# 510(k) Summary: Amphetamines II Assay for Integra Family of Analyzers

**Introduction**

According to the requirements of 21 CFR 807.92, the following information provides sufficient detail to understand the basis for a determination of substantial equivalence.

**Submitter Name, Address, Contact**

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JUL 2 8 2010

K093664

**Device Name**

Proprietary name: Amphetamines II

Common name: Amphetamine metabolite test system

Classification name: Enzyme Immunoassay, Amphetamine

Product Code: DKZ

**Device Description**

The Amphetamines II test is an immunoassay for use on automated clinical chemistry analyzers. The device consists of two wet reagents; a soluble drug-conjugate, and an antibody-bound microparticle solution. During the assay, in the absence of sample drug in urine, soluble drug-conjugates bind to antibody-bound microparticles, causing the formation of particle aggregates. When a urine sample contains the drug in question, this drug competes with the drug derivative conjugate for microparticle-bound antibody. Antibody bound to sample drug is no longer available to promote particle aggregation, and subsequent particle lattice formation is inhibited. The rate of absorbance change is proportional to the concentration of drug in the sample. Calibrators, ranging in concentration from 0-5000 ng/mL depending on cutoff and test mode, are run with the assay. Concentrations of controls and unknowns are calculated from the standard curve in semi-quantitative mode. Results for controls or calibrators are determined as preliminary positive or negative relative to the cutoff in qualitative mode.

C.f.a.s. DAT Qualitative Clinical, C.f.a.s. DAT Qualitative Plus, C.f.a.s. DAT Qualitative Plus Clinical, Preciset DAT Plus I Calibrators, and Preciset DAT Plus II Calibrators are ready to use, multianalyte calibrators prepared by the quantitative addition of drug or drug metabolite to drug-free human urine.

Control Set DAT I, II, and III, and Control Set DAT Clinical are ready to use multianalyte controls prepared by the quantitative addition of drug or drug metabolite to drug-free urine.

**Intended Use**

Amphetamines II (AMPII) is an in vitro diagnostic test for the qualitative and semiquantitative detection of amphetamines and methamphetamines on COBAS INTEGRA systems in human urine at cutoff concentrations of 300 ng/mL, 500 ng/mL and 1000 ng/mL when calibrated with *d*-methamphetamine. Semiquantitative test results may be obtained that permit laboratories to assess assay performance as part of a quality control program. . Semiquantitative assays are intended to determine an appropriate dilution of the specimen for confirmation by a confirmatory method such as gas chromatography/mass spectrometry (GC/MS).

**Amphetamines II provides only a preliminary analytical test result. A more specific alternate chemical method must be used in order to obtain a confirmed analytical result. Gas chromatography/mass spectrometry (GC/MS) is the preferred confirmatory method.[1] Clinical consideration and professional judgement should be applied to any drug of abuse test result, particularly when preliminary positive results are used.**

**Comparison to the Predicate Device**

The ONLINE Amphetamines II assay is substantially equivalent to other products in commercial distribution intended for similar use. Most notably, we claim substantial equivalence to the currently marketed Amphetamines II assay (K083764).

The ONLINE Amphetamines II assay contains an in vitro diagnostic reagent system intended for use on the Roche Integra analyzers for the semi-quantitative and qualitative detection of amphetamines in human urine at cutoff concentrations of 300, 500, and 1000 ng/mL.

The Roche ONLINE Amphetamines II assay utilizes a modified KIMS technology relative to the currently marketed Abuscreen OnLine Amphetamines assay. Differences between this application and the cleared assay include:

1. use of amphetamine and methamphetamine, and monoclonal antibodies attached to microparticles in solution
2. a soluble drug-polymer conjugate
3. increased sensitivity to "designer drugs"and their metabolites, and
4. addition of 300 and 500 ng/mL cutoff concentrations.

The recommended calibrators to be used with the proposed ONLINE Amphetamines II assay are the Preciset DAT Plus I, Preciset DAT Plus II and Cfas DAT Qualitative Plus Calibrators (K060645).

The recommended controls to be used with the proposed ONLINE Amphetamines II assay are the Control Set DAT I, Control Set DAT II, Control Set DAT III (K080183).

| Comparison of Technological Characteristics | | |
|---|---|---|
| **Feature** | **Amphetamines II Assay, Integra 800** | **Predicate Device: Amphetamines II Assay, Hitachi 917 (K083764)** |
| Methodology | Same | KIMS, Kinetic interaction of microparticles in solution |
| Sample Type | Same | Urine |
| Intended Use | Same | Qualitative and semi-quantitative detection of amphetamine and methamphetamine |
| Reagent | Same | 1. Conjugate Working Solution: Conjugated amphetamine and methamphetamine derivatives in buffer with bovine serum albumin(BSA) and 0.09% sodium azide.<br>2. Antibody/Microparticle Working Solution: Microparticles attached to amphetamine and methamphetamine antibodies (mouse monoclonal) in buffer with bovine serum albumin (BSA) and 0.09% sodium azide. |
| Cutoff | Same | 300, 500, 1000 ng/mL |
| Calibrator/Control Matrix | Same | Human urine |

# AMPII

**Amphetamines II**

## Order information

| | | |
|---|---|---|
| COBAS INTEGRA<br>ONLINE DAT<br>Amphetamines II | 200 Tests | Cat. No. **04512936** 190<br>System-ID 07 6835 9 |
| Preciset DAT Plus I calibrators<br>CAL 1-6 | 6 × 5 mL | Cat. No. **03304671** 190 |
| Preciset DAT Plus II calibrators<br>CAL 1-6 | 6 × 5 mL | Cat. No. **03304680** 190 |
| C.f.a.s. DAT Qualitative Plus | 6 x 5 mL | Cat. No. **03304698** 190 |
| C.f.a.s. DAT Qualitative Plus Clinical | 3 x 5 mL | Cat. No. **04590856** 190 |
| Control Set DAT II<br>(for 300 ng/mL assay) | | Cat. No. **03312968** 190 |
| PreciPos DAT Set II | 2 x 10 mL | |
| PreciNeg DAT Set II | 2 x 10 mL | |
| Control Set DAT I<br>(for 500 ng/mL assay) | | Cat. No. **03312950** 190 |
| PreciPos DAT Set I | 2 x 10 mL | |
| PreciNeg DAT Set I | 2 x 10 mL | |
| Control Set DAT Clinical<br>(for 500 ng/mL assay) | | Cat. No. **04500873** 190 |
| PreciPos DAT Clinical | 2 x 10 mL | |
| PreciNeg DAT Clinical | 2 x 10 mL | |
| Control Set DAT III<br>(for 1000 ng/mL assay) | | Cat. No. **03312976** 190 |
| PreciPos DAT Set III | 2 x 10 mL | |
| PreciNeg DAT Set III | 2 x 10 mL | |

## System information

Test **AM3S2:** 0-358: for semiquantitative assay, 300 ng/mL
Test **AM5S2:** 0-359: for semiquantitative assay, 500 ng/mL
Test **AM1S2:** 0-360: for semiquantitative assay, 1000 ng/mL
Test **AM3Q2:** 0-330: for qualitative assay, 300 ng/mL
Test **AM5Q2:** 0-340: for qualitative assay, 500 ng/mL
Test **AM1Q2:** 0-350: for qualitative assay, 1000 ng/mL
Test **AM5QC:** 0-341: for qualitative assay, 500 ng/mL; using C.f.a.s. DAT Qualitative Plus Clinical

## Intended use

Amphetamines II (AMPII) is an in vitro diagnostic test for the qualitative and semiquantitative detection of amphetamines and methamphetamines in human urine on COBAS INTEGRA systems at cutoff concentrations of 300 ng/mL, 500 ng/mL and 1000 ng/mL when calibrated with *d*-methamphetamine. Semiquantitative test results may be obtained that permit laboratories to assess assay performance as part of a quality control program. Semiquantitative assays are intended to determine an appropriate dilution of the specimen for confirmation by a confirmatory method such as gas chromatography/mass spectrometry (GC/MS).

