |
| Oxazepam (ng/mL) | 0.225-225 | 17.8% | 0.262 |

**Table 1.** Measured range, percent coefficient of variation (%CV) at lower-limit of quantitation (LLOQ), and functional sensitivity for each compound evaluated.

**Functional Sensitivity is defined here as the lowest concentration achieving a CV of 20%.*

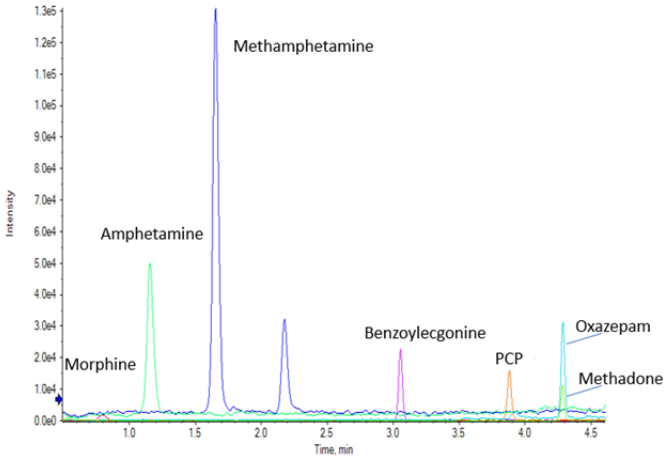


**Figure 1.** Chromatogram for all evaluated compounds using the Topaz System following the sample preparation and LC-MS/MS conditions.

