

ADME-Tox ASSAYS

SOLUTION PROPERTIES

| Family/assay | Ref. |
|--|------|
| AQUEOUS SOLUBILITY | |
| aqueous solubility (PBS, pH 7.4) | 0435 |
| aqueous solubility (simulated gastric fluid) | 2061 |
| aqueous solubility (simulated intestinal fluid) | 2062 |
| PARTITION COEFFICIENT (LOG D) | |
| partition coefficient (log D, n-octanol/PBS, pH 7.4) | 0417 |
| partition coefficient (log D, cyclohexane/PBS, pH 7.4) | 1186 |
| pKa (IONIZATION CONSTANT) | |
| pKa (ionization constant) | 1359 |
| PLASMA/SERUM PROTEIN BINDING - BLOOD PARTITIONING | |
| plasma protein binding (equilibrium dialysis) | 2194 |
| human serum albumin (HSA) binding (equilibrium dialysis) | 0736 |
| α -1 acid glycoprotein (AGP) binding (equilibrium dialysis) | 0925 |
| microsomal protein binding (equilibrium dialysis) | 2385 |
| blood partitioning | 1371 |

| Family/assay | Ref. |
|---|------|
| CHEMICAL / PLASMA / BLOOD STABILITY | |
| CHEMICAL STABILITY (2 time-points) | |
| chemical stability (PBS, pH 7.4) | 0785 |
| chemical stability (HBSS, pH 7.4) | 1533 |
| chemical stability (simulated gastric fluid) | 1453 |
| chemical stability (simulated intestinal fluid) | 1454 |
| CHEMICAL STABILITY (5 time-points) | |
| half-life (PBS, pH 7.4) | 0600 |
| half-life (HBSS, pH 7.4) | 1535 |
| half-life (simulated gastric fluid) | 1451 |
| half-life (simulated intestinal fluid) | 1452 |
| PLASMA/BLOOD STABILITY (2 time-points) | |
| plasma stability | 0719 |
| blood stability | 1366 |
| PLASMA/BLOOD STABILITY (5 time-points) | |
| half-life (plasma) | 0887 |
| half-life (blood) | 1437 |

IN VITRO DRUG ABSORPTION/DRUG TRANSPORT

| Family/assay | Ref. |
|--|------|
| A-B PERMEABILITY | |
| A-B permeability (TC7, pH 6.5/7.4) | 0423 |
| A-B permeability (TC7, pH 7.4/7.4) | 1187 |
| A-B permeability (MDRI-MDCKII, pH 7.4/7.4) | 1809 |
| A-B permeability (MDCKII, pH 7.4/7.4) | 1805 |
| B-A PERMEABILITY | |
| B-A permeability (TC7, pH 6.5/7.4) | 0523 |
| B-A permeability (TC7, pH 7.4/7.4) | 1188 |
| B-A permeability (MDRI-MDCKII, pH 7.4/7.4) | 1810 |
| B-A permeability (MDCKII, pH 7.4/7.4) | 1806 |

| Family/assay | Ref. |
|--|------|
| P-GLYCOPROTEIN INTERACTIONS | |
| P-gp substrate assessment (A-B/B-A permeability, pH 7.4/7.4 \pm verapamil) - Option I | 2446 |
| P-gp substrate assessment (A-B/B-A permeability, pH 7.4/7.4 \pm verapamil) - Option II | 2447 |
| P-gp substrate evaluation/FDA | 2794 |
| P-gp inhibitor evaluation/FDA | 2795 |
| P-glycoprotein inhibition (3 H]-digoxin substrate) | 1224 |
| P-glycoprotein inhibition (calcein AM substrate) | 1324 |

