Physiology and Pharmacology of the Gut Nutrient Perception

Brain Activation by Umami Substances via Gustatory and Visceral Signaling Pathways, and Physiological Significance

Takashi Kondoh* and Kunio Torii

Institute of Life Sciences, Ajinomoto Co., Inc.; 1–1 Suzuki-cho, Kawasaki-ku, Kawasaki 210–8681, Japan.
Received June 11, 2008

Monosodium L-glutamate (MSG) elicits a unique taste termed umami and is widely used as a flavor enhancer in a variety of cuisines. Recent studies suggest the existence of L-glutamate (GLU) receptors and its transduction molecules in the gut mucosa as well as in the oral cavity. The vagal gastric afferent fibers respond specifically to the luminal stimulation of GLU in the stomach. GLU administration in the stomach also activates several brain areas (insula cortex, basal ganglia, limbic system, and hypothalamus). Ingestion of MSG enhanced secretion of digestive juices and insulin. Spontaneous ingestion of an MSG solution at the most preferred concentration (1% (w/v)) reduced weight gain, fat deposition, and plasma leptin levels without affecting food intake, naso-anal length (an index of somatic development), and lean mass in rats. These results suggest that umami signaling via gustatory and visceral pathways may play an important role in the process of digestion, absorption, metabolism, and other physiological functions via activation of the brain.

Key words monosodium L-glutamate; umami; brain activation; vagus nerve; gastrointestinal tract; energy homeostasis

1. INTRODUCTION

L-Glutamate (GLU) is a multifunctional amino acid in our body. As an essential substrate in the intermediary metabolism, free GLU is present in the most organs and tissues (skeletal muscles, brain, kidneys, and liver) in substantial concentrations.1,2) GLU plays an important role in the energy metabolism and synthesis of other amino acids, glutathione, and proteins. In the brain, GLU acts as a major excitatory neurotransmitter and its activity regulates synaptic plasticity, learning, memory, motor activity, and neural development. On the other hand, GLU in foods elicits unique taste termed umami, which is generally thought as a cue signal of protein ingestion.

In addition to the gustatory roles of GLU, recent studies have unveiled the significance of postdigestive GLU on various physiological functions such as digestion, absorption, metabolism, and energy homeostasis through brain activation. Especially, GLU suppresses obesity, fat deposition, and plasma leptin levels through enhancement of energy expenditure. These effects might possibly be mediated via gut GLU receptors functionally linked to the afferent branches of the vagus nerve, or the afferent sensory nerves in the oral cavity.

In this review, we describe the physiological significance of dietary GLU on the maintenance of homeostasis.

2. UMAMI TASTE AND PLEASANTNESS OF FOOD INTAKE

Umami Substances Umami is a characteristic taste on natural food materials, such as sea weed, dried bonito fish, and dried mushroom, which are frequently used for traditional Japanese cuisines. Typical umami substances are classified into two groups based on the chemical structure, i.e., L-α-amino acids represented by GLU and the other 5′-ribonucleotides represented by inosine 5′-monophosphate (5′-IMP) and guanosine 5′-monophosphate (5′-GMP). Free GLU is found in many foods such as sea tangle, cheese, raw ham, and vegetables (tomato, green peas, corn, asparagus, and broccoli). 5′-IMP is found in dried bonito, dried fish, and meat, and 5′-GMP in dried mushroom.

All these original forms of umami substances are acidic and hence elicit sour taste and are less soluble in water. In contrast, sodium salts of these substances show more umami taste with no sour taste and are highly soluble in water. Therefore, sodium salts of umami substances are generally used as “umami seasonings” worldwide.

Enhancement of Umami Taste Umami neither enhances the other four basic tastes (sweet, salty, sour, and bitter) nor is enhanced by them.3) However, the combination of monosodium GLU (MSG) with 5′-ribonucleotide (5′-IMP or 5′-GMP) enhances umami taste more than their simple addition (synergistic action).4,5) This is known as a common knowledge in Japanese cuisine for preparation of dashi soup with the combination of sea tangle and dried bonito or dried mushroom, which results in more savory taste than the soup made with sea tangle alone. Combination of vegetables with animal materials (meat, fish, and shell) is also reasonable for an enhancement of umami taste and maintenance of nutritional balance. The synergistic action of umami taste is commonly observed in mammals.

