Gustatory Signaling in the Periphery: Detection, Transmission, and Modulation of Taste Information

Article *in* Biological & Pharmaceutical Bulletin · November 2010

DOI: 10.1248/bpb.33.1772 · Source: PubMed

4 authors, including:

Ryusuke Yoshida
Kyushu University
109 PUBLICATIONS **3,038** CITATIONS

SEE PROFILE

Shingo Takai
Kyushu University
18 PUBLICATIONS **324** CITATIONS

SEE PROFILE

Yuzo Ninomiya
Monell Chemical Senses Center, USA/ Kyushu University, Japan
307 PUBLICATIONS **11,017** CITATIONS

SEE PROFILE

Current Topics

Taste and Health: Nutritional and Physiological Significance of Taste Substances in Daily Foods

Gustatory Signaling in the Periphery: Detection, Transmission, and Modulation of Taste Information

Mayu Niki, Ryusuke Yoshida, Shingo Takai, and Yuzo Ninomiya*

Section of Oral Neuroscience, Graduate School of Dental Sciences, Kyushu University; 3–1–1 Maidashi, Higashi-ku, Fukuoka 812–8582, Japan.

Received July 28, 2010

Gustatory signaling begins with taste receptor cells that express taste receptors. Recent molecular biological studies have identified taste receptors and transduction components for basic tastes (sweet, salty, sour, bitter, and umami). Activation of these receptor systems leads to depolarization and an increase in $[Ca^{2+}]_i$ in taste receptor cells. Then transmitters are released from taste cells and activate gustatory nerve fibers. The connection between taste cells and gustatory nerve fibers would be specific because there may be only limited divergence of taste information at the peripheral transmission. Recent studies have demonstrated that sweet taste information can be modulated by hormones or other endogenous factors that could act on their receptors in a specific group of taste cells. These peripheral modulations of taste information may influence preference behavior and food intake. This paper summarizes data on molecular mechanisms for detection and transduction of taste signals in taste bud cells, information transmission from taste cells to gustatory nerve fibers, and modulation of taste signals at peripheral taste organs, in particular for sweet taste, which may play important roles in regulating energy homeostasis.

Key words taste receptor; taste coding; modulation; leptin; endocannabinoid

1. INTRODUCTION

Gustatory system plays important roles in maintaining homeostasis in animals. If animals lack essential nutrients for their survival such as sugars, minerals, and essential amino acids, they find out these insufficient nutrients by using gustatory clues. In contrast, spoiled foods and poisonous items that are deleterious to animal health have an unpleasant taste sensation. In general, sweet, salty, umami, sour, and bitter are considered to be basic taste qualities. Recent molecular biological studies have proposed candidate receptors for these five basic tastes.[1,2] These receptors are divided into two groups: G-protein coupled receptors (GPCRs) and channel type receptors. The expression patterns of these receptors suggest that each taste quality may be encoded by a separate population of taste bud cells. Many taste cells respond to one of five basic taste stimuli and may be responsible for detection and transmission of each taste quality.[3]

Activation of taste cells lead to transmitter release, then activation of gustatory nerve fibers. Processing of taste information may occur during this transmission. If the gustatory fiber receives inputs from taste cells that have different response properties, the fiber would respond to multiple taste qualities. However, this may not be the case because response characteristics of taste cells and gustatory nerve fibers are very similar and many gustatory nerve fibers may selectively innervate their corresponding types of taste cells.[4]

Recent studies have shown that sensitivities of taste cells can be modulated by hormones and other endogenous factors.[5] For example, leptin, an anorexigenic mediator that reduces food intake by acting on hypothalamic receptors, selectively suppresses sweet taste sensitivity of taste cells *via* leptin receptor (Ob-Rb). In contrast, endocannabinoids, orexigenic mediators that induce appetite and stimulate food intake *via* cannabinoid receptors (CB_1) mainly in the hypothalamus, enhance sweet taste sensitivity of taste cells *via* CB_1 receptor. Such reciprocal regulation of sweet taste by leptin and endocannabinoids may have important roles in maintaining energy homeostasis in animals.

In this paper, we summarize recent progress of studies on the molecular mechanisms for detection and transduction of taste in taste bud cells, connections between taste cells and gustatory fibers, and regulatory mechanisms of taste information at the periphery.