IN VITRO DRUG METABOLISM

| Family/assay | Ref. |
|---|------|
| METABOLIC STABILITY /2 TIME-POINTS (microsomes, S9, hepatocytes) | |
| metabolic stability (liver microsomes) | 0416 |
| metabolic stability (liver S9) | 0602 |
| metabolic stability (intestinal microsomes) | 0694 |
| metabolic stability (intestinal S9) | 1412 |
| metabolic stability (cryopreserved hepatocytes) | 1417 |
| INTRINSIC CLEARANCE /5 TIME-POINTS (microsomes, S9, hepatocytes) | |
| intrinsic clearance (liver microsomes) | 0607 |
| intrinsic clearance (liver S9) | 0634 |
| intrinsic clearance (intestinal microsomes) | 1422 |
| intrinsic clearance (intestinal S9) | 1427 |
| intrinsic clearance (cryopreserved hepatocytes) | 1432 |
| CYP PHENOTYPING | |
| STEP I /STABILITY (human recombinant CYPs) | |
| metabolic stability (CYP1A2) - 2 time-points | 1604 |
| metabolic stability (CYP1A2) - 5 time-points | 1645 |

| Family/assay | Ref. |
|--|------|
| CYP PHENOTYPING (cont'd) | |
| STEP I /STABILITY (human recombinant CYPs) (cont'd) | |
| metabolic stability (CYP2A6) - 2 time-points | 3120 |
| metabolic stability (CYP2A6) - 5 time-points | 3161 |
| metabolic stability (CYP2B6) - 2 time-points | 2226 |
| metabolic stability (CYP2B6) - 5 time-points | 2387 |
| metabolic stability (CYP2C8) - 2 time-points | 2043 |
| metabolic stability (CYP2C8) - 5 time-points | 2384 |
| metabolic stability (CYP2C9) - 2 time-points | 1602 |
| metabolic stability (CYP2C9) - 5 time-points | 1646 |
| metabolic stability (CYP2C19) - 2 time-points | 1603 |
| metabolic stability (CYP2C19) - 5 time-points | 1647 |
| metabolic stability (CYP2D6) - 2 time-points | 0716 |
| metabolic stability (CYP2D6) - 5 time-points | 1648 |
| metabolic stability (CYP2E1) - 2 time-points | 2706 |

■ new assay
 ■ new protocol
 ■ human
 ■ human & other species

IN VITRO DRUG METABOLISM ■ (cont'd)

| Family/assay | Ref. | Family/assay | Ref. |
|--|--------|--|---------|
| CYP PHENOTYPING (cont'd) | | UGT PHENOTYPING/2 OR 5 TIME-POINTS (cont'd) | |
| STEP I/STABILITY (human recombinant CYPs) (CONT'D) | | ☐ metabolic stability (UGT1A8) - 5 time-points | 🇫 3165 |
| metabolic stability (CYP2E1) - 5 time-points | 🇫 2707 | ☐ metabolic stability (UGT1A9) - 2 time-points | 🇫 3088 |
| metabolic stability (CYP3A4) - 2 time-points | 🇫 0715 | ☐ metabolic stability (UGT1A9) - 5 time-points | 🇫 3166 |
| metabolic stability (CYP3A4) - 5 time-points | 🇫 1649 | ☐ metabolic stability (UGT1A10) - 2 time-points | 🇫 3148 |
| metabolic stability (CYP3A5) - 2 time-points | 🇫 1362 | ☐ metabolic stability (UGT1A10) - 5 time-points | 🇫 3167 |
| metabolic stability (CYP3A5) - 5 time-points | 🇫 1650 | ☐ metabolic stability (UGT2B4) - 2 time-points | 🇫 3149 |
| STEP II - STABILITY (HLM with CYP selective inhibitors) | | ☐ metabolic stability (UGT2B4) - 5 time-points | 🇫 3168 |
| ☐ metabolic stability (HLM with furafylline) | 🇫 3128 | metabolic stability (UGT2B7) - 2 time-points | 🇫 1858 |
| ☐ metabolic stability (HLM with tranlycypromine) | 🇫 3129 | metabolic stability (UGT2B7) - 5 time-points | 🇫 1860 |
| ☐ metabolic stability (HLM with ticlopidine) | 🇫 3130 | ☐ metabolic stability (UGT2B15) - 2 time-points | 🇫 3150 |
| ☐ metabolic stability (HLM with quercetin) | 🇫 3131 | ☐ metabolic stability (UGT2B15) - 5 time-points | 3169 |
| ☐ metabolic stability (HLM with sulphaphenazole) | 🇫 3132 | PLASMA/BLOOD STABILITY | |
| ☐ metabolic stability (HLM with nookatone) | 🇫 3133 | 2 TIME-POINTS | |
| ☐ metabolic stability (HLM with quinidine) | 🇫 3134 | plasma stability | 🇫🇺 0719 |
| ☐ metabolic stability (HLM with clomethiazole) | 🇫 3135 | blood stability | 🇫🇺 1366 |
| ☐ metabolic stability (HLM with ketoconazole) | 🇫 3136 | 5 TIME-POINTS | |
| UGT PHENOTYPING/2 OR 5 TIME-POINTS | | half-life (plasma) | 🇫🇺 0887 |
| metabolic stability (UGT1A1) - 2 time-points | 🇫 1859 | half-life (blood) | 🇫🇺 1437 |
| metabolic stability (UGT1A1) - 5 time-points | 🇫 1861 | METABOLITE IDENTIFICATION | |
| ☐ metabolic stability (UGT1A3) - 2 time-points | 🇫 3086 | ☐ metabolite screen | 🇫🇺 3018 |
| ☐ metabolic stability (UGT1A3) - 5 time-points | 🇫 3162 | metabolite detection (microsomes, S9, CYP, plasma) | 🇫🇺 0434 |
| ☐ metabolic stability (UGT1A6) - 2 time-points | 🇫 3146 | metabolite detection (hepatocytes, cryopreserved, blood) | 🇫🇺 0434 |
| ☐ metabolic stability (UGT1A6) - 5 time-points | 🇫 3163 | metabolite characterization | 🇫🇺 0608 |
| ☐ metabolic stability (UGT1A7) - 2 time-points | 🇫 3147 | CONJUGATE DETECTION | |
| ☐ metabolic stability (UGT1A7) - 5 time-points | 🇫 3164 | glutathione conjugate detection (liver S9) | 🇫🇺 0530 |
| ☐ metabolic stability (UGT1A8) - 2 time-points | 🇫 3087 | glucuronide conjugate detection (liver microsomes) | 🇫🇺 0610 |