Taste Sensation and Satisfaction for Food Intake We experience strong satisfaction after ingestion of delicious foods in hungry condition. This sensation, satisfaction or satisfaction, does not occur by taste sensation alone but also requires ingestion of food substances and absorption of nutrients as required. Sensory input of foods such as color, shape (visual), smell (olfactory), sound (auditory), texture and temperature (trigeminal) are also important determinant for satisfaction. During ingestion and digestion processes, sensory information is transmitted to the brain and then integrated

* To whom correspondence should be addressed. e-mail: takashi_kondoh@ajinomoto.com © 2008 Pharmaceutical Society of Japan
with past food memory, health condition and hunger/satiety condition. If we experience abdominal pains or diarrhea after meal, it is memorized as unpleasant (aversive) food. The sensation of satisfaction or aversion occurs in every meal and develops as preference and aversion for each food.

**Umami Taste Is a Cue Signal of Protein Ingestion**

Protein is an essential nutrient for growth and maintenance of our body. It constitutes about 20% of our body weight. Amino acids are the building blocks of protein and hence important for synthesis of muscles and tendons as well as functional protein such as receptors, channels, transporters, enzymes, hormones, and immunoproteins. Protein itself produces no taste but each amino acid elicits characteristic taste. As food materials that contain protein have free amino acids together, the taste of amino acids comes to be a signal of protein in foods. Contents of bound GLU in various proteins reach between 11% and 23% in animal and between 20% and 45% in plants. \(^1\) In addition, 5'-ribonucleotides exist in cells, which produce synergistic effect with GLU. Therefore, it is reasonable to mark umami substances as a source of dietary protein.

**Umami Preference Is Linked to Protein Nutrition**

Amount of protein requirement varies dynamically depending on the growth stage. In general, animals in the early stage of development grow rapidly and hence require high amount of protein in foods. In contrast adult or aged animals require relatively small amount of protein for maintenance of their body. For example, protein requirement in diets for normal growth in growing rats is more than 15% in 4—8 weeks-old, 10% for 8—12 weeks-old, and 5% in 12—16 weeks-old. \(^6\)

Taste preference is largely affected by protein nutrition. In rats fed low or no protein diet, their growth was suppressed and they showed strong preference for NaCl. Preference for umami (MSG) is very low in this condition. The same phenomenon occurs when amino acid imbalanced diets, such as lysine-deficient diet, were offered (Fig. 1). In contrast, the preference for umami increases and preference for NaCl decreases in reverse when protein levels in diet increases and growth recovered. Therefore, umami preference is a marker of protein nutrition because it is enhanced only when protein intake exceeds the requirement.

### 3. TASTE MECHANISMS

Torii and Cagan (1980) first reported the existence of GLU receptors in the taste epithelium of bovine circumvallate papilla. \(^7\) The binding affinity of the taste membrane to GLU \((K_D=20—30 \text{ mM})\) is lower than the affinity of GLU receptors in the brain \((50\% \text{ effective concentration}=1—20 \mu \text{m})\) \(^8\) but consistent with the behavioral threshold of MSG in rats \((1—10 \text{ mM})\). \(^9\)

Now, several types of GLU receptors are found to exist at the apical membrane of the taste cells (type II taste cells) including the T1R1/T1R3, metabotropic GLU receptor type 1 and 4 (mGluR1 and mGluR4), taste mGluR1, and taste mGluR4. \(^10—14\) Activation of receptors produces changes in intracellular second messenger levels and open/close state of membrane channels to cause excitation of taste cells. The activation of taste cells is transmitted to the taste nerves through the release of a transmitter ATP. \(^15\) The activation of the taste nerves is finally conveyed to higher brain regions, and then sense of taste is perceived.

