

# Amino acid and carbohydrate preferences in C57BL/6ByJ and 129P3/J mice

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## Abstract

Compared with mice from the 129P3/J (129) inbred strain, mice from the C57BL/6ByJ (B6) inbred strain have higher consumption of several sweet-tasting amino acids and carbohydrates. To examine the relative contribution of taste and nutritive properties in these strain differences, we measured responses of B6 and 129 mice to eight sweet and non-sweet amino acids and carbohydrates in two-bottle preference tests with water. Mice from the two strains did not differ in consumption of non-sweet L-valine and L-histidine. Compared with 129 mice, B6 mice had higher consumption and lower preference thresholds for sweet amino acids L-glutamine, L-alanine and L-threonine, monosaccharides glucose and fructose, and maltooligosaccharide. These data suggest that differences in gustatory responsiveness are an important factor underlying higher consumption of some amino acids and carbohydrates by B6 mice compared with 129 mice. It is likely that in B6 mice, higher sweet taste responsiveness results in increased consumption of sweet-tasting amino acids and sugars, and higher taste responsiveness to complex carbohydrates results in increased consumption of maltooligosaccharide. However, postingestive processes also influence nutrient consumption and may be responsible for higher intake of carbohydrates compared with sweet-tasting amino acids. Results of this study set the stage for genetic analysis of differences between B6 and 129 mice in taste responsiveness and macronutrient consumption.

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## 1. Introduction

Variation among inbred mouse strains provides a tool to detect genetic loci underlying variable traits and to identify corresponding polymorphic genes. Mouse strains differ in consummatory responses to various taste stimuli and nutrients, including sweeteners, carbohydrates and amino acids [1–10]. Mice from the C57BL/6 and 129 inbred strains have been extensively studied for differences in sweetener and nutrient consumption [5–7,11–15]. Compared with 129 mice, C57BL/6 mice have higher consumption of several different sweet-tasting amino acids and carbohydrates [2,3,5,6,11–13,16].

Ingestive responses to amino acids and carbohydrates depend on both their taste properties and their postingestive effects. The goal of the study was to examine the relative contribution of taste and nutritive properties in differential ingestive responses of C57BL/6ByJ (B6) and 129P3/J (129) mice. To achieve this, we compared responses of B6 and 129 mice to sweet and non-sweet

amino acids and carbohydrates in two-bottle preference tests. We hypothesized that if mice from these two strains differ in postingestive responses to nutrients, then they will differ in consumption of nutrients regardless of their sensory properties. If these strains differ in sweetness perception, then they will differ in consumption of only sweet-tasting nutrients.

We have chosen for this study 8 compounds that include five amino acids and three carbohydrates. To humans, three of the amino acids (L-glutamine, L-alanine and L-threonine) have a prominent sweet taste, while two other amino acids (L-valine and L-histidine) lack a strong sweet component [17–21]. This is consistent with available data on preferences, conditioned taste aversion generalization, and single fiber recordings from gustatory nerves for these compounds in rodents [22–29]. Sweet L-threonine and non-sweet L-valine are essential amino acids. Sweet L-glutamine and L-alanine, and non-sweet L-histidine are non-essential amino acids.

The three carbohydrates were glucose, fructose and maltooligosaccharide. To humans, the monosaccharides glucose and fructose taste sweet [21], and are qualitatively very similar to sucrose [30,31]. Behavioral and neurophysiological

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data in mice also support sucrose-like taste of glucose and fructose [27,28]. Maltooligosaccharides are glucose polymers containing 2 to 10 glucose units. The maltooligosaccharide used in this study contains predominantly polymers with 3 to 6 glucose units, no glucose monomers, and only 2% maltose and polymers with greater than 7 glucose units. Another commonly used maltodextrin preparation, Polycose, contains a higher proportion of sugars (9% of glucose and maltose) and polymers with greater than 7 glucose units (43%). Behavioral and neurophysiological studies in rats have suggested that the taste of polysaccharides is qualitatively different from the taste of sugars or starch [32–35]. We have found no published data on human perception of maltooligosaccharide taste.

## 2. Materials and methods

### 2.1. Animals

Male mice from the C57BL/6ByJ (B6, *n*=17) and 129P3/J (129, *n*=26) inbred strains were obtained from the Jackson Laboratory (Bar Harbor, ME). Group 1 included 10 B6 and 18 129 mice that were 8.7–11.1 months old ( $10.6 \pm 0.9$  months, *M*±*SD*) when testing began. Group 2 included 7 B6 and 8 129 mice that were 3.7–4.2 months old ( $4.0 \pm 0.2$  months, *M*±*SD*) when testing began. Although mice from these two groups differed in age, it is unlikely that age variation affected results because in our previous experiments taste preferences of B6 and 129 mice remained stable over a period spanning more than two years [36]. During the experiments, the mice were housed in individual cages in a temperature-controlled room at 23 °C on a 12-h light: 12-h dark cycle (7:00 a.m. on, 7:00 p.m. off). They had free access to Teklad Rodent Diet 8604 (Harlan Teklad, Madison, WI; 24.5% protein, 50.3% carbohydrate and 4.4% fat; 3.93 Kcal/g gross energy; 0.31% sodium, 0.99% potassium and 1.46% calcium).

