The objective of this study was to develop a rat model to differentiate the roles of absorption and metabolism in limiting the oral bioavailability of certain drugs and to discern the importance of first-pass metabolism in gut vs. liver. Rats were dosed with triazolam (TRZ), dextromethorphan (DM), and atenolol (AT) as a capsule IV (1-10 mg/kg) or PO (3-20 mg/kg), with or without pre-treatment with 1-aminobenzotriazole (ABT) IV (50 mg/kg) or IP+PO (100, 50 mg/kg) and via intraperitoneal vein (IPV) without ABT pre-treatment. PK parameters were estimated based on plasma samples collected from the jugular vein. Following oral dosing, the systemic exposure (AUC) of TRZ and DM increased 36- and 22-fold, respectively, in rats pre-treated with ABT IP+PO and 17- and 8-fold, respectively, in rats pre-treated with ABT IV. Following IV dosing, the systemic exposure of TRZ and DM increased significantly in rats pre-treated with ABT, either IP+PO or IV. There was no significant change in the rate and extent of absorption or metabolism of atenolol in ABT pre-treated vs. untreated rats. Low systemic exposure in ABT pre-treated rats implies absorption-limited systemic exposure, whereas large increases in exposure with ABT pre-treatment suggest that first-pass metabolism is the limiting factor. The current study demonstrates the utility and limitations of the model in studying first-pass gut and hepatic loss of drugs with estimation of Φ(x,F) and Φ(F).

Methods

Study Design and Dosing: ABT was dosed as a solution in saline and all the test formulations were prepared and dosed in male Sprague-Dawley rats as a capsule in 15% Solutol : NMP (1:2) in normal saline. Samples were collected from JVC following IV, IPV, and PO dosing.

Sample analysis: Plasma samples were extracted with acetonitrile and test compounds were quantified by LC-MS/MS. LLOQ was 1 ng/mL in each case.

Pharmacokinetic (PK) analysis: Noncompartmental PK parameters were calculated using WinNonlin 4.1. Bioavailability (%F) was calculated as the ratio of the mean dose-normalized AUC values for oral and IV groups.

Calculation of Φ Φ(x,F) and Φ(F): Fraction escaped from the liver (Φ(x,F); hepatic first-pass), fraction escaped from the gut (Φ(x,F); intestinal first-pass), overall first-pass (Φ(x,F); and fraction absorbed (Φ(F)) were calculated using dose-normalized AUCs. Exposure ratio of test compounds after PO and IV dosing was the ratio of AUCs in presence and absence of ABT. Hepatic clearance (CLH) was estimated as the product of (1-Φ(x,F)) x QH where QH is the hepatic blood flow in rats, reported as 3.3 L/h/kg.

Statistical analysis: All statistical tests were performed using GraphPad Prism (Version 4; San Diego, CA). One-way analysis of variance was performed, and a minimum P value of 0.05 was used as the significance level for all tests. Pair-wise comparisons were performed by the Student-Newman-Keuls method.

Conclusion

- Rapid clearance of TRZ and DM following PO and IV dosing, due to extensive extraction in the gut and liver, decreased significantly in ABT pretreated animals.
- AT has incomplete absorption and low hepatic clearance.
- ABT is useful for distinguishing poor absorption vs. rapid first-pass metabolism.
- IPV vs. IV dosing is superior to IV vs. IP+PO pre-treatment with ABT to assess the extent of hepatic metabolism.