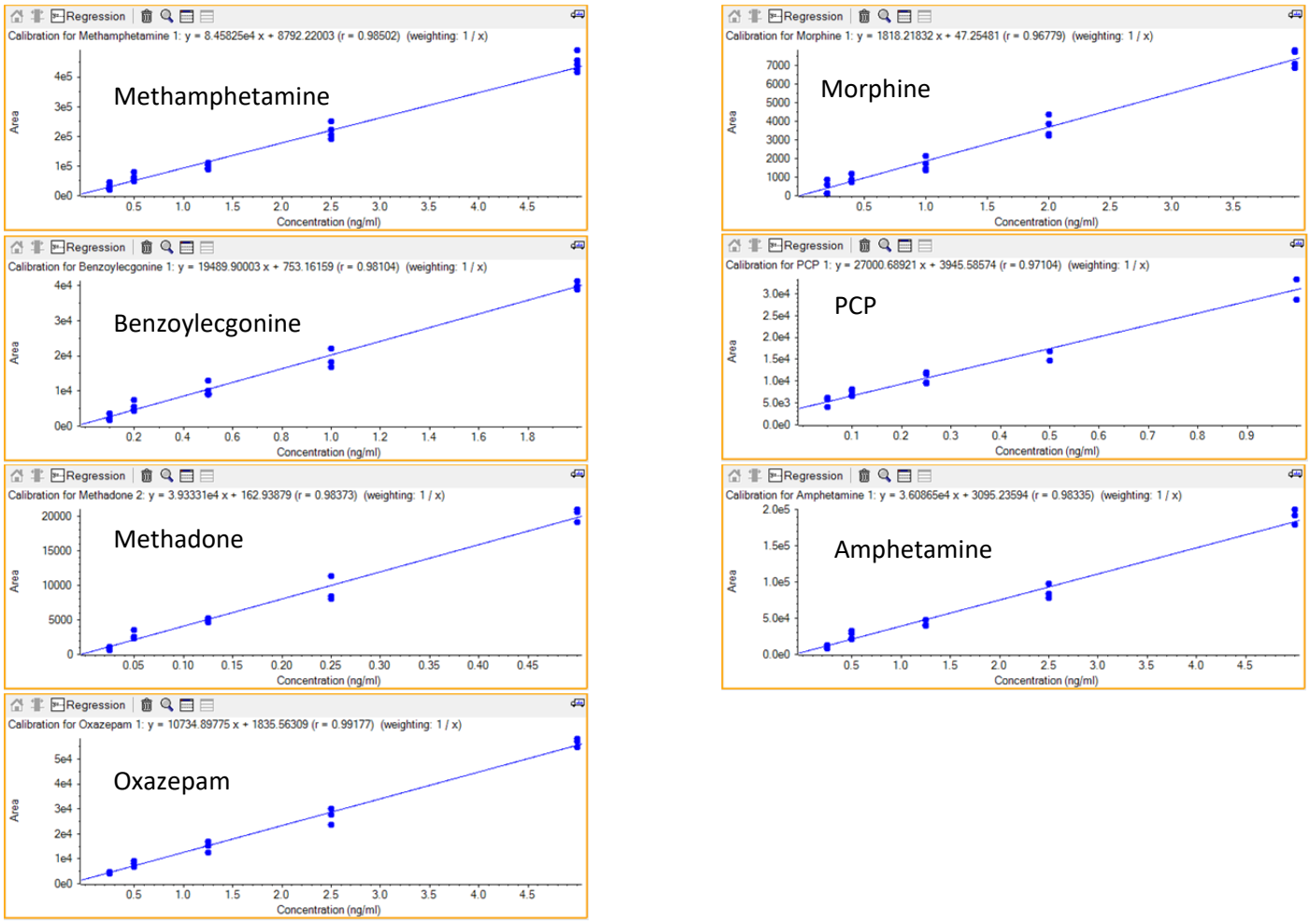


**Figure 2.** Calibration curves using ordinary least-squares regression and 1/x weighting for all the compounds evaluated over the defined concentration ranges in Table 1.

## Conclusions

Based on the above performance testing, the following results were obtained:

**Assay Linearity:** All compounds exhibited a linear dynamic range of at least 3 orders of magnitude.

**Reproducibility:** At each LLOQ, the precision (%CV) was <18% for all compounds evaluated as determined by n=6 replicates. CV data for all compounds evaluated is based on calculated concentration without internal standard.

**Sensitivity:** Functional sensitivity, as defined by a CV of <20%, was <0.8 ng/mL for all evaluated compounds.

The Topaz System exhibited capability to deliver sensitive and reproducible analytical performance for Methamphetamine, Morphine, Benzoylecgonine, Methadone, Phencyclidine, Amphetamine, and Oxazepam in oral fluid matrix.

# Topaz™ System Analytical Performance Data Sheet

**Illustration of analytical performance for Capsofungin, Itraconazole, Hydroxyitraconazole, Voriconazole, and Fluconazole**

The SCIEX Topaz LC-MS/MS System is intended to identify inorganic or organic compounds in human specimens. All laboratory-developed tests must be developed, verified, and validated in accordance with applicable laws and regulations prior to their use for clinical diagnostic purposes.

This document describes a test of the analytical performance of the SCIEX Topaz LC-MS/MS System to analyze the compounds Capsofungin, Itraconazole, Hydroxyitraconazole, Voriconazole, and Fluconazole in serum matrix.

The analytical performance data presented here is for illustrative purposes only to demonstrate the potential capabilities of the system. Performance in individual laboratories may differ due to a number of factors, including system configuration, laboratory methods, and operator technique. This document does not constitute a warranty of merchantability or fitness for any particular purpose, express or implied, including for the testing of the compounds analyzed in this experiment.