### 4. VISCERAL MECHANISMS

Recent studies have demonstrated the existence of a GLU-sensing system in the gut linked to activation of vagal afferent nerve that transmits food signals to the brain.

**Blood and Brain Concentration of GLU** GLU is the single major source of energy and an important substrate for the synthesis of other amino acids, glutathione, and protein in the intestinal mucosa. \(^2,16\) Because intestinal mucosa cells metabolize virtually all enteral GLU during absorption, blood levels of GLU do not rise appreciably after food ingestion. Figure 2 shows diurnal levels of GLU in the blood and brain of rats fed 30% casein protein diet. \(^17\) Although rats consumed the most food during the dark period, the GLU levels in the blood and brain were invariable within a day. The results suggest that the blood GLU levels do not provide essential information about the ingested protein. If the brain monitors amount of ingested protein through GLU, the sensing system of GLU should work before absorption.

**Gut Receptors for GLU** Recently, several taste recep-
tors and its transduction elements are found in the gut epithelium. For example, mGluR1 is found in the chief cells (pepsinogen secreting cells) in the rat stomach\(^{18}\) and both T1R1 and T1R3 are found in the stomach, small intestine, and colon in mice and humans.\(^{19}\) Transduction elements are also found in the small intestine.\(^{19}\) Considering the location of mGluR1 in the chief cells, GLU may stimulate the release of pepsinogen in the stomach. Identification of receptor types and determination of the function including vagus activation should be clarified in the future experiments.

**Activation of Vagal Afferents by Ingested GLU** Abdominal vagus nerve bundle consists of 3 branches, i.e., gastric, celiac, and hepatic branches. Administration of MSG in the stomach, duodenum and portal vein activates the vagal afferents in the gastric, celiac, and hepatic branches, respectively,\(^{20}\) suggesting the existence of GLU-sensing mechanisms, at least, in the stomach, duodenum, and portal vein (or liver).

**Effects of Abdominal Vagotomy on Ingestive Behavior of MSG Solution** If the vagus nerve plays some significant roles on taste preference, abdominal vagotomy would alter ingestive behavior for MSG solutions. As expectedly, vagotomy reduces ingestion (acceptance) of MSG solutions at high concentrations (240—600 mM) in the order of total vagotomy (TVX)>gastric vagotomy (GVX)>celiac vagotomy (CVX)>hepatic vagotomy (HVX)=intact controls (Fig. 3).\(^{21}\) The MSG ingestion is unaffected by hepatic vagotomy (HVX), especially by gastric vagotomy in Sprague-Dawley (SD) rats. The extents of inhibitory effects were: total vagotomy (TVX)=gastric vagotomy (GVX)>celiac vagotomy (CVX)>hepatic vagotomy (HVX)=intact. Data were shown as percentage for the water intake. Figure was cited from Torii\(^{17}\) with modification.

**Mechanisms of Vagal Activation** Nerve terminals of the vagus are located away from the epithelium of the gut. Then how is the activation of GLU receptors linked to the activation of the vagus nerves? We found that the activation of the vagal gastric afferents by intragastric administration of MSG is suppressed either by 1) luminal perfusion of lidocaine (a local anesthetic), 2) pretreatment of p-chlorophenylalanine (a depleter of endogenous serotonin), 3) administration of granisetron (a selective antagonist for serotonin receptor type 3), or 4) administration of N\(^{\text{6}}\)-nitro-L-arginine methyl ester (L-NAME, a nonselective inhibitor for nitric oxide [NO] synthases).\(^{22}\) Moreover, the neural response is mimicked by intragastric administration of sodium nitroprusside (an NO donor) and the response is again abolished by granisetron. Perfusion of the stomach lumen with serotonin itself does not activate the nerve. These results suggest that luminal GLU activates vagal nerve through production of bioactive substances such as NO and serotonin.