**Amphetamines II provides only a preliminary analytical test result. A more specific alternate chemical method must be used in order to obtain a confirmed analytical result. GC/MS is the preferred confirmatory method.[1] Clinical consideration and professional judgement should be applied to any drug of abuse test result, particularly when preliminary positive results are used.**

## Summary

The amphetamines are known as the sympathomimetic amines as they mimic the effects of stimulation of the sympathetic nervous system. These small molecules, based on ß-phenylethylamine, structurally resemble the bodies own catecholamines. A wide variety have been created via substitutions anywhere on the structure. The amphetamines are potent central nervous stimulants. As such they can increase wakefulness, physical activity, and decrease appetite. The amphetamines have some limited indications and approval for use in ADHD, narcolepsy, and obesity. However, because these CNS stimulants convey a sense of self-confidence, well being, and euphoria, they are highly addictive, widely abused, and consequently controlled substances.[2] Abuse can lead to medical, psychological, and social consequences. Adverse health effects include memory loss, aggression, psychotic behavior, heart damage, malnutrition, and severe dental problems.[3] Amphetamine may be self-administered either orally or by intravenous injection in amounts of up to 2000 mg daily by tolerant addicts. It is a metabolite of a number of other drugs including methamphetamine. Normally about 30 % is excreted unchanged in the 24 hour urine, but this may change to as much as 74 % in acid urine and may decrease to 1 % in alkaline urine.[4]
Amphetamines II is calibrated with *d*-methamphetamine and therefore the sensitivity towards amphetamines is different than *d*-methamphetamine, as indicated in the "Analytical specificity" section.

## Test principle

The assay is based on the kinetic interaction of microparticles in a solution (KIMS)[5],[6] as measured by changes in light transmission. In the absence of amphetamine or methamphetamine, soluble drug conjugates bind to antibody-bound microparticles, causing the formation of particle aggregates. As the aggregation reaction proceeds in the absence of amphetamine or methamphetamine, the absorbance increases.

When a urine sample contains amphetamine or methamphetamine,, this drug competes with the drug derivative conjugate for microparticle-bound antibody. Antibody bound to amphetamine or methamphetamine is no longer available to promote particle aggregation, and subsequent particle lattice formation is inhibited. The presence of amphetamine or methamphetamine diminishes the increasing absorbance in proportion to the concentration of drug in the sample. Amphetamine or methamphetamine content is determined relative to the value obtained for a known cutoff concentration of *d*-methamphetamine.[7]

## Reagents - working solutions

**R1** Conjugated amphetamine and methamphetamine derivatives; buffer; bovine serum albumin; 0.09 % sodium azide

**SR** Microparticles attached to amphetamine and methamphetamine antibodies (mouse monoclonal); buffer; bovine serum albumin; 0.09 % sodium azide

## Precautions and warnings

Pay attention to all precautions and warnings listed in this Method Manual, Chapter 1, Introduction.

## Reagent handling

COBAS INTEGRA 800 analyzers

The reagent is automatically mixed for 1 minute after **cobas c** pack puncture and for half a minute during Begin of Day.

## Storage and stability

| | |
|---|---|
| Shelf life at 2 to 8 °C | See expiration date on **cobas c pack label** |
| COBAS INTEGRA 800 analyzers | |
| On-board in use at 8 °C | 84 days |

The on-board in use stability period begins at the time of **cobas c** pack puncture. Do not freeze reagents. Reagents that have been frozen should be discarded.

## Specimen collection and preparation

Only the specimens listed below were tested and found acceptable.

Urine: Collect urine samples in clean glass or plastic containers. Fresh urine samples do not require special handling or pretreatment, but an effort should be made to keep pipetted samples free of gross debris. Samples should be within the normal physiological pH range of 5 to 8. No additives or preservatives are required. It is recommended that urine specimens be stored at 2 to 8 °C and tested within 5 days of collection.[8] Centrifuge highly turbid specimens before testing. Adulteration or dilution of the sample can cause erroneous results. If adulteration is suspected, another sample should be collected. Specimen validity testing is required for specimens collected under the *Mandatory Guidelines for Federal Workplace Drug Testing Programs.*[9] Specimens containing human-sourced materials should be handled as if potentially infectious using safe laboratory procedures such as those outlined in *Biosafety in Microbiological and Biomedical Laboratories* (HHS Publication Number [CDC] 93-8395).

### Caution

Specimen dilutions should only be used as an estimation for GC/MS and are not intended for patient values. Dilution procedures, when used, should be validated.

## Materials provided

See "Reagents - working solutions" section for reagents.

## Assay

For optimum performance of the assay follow the directions given in this document for the analyzer concerned. Refer to the appropriate operator's manual for analyzer-specific assay instructions.

## Application for urine

### COBAS INTEGRA 800 test definition

| | *Semiquantitative* | *Qualitative* |
|---|---|---|
| Measuring mode | Absorbance | Absorbance |
| Abs. calculation mode | Endpoint | Endpoint |
| Reaction mode | R1-S-SR | R1-S-SR |
| Reaction direction | Increase | Increase |
| Reaction start | SR | SR |
| Wavelength A | 659 nm | 659 nm |
| Test range | | |
| *AM3S2* | 0-2000 ng/mL | 0-2000 |
| *AM5S2, AM1S2* | 0-5000 ng/mL | |
| with postdilution | | |
| *AM3S2* | 0-20000 ng/mL | |
| *AM5S2, AM1S2* | 0-50000 ng/mL | |
| Postdilution factor | 10 recommended [b] | No |
| Calc. first/last | 46/86 | 46/86 |
| Unit | ng/mL | |

b) For use when estimating concentration in preparation for GC/MS analysis.

### Pipetting parameters

| | | |
|---|---|---|
| *AM3S2, AM3Q2* | | Diluent (H2O) |
| R1 | 60 µL | 20 µL |
| Sample | 10 µL | 5 µL |
| SR | 47 µL | 3 µL |
| Total volume | 145 µL | |
| | | Diluent (H2O) |
| *AM5S2, AM5Q2, AM5QC* | | |
| R1 | 60 µL | 20 µL |
| Sample | 7.5 µL | 5 µL |
| SR | 47 µL | 3 µL |
| Total volume | 142.5 µL | |
| *AM1S2, AM1Q2* | | Diluent (H2O) |
| R1 | 60 µL | 20 µL |
| Sample | 5 µL | 5 µL |
| SR | 47 µL | 3 µL |
| Total volume | 140 µL | |

## Calibration

| | | |
|---|---|---|
| Calibrators | | *Semiquantitative applications* |
| | *AM3S2, 0-358* | Preciset DAT Plus II calibrators<br>6 Calibrators (300 cutoff, DATS8, 07 6796 [c])<br>0, 150, 300, 600, 1000, 2000 ng/mL<br>*d-methamphetamine* |
| | *AM5S2, 0-359;*<br>*AM1S2, 0-360* | Preciset DAT Plus I calibrators<br>6 Calibrators (500 and 1000 cutoffs, DATS2, 07 6764 6 [c])<br>0, 250, 500, 1000, 3000, 5000 ng/mL<br>*d-methamphetamine* |
| | | *Qualitative applications* |
| | *AM3Q2, 0-330* | 0 ng/mL (Preciset DAT Plus II calibrators, CAL 1) *or* deionized water and 300 ng/mL (Preciset DAT Plus II calibrators, CAL 3)<br>2 Calibrators (300 cutoff, DATQ3, 07 6770 0 [c])<br>For qualitative applications, the cutoff value is assigned as 1000. |
| | *AM5Q2, 0-340* | 0 ng/mL (Preciset DAT Plus I calibrators, CAL 1) *or* deionized water and 500 ng/mL (C.f.a.s. DAT Qualitative Plus)<br>2 Calibrators (500 cutoff, DATQ1, 07 6744 1 [c])<br>For qualitative applications, the cutoff value is assigned as 1000. |
| | *AM5QC, 0-341* | 0 ng/mL (Preciset DAT Plus I or II calibrators, CAL 1) *or* deionized water and 500 ng/mL (C.f.a.s. DAT Qualitative Plus Clinical)<br>2 Calibrators (500 cutoff, DATQ5, 07 6880 4 [c])<br>For qualitative applications, the cutoff value is assigned as 1000. |
| | *AM1Q2, 0-350* | 0 ng/mL (Preciset DAT Plus I calibrators, CAL 1) *or* deionized water and 1000 ng/mL (Preciset DAT Plus I calibrators, CAL 4)<br>2 Calibrators (1000 cutoff, DATQ2, 07 6768 9 [c])<br>For qualitative applications, the cutoff value is assigned as 1000. |

c) Short name and ID used on the COBAS INTEGRA systems.