## Materials and Methods

Commercially available serum calibrators and quality controls (UTAK P/N 43011) containing all compounds of interest were processed using the following approach:

**Sample Prep Conditions**
Protein Crash with Methanolic Zinc Sulphate solution using 100ul plasma, centrifugation and direct injection

**Liquid Chromatography Conditions**
Column: Phenomenex® Kinetex® Biphenyl
Mobile Phase A: Water/0.1% Formic Acid
Mobile Phase B: Methanol/0.1% Formic Acid
Flow Rate: 0.6ml/min
Injection Volume: 25ul
Gradient: Linear 5-98%B over 5 minutes
Retention Time: Compound dependent 1.5-3 minutes

**Mass Spectrometry Conditions**
Method Duration: 5 minutes
Polarity: Positive electrospray
Transitions: Compound dependent
Source Conditions: Flow rate-optimized

## Results

Analytical performance statistics including the concentration range evaluated, precision experiments (n=6 replicates), and functional sensitivity are shown in Table 1. Chromatogram of the compounds evaluated utilizing the described method is shown in Figure 1. Calibration curves over the defined concentration ranges for each compound are illustrated in Figure 2.

| Compound (units) | Range | %CV (at LLOQ) | Functional Sensitivity* |
|---|---|---|---|
| Capsofungin (µg/mL) | 0.09-1.8 | 10.6% | 0.078 |
| Fluconazole (µg/mL) | 0.2-10 | 16.5% | 0.142 |
| Hydroxyitraconazole (µg/mL) | 0.05-1 | 12.0% | 0.067 |
| Itraconazole (µg/mL) | 0.05-1 | 18.3% | 0.174 |
| Voriconazole (µg/mL) | 0.005-2.5 | 7.6% | 0.033 |

**Table 1.** Measured range, percent coefficient of variation (%CV) at lower-limit of quantitation (LLOQ), and functional sensitivity for each compound evaluated.

**Functional Sensitivity is defined here as the lowest concentration achieving a CV of 20%.*

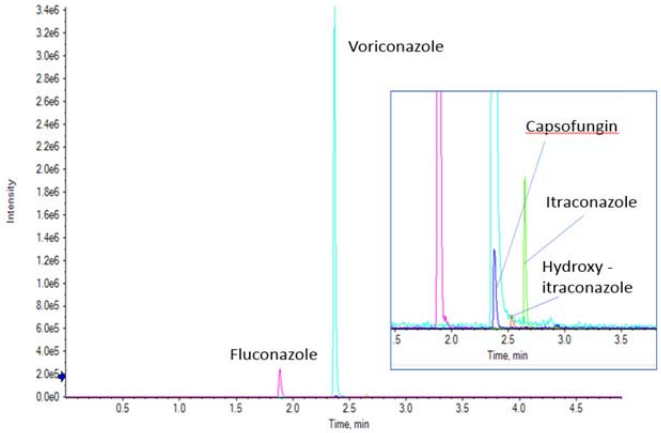


**Figure 1.** Chromatogram for all evaluated compounds using the Topaz System following the sample preparation and LC-MS/MS conditions.