5. BRAIN MECHANISMS

**Specific Neuronal Responses to GLU in the Lateral Hypothalamic Area** The lateral hypothalamic area (LHA) regulates instinct behaviors (such as feeding and drinking) and autonomic nervous systems. Multimodal responsive neurons that respond to external sensory stimuli (olfactory, visual, gustatory, auditory, and trigeminal stimuli) and internal humoral environment (blood levels of glucose, insulin, and free fatty acids) exist in the LHA to maintain homeostasis.

When rats are given a diet deficient in lysine, an essential amino acid, a great preference for NaCl solution appears and the rats drink little umami solution.\(^{10}\) If they are offered a lysine solution (bitter taste) for this condition, the preference for lysine develops gradually and finally they learn to drink lysine solution in a quantitative manner.

The LHA is one of the essential brain areas that are responsible for dietary amino acids. Functional magnetic reso-
Forebrain Activation by Intragastric Administration of GLU

Does the brain respond to dietary GLU after ingestion? Recording of the vagal efferent activity showed that stimulation of MSG in the oral cavity, stomach, duodenum, and portal vein enhances vagal activity in the gastric and pancreatic efferent fibers (Fig. 4).20) The results suggest that the brain should be activated by GLU stimulation in the gut. Therefore, we measured the brain responses to intragastric administration of taste substances (glucose, MSG, and NaCl at 60 mM) by using fMRI in rats (Fig. 5). The results clearly demonstrated the forebrain activation in a variety of areas including insular cortex, basal ganglia, limbic system, and hypothalamus.26) Notably, the medial preoptic area (mPOA), dorsomedial nucleus of the hypothalamus (DMH), and habenular nucleus (Hb) are activated by MSG alone. On the other hand, the nucleus accumbens (NAC) is activated by glucose alone. The amygdala is activated by both glucose and MSG. Other areas such as the insular cortex (ICx), anterior cingulate cortex (ACC), caudate-putamen (CPu), hippocampus (HIP), and LHA are activated by glucose, MSG, and NaCl. These results suggest that GLU may have some essential role on the thermoregulation, energy homeostasis, and emotional behavior.

Temporal patterns of brain response are distinct among glucose, MSG, and NaCl. Glucose-induced response develops slowly with the peak at 20—30 min after the start of infusion and the response is maintained up to 60 min.26) The response pattern is consistent to the changes in blood glucose levels. On the other hand, MSG-induced response develops rapidly with the peak at 10—12 min (just after the cessation of infusion) and reduces rapidly thereafter to the pre-administration basal levels at 30 min. NaCl-induced response shows similar pattern to MSG-induced one but the activation area is very small. Another experiments using isotonic NaCl (150 mM) produces little response suggesting that the response to 60 mM NaCl is not due to chemical component of NaCl but due to other factors, i.e., hypo-osmotic stimulation. Analysis of MSG minus NaCl response shows results similar to MSG alone. These results suggest that the sensing of GLU may be located close to the administration area, i.e., in the stomach or proximal intestine (duodenum).
6. PHYSIOLOGICAL SIGNIFICANCE

Gustofacial Reflex The taste sensation contributes to the pleasure dimension or hedonics. In perinatal human infant, gustatory stimulation can trigger oral-facial motor reactions termed gustofacial reflex.27 The typical motor features comprising the sweet response are lip licking, smacking, puckering of the lips, and sucking, as well as the occasional smile. The bitter-induced responses are closed eyes, depressed mouth corners, wide gaping with flat and protruding tongue, excessive drooling, and occasional wretching or sometimes vomiting. These features are stereotypical and fixed. The slight aversion to the clear vegetable soup without addition of GLU is changed to acceptance response after the addition of GLU (sodium or potassium salts at 0.1—0.5%) in the neonate human infant.27 The facial responses induced by umami taste are lip licking, sucking, smacking, relaxation of the face, which indicates acceptance or appetitive behavior.

