

R1000.IS9 Lot No. 1910195

Sprague Dawley (SD) Rat Intestine S9 Fraction Untreated, Male, Pool of 235
1.0 mL at 4 mg protein / mL

Suspension medium: 50 mM Tris·HCl, 150 mM KCl, 1 mM EDTA, 20% glycerol,

heparin, PMSF, leupeptin, DTT, aprotinin

Enzyme Activities		Rate
NADPH-cytochrome <i>c</i> reductase Midazolam 1'-hydroxylation Testosterone 6β-hydroxylation Glucuronidation of 4-Methylumbelliferone	(nmol/mg protein/min) (pmol/mg protein/min) (pmol/mg protein/min) (nmol/mg protein/min)	32.8 ± 0.3 25.1 ± 1.3 65.2 ± 5.4 89.5

Values for enzyme activities were determined at a single substrate concentration and are mean ± standard deviation of three or more determinations.

Aprotinin, Leupeptin, and Phenylmethylsulfonyl-fluoride were used in the preparation of this S9 fraction. Subcellular fractions were prepared from duodenal and jejunal tissue.

To measure cytochrome P450 (CYP) activity, intestine S9 samples (0.2 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 MgCl₂ (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 testosterone (250 μ M) or midazolam (30 μ M), at the final concentrations indicated. Metabolite formation was determined by validated LC-MS/MS methods with deuterated metabolites as internal standards.

To measure UDP-glucuronosyltransferase (UGT) activity, intestine S9 samples (0.2 mg/mL) were incubated in triplicate at $37 \pm 2^{\circ}$ C for 10 minutes in Tris-HCl (100 mM, pH 7.7 at 37° C), CHAPS (0.5 mM), EDTA (1.0 mM), MgCl₂ (10 mM), D-saccharic acid 1,4-lactone (100 μ M), uridine diphosphate-glucuronic acid (10.0 mM) and 4-methylumbelliferone (1 mM), at the final concentrations indicated. Metabolite formation was determined by validated LC-MS/MS methods with deuterated metabolites as internal standards.

Animal Information

Species: Rat

Strain: International Genetic Standard (IGS), Sprague Dawley

Sex: Male Age: ~8 weeks

Vendor: Charles River, Raleigh, NC

Animals were housed in an AAALAC-accredited facility and allowed to acclimate ≥ seven days before use.

Food: Purina 5L79 (ad libitum)

Water: Automatic watering system (ad libitum)

Light/dark cycle: 5:00 am - 5:00 pm, light; 5:00 pm - 5:00 am, dark (12-hour light/dark)

Temperature: $70^{\circ}\text{F} \pm 2^{\circ}\text{F}$ Humidity: $30-70^{\circ}$ %

Bedding: Beta Chip (hardwood), NEPCO, Warrensburg, NY Cage: Polycarbonate Shoebox Cage, conventional cage



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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