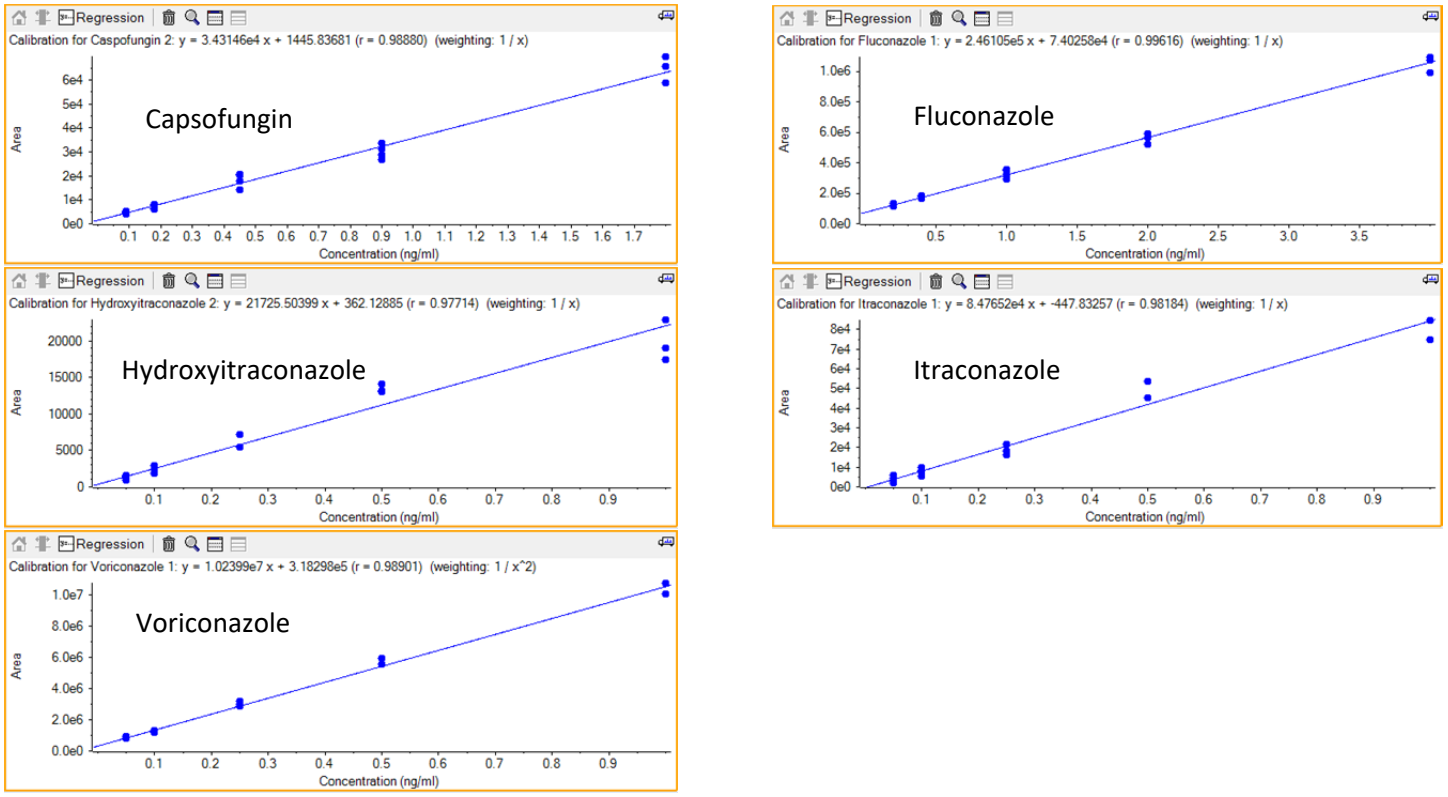


**Figure 2.** Calibration curves using ordinary least-squares regression and 1/x weighting for all the compounds evaluated over the defined concentration ranges in Table 1.

## Conclusions

Based on the above performance testing, the following results were obtained:

**Assay Linearity:** All compounds exhibited a linear dynamic range of at least 3 orders of magnitude.

**Reproducibility:** At each LLOQ, the precision (%CV) was <19% for all compounds evaluated as determined by n=6 replicates. CV data for all compounds evaluated is based on calculated concentration without internal standard.

**Sensitivity:** Functional sensitivity, as defined by a CV of <20%, was <0.2 µg/mL for all evaluated compounds.

The Topaz System exhibited capability to deliver sensitive and reproducible analytical performance for Capsofungin, Itraconazole, Hydroxyitraconazole, Voriconazole, and Fluconazole in serum matrix.

# Topaz™ System Analytical Performance Data Sheet

**Illustration of analytical performance for Cyclosporin A, Tacrolimus, Sirolimus, and Everolimus**

The SCIEX Topaz LC-MS/MS System is intended to identify inorganic or organic compounds in human specimens. All laboratory-developed tests must be developed, verified, and validated in accordance with applicable laws and regulations prior to their use for clinical diagnostic purposes.

This document describes a test of the analytical performance of the SCIEX Topaz LC-MS/MS System to analyze the compounds Cyclosporin A, Tacrolimus, Sirolimus, and Everolimus in whole blood matrix.

