Pharmacokinetic Evaluation of Nicotinamide Riboside in Rats
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Purpose
Nicotinamide riboside (NR), 1-(β-D-ribofuranosyl)nicotinamide, is a precursor for biosynthesis of nicotinamide adenine dinucleotide (NAD+). Biological functions of NR are closely resembled to vitamin B3 [1]. In order to understand its metabolic pathway and assessment as a source of nicotinamide, pharmacokinetic studies were designed and performed in male and female Sprague Dawley rats following oral administration of NR and nicotinamide.

Methods
Fast and simple liquid chromatography-tandem mass spectrometry methods were developed and validated for quantification of nicotinamide and its metabolite (N-methyl nicotinamide; MeNMN) in rat plasma. Acquity UPLC HSS CYANO column (3.0x50mm, 1.8 μm) using mobile phase [acetonitrile : aqueous ammonium acetate (0.01 M) buffer (60 : 40, %v/v)] was employed for chromatographic separation of nicotinamide. A gradient elution using Acquity UPLC BEH HILIC column (2.1x50mm, 1.7 μm) was used for MeNMN. Deuterium labeled (nicotinamideD4 and MeNMN-D3) surrogates were used as internal standards. Bioanalytical methods were validated for linearity range of 0.5-100 μg/mL and 200-4000 ng/mL, respectively, for nicotinamide and MeNMN following the FDA guidelines. In brief, rats were divided in two groups (male and female, 9 each) and an oral dose (1 g/kg) of NR was given to first group (G6), whereas a molar equivalent of nicotinamide (420 mg/kg) was dosed to second group (G7). Sparse sampling technique was implemented for blood collection and test samples were analyzed employing calibration and quality control standards. Concentration—time data was subjected to non-compartmental analysis using Phoenix WinNonlin (version 6.3; Certara Inc, St. Louis, MO, USA).

Results
The developed bioanalytical methods for nicotinamide and MeNMN were found to be linear (r ≥0.99) and adequate to quantify test samples up to 24 h. Methods were validated in terms of accuracy, precision, recovery, dilution integrity and stability (long term, short term, stock solution, bench top, autosampler and freeze-thaw stability). Precision (%RSD; nicotinamide, -6.0 to 12.4; MeNMN, 2.5 to 6.9) and accuracy (%bias; nicotinamide, -9.3 to -0.5; MeNMN, -10.6 to 6.8) of the methods were observed within the permissible limit. As demonstrated by change in area under concentration time curve (AUC), systemic exposure of nicotinamide was always higher in animal treated with pure nicotinamide (G7) compared to animals treated with NR (G6). The %systemic availability [(AUCG6/AUCG7)*100] of nicotinamide was about 43.5-47.7% when administered as NR, suggesting it would be used as a source of nicotinamide. The time to reach maximum plasma concentration (tmax) of nicotinamide was always higher in G6 than G7 group of rats, evidencing the sustained availability of nicotinamide followed by administration of NR.

Conclusion
The lower systemic availability (%) of nicotinamide up on administration of NR evidenced that it doesn’t solely act as a prodrug of nicotinamide, however, it would have additional pathway to change NAD+ levels in biological system.