2. TASTE RECEPTORS AND TRANSDUCTION IN TASTE BUD CELLS

Sweet, and umami taste are mediated by T1R family (T1R1, T1R2, T1R3) that belongs to family C of GPCRs including metabotropic glutamate receptors (mGluRs), calcium sensing receptors, and V2r pheromone receptors[6—15] (Fig. 1). T1Rs assemble into heterodimeric receptor complexes to function as sweet (T1R2+T1R3) or umami (T1R1+T1R3) taste receptors.[11,12] In heterologous expression system, T1R2+T1R3 heterodimer is activated by various sweeteners, such as sugars, artificial sweeteners, sweet amino acids, and sweet proteins, whereas T1R1+T1R3 heterodimer is activated by glutamate (human) and amino acids (mouse).[12,14] The characteristic feature of umami is a synergism, in which the taste intensity of monosodium glutamate (MSG) is enhanced by 5′-ribonucleotide monophosphates such as inosine-5′-monophosphate (IMP) and guanosine-5′-monophosphate (GMP).[16,17] Umami synergism is observed in re-

* To whom correspondence should be addressed. e-mail: yuninom@dent.kyushu-u.ac.jp

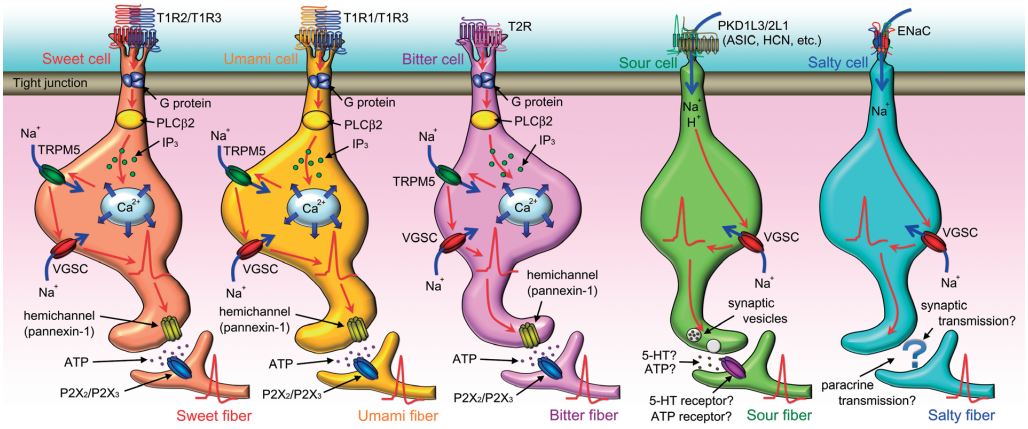

Fig. 1. Receptors and Transduction Mechanisms for Each Basic Taste

Details are described in the main text. VGSC: voltage gated sodium channel. 5-HT: serotonin.

sponses of human embryo kidney (HEK) cells expressing both T1R1 and T1R3[14,18,19] and is shown to occur through activation of venus-fly trap extracellular domain of T1R1 to which both MSG and 5′-ribonucleotide can bind cooperatively.[18] Although T1R3 contributes to both sweet and umami tastes, mice genetically lacking T1R3 showed diminished but not abolished behavioral and nerve responses to sugars and umami compounds.[20] The existence of the residual responses to the stimuli indicates that T1R3-independent sweet- and umami-responsive receptors and/or pathways may exist in taste cells. Potential candidates for umami taste receptors other than T1R1+T1R3 are mGluR variants such as taste mGluR1 and 4, which have been shown to be expressed in taste cells.[21,22] It has also been reported that antagonists for mGluR1 [1-aminoindan-1,5-dicarboxylic acid (AIDA)] and mGluR4 [(*RS*)-alpha-cyclopropyl-4-phosphonophenylglycine (CPPG)] reduced taste cell, gustatory nerve and behavioral responses to umami substances in mice.[23]

Bitter taste is mediated by the T2R family that belongs to family A of GPCRs including V1r pheromone receptors and opsin receptors (Fig. 1).[24—26] In humans, 25 members of the T2R family may function as bitter taste receptors although bitter ligands for five of the 25 T2R members (T2R41, T2R42, T2R45, T2R48, T2R60) are still unknown.[27] T2Rs are coexpressed in a subpopulation of taste receptor cells,[24,28] raising a possibility that T2Rs form hetero-oligomers like T1Rs for sweet and umami taste. One study demonstrated that the vast majority of T2R pairs form oligomers, but functional significance of oligomerization has not been elucidated.[29] In humans, individual variation in sensitivity to the bitter compound phenylthiocarbamide (PTC) is well known.[30] This variation is correlated with a genetic variation in T2R38, which is a receptor for PTC and 6-*n*-propylthiouracil (PROP).[31]

Sweet, umami, and bitter tastes are mediated by different GPCRs but use a common signaling pathway after activation of these receptors. Tastant binding to sweet, umami, and bitter receptors activates heteromeric G-protein, α-gustducin,[32] and subsequent stimulation of phospholipase Cβ2 (PLCβ2).[33] Activation of PLCβ2 produces inositol-1,4,5-triophosphate, a ligand for inositol-1,4,5-triophosphate receptor type 3 (IP_3R3)[34] in the Ca^{2+} stores. Then Ca^{2+} is released from these stores and stimulates transient receptor potential channel M5 (TRPM5)[33,35] to depolarize the taste cells. Such depolarization leads to the generation of action potentials in taste cells (Fig. 1).