### 2.2. Taste solutions

Taste solutions were prepared in deionized water. All chemicals were purchased from Sigma Chemical Company (St. Louis, MO), except for maltooligosaccharide purchased from Pfanstiehl Laboratories, Inc. (Waukegan, IL). The maltooligosaccharide used in this study contains no measurable glucose, 1.5% maltose, 97% polymers with 3 to 6 glucose units, and 0.5% polymers with greater than 7 glucose units, with an average degree of polymerization of 4.4 [32].

### 2.3. Two-bottle preference tests

Construction of the drinking tubes has been described previously [37] and is given in detail on the Monell Mouse Taste Phenotyping Project web site ([www.monell.org/MMTPP](http://www.monell.org/MMTPP); [36]). Individually housed mice were presented with one tube containing a taste solution in deionized water, and the other tube containing deionized water. Daily measurements were made in the middle of the light period by reading fluid volume to the nearest 0.1 ml. Each concentration of a taste solution was tested for 48 h, with the positions of the tubes containing taste solution and water switched

after 24 h to control for side preferences. The solutions were tested in the increasing order of concentration, with the concentrations changing by approximately half log-steps. There were no breaks between testing different concentrations of the same compound, but between testing different compounds the mice received deionized water in both drinking tubes for at least three days.

Mice from Group 1 were tested with 0.1, 0.3, 1, 3, 10, 30, 100 and 300 mM L-valine and 0.1, 0.3, 1, 3, 10, 30, 100 and 300 mM L-histidine, in the order listed. Mice from Group 2 were tested with 1, 3, 10, 30, 100 and 300 mM L-glutamine; 1, 3, 10, 30, 100 and 300 mM L-alanine; 1, 3, 10, 30, 100 and 300 mM L-threonine; 0.1, 0.3, 1, 3, 10 and 30% maltooligosaccharide; 10, 30, 100, 300 and 1000 mM glucose; and 10, 30, 100, 300 and 1000 mM fructose, in the order listed. Body weight (BW) was measured (to the nearest 0.1 g) at the beginning of each taste solution concentration series, and at the end of the experiment.

### 2.4. Data analyses

Average daily (24-h) fluid intakes were calculated for each mouse for each solution concentration. Preference scores were calculated as the ratio of the average daily solution intake to the average daily total fluid (solution+water) intake, in percent.

The B6 mice were heavier than were 129 mice: the average body weight measured throughout the experiment in Group 1 was  $33.1 \pm 0.9$  g for B6 mice and  $27.1 \pm 0.3$  g for 129 mice (*M*±*SE*; *p*<0.001, *t*-test); in Group 2 it was  $32.9 \pm 1.4$  g and  $28.6 \pm 0.5$  g, respectively (*p*<0.05). To account for the strain

Table 1  
ANOVA results for two-bottle preference tests of B6 and 129 mice

Taste compound	Effect	<i>d.f.</i>	F values	
			Solution intake/30 g BW	Preference score
L-valine	Strain	1, 26	0.1	0.7
	Concentration	7, 182	1.8	0.5
	Strain × concentration	7, 182	4.6*	3.4*
L-histidine	Strain	1, 26	0.4	0.1
	Concentration	7, 182	13.6*	10.0*
	Strain × concentration	7, 182	0.6	1.1
L-glutamine	Strain	1, 13	58.8*	18.9*
	Concentration	5, 65	68.6*	32.7*
	Strain × concentration	5, 65	1.3	0.9
L-alanine	Strain	1, 13	18.6*	10.4*
	Concentration	5, 65	50.0*	27.0*
	Strain × concentration	5, 65	3.1*	1.8
L-threonine	Strain	1, 13	10.3*	2.4
	Concentration	5, 65	31.2*	18.4*
	Strain × concentration	5, 65	4.6*	1.6
Maltooligosaccharide	Strain	1, 13	29.6*	12.2*
	Concentration	5, 65	117.3*	17.2*
	Strain × concentration	5, 65	24.9*	4.5*
Glucose	Strain	1, 13	14.2*	8.4*
	Concentration	4, 52	67.1*	12.7*
	Strain × concentration	4, 52	17.1*	4.8*
Fructose	Strain	1, 13	10.6*	14.9*
	Concentration	4, 52	61.7*	6.5*
	Strain × concentration	4, 52	1.8	1.3

\**P*<0.05, ANOVA.

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