The analytical performance data presented here is for illustrative purposes only to demonstrate the potential capabilities of the system. Performance in individual laboratories may differ due to a number of factors, including system configuration, laboratory methods, and operator technique. This document does not constitute a warranty of merchantability or fitness for any particular purpose, express or implied, including for the testing of the compounds analyzed in this experiment.

## Materials and Methods

The Topaz LC-MS/MS System was controlled and data processed using ClearCore™ MD Software (ver. 1.1). Commercially available whole blood calibrators and quality controls (Chromsystems, P/N 93900) containing the compounds of interest were processed with the conditions:

**Sample Prep Conditions**
Precipitation with kit extraction solutions using 50µL whole blood, centrifugation and direct injection

**Liquid Chromatography Conditions**
Column: Commercial kit
Mobile Phase A: Commercial kit
Mobile Phase B: Commercial kit
Flow Rate: 1-2.2mL/min
Injection Volume: 25µL
Gradient: Linear 9-100%B over 1.65 min
Retention Time: Compound dependent 1-1.2 min

**Mass Spectrometry Conditions**
Method Duration: 1.65 minutes
Polarity: Positive electrospray
Transitions: Compound dependent
Source Conditions: Flow rate-optimized

## Results

Analytical performance statistics including the concentration range evaluated, precision experiments (n=4 replicates, analyzed in triplicate), and functional sensitivity are shown in Table 1. Chromatogram of the compounds evaluated utilizing the described method is shown in Figure 1. Calibration curves over the defined concentration ranges for each compound are illustrated in Figure 2.

| Compound (units) | Range | %CV (at LLOQ) | Functional Sensitivity* |
|---|---|---|---|
| Cyclosporin A (µg/L) | 4-2000 | 4.2% | 1.1 |
| Tacrolimus (µg/L) | 0.4-100 | 5.1% | 0.43 |
| Sirolimus (µg/L) | 0.4-100 | 6.6% | 0.47 |
| Everolimus (µg/L) | 0.4-100 | 5.8% | 0.14 |

**Table 1.** Measured range, percent coefficient of variation (%CV) at lower-limit of quantitation (LLOQ), and functional sensitivity for each compound evaluated.

**Functional Sensitivity is defined here as the lowest concentration achieving a CV of 20%.*

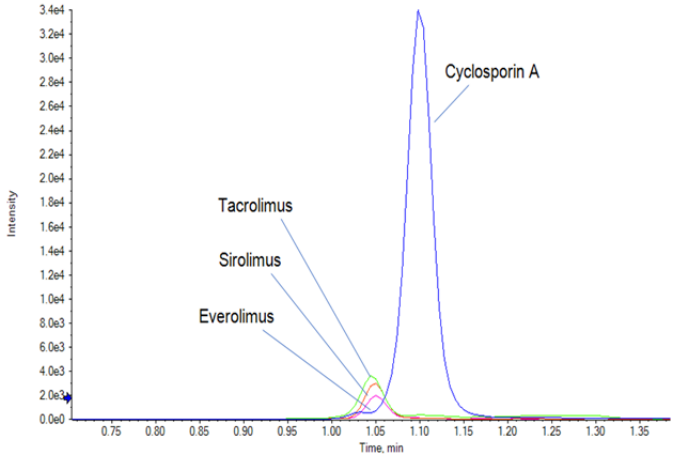


**Figure 1.** Chromatogram for all evaluated compounds using the Topaz System following the sample preparation and LC-MS/MS conditions.

