Biotransformation of BOF-4272, a sulfoxide-containing drug, in the cynomolgus monkey

S. NAITO¹, M. NISHIMURA¹ and Y. JIN²

¹Laboratory of Drug Metabolism Research, Naruto Research Institute, Otsuka Pharmaceutical Factory, Inc., Naruto, Tokushima, Japan
²Tokai Research Laboratories, Daiichi Pure Chemicals Co., Ltd, Ibaraki, Japan

Received for publication: September 13, 1999

Keywords: Metabolism, cynomolgus monkey, xanthine oxidase inhibitor, sulfoxide-containing drug, BOF-4272

SUMMARY

BOF-4272, (±)-8-(3-methoxy-4-phenylsulfinylphenyl) pyrazolo[1,5-a]-1,3,5-triazine-4(1H)-one, is a new drug intended for the treatment of hyperuricemia. This report describes the pharmacokinetics and detailed metabolic pathways of BOF-4272 in the cynomolgus monkey, which were investigated using the metabolites found in plasma, urine, and faeces after intravenous and oral administration. M-4 was the main metabolite in plasma after intravenous administration. M-3 and M-4 were the main metabolites in plasma after oral administration. The Cₘₐₓ and AUC₀-t of M-4 were the highest of all the metabolites after intravenous administration. The Cₘₐₓ and AUC₀-t of M-3 were the highest of all the metabolites, and those of M-4 were the second highest, after oral administration. M-4 and M-3 were the main metabolites detected in urine and faeces, respectively, after intravenous administration, with M-4 and M-3 at 47.2% in urine and 19.1% in faeces, respectively, within 120 h after administration. M-4 was the only metabolite detected in urine after oral administration, at about 5% within 120 h after administration. M-3 was detected in faeces at 17.0% within 120 h after oral administration. These results suggest that, in the cynomolgus monkey, BOF-4272 is rapidly biotransformed to a main metabolite (M-4, a sulfoxide-containing metabolite of BOF-4272) and that M-4 is mainly excreted in urine and possibly also in bile, with subsequent conversion to M-3 by the intestinal flora. It is expected that the biotransformation of BOF-4272 would be similar in healthy human volunteers.

INTRODUCTION

BOF-4272, a derivative of pyrazolotriazine, is a new drug that has been developed for the treatment of hyperuricemia and ischemic reperfusion injury (1–4). BOF-4272 inhibits the de novo biosynthesis of uric acid by blocking the xanthine oxidase/xanthine dehydrogenase system, which catalyses the last step of purine catabolism (1,4). The mechanism of the inhibitory action of BOF-4272 has been elucidated by an in vitro study using milk xanthine oxidase/xanthine dehydrogenase (5). BOF-4272 significantly decreases the concentration of the free radicals generated by xanthine oxidase and consequently reduces cellular necrosis (6). The pharmacological activity of BOF-4276 is weaker than that of BOF-4272, whereas BOF-4269 has no activity (7). Recent studies have demonstrated that the hepatic elimination of BOF-
280

Eur. J. Drug Metab. Pharmacokinet. 1999, No. 3

4272 is quite substantial (8–10). It has also been demonstrated that the linear range of absorption and/or elimination of BOF-4272 is very wide in the mouse and the rat (11). It has been shown that BOF-4272 is rapidly biotransformed to a main metabolite (M-4, a sulfoxide-containing metabolite of BOF-4272), that M-4 is then mainly excreted in urine, and that BOF-4269 is a minor metabolite in plasma after oral administration to healthy volunteers (4). It has been suggested that we should clarify the metabolic pathways of BOF-4272 in the rat (12) and the dog (13). However, M-4, which is a metabolite of BOF-4272, is not excreted in urine but in bile, and BOF-4272 is mainly metabolised to BOF-4269 by the intestinal flora in the rat (12). BOF-4272 is not metabolised to M-4, and BOF-4269, which may be metabolised by the intestinal flora, is the main metabolite of BOF-4272 in the dog (13). This paper describes the detailed biotransformation of BOF-4272 in the cynomolgus monkey. We expect that the biotransformation of BOF-4272 in the cynomolgus monkey should be similar to that in healthy human volunteers.