IN VIVO PK/BBB ■

| Family/assay | Ref. | Family/assay | Ref. |
|--|------|--------------------------------|------|
| PHARMACOKINETICS | | BLOOD-BRAIN BARRIER | |
| <i>in vivo</i> rat PK (serial sampling) - IV | 1280 | <i>in vivo</i> BBB (rat, IV) | 1150 |
| <i>in vivo</i> rat PK (serial sampling) - PO | 1278 | <i>in vivo</i> BBB (mouse, IV) | 0790 |
| <i>in vivo</i> mouse PK (parallel sampling) - IV | 0809 | EXCRETION | |
| <i>in vivo</i> mouse PK (parallel sampling) - PO | 0808 | biliary excretion (rat, IV) | 2390 |
| <i>in vivo</i> mouse PK (serial sampling) - IV | 2388 | renal excretion (rat, IV) | 2391 |
| <i>in vivo</i> mouse PK (serial sampling) - PO | 2389 | | |

BIOANALYTICAL SUPPORT ■

| Family/assay | Ref. | Family/assay | Ref. |
|---|---------|--|---------|
| ANALYTICAL METHOD DEVELOPMENT | | BIOANALYTICAL SUPPORT OF PHARMACOKINETIC STUDIES (cont'd) | |
| HPLC-MS screen | 0881 | quantitative bioanalysis | 🇫🇺 1218 |
| HPLC-MS optimization | 1265 | DRUG EXPOSURE MEASUREMENT/DOSE SOLUTION ANALYSIS | |
| BIOANALYTICAL SUPPORT OF PHARMACOKINETIC STUDIES | | drug exposure measurement | 2448 |
| linearity | 🇫🇺 1615 | hERG dose solution analysis (non GLP) | 1978 |
| recovery | 🇫🇺 2444 | | |