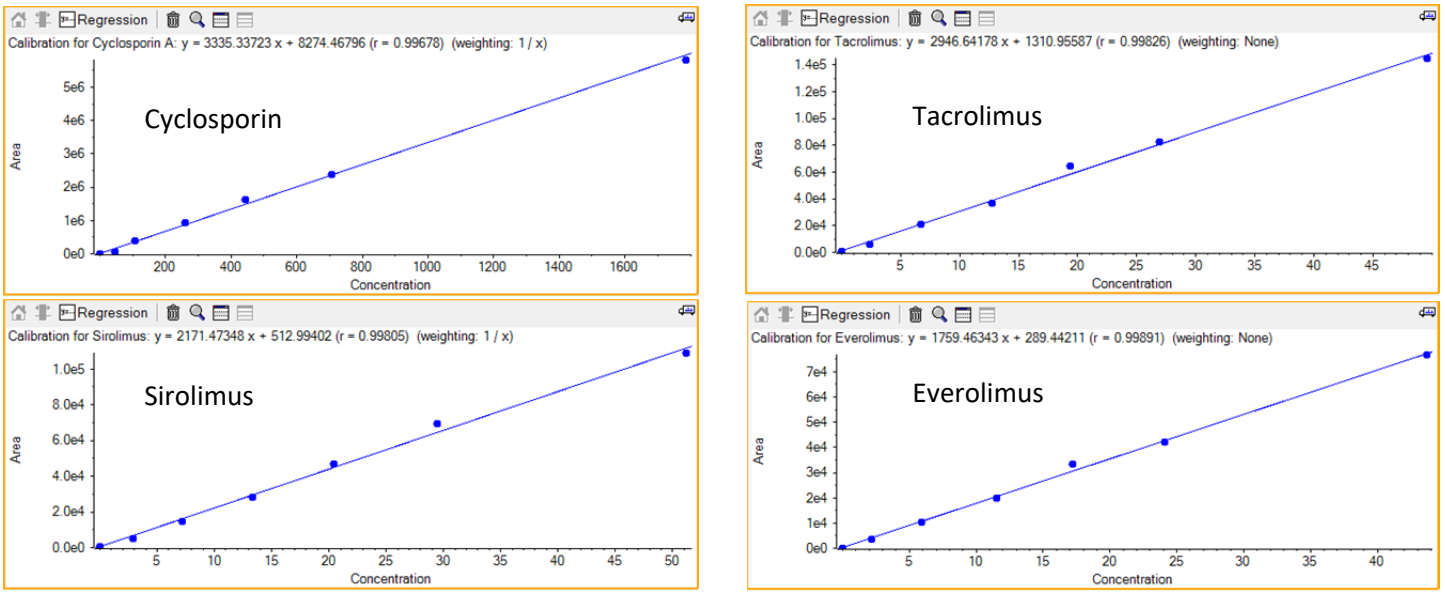


**Figure 2.** Calibration curves using ordinary least-squares regression and 1/x weighting for all the compounds evaluated over the defined concentration ranges in Table 1.

## Conclusions

Based on the above performance testing, the following results were obtained:

**Assay Linearity:** All compounds exhibited a linear dynamic range of at least 3 orders of magnitude.

**Reproducibility:** At each LLOQ, the precision (%CV) was <7% for all compounds evaluated as determined by n=4 replicates, analyzed in triplicate. CV data for all compounds evaluated is based on calculated concentration with internal standard.

**Sensitivity:** Functional sensitivity, as defined by a CV of <20%, was <1.2 µg/L for all evaluated compounds.

The Topaz System exhibited capability to deliver sensitive and reproducible analytical performance for Cyclosporin A, Tacrolimus, Sirolimus, and Everolimus in whole blood matrix.

# Topaz™ System Analytical Performance Data Sheet

## Illustration of Analytical Performance for Vitamin B1 & Vitamin B6 in Whole Blood

***The SCIEX Topaz LC-MSMS System is intended to identify inorganic or organic compounds in human specimens. All laboratory-developed tests must be developed, verified, and validated in accordance with applicable laws and regulations prior to their use for clinical diagnostic purposes.***

This document describes a test of the analytical performance of the SCIEX Topaz LC-MSMS System to analyze the compounds Vitamin B1 & Vitamin B6 in whole blood matrix.

