Note

Comparison of the Enzyme Activities in the Tryptophan-NAD Pathway between the Wistar and Sprague Dawley Strains of Rats

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During experiments on tryptophan-NAD metabolism, when rats were fed a nicotinic acid deficient-diet of an 8.0% amino acid mixture simulating rice protein supplemented with some limiting amino acids except for tryptophan,¹⁾ we have found that there was a difference in growth rate between the rats of the Wistar and Sprague Dawley strains²⁾ and that total amounts of tryptophan

metabolites in liver, kidneys, and urine of the rats of the Sprague Dawley strain were higher than those of the rats of the Wistar strain.³⁾ These findings spur us into comparing enzyme activities in the tryptophan-NAD pathway between the two strains of rats.

Six-week-old rats of the Sprague Dawley and Wistar strains were obtained from Clea Japan Inc. The rats of the two strains were separately kept in wire-bottomed cages, and a CE-2 diet (Clea Japan Inc.) and water were supplied *ad libitum*. The room temperature was $22 \pm 2^\circ\text{C}$. The lighting schedule was 06:00~18:00 hr (light) and 18:00~06:00 hr (dark). On the 8th day, the rats (body weight: Sprague Dawley, 214.2 ± 6.04 g ($n=6$); Wistar, 202.5 ± 7.83 g ($n=6$)) were intraperitoneally injected with sodium pentobarbital and their livers were removed. Each liver (Sprague Dawley, 8.65 ± 0.30 g; Wistar, 8.44 ± 0.19 g) was minced and homogenized in five volumes of cold 50 mM potassium phosphate buffer, pH 7.0, using a Yamato ultradisperser model LK-21. These homogenates were used as enzyme sources for measuring the activities of tryptophan oxygenase (EC 1.13.11.11), kynureninase (EC 3.7.1.3), quinolinate phosphoribosyltransferase (EC 2.4.2.19), nicotinate phosphoribosyltransferase (EC 2.4.2.11), nicotinamide mononucleotide adenylyltransferase (EC 2.7.7.1), NAD synthetase (EC 6.3.5.1), nicotinamide methyltransferase (EC 2.1.1.1), and kynurenine aminotransferase (EC 2.6.1.7). One part of each homogenate was centrifuged at $10,000 \times g$ for 20 min to measure the activities of 3-hydroxyanthranilic acid oxygenase (EC 1.13.11.6) and aminocarboxymuconate-semialdehyde de-

TABLE I. COMPARISON OF THE ENZYME ACTIVITIES IN THE TRYPTOPHAN NAD PATHWAY BETWEEN THE LIVERS OF THE WISTAR AND SPRAGUE DAWLEY STRAINS OF RATS

	Wistar	Sprague Dawley
	(μmol/hr/g fresh weight)	
Tryptophan oxygenase	1.57 ± 0.08^a	2.06 ± 0.15^b
Kynureninase	0.324 ± 0.012^a	0.259 ± 0.024^b
3-Hydroxyanthranilic acid oxygenase	491.38 ± 36.50^a	693.35 ± 28.29^b
Aminocarboxymuconate-semialdehyde decarboxylase	1.42 ± 0.20	0.89 ± 0.15
Quinolinate phosphoribosyltransferase	0.267 ± 0.022	0.365 ± 0.043
Nicotinate phosphoribosyltransferase	0.049 ± 0.005^a	0.104 ± 0.017^b
Nicotinamide mononucleotide adenylyltransferase	5.50 ± 0.15^a	6.57 ± 0.22^b
NAD synthetase	0.88 ± 0.04	0.91 ± 0.07
Nicotinamide methyltransferase	0.724 ± 0.030	0.701 ± 0.014
Kynurenine aminotransferase	21.24 ± 1.69	26.38 ± 1.76

Values are mean $\pm$ SE of six rats; means in the same column not sharing a common superscript differ significantly ($p < 0.05$). Significant differences between means having the same variance or different variance when analyzed by the F-test were evaluated by Student's *t*-test and Welch's test, respectively.

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carboxylase (EC 4.1.1.45). To measure the enzyme activities we used, for tryptophan oxygenase, the method of Knox *et al.*⁴; kynureninase, the method of Takeuchi *et al.*⁵; 3-hydroxyanthranilic acid oxygenase, the method of Decker *et al.*⁶; aminocarboxymuconate-semialdehyde decarboxylase, the method of Ichiyama *et al.*⁷; quinolinate phosphoribosyltransferase, the method of Iwai and Taguchi⁸; nicotinate phosphoribosyltransferase, the method of Honjo⁹; nicotinamide mononucleotide adenylyltransferase, the method of Kurokawa *et al.*¹⁰; NAD synthetase, the method of Yu and Dietrich¹¹; nicotinamide methyltransferase, the method of Cantoni¹²; and for kynurenine aminotransferase, the method of Tobes and Mason.¹³

Table I shows the activities of ten enzymes that are involved in tryptophan-NAD metabolism in the livers of the Wistar and Sprague Dawley strains. The activities of tryptophan oxygenase, 3-hydroxyanthranilic acid oxygenase, and nicotinate phosphoribosyltransferase in the liver of the Sprague Dawley strain were significantly higher than those of the Wistar strain. The activities of NAD synthetase, nicotinamide methyltransferase, and kynurenine aminotransferase were almost the same between the two strains. The activity of quinolinate phosphoribosyltransferase tended to be higher (1.4 fold) in the Sprague Dawley strain than the Wistar strain, though it was not significantly different. The activity of aminocarboxymuconate-semialdehyde decarboxylase tended to be lower (0.6 fold) in the Sprague Dawley strain than the Wistar strain, though it was not significantly different. The lower activity of aminocarboxymuconate-semialdehyde decarboxylase means that quinolinic acid formation from tryptophan is higher in the Sprague Dawley strain than the Wistar strain. Generally speaking, the higher enzyme activities of the tryptophan-NAD pathway in the livers of the Sprague Dawley strain were more in favor of NAD formation from tryptophan than those of the Wistar strain. The higher production of the tryptophan metabolites and the higher growth rate of the Sprague Dawley strain than of the Wistar strain² was due to the higher enzyme activities of the tryptophan-NAD

pathway in the Sprague Dawley strain. Therefore, the Sprague Dawley strain of rats has greater resistance to a low nicotinic acid diet than the Wistar strain. The strains of rats should be considered when referring to the results from different papers.

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