CYP-MEDIATED DRUG-DRUG INTERACTION ■

| Family/assay | Ref. | Family/assay | Ref. |
|---|--------|--|--------|
| CYTOCHROME P450 INHIBITION / HLM | | CYTOCHROME P450 INHIBITION / HLM (cont'd) | |
| ☐ cocktail CYP inhibition (HLM, 5 substrates) | 🇫 3090 | CYP2E1 inhibition (HLM, chlorzoxazone substrate) | 🇫 2068 |
| CYP1A inhibition (HLM, phenacetin substrate) | 🇫 2064 | CYP3A inhibition (HLM, midazolam substrate) | 🇫 1770 |
| ☐ CYP2A6 (HLM, coumarin substrate) | 🇫 3119 | CYP3A inhibition (HLM, testosterone substrate) | 🇫 1769 |
| CYP2B6 inhibition (HLM, bupropion substrate) | 🇫 2065 | CYTOCHROME P450 INHIBITION / RECOMBINANT, DRUG PROBES | |
| CYP2C8 inhibition (HLM, paclitaxel substrate) | 🇫 2244 | ☐ cocktail CYP inhibition (5 rCYPs, 5 substrates) | 🇫 3091 |
| CYP2C9 inhibition (HLM, diclofenac substrate) | 🇫 2066 | CYP1A2 inhibition (recombinant, phenacetin substrate) | 🇫 1958 |
| CYP2C19 inhibition (HLM, omeprazole substrate) | 🇫 1772 | CYP2C9 inhibition (recombinant, diclofenac substrate) | 🇫 2067 |
| CYP2D6 inhibition (HLM, dextromethorphan substrate) | 🇫 1838 | | |

☐ new assay 🇫 new protocol 🇫 human 🇫🇺 human & other species 🇫 binding assay

CYP-MEDIATED DRUG-DRUG INTERACTION ■ (cont'd)

| Family/assay | Ref. | Family/assay | Ref. |
|--|------|---|------|
| CYP450 INHIBITION / RECOMBINANT, DRUG PROBES (cont'd) | | CYTOCHROME P450 TIME-DEPENDENT INHIBITION / HLM | |
| CYP2C19 inhibition (recombinant, omeprazole subst.) | 1184 | time-dependent CYP1A inhibition (HLM, phenacetin subst.) | 0636 |
| CYP2D6 inhibition (recombinant, dextromethorphan subst.) | 0988 | time-dependent CYP2C9 inhibition (HLM, diclofenac subst.) | 0635 |
| CYP3A4 inhibition (recombinant, midazolam substrate) | 1183 | time-dependent CYP2C19 inhib. (HLM, omeprazole subst.) | 2395 |
| CYP3A4 inhibition (recombinant, testosterone substrate) | 0940 | time-dependent CYP2D6 inhib. (HLM, dextromethorphan subst.) | 2153 |
| CYP450 INHIBITION / RECOMBINANT, FLUORESCENT PROBES | | CYTOCHROME P450 INDUCTION | |
| CYP1A2 inhibition (recombinant, CEC substrate) | 0389 | CYP1A induction (human hepatocytes) - 1 donor | 1919 |
| CYP2B6 inhibition (recombinant, EFC substrate) | 0415 | CYP1A induction (human hepatocytes) - 3 donors | 2401 |
| CYP2C8 inhibition (recombinant, DBF substrate) | 1201 | CYP2B6 induction (human hepatocytes) - 1 donor | 2383 |
| CYP2C9 inhibition (recombinant, MFC substrate) | 0412 | CYP2B6 induction (human hepatocytes) - 3 donors | 2403 |
| CYP2C19 inhibition (recombinant, CEC substrate) | 0390 | CYP3A induction (human hepatocytes) - 1 donor | 1833 |
| CYP2D6 inhibition (recombinant, MFC substrate) | 1338 | CYP3A induction (human hepatocytes) - 3 donors | 2402 |
| CYP2E1 inhibition (recombinant, EC substrate) | 0414 | CYP1A induction (rat hepatocytes) - pool of 3-5 rats | 1818 |
| CYP3A4 inhibition (recombinant, BFC substrate) | 0391 | CYP3A induction (rat hepatocytes) - pool of 3-5 rats | 1832 |
| CYP3A4 inhibition (recombinant, BzRes substrate) | 0939 | UDP-GLUCURONOSYL TRANSFERASE INHIBITION | |
| CYP3A5 inhibition (recombinant, BFC substrate) | 0786 | UGT1A1 inhibition (recombinant, estradiol substrate) | 1864 |