| | |
|---|---|
| Calibration mode | *Semiquantitative applications* |
| COBAS INTEGRA 800 analyzers | Logit/log 4<br>*Qualitative applications*<br>Linear regression |
| Calibration replicate | Duplicate recommended |
| Calibration interval | |
| COBAS INTEGRA 800 analyzers | Each lot, every 4 weeks, and as required following quality control procedures |

A calibration curve is generated using the calibrators. Calibrators must be placed from the highest concentration first to the lowest last on the CAL/QC rack. This curve is retained in memory by the COBAS INTEGRA systems and recalled for later use.

Traceability: This method has been standardized against GC/MS.

## Quality control

| | |
|---|---|
| Quality control | *300 ng/mL cutoff*<br>*AM3S2 and AM3Q2:*<br>Control Set DAT II<br>PreciPos DAT Set II<br>(DAT2P, 07 6771 9 [d])<br>PreciNeg DAT Set II<br>(DAT2N, 07 6772 7 [d])<br><br>*500 ng/mL cutoff*<br>*AM5S2 and AM5Q2:*<br>Control Set DAT I<br>PreciPos DAT Set I<br>(DAT1P, 07 6753 0 [d])<br>PreciNeg DAT Set I<br>(DAT1N, 07 6754 9 [d])<br><br>*AM5QC:*<br>Control Set DAT Clinical<br>PreciPos DAT Clinical<br>(DATCP, 07 6879 0 [d])<br>PreciNeg DAT Clinical<br>(DATCN, 07 6878 2 [d])<br><br>*1000 ng/mL cutoff*<br>*AM1S2 and AM1Q2:*<br>Control Set DAT III<br>PreciPos DAT Set III<br>(DAT3P, 07 6773 5 [d])<br>PreciNeg DAT Set III<br>(DAT3N, 07 6774 3 [d]) |
| Control sequence | User defined |
| Control after calibration | Recommended |

d) Short name and ID used on the COBAS INTEGRA systems.

For quality control, use control materials as listed in the Order information section. Other suitable control material can be used in addition.
Drug concentrations of the controls have been verified by GC/MS.
The control intervals and limits should be adapted to each laboratory's individual requirements.
Values obtained should fall within the defined limits.
Each laboratory should establish corrective measures to be taken if values fall outside the limits.
Follow the applicable government regulations and local guidelines for quality control.

## Results

COBAS INTEGRA systems report results with the following test flags:
***Semiquantitative result reporting***
*AM3S2 (300 ng/mL cutoff)*

| Flag | COBAS INTEGRA | Value range |
|---|---|---|
| No flag | Negative | < 300 ng/mL |
| <TEST RNG | Negative | < 0 ng/mL |
| >TEST RNG | Positive | > 2000 ng/mL |
| POS 300 | Positive | ≥ 300 ng/mL |

Value ranges listed above are based on a cutoff value of 300 ng/mL.

***Semiquantitative result reporting***

*AM5S2 (500 ng/mL cutoff)*

| Flag | COBAS INTEGRA | Value range |
|---|---|---|
| No flag | Negative | < 500 ng/mL |
| <TEST RNG | Negative | < 0 ng/mL |
| >TEST RNG | Positive | > 5000 ng/mL |
| POS 500 | Positive | ≥ 500 ng/mL |

Value ranges listed above are based on a cutoff value of 500 ng/mL.

***Semiquantitative result reporting***

*AM1S2 (1000 ng/mL cutoff)*

| Flag | COBAS INTEGRA | Value range |
|---|---|---|
| No flag | Negative | < 1000 ng/mL |
| <TEST RNG | Negative | < 0 ng/mL |
| >TEST RNG | Positive | > 5000 ng/mL |
| POS 1000 | Positive | ≥ 1000 ng/mL |

Value ranges listed above are based on a cutoff value of 1000 ng/mL.

***Qualitative result reporting***

*AM3Q2 (300 ng/mL cutoff)*

| Flag | COBAS INTEGRA | Value range |
|---|---|---|
| No flag | Negative | < 1000 |
| <TEST RNG | Negative | < 0 |
| >TEST RNG | Positive | > 2000 |
| POS 1000 | Positive | ≥ 1000 |

Value ranges above are based on assigning the cutoff of 300 ng/mL a value of 1000.

***Qualitative result reporting***

*AM5Q2, AM5QC (500 ng/mL cutoff)*

| Flag | COBAS INTEGRA | Value range |
|---|---|---|
| No flag | Negative | < 1000 |

| | | |
|---|---|---|
| <TEST RNG | Negative | < 0 |
| >TEST RNG | Positive | > 2000 |
| POS 1000 | Positive | ≥ 1000 |

Value ranges above are based on assigning the cutoff of 500 ng/mL a value of 1000.

***Qualitative result reporting***

*AM1Q2 (1000 ng/mL cutoff)*

| Flag | COBAS INTEGRA | Value range |
|---|---|---|
| No flag | Negative | < 1000 |
| <TEST RNG | Negative | < 0 |
| >TEST RNG | Positive | > 2000 |
| POS 1000 | Positive | ≥ 1000 |

Value ranges above are based on assigning the cutoff of 1000 ng/mL a value of 1000.

## Semiquantitative result reporting

The semiquantitation of preliminary positive results should only be used by laboratories to determine an appropriate dilution of the specimen for confirmation by a confirmatory method such as GC/MS. It also permits the laboratory to establish quality control procedures and assess control performance.

**Note:** When using the post-dilution function (1:10 dilution), to ensure the sample was not over-diluted, the diluted result must be at least half the analyte cutoff value times 10. If the diluted result falls below half the analyte cutoff value times 10, repeat the sample with a smaller dilution. A dilution that produces a result closest to the analyte cutoff is the most accurate estimation. To estimate the positive sample's concentration, multiply the result by the appropriate dilution factor. Dilutions should only be used as an estimation for GC/MS.

## Limitations[10]

See the Analytical specificity section of this method sheet for information on substances tested for cross-reactivity in this assay. There is the possibility that other substances and/or factors may interfere with the test and cause erroneous results (e.g., technical or procedural errors).

A preliminary positive result with this assay indicates the presence of amphetamine or methamphetamine in urine. It does not measure the level of intoxication.

## ACTION REQUIRED

***Special wash programming:*** The use of special wash steps is mandatory when certain test combinations are run together on COBAS INTEGRA 800 analyzer. Refer to the Method Manual, Introduction, Extra Wash Cycles for further instructions.

**Where required, special wash/carry-over evasion programming must be implemented prior to reporting results with this test.**

## Expected values

No drug should be present in individuals that have not ingested amphetamines.

## Specific performance data

Representative performance data on the COBAS INTEGRA 800 analyzer are given below. Results obtained in individual laboratories may differ.