Sour and salty taste may be mediated by channel type receptors (Fig. 1). In the case of sour taste, many candidate receptors have been implicated in detection such as acid-sensing ion channels (ASICs),[36] hyperpolarization-activated cyclic nucleotide-gated potassium channels (HCNs),[37] potassium channels,[38,39] 5-nitro-2-(3-phenylpropylamino)-benzoic acid (NPPB)-sensitive Cl^- channels,[40] and polycystic kidney disease 1L3 and 2L1 heterodimer (PKD1L3+PKD2L1).[41,42] Genetic elimination of cells expressing PKD2L1 substantially reduces gustatory nerve responses to sour taste stimuli, suggesting that PKD2L1 expressing cells are sour sensitive taste receptor cells.[41] Patients with sour ageusia (taste blind), but not sour normal individuals, lack the expression of mRNAs for PKD1L3, PKD2L1, and ASIC subunits in the anterior part of the tongue,[43] suggesting that the PKDs and ASICs may have a role in sour taste sensation in humans. However, the *in vivo* function of these candidates for the sour sensation must be elucidated in future studies. In the case of salt taste, epithelial sodium ion channel (ENaC) is believed to be a receptor because amiloride, an epithelial sodium channel blocker, reduces taste cell, neural, and behavioral responses to NaCl.[44—47] A recent study has demonstrated that mice lacking ENaC α-subunit in taste cells showed total loss of amiloride-sensitive NaCl responses, indicating that ENaC mediates amiloride-sensitive NaCl responses in mice.[48] Amiloride-insensitive components of NaCl responses are suggested to be mediated by a transient receptor potential channel V1 (TRPV1) variant.[49] However, TRPV1-knockout mice are able to detect NaCl with or without amiloride, suggesting that additional mechanisms must contribute to the amiloride-insensitive NaCl response.[50] When channel type receptors are activated by taste compounds, taste cells are depolarized and elicit action potentials (Fig. 1).

3. TASTE CODING AND TRANSMISSION OF TASTE INFORMATION AT THE PERIPHERY

Expression patterns of taste receptors in taste buds imply that different taste bud cells define the different taste modalities. For example, T1R3 and T2Rs are not coexpressed in taste bud cells.[11] PKD2L1 is not coexpressed with T1R3 and T2Rs.[41] In addition, ENaC α subunit is expressed in a unique subset of "ENaC alone" taste bud cells, although some cells expressing Car 4, a sour cell marker, coexpress ENaC α subunit.[48] Physiological studies in mouse taste cells demonstrated that the majority (60—70%) of taste cells respond to one of basic taste qualities.[51,52] In mouse fungiform taste buds on the anterior part of the tongue, identified taste cells respond more specifically to basic taste compounds; the majority of gustducin-expressing cells respond to sweet, bitter, or umami compounds and many GAD67-expressing cells that may also express sour taste receptor candidates specifically respond to sour taste stimuli.[53] Thus, taste qualities may be discriminated at the taste receptor cell level. However, a significant portion of taste cells respond to multiple taste qualities. These cells may contribute to the discrimination of more slight differences between taste compounds.