CARDIAC TOXICITY ■

| Family/assay | Ref. | Family/assay | Ref. |
|---|------|---|------|
| K⁺ CHANNEL / hERG | | K⁺ CHANNEL / K_v1.5 (cont'd) | |
| hERG (automated patch-clamp) - 3 conc., n=2 | 2245 | K _v 1.5 (automated patch-clamp) - 5 conc., n=2 | 2392 |
| hERG (automated patch-clamp) - 5 conc., n=2 | 2245 | K _v 1.5 (conventional patch-clamp) - 1 conc., n=2 | 1865 |
| hERG (conventional patch-clamp) - 1 conc., n=2 | 2027 | K _v 1.5 (conventional patch-clamp) - 5 conc., n=2 | 1865 |
| hERG (conventional patch-clamp) - 5 conc., n=2 | 2027 | NA⁺ CHANNEL / NA_v1.5 | |
| hERG (conventional patch-clamp) - GLP compliant | 2063 | NA _v 1.5 (automated patch-clamp) - 3 conc., n=2 | 2393 |
| hERG (membrane preparation) - antagonist radioligand | 1868 | NA _v 1.5 (automated patch-clamp) - 5 conc., n=2 | 2393 |
| hERG dose solution analysis (non GLP) | 1978 | NA _v 1.5 (conventional patch-clamp) - 1 conc., n=2 | 1938 |
| K⁺ CHANNEL / K_v1.5 | | NA _v 1.5 (conventional patch-clamp) - 5 conc., n=2 | |
| K _v 1.5 (automated patch-clamp) - 3 conc., n=2 | 2392 | | |

IN VITRO TOXICITY ■

| Family/assay | Ref. | Family/assay | Ref. |
|----------------------------|------|------------------------------------|------|
| cytotoxicity panel (HepG2) | 1537 | cell viability (HepG2) | 0430 |
| phospholipidosis (HepG2) | 2396 | cell viability (human hepatocytes) | 0533 |
| neurite outgrowth (PC12) | 2397 | cell viability (rat hepatocytes) | 0789 |

GENETIC TOXICITY ■

| Family/assay | Ref. |
|---|------|
| Ames test -TA98/TA100/TA1535 (± S9) | 1899 |
| <i>in vitro</i> micronucleus in CHO-K1 (± S9) | 1900 |

ASSAY CATALOG REFERENCES

In 2009, Cerep finalized one of the milestones in the industrialization process: the implementation of new referencing and supply chain management systems. The references of each of the assays have thus been simplified: **each reference will now be displayed as 4 digits.**

A correlation table between old and new assay references is available at www.cerep.com **CATALOG ONLINE**
<<http://www.cerep.com/Cerep/Users/pages/Catalog/Assay/catalog.asp>>

QUESTIONS OR CONCERNS ?

Please contact us: sales@cerep.com



TESTING CONDITIONS

REQUESTED COMPOUND INFORMATION

To reduce the registration time and ensure that all the appropriate information is available to start the study in a shortest possible timeframe, please use **Cerep compound submission form** ¹ or MS Excel file ², and provide the following **compound information**:

- **Name (compound ID) / Batch # / Molecular weight** ³ / **Formula weight** ⁴ / **Stock concentration** / **Stock solvent** / **Quantity** / **Unit** / **Form** / **Storage conditions** / **Solubility**, as well as **Plate ID/plate position** for compounds delivered in plates, **Comments** ⁵, and **Quotation number**.

NOTE: Impurity and colored compounds might affect the results (compound color information is mentioned in the study report).