## Precision

A *d*-methamphetamine (MAMP) solution (1 mg/mL) was added to 9 samples obtained from a human urine sample pool to achieve concentrations at approximately - 100 %, - 75 %, - 50 %, - 25 %, +/- 0 %, + 25 %, + 50 %, + 75 %, and + 100 % of the cutoff value. These samples were tested for precision in qualitative and semiquantitative modes. Following a CLSI (EP5-A2) precision protocol, samples were tested in 2 replicates per run, 2 runs per day for 21 days, total N = 84.

***Qualitative - 300 ng/mL Cutoff***

| Drug | Concentration of Sample | Number of Determinations | Results # Neg / # Pos |
|---|---|---|---|
| MAMP | zero drug | 84 | 84 Neg / 0 Pos |
| MAMP | - 75 % | 84 | 84 Neg / 0 Pos |
| MAMP | - 50 % | 84 | 84 Neg / 0 Pos |
| MAMP | - 25 % | 84 | 83 Neg / 1 Pos |
| MAMP | cutoff | 84 | 27 Neg / 57 Pos |
| MAMP | + 25 % | 84 | 0 Neg / 84 Pos |
| MAMP | + 50 % | 84 | 0 Neg / 84 Pos |
| MAMP | + 75 % | 84 | 0 Neg / 84 Pos |
| MAMP | + 100 % | 84 | 0 Neg / 84 Pos |

***Qualitative - 500 ng/mL Cutoff***

| Drug | Concentration of Sample | Number of Determinations | Results # Neg / # Pos |
|---|---|---|---|
| MAMP | zero drug | 84 | 84 Neg / 0 Pos |
| MAMP | - 75 % | 84 | 84 Neg / 0 Pos |
| MAMP | - 50 % | 84 | 84 Neg / 0 Pos |
| MAMP | - 25 % | 84 | 84 Neg / 0 Pos |
| MAMP | cutoff | 84 | 28 Neg / 56 Pos |
| MAMP | + 25 % | 84 | 0 Neg / 84 Pos |
| MAMP | + 50 % | 84 | 0 Neg / 84 Pos |
| MAMP | + 75 % | 84 | 0 Neg / 84 Pos |
| MAMP | + 100 % | 84 | 0 Neg / 84 Pos |

***Qualitative - 1000 ng/mL Cutoff***

| Drug | Concentration of Sample | Number of Determinations | Results # Neg / # Pos |
|---|---|---|---|
| MAMP | zero drug | 84 | 84 Neg / 0 Pos |
| MAMP | - 75 % | 84 | 84 Neg / 0 Pos |
| MAMP | - 50 % | 84 | 84 Neg / 0 Pos |
| MAMP | - 25 % | 84 | 84 Neg / 0 Pos |
| MAMP | cutoff | 84 | 13 Neg / 71 Pos |
| MAMP | + 25 % | 84 | 0 Neg / 84 Pos |
| MAMP | + 50 % | 84 | 0 Neg / 84 Pos |
| MAMP | + 75 % | 84 | 0 Neg / 84 Pos |
| MAMP | + 100 % | 84 | 0 Neg / 84 Pos |

***Semiquantitative - 300 ng/mL Cutoff***

| Drug | Sample Conc. | Results # Neg / # Pos | Repeatability* | | Intermediate Precision** | |
|---|---|---|---|---|---|---|
| | | | SD, ng/mL | CV, % | SD, ng/mL | CV, % |
| MAMP | zero drug | 84 / 0 | 9.4 | 95.4 | 12.2 | 123.8 |
| MAMP | - 75 % | 84 / 0 | 12.9 | 15.5 | 15.7 | 18.8 |
| MAMP | - 50 % | 84 / 0 | 14.3 | 9.0 | 17.7 | 11.2 |
| MAMP | - 25 % | 84 / 0 | 13.2 | 5.4 | 19.8 | 8.2 |
| MAMP | cutoff | 34 / 50 | 23.2 | 7.6 | 23.5 | 7.7 |
| MAMP | + 25 % | 0 / 84 | 20.9 | 5.2 | 24.4 | 6.1 |
| MAMP | + 50 % | 0 / 84 | 22.5 | 5.0 | 29.5 | 6.6 |
| MAMP | + 75 % | 0 / 84 | 27.4 | 5.0 | 36.5 | 6.6 |
| MAMP | + 100 % | 0 / 84 | 35.7 | 5.7 | 40.8 | 6.5 |

***Semiquantitative - 500 ng/mL Cutoff***

| Drug | Sample Conc. | Results # Neg / # Pos | Repeatability* | | Intermediate Precision** | |
|---|---|---|---|---|---|---|
| | | | SD, ng/mL | CV, % | SD, ng/mL | CV, % |
| MAMP | zero drug | 84 / 0 | 9.6 | 117.3 | 11.0 | 134.1 |
| MAMP | - 75 % | 84 / 0 | 14.6 | 10.5 | 20.6 | 14.9 |
| MAMP | - 50 % | 84 / 0 | 20.0 | 7.9 | 21.9 | 8.6 |
| MAMP | - 25 % | 84 / 0 | 24.1 | 6.2 | 27.2 | 7.0 |
| MAMP | cutoff | 39 / 45 | 28.2 | 5.6 | 39.4 | 7.8 |
| MAMP | + 25 % | 0 / 84 | 31.5 | 4.9 | 48.5 | 7.5 |
| MAMP | + 50 % | 0 / 84 | 34.5 | 4.8 | 49.5 | 6.8 |
| MAMP | + 75 % | 0 / 84 | 41.6 | 5.0 | 62.3 | 7.5 |
| MAMP | + 100 % | 0 / 84 | 46.7 | 4.6 | 72.4 | 7.1 |

***Semiquantitative - 1000 ng/mL Cutoff***

| Drug | Sample Conc. | Results # Neg / # Pos | Repeatability* | | Intermediate Precision** | |
|---|---|---|---|---|---|---|
| | | | SD, ng/mL | CV, % | SD, ng/mL | CV, % |
| MAMP | zero drug | 84 / 0 | 20.2 | 97.2 | 24.9 | 119.5 |
| MAMP | - 75 % | 84 / 0 | 25.3 | 9.3 | 30.7 | 11.2 |
| MAMP | - 50 % | 84 / 0 | 33.3 | 6.4 | 39.4 | 7.5 |
| MAMP | - 25 % | 84 / 0 | 34.3 | 4.6 | 52.1 | 7.0 |
| MAMP | cutoff | 25 / 59 | 56.9 | 5.5 | 71.3 | 6.9 |
| MAMP | + 25 % | 0 / 84 | 56.6 | 4.2 | 79.3 | 5.9 |
| MAMP | + 50 % | 0 / 84 | 63.6 | 4.0 | 81.3 | 5.2 |
| MAMP | + 75 % | 0 / 84 | 60.3 | 3.5 | 101.6 | 5.9 |
| MAMP | + 100 % | 0 / 84 | 108.1 | 5.1 | 126.6 | 6.0 |

*repeatability = within-run precision

**intermediate precision = total precision / between run precision / between day precision

The same precision experiment was repeated utilizing *d*-amphetamine (AMP) as the target analyte instead of *d*-methamphetamine. The following tables show the results obtained on a COBAS INTEGRA 800 analyzer.

