

## R1063 Lot No. 2010206

Sprague Dawley (SD) Rat Liver Microsomes Clofibric Acid-treated Male, Pool of 25 0.5 mL at 20 mg protein / mL Suspension medium: 250 mM Sucrose

| Specific Content and Enzyme Activities                           | Content / Rate                                 |                         |
|--|--|-------------------------|
| Cytochrome P450 content<br>Cytochrome b₅ content                 | (nmol/mg protein)<br>(nmol/mg protein)         | 1.308<br>0.613          |
| NADPH-cytochrome <i>c</i> reductase Lauric acid 12-hydroxylation | (nmol/mg protein/min)<br>(pmol/mg protein/min) | 320 ± 16<br>13050 ± 570 |

Background: Treatment of rats with the peroxisome proliferator, clofibric acid, causes a marked induction (>7-fold) of liver microsomal CYP4A levels, which is associated with an increase in lauric acid 12-hydroxylation. The above results confirm the anticipated induction of CYP4A activity. Note: Clofibric acid is not the same as clofibrate, although both compounds are peroxisome proliferators and CYP4A inducers. Clofibrate is the ethyl ester of clofibric acid and is not readily soluble in water, in contrast to the free acid, clofibric acid.

To measure cytochrome P450 (CYP) activity, treated liver microsome samples (0.05 mg/mL) were incubated in triplicate at  $37 \pm 2^{\circ}$ C for 10 minutes in potassium phosphate buffer (50 mM, pH 7.4), containing MgCl<sub>2</sub> (3.0 mM), EDTA (1.0 mM), NADP (1.0 mM), glucose-6-phosphate (5.0 mM), glucose-6-phosphate dehydrogenase (1 Unit/mL) and lauric acid (100  $\mu$ M), at the final concentrations indicated. Metabolite formation was determined by validated LC-MS/MS methods with deuterated metabolites as internal standards.

| Animal Information   |   |   |   |  |
|--|---|---|---|--|
| Species:<br>Strain:<br>Sex:<br>Age:  | Rat<br>* IGS Sprague Dawley<br>Male<br>~ 8 weeks  | Treatment:<br>Source:<br>Vehicle:<br>Concentration: | Clofibric acid<br>Sigma (Cat. No. 197777)<br>Saline, pH adjusted to 7 with NaOH<br>40 mg/mL |  |
| Vendor:  | Charles River, Raleigh, NC  | Regimen:  | 200 mg/kg body weight once per day on days 1-4,<br>liver microsomes prepared on day 5       |  |
| *International Ger<br>Animals were hou   | ietic Standard<br>sed in an AAALAC-accredited fac   | ility and allowed to accli                          | mate <u>&gt;</u> seven days before use.   |  |
| Food:<br>Water:<br>Light/dark cycle:<br>Temperature:<br>Humidity:<br>Bedding:<br>Cage: | Purina 5L79 ( <i>ad libitum</i> )<br>Automatic watering system<br>5:00 am - 5:00 pm, light; 5<br>70°F ± 2°F<br>30-70 %<br>Beta Chip (hardwood), NEF<br>Polycarbonate Shoebox Ca | :00 pm - 5:00 am, dark<br>PCO, Warrensburg, N       | (   |  |



## Store at -80°C

CAUTION: This sample should be considered as a potential biohazard and universal precautions should be followed. Intended for *in vitro* use only.

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