► General remarks:

- If compound(s) are supplied as a stock solution in plate(s) (preferred format for any submission of 10 or more compounds), please leave columns 1 and 12 empty in a 96W plate. The 384W plate format is also acceptable with columns 1, 2, 23 and 24 empty. For any other plate format, please inquire.
- If compound(s) are not soluble in 100% DMSO, please provide any useful information concerning the solubility of the compound. The following solvents are compatible with most of our assays: DMSO (Cerep standard), H₂O, Methanol, Tris/HCl 10 mM pH 7.4.
- Organic solvents such as acetone, chloroform, ether, acetonitrile (except for CYP assays), tetrahydrofuran and trifluoroacetic acid are not recommended as they will significantly affect the results from many *in vitro* assays, even at very low concentrations.
- Any study including mass spectrometry (MS) assays (such as stability, permeability, protein binding) requires both molecular weight (MW) and formula weight (FW) of each tested compound, even if they are the same. The FW is required to prepare the stock solution of each compound at the correct concentration. The MW gives indication on which expected molecular ion to look for in our LC-MS screen assay.
- Chemical structure is required for Metabolite characterization.
- Chemical structure or list of the ionizable groups is required for pKa.
- Solubility information is useful and recommended for pKa and genotoxicity testing (due to high test concentrations).

WARNING: Cerep will apply the standard solubilization process when compounds are received at the testing site, unless special instructions are provided with the compounds.

Customized handling procedure of compounds can be accommodated, please inquire for pricing conditions.

¹ Cerep compound submission form will be emailed to you with your quotation. A copy can be requested from sales@cerep.com, or downloaded from Cerep website: www.cerep.com/Catalog Online

² Systematically required for studies of 10 compounds or more.

³ Molecular weight (MW) of free acid or base form.

⁴ Formula weight (FW) including salt form and/or hydrate form if applicable.

⁵ e.g. useful information such as sensitivity to light, stability or hygroscopicity issues.

SAMPLE SIZE

Assuming a molecular weight ≤ 500 g/mol and assays tested at default concentrations (including retest).

| | WEIGHT (pre-weighed) | VOLUME (100% DMSO) |
|---|-------------------------|----------------------------|
| INDIVIDUAL ADME-TOX CATALOG ASSAYS | | |
| 1 to 20 assays | 1 - 2 mg | 200 µL @ 10 mM (50 µL min) |
| 1 to 10 IC ₅₀ follow-up assays | 1.5 - 2 mg | 250 µL @ 10 mM (50 µL min) |
| Weights needed in addition to the general weight required for 1 to 20 ADME-Tox assays: | | |
| . pKa | 3 x 1.5 mg | - |
| . CYP1A, 2B6 & 3A induction (3 donors) | 6 mg (2 mg/CYP) | 120 µL @ 100 mM |
| . hERG (conventional patch-clamp)-GLP compliant | 8 - 10 mg | - |
| . Ames (3 strains) | 4 mg | 800 µL @ 10 mM |
| . <i>in vitro</i> micronucleus | 1 mg | 40 µL @ 50 mM |
| . typical rat PK ¹ | 25 mg | - |
| . typical mouse PK (serial sampling) ¹ | 10 mg | - |
| . typical mouse PK (parallel sampling) ¹ | 20 mg | - |
| . typical rat BBB ² | 15 mg | - |
| . typical mouse BBB ² | 10 mg | - |
| . Plasma sample for quantitative bioanalysis | - | 100 µL |

¹ 2 routes, 1 dose: 1 mg/kg for IV and 5 mg/kg for PO, n=3

² 1 route, 1 dose: 1 mg/kg for IV, 3 time points, n=3



FRANCE
Le Bois l'Évêque
86600 CELLE L'ÉVESCAULT
tel. +33 (0)5 49 89 30 00

(Headquarters)
155 boulevard Haussmann
75008 PARIS
tel. +33 (0)1 45 64 44 60

USA
15318 N.E. 95th Street
REDMOND, WA 98052
tel. +1 (425) 895 8666

JAPAN
Namiki Shoji Co., Ltd.
Kenseishinjuku Bldg. 5-5-3
Shinjuku, Shinjuku-ku
TOKYO, 160-0022
tel. +81 (0)3 3354 4026
fax +81 (0)3 3352 2196

CHINA
Ai Di Sheng (Edison) Road 326,
302-1 room
Zhangjiang High-Tech Park
SHANGHAI
tel. +86 18702160370