***Qualitative - 300 ng/mL Cutoff***

| Drug | Concentration of Sample | Number of Determinations | Results # Neg / # Pos |
|---|---|---|---|
| AMP | zero drug | 84 | 84 Neg / 0 Pos |
| AMP | - 75 % | 84 | 84 Neg / 0 Pos |
| AMP | - 50 % | 84 | 84 Neg / 0 Pos |
| AMP | - 25 % | 84 | 83 Neg / 1 Pos |
| AMP | cutoff | 84 | 2 Neg / 82 Pos |
| AMP | + 25 % | 84 | 0 Neg / 84 Pos |
| AMP | + 50 % | 84 | 0 Neg / 84 Pos |
| AMP | + 75 % | 84 | 0 Neg / 84 Pos |
| AMP | + 100 % | 84 | 0 Neg / 84 Pos |

***Qualitative - 500 ng/mL Cutoff***

| Drug | Concentration of Sample | Number of Determinations | Results # Neg / # Pos |
|---|---|---|---|
| AMP | zero drug | 84 | 84 Neg / 0 Pos |
| AMP | - 75 % | 84 | 84 Neg / 0 Pos |
| AMP | - 50 % | 84 | 84 Neg / 0 Pos |
| AMP | - 25 % | 84 | 82 Neg / 2 Pos |
| AMP | cutoff | 84 | 6 Neg / 78 Pos |
| AMP | + 25 % | 84 | 0 Neg / 84 Pos |
| AMP | + 50 % | 84 | 0 Neg / 84 Pos |
| AMP | + 75 % | 84 | 0 Neg / 84 Pos |
| AMP | + 100 % | 84 | 0 Neg / 84 Pos |

***Qualitative - 1000 ng/mL Cutoff***

| Drug | Concentration of Sample | Number of Determinations | Results # Neg / # Pos |
|---|---|---|---|
| AMP | zero drug | 84 | 84 Neg / 0 Pos |
| AMP | - 75 % | 84 | 84 Neg / 0 Pos |
| AMP | - 50 % | 84 | 84 Neg / 0 Pos |
| AMP | - 25 % | 84 | 82 Neg / 2 Pos |
| AMP | cutoff | 84 | 7 Neg / 77 Pos |
| AMP | + 25 % | 84 | 1 Neg / 83 Pos |
| AMP | + 50 % | 84 | 0 Neg / 84 Pos |
| AMP | + 75 % | 84 | 0 Neg / 84 Pos |
| AMP | + 100 % | 84 | 0 Neg / 84 Pos |

***Semiquantitative - 300 ng/mL Cutoff***

| Drug | Sample Conc. | Results # Neg / # Pos | Repeatability* | | Intermediate Precision** | |
|---|---|---|---|---|---|---|
| | | | SD, ng/mL | CV, % | SD, ng/mL | CV, % |
| AMP | zero drug | 84 / 0 | 8.6 | 110.0 | 10.6 | 135.7 |
| AMP | - 75 % | 84 / 0 | 10.4 | 16.8 | 13.6 | 21.9 |
| AMP | - 50 % | 84 / 0 | 14.6 | 10.0 | 19.9 | 13.6 |
| AMP | - 25 % | 84 / 0 | 15.7 | 6.6 | 20.6 | 8.7 |
| AMP | cutoff | 4 / 80 | 25.8 | 7.4 | 28.5 | 8.2 |
| AMP | + 25 % | 0 / 84 | 30.7 | 7.1 | 32.2 | 7.4 |
| AMP | + 50 % | 0 / 84 | 31.1 | 6.1 | 37.6 | 7.3 |
| AMP | + 75 % | 0 / 84 | 29.1 | 4.9 | 41.2 | 7.0 |
| AMP | + 100 % | 0 /.84 | 36.2 | 5.3 | 44.6 | 6.5 |

***Semiquantitative - 500 ng/mL Cutoff***

| Drug | Sample Conc. | Results # Neg / # Pos | Repeatability* | | Intermediate Precision** | |
|---|---|---|---|---|---|---|
| | | | SD, ng/mL | CV, % | SD, ng/mL | CV, % |
| AMP | zero drug | 84 / 0 | 12.6 | 117.0 | 13.2 | 122.5 |
| AMP | - 75 % | 84 / 0 | 21.4 | 16.9 | 25.2 | 20.0 |
| AMP | - 50 % | 84 / 0 | 18.2 | 6.9 | 25.6 | 9.8 |
| AMP | - 25 % | 83 / 1 | 25.6 | 5.7 | 29.7 | 6.6 |
| AMP | cutoff | 0 / 84 | 29.1 | 4.7 | 38.3 | 6.2 |
| AMP | + 25 % | 0 / 84 | 42.0 | 5.5 | 51.7 | 6.8 |
| AMP | + 50 % | 0 / 84 | 40.3 | 4.6 | 49.9 | 5.7 |
| AMP | + 75 % | 0 / 84 | 58.7 | 5.9 | 59.7 | 6.0 |
| AMP | + 100 % | 0 / 84 | 79.0 | 7.0 | 86.3 | 7.6 |

***Semiquantitative - 1000 ng/mL Cutoff***

| Drug | Sample Conc. | Results # Neg / # Pos | Repeatability* | | Intermediate Precision** | |
|---|---|---|---|---|---|---|
| | | | SD, ng/mL | CV, % | SD, ng/mL | CV, % |
| AMP | zero drug | 84 / 0 | 15.9 | 111.1 | 19.0 | 132.6 |
| AMP | - 75 % | 84 / 0 | 21.9 | 9.2 | 29.8 | 12.5 |
| AMP | - 50 % | 84 / 0 | 30.3 | 5.4 | 40.9 | 7.3 |
| AMP | - 25 % | 84 / 0 | 44.3 | 5.2 | 55.0 | 6.4 |
| AMP | cutoff | 6 / 78 | 53.4 | 4.8 | 84.3 | 7.6 |
| AMP | + 25 % | 0 / 84 | 72.5 | 5.6 | 87.5 | 6.7 |
| AMP | + 50 % | 0 / 84 | 65.7 | 4.4 | 96.6 | 6.5 |
| AMP | + 75 % | 0 / 84 | 74.3 | 4.5 | 105.2 | 6.4 |
| AMP | + 100 % | 0 / 84 | 106.7 | 5.9 | 131.4 | 7.3 |

*repeatability = within-run precision

**intermediate precision = total precision / between run precision / between day precision

## Accuracy

The accuracy of this assay was determined against *d*-methamphetamine or *d*-amphetamine GC/MS results. The evaluated cutoff concentrations for the GC/MS testing were the same as the assay screening cutoffs. For both *d*-methamphetamine and *d*-amphetamine, 36 urine samples, obtained from a clinical laboratory were confirmed to be negative by GC/MS and were evaluated with Amphetamines II. 100 % of these normal urines were negative with both the semiquantitative and qualitative assay relative to the 300 ng/mL, 500 ng/mL, and 1000 ng/mL cutoffs. 4 additional unaltered samples, containing *d*-methamphetamine between 85-204 ng/mL (GC/MS confirmed), were tested with the 500 ng/mL cutoff and gave negative results.

36 samples obtained from a clinical laboratory, where they screened positive with a commercially available enzyme immunoassay and were subsequently confirmed positive by GC/MS to contain *d*-methamphetamine or *d*-amphetamine, were evaluated with Amphetamines II. 100 % of these samples were positive relative to the 300 ng/mL, 500 ng/mL, and 1000 ng/mL cutoffs.

In addition, several unaltered near cutoff samples were run for each cutoff. These samples fell in the near cutoff negative range (between - 50 % and cutoff) and the near cutoff positive range (between cutoff and + 50 %). For each cutoff, 4 negative near cutoff samples and 4 positive near cutoff samples were assayed.

Data from the accuracy studies described above were combined with data generated from the unaltered near cutoff urine samples. The following results were obtained with the Amphetamines II assay on the COBAS INTEGRA 800 analyzer relative to the GC/MS values for both *d*-methamphetamine and *d*-amphetamine.