Secretion of Digestive Juices and Insulin Umami taste stimulates salivary flow in human.28,29 Umami taste also stimulates cephalic phase pancreatic exocrine secretions and insulin release.11 Furthermore, addition of umami substances (mixture of 92% MSG and 8% 5′-IMP) to meat enhances secretion of gastric juice by 60% in dogs.30 In patients of atrophic gastritis, addition of 2—3 g MSG to food substrates per day ameliorates both the basal release of gastric juice and maximal release of gastric acid.31 These umami effects resulted in a reduction of dissatisfaction to meal. Therefore, umami substances stimulate secretion of digestive juices (saliva, gastric juice, gastric acid, and pancreatic juice) and insulin.

Importance as an Intermediary Metabolite Why is GLU such an abundant biomolecule in our body? Although GLU is classified as a nonessential amino acid, dietary GLU has an “essential” functional role, especially in the intestinal mucosa.16 For example, GLU is: 1) the single largest contributor to energy production, 2) the specific precursor for the biosynthesis of two conditionally essential amino acids (proline, alanine) and glutathione (an antioxidant), and 3) the essential oxidative substrate in intermediary metabolism.

Consumption of a diet completely devoid of GLU and glutamine (an interchangeable amino acid to GLU) produces lower weight gain and rapid development of preference for GLU and glutamine solutions in rats.31 Therefore, GLU has an essential role in growth.

Suppression of Obesity Addition of MSG to foods enhances its palatability and acceptability. Then, does MSG enhance amount of food intake and cause obesity? In human experiments in healthy elderly people and diabetic patients, addition of MSG to meals changes food selection to be more healthier but the total caloric intake does not alter, suggesting that umami substance not causing obesity in human.

The relationship between MSG intake and obesity were further tested in rats. They were given free access to a 1% MSG solution (the most preferred concentration of MSG for rats) and water (MSG group) or water alone (water group) as drinking solution.35 The fat mass and lean mass in the abdomen were measured by MRI. Rats given free access to MSG and water show a high preference (93—97%) for the MSG solution. Rats ingesting MSG have a significantly smaller weight gain, reduced abdominal fat mass, and lower plasma leptin levels, compared to rats ingesting water alone (Fig. 6). Naso-anal length, lean mass, food and energy intakes, blood pressure, blood glucose, and plasma levels of insulin, triglyceride, total cholesterol, albumin, and GLU are not influenced by the ingestion of the MSG solution. Together, these results suggest that MSG ingestion reduces weight gain, body fat mass, and plasma leptin levels. Moreover, these changes are likely to be mediated by increased energy expenditure, not reduced energy intake or delayed development. Conceivably, these effects of MSG might be mediated via gut GLU receptors functionally linked to afferent branches of the vagus nerve in the gut, or the afferent sensory nerves in the oral cavity.

Then, how does GLU could enhance energy expenditure to produce reduced weight gain? One possible mechanism is the enhanced heat generation by umami substances. It is reported that the ingestion of an MSG solution enhances diet-induced heat production in rats.37 The ingestion of MSG alone does not affect background metabolism, but does enhance the thermogenesis produced by food consumption.38,39 Taste stimulation by MSG enhances activation of sympathetic nerve that innervates white adipose tissue.40 Conceivably, an enhancement of diet-induced thermogenesis and fat metabolism might constitute part or all of the mechanisms by which MSG ingestion diminished weight gain.

7. CONCLUSIONS

Dietary free GLU activates a variety of brain areas (cortex, basal ganglia, limbic system, and hypothalamus) through activation of taste and vagus nerves. Notably, luminal GLU in the stomach activates the vagal gastric afferent fibers through production of bioactive substances such as NO and serotonin. Ingestion of umami substances enhances secretion of digestive juices and insulin. Spontaneous ingestion of an MSG so-
lution suppresses obesity through enhanced energy expenditure. These results suggest the physiological significance of umami substances in the process of digestion, absorption, metabolism, and other functions via activation of the brain.

REFERENCES

16) Torii, K., Metabolism, 26, 195—201 (1989).