MATERIALS AND METHODS

Materials

[^14C]-labelled BOF-4272 (3.89 MBq/mg) was obtained from Amersham International plc (Buckinghamshire, UK). The radiochemical purity was greater than 97.0%. The BOF-4272 and its metabolites used in this study were synthesised at Otsuka Pharmaceutical Factory, Inc. (Tokushima, Japan). The chemical purity was greater than 99.0%. Carboxymethyl cellulose sodium salt (CMC), acetonitrile, and ethyl acetate were purchased from Wako Pure Chemical Industries, Ltd (Osaka, Japan). Polyethylene glycol 400 (PEG 400) was purchased from Nacalai Tesque, Inc. (Kyoto, Japan).

All other chemicals and reagents used were of analytical reagent grade.

Animals

The animals used were male cynomolgus monkeys weighing 4.06–6.52 kg (n = 12) purchased from Charles River Japan (Kanagawa, Japan) and Japan SLC, Inc. (Shizuoka, Japan). During the experiment, the monkeys were housed individually in metabolic cages at a temperature of 23 ± 2°C and a relative humidity of 55 ± 10% with a 12 h night/day cycle. The animals were allowed free access to food (#5048, Ralston Purina Company, USA), banana, apple, sweet potato, and lemon, except during periods of fasting. The animals were allowed free access to drinking water all the time.

Chromatography

BOF-4272 concentrations were measured using high-performance liquid chromatography (HPLC) systems (CCP & 8010 Series, Tosoh Co., Tokyo, Japan) with a stationary phase of TSKgel ODS-120T (250 x 4.6 mm ID, Tosoh Co.). The HPLC system consisted of a system controller (PX-8010), pump (CCPM), autosampler (AS-48), UV detector (UV-8010), and integrated data analyser (C-R4AX Chromatopac, Shimadzu Co., Kyoto, Japan). The detector wavelength and the flow rate were 323 nm and 1.0 ml/min, respectively. The column temperature was ambient. The mobile phase was a mixture of solution A (10 mM NH₄H₂PO₄, pH 3.0) and solution B (acetonitrile and solution A, 80:20 v/v), with a gradient from 70%/30% to 0%/100% over 31 min.

Animal studies

The monkeys were fasted from 20 h before to 4 h after the intravenous or oral administration of the test drug. BOF-4272 was dissolved in 50% PEG 400 for intravenous administration and in 0.5% CMC solution for oral administration. BOF-4272 was administered to the monkeys intravenously at a dose of 5 mg/kg (0.70 MBq/kg) and orally at a dose of 25 mg/kg (1.63 MBq/kg).

Blood samples were obtained from the median cephalic vein into a heparinised syringe. Blood samples were drawn into test tubes at 10 time points (0 [pre-dose], 0.083, 0.167, 0.5, 1, 2, 4, 6, 8, and 12 h) after bolus intravenous injection and at 11 time points (0 [pre-dose], 0.5, 1, 2, 4, 6, 8, 12, 24, 48, and 72 h) after oral administration. All blood samples were immediately centrifuged to obtain plasma.

Urine and faeces samples were collected at room temperature for 120 h after intravenous or oral administration.

Sample analysis

Total[^14C]-concentrations were measured. Aliquots of the plasma (0.2 ml) and urine (0.5 ml) samples obtained were each mixed with 0.5 ml of water and 16 ml of ACS II® (Amersham). The total[^14C]-concentration in each mixture was determined using a liquid scintillation analyser (TRI-CARB Model 4530, Packard Instruments Co., Meriden, CT, USA). Each faeces sample was added to