**Amphetamines II Qualitative Assay Results (MAMP)**

| Roche ONLINE DAT AMPII assay | Low Neg | Near Cutoff Negative by GC/MS (between -50 % and cutoff) | Near Cutoff Positive by GC/MS (between cutoff and + 50 %) | High Positive by GC/MS (greater than + 50 %) | Percent Agreement with GC/MS (MAMP) |
|---|---|---|---|---|---|
| *300 ng/mL Cutoff* | | | | | |
| Positive | 0 | 4 | 4 | 36 | 100 % |
| Negative | 36 | 0 | 0 | 0 | 90 % |
| *500 ng/mL Cutoff* | | | | | |
| Positive | 0 | 4 | 4 | 36 | 100 % |
| Negative | 40 | 0 | 0 | 0 | 91 % |
| *1000 ng/mL Cutoff* | | | | | |
| Positive | 0 | 4 | 4 | 36 | 100 % |
| Negative | 36 | 0 | 0 | 0 | 90 % |

**Amphetamines II Semiquantitative Assay Results (MAMP)**

| Roche ONLINE DAT AMPII assay | Low Neg | Near Cutoff Negative by GC/MS (between -50 % and cutoff) | Near Cutoff Positive by GC/MS (between cutoff and + 50 %) | High Positive by GC/MS (greater than + 50 %) | Percent Agreement with GC/MS (MAMP) |
|---|---|---|---|---|---|
| *300 ng/mL Cutoff* | | | | | |
| Positive | 0 | 4 | 4 | 36 | 100 % |
| Negative | 36 | 0 | 0 | 0 | 90 % |
| *500 ng/mL Cutoff* | | | | | |
| Positive | 0 | 4 | 4 | 36 | 100 % |
| Negative | 40 | 0 | 0 | 0 | 91 % |
| *1000 ng/mL Cutoff* | | | | | |
| Positive | 0 | 4 | 4 | 36 | 100 % |
| Negative | 36 | 0 | 0 | 0 | 90 % |

Accuracy samples were categorized based upon the *d*-methamphetamine GC/MS concentration only. The table below identifies those samples with a *d*-methamphetamine concentration below the cutoff, in which the observed result on a COBAS INTEGRA 800 analyzer was positive. The expected results column identifies the result expected with the Amphetamines II assay based upon the *d*-methamphetamine (MAMP) value relative to the cutoff.

**GC/MS Summary of Discrepant Results (MAMP)**

| Cutoff Value (ng/mL) | Roche ONLINE DAT AMP II OBSERVED Result | Roche ONLINE DAT AMP II EXPECTED Result | GC/MS (ng/mL) | Drug / Metabolite |
|---|---|---|---|---|
| 300 (SQ, Q) | Positive | Positive | 173<br>181 | MAMP<br>AMP |
| 300 (SQ, Q) | Positive | Positive | 278<br>101 | MAMP<br>AMP |
| 300 (SQ, Q) | Positive | Positive | 171<br>220 | MAMP<br>AMP |
| 300 (SQ, Q) | Positive | Positive | 291<br>145 | MAMP<br>AMP |
| 500 (SQ, Q) | Positive | Positive | 488<br>466 | MAMP<br>AMP |
| 500 (SQ, Q) | Positive | Negative | 325<br>171 | MAMP<br>AMP |
| 500 (SQ, Q) | Positive | Negative | 291<br>145 | MAMP<br>AMP |
| 500 (SQ, Q) | Positive | Positive | 472<br>650 | MAMP<br>AMP |
| 1000 (SQ, Q) | Positive | Positive | 706<br>443 | MAMP<br>AMP |
| 1000 (SQ, Q) | Positive | Positive | 540<br>693 | MAMP<br>AMP |
| 1000 (SQ, Q) | Positive | Positive | 769<br>395 | MAMP<br>AMP |
| 1000 (SQ, Q) | Positive | Positive | 572<br>432 | MAMP<br>AMP |

**Amphetamines II Qualitative Assay Results (AMP)**

| Roche ONLINE DAT AMPII assay | Low Neg | Near Cutoff Negative by GC/MS (between -50 % and cutoff) | Near Cutoff Positive by GC/MS (between cutoff and + 50 %) | High Positive by GC/MS (greater than + 50 %) | Percent Agreement with GC/MS (AMP) |
|---|---|---|---|---|---|
| *300 ng/mL Cutoff* | | | | | |
| Positive | 0 | 3 | 4 | 36 | 100 % |
| Negative | 36 | 1 | 0 | 0 | 92.5 % |
| *500 ng/mL Cutoff* | | | | | |
| Positive | 0 | 3 | 4 | 36 | 100 % |
| Negative | 36 | 1 | 0 | 0 | 92.5 % |
| *1000 ng/mL Cutoff* | | | | | |
| Positive | 0 | 1 | 4 | 36 | 100 % |
| Negative | 36 | 3 | 0 | 0 | 97.5 % |

**Amphetamines II Semiquantitative Assay Results (AMP)**

| Roche ONLINE DAT AMPII assay | Low Neg | Near Cutoff Negative by GC/MS (between -50 % and cutoff) | Near Cutoff Positive by GC/MS (between cutoff and + 50 %) | High Positive by GC/MS (greater than + 50 %) | Percent Agreement with GC/MS (AMP) |
|---|---|---|---|---|---|
| *300 ng/mL Cutoff* | | | | | |
| Positive | 0 | 3 | 4 | 36 | 100 % |
| Negative | 36 | 1 | 0 | 0 | 92.5 % |
| *500 ng/mL Cutoff* | | | | | |
| Positive | 0 | 3 | 4 | 36 | 100 % |
| Negative | 36 | 1 | 0 | 0 | 92.5 % |
| *1000 ng/mL Cutoff* | | | | | |
| Positive | 0 | 2 | 4 | 36 | 100 % |
| Negative | 36 | 2 | 0 | 0 | 95 % |

Accuracy samples were categorized based upon the *d*-amphetamine GC/MS concentration only. The table below identifies those samples with a *d*-amphetamine concentration below the cutoff, in which the observed result on a COBAS INTEGRA 800 analyzer was positive. The expected results column identifies the result expected with the Amphetamines II assay based upon the *d*-amphetamine (AMP) value relative to the cutoff.

**GC/MS Summary of Discrepant Results (AMP)**

| Cutoff Value (ng/mL) | Roche ONLINE DAT AMP II OBSERVED Result | Roche ONLINE DAT AMP II EXPECTED Result | GC/MS (ng/mL) | Drug / Metabolite |
|---|---|---|---|---|
| 300 (SQ, Q) | Positive | Positive | 157<br>363 | AMP<br>MAMP |
| 300 (SQ, Q) | Positive | Positive | 181<br>173 | AMP<br>MAMP |
| 300 (SQ, Q) | Positive | Positive | 220<br>171 | AMP<br>MAMP |
| 500 (SQ, Q) | Positive | Positive | 438<br>121 | AMP<br>MAMP |
| 500 (SQ, Q) | Positive | Positive | 457<br>1152 | AMP<br>MAMP |
| 500 (SQ, Q) | Positive | Positive | 443<br>706 | AMP<br>MAMP |
| 1000 (SQ) | Positive | Negative | 920 | AMP |
| 1000 (SQ, Q) | Positive | Positive | 837<br>1163 | AMP<br>MAMP |

**Analytical specificity**

The specificity of Amphetamines II, on the COBAS INTEGRA 800 analyzer, for various phenethylamines and structurally similar compounds was determined by generating inhibition curves for each of the compounds listed for both semiquantitative and qualitative modes and determining the approximate quantity of each compound that is equivalent in assay reactivity to the 300 ng/mL, 500 ng/mL, and 1000 ng/mL *d*-methamphetamine assay cutoff. The tables below show the semiquantitative results of the study for each assay cutoff. The same samples were run in the qualitative mode and all recovered appropriately negative or positive, based on the calculated cross-reactivity.