Taste information discriminated by taste receptor cells would be straightforwardly relayed to the gustatory nerve fibers because response profiles of taste cells are well conserved among gustatory nerve fibers.[3,52] To do this, the selective connection must be formed between them. There is no direct evidence showing the selective or specific connection between taste cells and gustatory fibers, but several nerve regeneration studies may provide insights into this problem. As noted above, taste responses to NaCl are divided into two components, amiloride-sensitive (AS) and -insensitive (AI) components. In general, amiloride primarily inhibits NaCl (and LiCl) responses of gustatory fibers that selectively respond to sodium and lithium salts (labeled N-type), whereas it hardly affects NaCl responses of fibers that show broad sensitivity to electrolytes (labeled E- or H-type). The chorda tympani (CT) nerve innervating the anterior part of the tongue contains both types of fibers almost equally. In contrast, the glossopharyngeal (GL) nerve innervating the posterior part of the tongue contains primarily E-type fibers but only a very few if at all N-type fibers.[54] This relative abundance of the E- and N-type fibers was not altered by cross-regeneration of the two gustatory nerves into the reverse tongue regions,[55] suggesting that regenerated taste axons selectively recoupled with the appropriate type of receptor cells. In the nerve regeneration study for salt taste,[56] NaCl responses of the CT nerve started to recover from *ca.* 3 weeks after the nerve crush, whereas amiloride inhibition of NaCl responses clearly reappeared from *ca.* 4 weeks onward. N- and E-type fibers were clearly distinguishable during the process of reformation of the AS and AI neural systems, suggesting that the AS and AI systems are independently reformed after the nerve crush. A similar result was obtained in the nerve regeneration study for sweet taste using gurmarin, a sweet-suppressing peptide in rodents.[57] In mice, there are two types of sweet-responsive CT fibers, gurmarin-sensitive (GS) and gurmarin-insensitive (GI) fibers.[58] After the CT nerve crush, recovery of GI responses preceded recovery of GS responses by *ca.* 1 week, and the GS and GI fibers could be distinguished during the course of CT regeneration. Thus, the two sweet-reception systems may be reconstituted independently during regeneration of the CT nerve. These data suggest that the selective connection may be formed between corresponding classes of taste cells and gustatory axons.

In taste buds, only a few taste bud cells have synaptic contact with nerve fibers.[59] Taste bud cells expressing receptors and transduction components for sweet, bitter and umami taste do not possess conventional synaptic structures, but they have close contact with sensory nerve fibers such as subsurface cisternae.[60] How do these cells transmit their signals to gustatory nerves? Regarding the signal transmission from taste cells to gustatory nerve fibers, ATP is the most likely candidate transmitter. Gustatory nerve fibers express ionotropic purinergic receptors, $P2X_2$ and $P2X_3$.[61] Mice lacking both $P2X_2$ and $P2X_3$ showed abolished gustatory nerve responses to taste stimuli and reduced behavioral responses to sweet, umami, and bitter substances.[62] Taste cells release ATP in response to serial depolarization[63] or sweet, bitter, and umami taste stimuli.[64,65] ATP release from taste cells was blocked by a hemichannel blocker, carbenoxolone.[64,65] Therefore the signal transmission from taste cells to gustatory nerve fibers for sweet, bitter, and umami taste may be following: 1) taste cells activated by sweet, bitter, or umami substances increase in $[Ca^{2+}]_i$ and generate action potentials; 2) Ca^{2+} and depolarization stimulate hemichannels (possibly the pannexin-1 hemichannel) to open and release ATP; and 3) released ATP activates $P2X_2/P2X_3$ receptors on the adjacent gustatory nerve fibers (Fig. 1). Taste bud cells expressing PKD2L1, which may be sour taste receptor cells, possess synaptic structures.[66] These cells also have serotonin,[66] and serotonin is released from taste cells in response to sour taste stimuli.[67] The role of serotonin in signal transmission is unclear, although sour-sensitive cells may use conventional synaptic transmission (Fig. 1). The mechanism for the signal transmission from salt-sensitive taste cells to gustatory nerve fibers is unknown. Further studies are required to elucidate the signal transmission from taste cells to gustatory nerve fibers.

4. MODULATION OF TASTE INFORMATION AT THE PERIPHERY

Sensory information on taste has a great impact on the feeding behavior of animals. There is growing evidence that taste information is modulated by internal and external factors at the peripheral taste organs to help maintain homeostasis. Sweet taste may have a role in detecting energy sources and is very attractive to animals. Recent studies have revealed that sweet taste sensitivity is modulated by orexigenic and anorexigenic mediators in the peripheral taste organs.

Leptin is an anorexigenic mediator that reduces food intake by acting on hypothalamic receptors.[68] There are five isoforms of leptin receptors (Ob-Ra—Ob-Re). Among them, Ob-Rb is a functional leptin receptor.[69] The *db*/*db* mice that have defects in the leptin receptor are hyperphagic, massively obese, and diabetic.[69] These mice showed greater gustatory nerve responses and behavioral responses to various sweet substances but not to salty, bitter, and sour substances than lean control mice.[70,71] Streptozotocin-induced diabetic mice did not exhibit greater sugar responses,[70] indicating that