The analytical performance data presented here is for illustrative purposes only to demonstrate the potential capabilities of the system. Performance in individual laboratories may differ due to a number of factors, including system configuration, laboratory methods, and operator technique. This document does not constitute a warranty of merchantability or fitness for any particular purpose, express or implied, including for the testing of the compounds analyzed in this experiment.

## Materials & Methods

The Topaz LC-MS/MS System was controlled and data processed using ClearCore™ MD Software (ver. 1.1). A commercially available kit for the analysis of Vitamin B1 and B6 in whole blood (Instruchemie B.V.) was processed using the following conditions:

**Sample Prep Conditions**
Liquid-Liquid Extraction with MTBE

**Liquid Chromatography Conditions**
Column: Phenomenex Luna C18
Mobile Phase A: Provided in Kit
Mobile Phase B: Provided in Kit
Flow Rate: 0.65 – 0.8 mL/min
Injection Volume: 20 µL
Gradient: Linear 10-97% B over 3 minutes
Retention Time: 0.95 min (B1), 1.05 min (B6)

**Mass Spectrometry Conditions**
Method Duration: 3 minutes
Polarity: Positive ESI
Transitions: Compound Dependent
Source Conditions: Flow Rate-Optimized

## Results

Analytical performance statistics including the concentration range evaluated, precision experiments (n=6 replicates), and functional sensitivity are shown in Table 1. Chromatogram of the compounds evaluated utilizing the described method is shown in Figure 1. Calibration curves over the defined concentration ranges for each compound are illustrated in Figure 2.

| Compound (units) | Range (nmol/L) | %CV (at LLOQ) | Functional Sensitivity* (nmol/L) |
|---|---|---|---|
| Vitamin B1 (TDP) | 39-890 | 1.3 | 4.45 |
| Vitamin B6 (PLP) | 21-550 | 5.9 | 6.31 |

**Table 1.** Measured range, percent coefficient of variation (%CV) at lower-limit of quantitation (LLOQ), and functional sensitivity for each compound evaluated.

**Functional Sensitivity is defined here as the lowest concentration achieving a CV of 20%.*

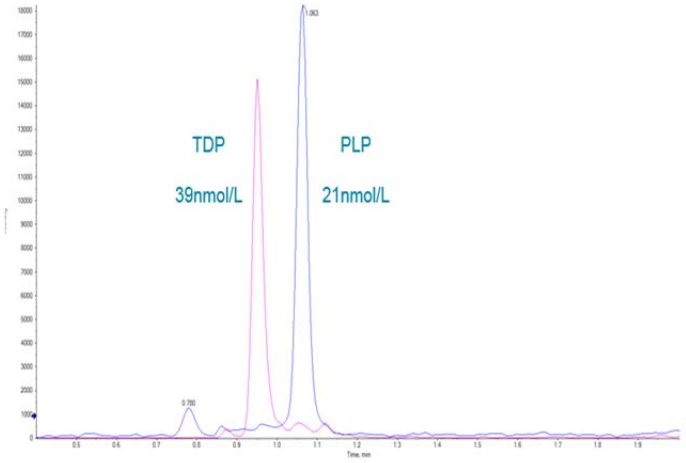


**Figure 1.** Chromatogram for all evaluated compounds using the Topaz System following the sample preparation and LC-MS/MS conditions.