| Compound | ng/mL Equivalent to 300 ng/mL MAMP | Approx. Percent Cross-reactivity |
|---|---|---|
| ± MDMA[e] | 114 | 264 |
| ± MDA[f] | 249 | 121 |
| ± MDEA[h] | 285 | 105 |
| *d*-Amphetamine | 311 | 96 |
| *d*-Methamphetamine | 327 | 92 |
| ± MBDB HCl[g] | 339 | 88 |
| ± BDB HCl[i] | 648 | 46 |
| *l*-Methamphetamine | 2754 | 11 |
| *l*-Amphetamine | 6443 | 5 |
| Phendimetrazine | 47473 | 0.63 |
| Phentermine | 66385 | 0.45 |
| Tyramine | 86271 | 0.35 |
| *d*-Pseudoephedrine | 87578 | 0.34 |
| *l*-Ephedrine | 94792 | 0.32 |
| *d,l*-Phenylpropanolamine HCl | 280374 | 0.11 |
| *d*-Ephedrine | 329670 | 0.09 |

| Compound | ng/mL Equivalent to 500 ng/mL MAMP | Approx. Percent Cross-reactivity |
|---|---|---|
| ± MDMA[e] | 173 | 289 |
| ± MDA[f] | 433 | 115 |
| *d*-Methamphetamine | 444 | 113 |
| *d*-Amphetamine | 460 | 109 |
| ± MDEA[h] | 494 | 101 |
| ± MBDB HCl[g] | 713 | 70 |
| ± BDB HCl[i] | 1209 | 41 |
| *l*-Methamphetamine | 4098 | 12 |
| *l*-Amphetamine | 11174 | 4 |
| Phendimetrazine | 72500 | 0.69 |
| Phentermine | 118483 | 0.42 |
| *d*-Pseudoephedrine | 132275 | 0.38 |
| Tyramine | 141243 | 0.35 |
| *l*-Ephedrine | 154321 | 0.32 |
| *d,l*-Phenylpropanolamine HCl | 390625 | 0.13 |
| *d*-Ephedrine | 413223 | 0.12 |

| Compound | ng/mL Equivalent to 1000 ng/mL MAMP | Approx. Percent Cross-reactivity |
|---|---|---|
| ± MDMA[e] | 446 | 224 |
| ± MDA[f] | 785 | 127 |
| *d*-Methamphetamine | 970 | 103 |
| *d*-Amphetamine | 1024 | 98 |
| ± MBDB HCl[g] | 1194 | 84 |
| ± MDEA[h] | 1203 | 83 |
| ± BDB HCl[i] | 2262 | 44 |
| *l*-Methamphetamine | 9008 | 11 |
| *l*-Amphetamine | 23445 | 4 |
| Phendimetrazine | 156740 | 0.64 |
| *d*-Pseudoephedrine | 269542 | 0.37 |
| Phentermine | 294118 | 0.34 |
| *l*-Ephedrine | 317460 | 0.32 |
| Tyramine | 323625 | 0.31 |
| *d*-Ephedrine | 793651 | 0.13 |
| *d,l*-Phenylpropanolamine HCl | 1111111 | 0.09 |

e) *d,l*-3,4-Methylenedioxymethamphetamine

f) *d,l*-3,4-Methylenedioxyamphetamine

g) *d,l*-N-Methyl-1-(3,4-methylenedioxyphenyl)-2-butanamine hydrochloride

h) *d,l*-3,4-Methylenedioxyethylamphetamine

i) *d,l*-3,4-Methylenedioxyphenyl-2-butanamine hydrochloride

### Interference and cross-reactivity with unrelated drugs

The following compounds were added at the listed concentrations to a human urine pool spiked with either *d*-amphetamine or *d*-methamphetamine at approximately the negative and positive control concentrations for each cutoff (+/- 25 % of assay cutoff). For each compound, the control level samples recovered properly for the 300 ng/mL, 500 ng/mL, and 1000 ng/mL cutoff in both semiquantitative and qualitative modes.

| | | **Semiquantitative All Cutoffs** | | **Qualitative All Cutoffs** | |
|---|---|---|---|---|---|
| **Compound** | **Conc. (ng/mL)** | **Low Ctrl** | **High Ctrl** | **Low Ctrl** | **High Ctrl** |
| Acetaminophen | 100000 | NEG | POS | NEG | POS |
| Acetylsalicylic acid | 100000 | NEG | POS | NEG | POS |
| Amitriptyline | 100000 | NEG | POS | NEG | POS |
| Aspartame | 40000 | NEG | POS | NEG | POS |
| Benzocaine | 100000 | NEG | POS | NEG | POS |
| Benzoylecgonine | 100000 | NEG | POS | NEG | POS |
| Caffeine | 100000 | NEG | POS | NEG | POS |
| Cannabidiol | 100000 | NEG | POS | NEG | POS |
| Cocaine | 100000 | NEG | POS | NEG | POS |
| Codeine | 100000 | NEG | POS | NEG | POS |
| Desipramine HCl | 100000 | NEG | POS | NEG | POS |
| Dextromethorphan | 100000 | NEG | POS | NEG | POS |
| Dextropropoxyphene | 100000 | NEG | POS | NEG | POS |
| Diazepam | 100000 | NEG | POS | NEG | POS |
| Digoxin | 100000 | NEG | POS | NEG | POS |
| Diphenhydramine | 100000 | NEG | POS | NEG | POS |
| Diphenylhydantoin | 100000 | NEG | POS | NEG | POS |
| Doxepin | 100000 | NEG | POS | NEG | POS |
| Ecgonine | 100000 | NEG | POS | NEG | POS |
| Ecgonine methyl ester | 100000 | NEG | POS | NEG | POS |
| Erythromycin | 100000 | NEG | POS | NEG | POS |
| Furosemide | 100000 | NEG | POS | NEG | POS |
| Guaiacol glycerol ether | 100000 | NEG | POS | NEG | POS |
| Hydrochlorothiazide | 100000 | NEG | POS | NEG | POS |
| Ibuprofen | 100000 | NEG | POS | NEG | POS |
| Ketamine | 100000 | NEG | POS | NEG | POS |
| Levothyroxine | 100000 | NEG | POS | NEG | POS |
| LSD | 2500 | NEG | POS | NEG | POS |
| Meperidine | 100000 | NEG | POS | NEG | POS |
| Methadone | 100000 | NEG | POS | NEG | POS |
| Methaqualone | 75000 | NEG | POS | NEG | POS |
| Morphine | 100000 | NEG | POS | NEG | POS |
| Naloxone | 100000 | NEG | POS | NEG | POS |
| Naltrexone | 100000 | NEG | POS | NEG | POS |
| Naproxen | 100000 | NEG | POS | NEG | POS |
| Niacinamide | 100000 | NEG | POS | NEG | POS |
| Nicotine | 100000 | NEG | POS | NEG | POS |

| | | | | | |
|---|---|---|---|---|---|
| Nifedipine | 100000 | NEG | POS | NEG | POS |
| Nordiazepam | 100000 | NEG | POS | NEG | POS |
| Omeprazole | 100000 | NEG | POS | NEG | POS |
| Oxazepam | 100000 | NEG | POS | NEG | POS |
| Penicillin G | 100000 | NEG | POS | NEG | POS |
| Phencyclidine | 40000 | NEG | POS | NEG | POS |
| Phenobarbital | 100000 | NEG | POS | NEG | POS |
| Quinine | 100000 | NEG | POS | NEG | POS |
| Secobarbital | 100000 | NEG | POS | NEG | POS |
| Tetracycline | 100000 | NEG | POS | NEG | POS |
| Δ9-THC | 10000 | NEG | POS | NEG | POS |

The compounds were additionally added to aliquots of pooled drug-free human urine at a concentration of 100000 ng/mL. None of these compounds gave values in the assay that were equal to or greater than 0.19 % cross-reactivity and no results were greater than the assay cutoffs (300 ng/mL, 500 ng/mL, and 1000 ng/mL), with the following exception.

The cross-reactivity for LSD was tested at a concentration of 2500 ng/mL. The results obtained were 0.32 % , and 0.71 %, for the 300 ng/mL and 1000 ng/mL assay cutoffs respectively.

### Interference with other substances

Interfering substances were added to urine containing *d*-methamphetamine (MAMP) at - 25 % and + 25 % of the cutoff level at the concentration listed below. The same substances were additionally added to urine containing *d*-amphetamine (AMP) at - 25 % and + 25 % of the cutoff level at the concentration listed below. All samples were tested and the following results were obtained on a COBAS INTEGRA 800 analyzer. The value in the table indicates the level at which no interference was found for samples containing either *d*-methamphetamine or *d*-amphetamine.

| ***Qualitative*** | | **300 ng/mL Cutoff** | | **500 ng/mL Cutoff** | | **1000 ng/mL Cutoff** | |
|---|---|---|---|---|---|---|---|
| **Compound** | **Cmpd. Conc.** | **Neg Level** | **Pos Level** | **Neg Level** | **Pos Level** | **Neg Level** | **Pos Level** |
| Acetone | 7.9 mg/mL | Neg | Pos | Neg | Pos | Neg | Pos |
| Ascorbic Acid | 10 mg/mL | Neg | Pos | Neg | Pos | Neg | Pos |
| Conjugated Bilirubin | 0.1 mg/mL | Neg | Pos | Neg | Pos | Neg | Pos |
| Creatinine | 5 mg/mL | Neg | Pos | Neg | Pos | Neg | Pos |
| Ethanol | 7.9 mg/mL | Neg | Pos | Neg | Pos | Neg | Pos |
| Glucose | 12 mg/mL | Neg | Pos | Neg | Pos | Neg | Pos |
| Hemoglobin | 1 mg/mL | Neg | Pos | Neg | Pos | Neg | Pos |
| Human serum albumin | 3 mg/mL | Neg | Pos | Neg | Pos | Neg | Pos |
| Oxalic Acid | 2 mg/mL | Neg | Pos | Neg | Pos | Neg | Pos |
| Sodium Chloride | 23 mg/mL | Neg | Pos | Neg | Pos | Neg | Pos |

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| Urea | 60 mg/mL | Neg | Pos | Neg | Pos | Neg | Pos |

The same experiment was performed in the semiquantitative mode for each cutoff. All negative and positive controls recovered properly, within 80 % - 120 %, in the presence of the interfering substance.

An additional protocol was executed in which samples containing either AMP or MAMP at control levels (± 25 % of cutoff) with specific gravities ranging from 1.001 to 1.034 were tested. As with the other interferences, there were no control cross-overs on any of the 3 assay cutoffs at either extreme specific gravity level.
Samples having a pH ranging from 4.5 to 8.0 and containing either AMP or MAMP at control levels (± 25 % of cutoff) were also tested. There were ≤ 5 % cross-overs on any of the 3 assay cutoffs.
Any modification of the instrument as set forth in this labeling requires validation by the laboratory.

## References


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DEPARTMENT OF HEALTH & HUMAN SERVICES

Food and Drug Administration
10903 New Hampshire Avenue
Document Mail Center – WO66-0609
Silver Spring, MD 20993-0002

JUL 2 8 2010

Roche Diagnostics
c/o Mrs. Michelle Neff
9115 Hague Road
Indianapolis, IN 46250

Re: k093664
Trade/Device Name: Amphetamines II Assay
Regulation Number: 21 CFR §862.3100
Regulation Name: Amphetamine Test System
Regulatory Class: Class II
Product Code: DKZ, LAF
Dated: July 2, 2010
Received: July 6, 2010

Dear Mrs. Neff:

We have reviewed your Section 510(k) premarket notification of intent to market the device referenced above and have determined the device is substantially equivalent (for the indications for use stated in the enclosure) to legally marketed predicate devices marketed in interstate commerce prior to May 28, 1976, the enactment date of the Medical Device Amendments, or to devices that have been reclassified in accordance with the provisions of the Federal Food, Drug, and Cosmetic Act (Act) that do not require approval of a premarket approval application (PMA). You may, therefore, market the device, subject to the general controls provisions of the Act. The general controls provisions of the Act include requirements for annual registration, listing of devices, good manufacturing practice, labeling, and prohibitions against misbranding and adulteration.

If your device is classified (see above) into class II (Special Controls), it may be subject to such additional controls. Existing major regulations affecting your device can be found in Title 21, Code of Federal Regulations (CFR), Parts 800 to 895. In addition, FDA may publish further announcements concerning your device in the Federal Register.

Please be advised that FDA's issuance of a substantial equivalence determination does not mean that FDA has made a determination that your device complies with other requirements of the Act or any Federal statutes and regulations administered by other Federal agencies. You must comply with all the Act's requirements, including, but not limited to: registration and listing (21 CFR Part 807); labeling (21 CFR Parts 801 and 809); medical device reporting (reporting of medical device-related adverse events) (21 CFR 803); and good manufacturing practice requirements as set forth in the quality systems (QS) regulation (21 CFR Part 820).

If you desire specific advice for your device on our labeling regulation (21 CFR Part 801), please contact the Office of *In Vitro* Diagnostic Device Evaluation and Safety at (301) 796-5450. Also, please note the regulation entitled, "Misbranding by reference to premarket notification" (21 CFR Part 807.97). For questions regarding postmarket surveillance, please contact CDRH's Office of Surveillance and Biometric's (OSB's) Division of Postmarket Surveillance at (301) 796-5760. For questions regarding the reporting of adverse events under the MDR regulation (21 CFR Part 803), please go to http://www.fda.gov/MedicalDevices/Safety/ReportaProblem/default.htm for the CDRH's Office of Surveillance and Biometrics/Division of Postmarket Surveillance.

You may obtain other general information on your responsibilities under the Act from the Division of Small Manufacturers, International and Consumer Assistance at its toll-free number (800) 638-2041 or ( 301 ) 796-5680 or at its Internet address http://www.fda.gov/MedicalDevices/ResourcesforYou/Industry/default.htm.

Sincerely yours,

Courtney C. Harper, Ph.D.
Director
Division of Chemistry and Toxicology
Office of *In Vitro* Diagnostic Device
Evaluation and Safety
Center for Devices and Radiological Health

Enclosure

# Indication for Use

510(k) Number (if known): K093664 JUL 28 2010

Device Name: **Amphetamines II**
Indication For Use:

Amphetamines II (AMPII) is an in vitro diagnostic test for the qualitative and semiquantitative detection of amphetamines and methamphetamines in human urine on COBAS INTEGRA systems at cutoff concentrations of 300 ng/mL, 500 ng/mL and 1000 ng/mL when calibrated with *d*-methamphetamine. Semiquantitative test results may be obtained that permit laboratories to assess assay performance as part of a quality control program. Semiquantitative assays are intended to determine an appropriate dilution of the specimen for confirmation by a confirmatory method such as gas chromatography/mass spectrometry (GC/MS).
**Amphetamines II provides only a preliminary analytical test result. A more specific alternate chemical method must be used in order to obtain a confirmed analytical result. Gas chromatography/mass spectrometry (GC/MS) is the preferred confirmatory method.[1] Clinical consideration and professional judgement should be applied to any drug of abuse test result, particularly when preliminary positive results are used.**

Prescription Use ___XXX___ And/Or Over the Counter Use ____
(21 CFR Part 801 Subpart D) (21 CFR Part 801 Subpart C)

(PLEASE DO NOT WRITE BELOW THIS LINE; CONTINUE ON ANOTHER PAGE IF NEEDED)

Concurrence of CDRH, Office of In Vitro Diagnostic Device Evaluation and Safety (OIVD)

Division Sign-Off
Office of In Vitro Diagnostic Device
Evaluation and Safety

510(k) K093664