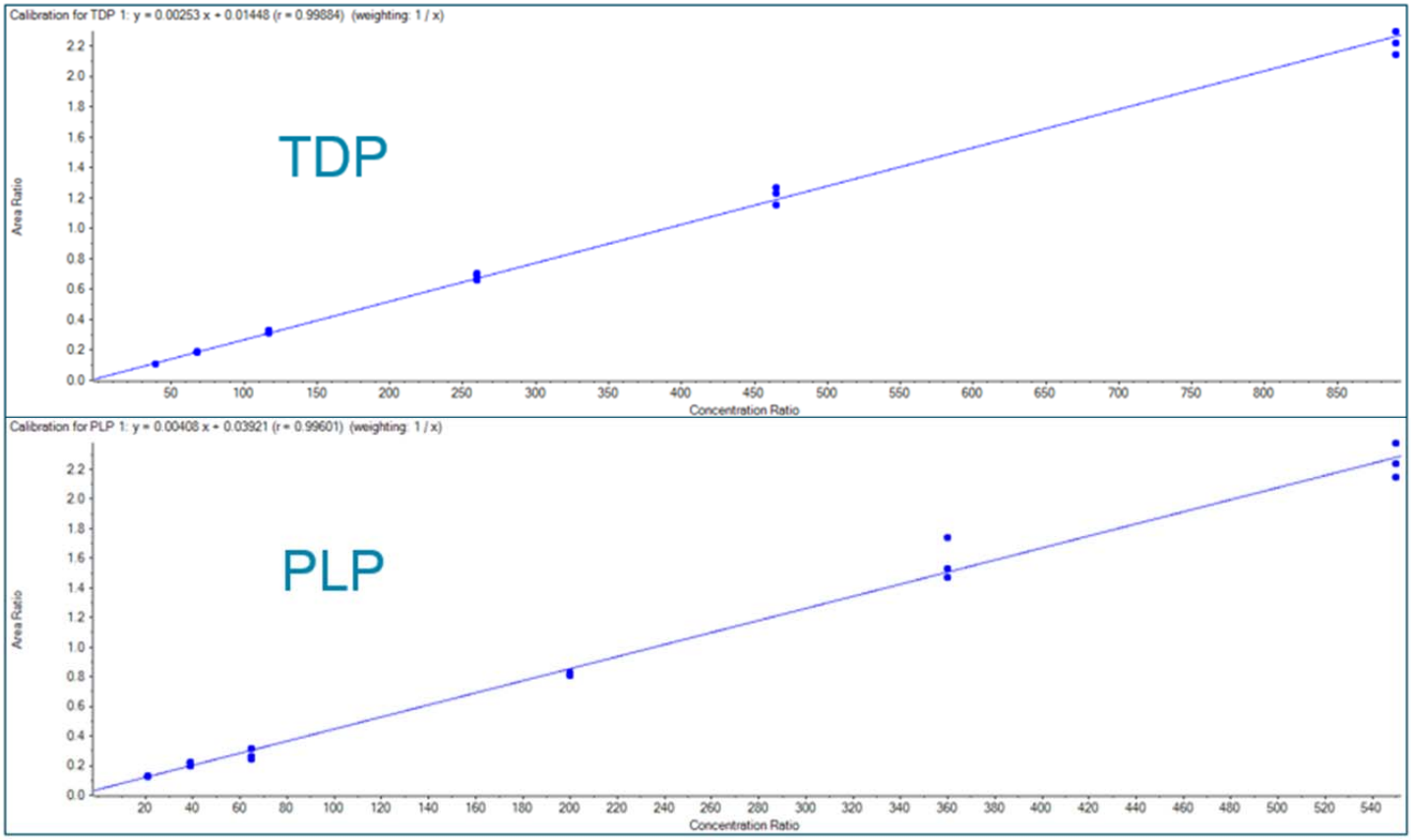


**Figure 2.** Calibration curves using ordinary least-squares regression and 1/x weighting for all the compounds evaluated over the defined concentration ranges in Table 1.

## Conclusions

Based on the above performance testing, the following results were obtained:

**Assay Linearity:** Compounds exhibited a linear dynamic range of approximately 3 orders of magnitude.

**Reproducibility:** At each LLOQ, the precision (%CV) was <6% for compounds evaluated as determined by n=3 replicates. CV data for all compounds evaluated is based on calculated concentration with internal standard.

**Sensitivity:** Functional sensitivity, as defined by a CV of <20%, was <7 nmol/L for evaluated compounds.

The Topaz System exhibited capability to deliver sensitive and reproducible analytical performance Vitamin B1 and B6 in whole blood matrix.



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