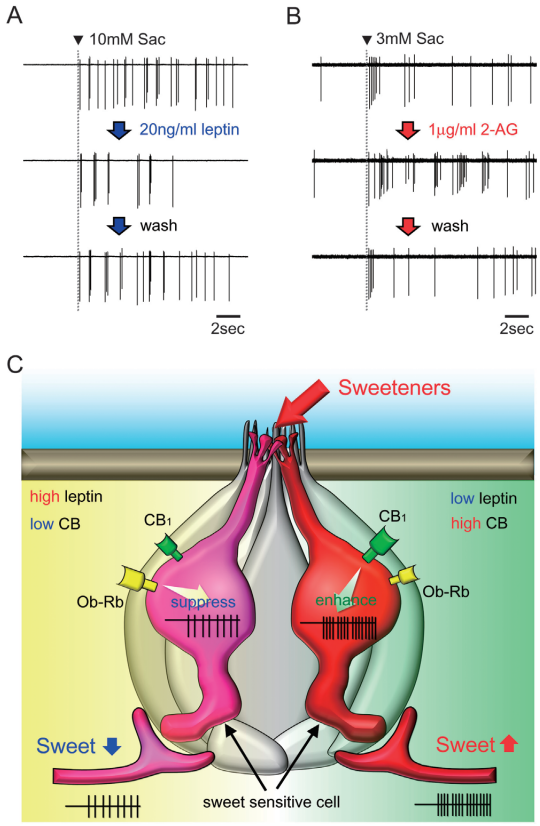

Fig. 2. Modulation of Sweet Taste Sensitivity by Leptin and Endocannabinoids

(A) Sample recordings demonstrating the effect of bath application of leptin on sweet responses of a taste cell. Bath application of 20 ng/ml leptin suppressed the response of the cell to 10 mM saccharin. After washout of leptin, the saccharin response was recovered to the control level. (B) Sample recordings demonstrating the effect of 2-AG on sweet responses of a taste cell. Bath application of 1 μg/ml 2-AG enhanced the response of the cell to 3 mM saccharin. After washout of 2-AG, saccharin response was returned to the control level. Dotted lines indicates the onset of taste stimuli. Recordings in A and B were obtained from different taste cells. (C) A schematic drawing showing the effects of leptin and endocannabinoids. In high-leptin tune (shown in yellow), responses of sweet-sensitive cells were suppressed *via* the leptin receptor Ob-Rb. In high-cannabinoid (CB) tune (shown in green), responses of sweet-sensitive cells were enhanced *via* the CB_1 receptor. These reciprocal regulations of peripheral sweet taste reception by leptin and endocannabinoids may play an important role in regulating energy homeostasis.

greater sensitivity to sweet substances was not induced by the diabetic state itself. A subsequent study[72] demonstrated that the administration of leptin into lean control mice suppressed CT nerve responses to sweet substances without affecting responses to sour, salty, and bitter substances. In addition, CT nerve responses to sucrose in lean control mice were negatively correlated with their plasma leptin levels. Thus, leptin selectively suppresses sweet taste sensitivity in mice. This effect may be mediated by leptin receptors, because suppression of sweet taste responses by leptin was not observed in *db*/*db* mice. Taste bud cells express Ob-Rb,[72—74] and taste cell responses to sweeteners were suppressed by the administration of leptin in about half of sweet-sensitive cells (Fig. 2; Yoshida *et al.*, unpublished data). Thus leptin reduces sweet taste information by acting on leptin receptors in sweet-sensitive taste cells.

There is a diurnal pattern in circulating leptin levels.[75,76] In humans, plasma leptin levels start rising before noon and peak between 23:00 and 01:00, after which the levels decline until morning.[77] To examine the relationship between plasma leptin levels and taste sensitivity in humans, the diurnal pattern of plasma leptin levels and taste recognition threshold were measured in healthy adult.[78] The recognition threshold for sweet compounds exhibited a diurnal variation that paralleled the variation in plasma leptin levels. Such diurnal variation is not observed for other tastes (salty, bitter, sour, and umami taste). The diurnal pattern of plasma leptin levels shows meal-related shifts.[79] When leptin levels were phase-shifted following imposition of one or two meals par day, the diurnal variation in the recognition threshold for sweet taste shifted in parallel. These synchronizations of diurnal variation in plasma leptin levels and recognition threshold for sweet taste strongly suggest a mechanistic connection between these two variables in humans.

Leptin selectively suppresses sweet taste responses in wild-type mice but not in leptin receptor-deficient *db*/*db* mice. This may be one major reason for greater responses to sweeteners in *db*/*db* mice. However, it is possible that another factor may also be involved in greater sweet responses in *db*/*db* mice. We hypothesized that bioactive substances that have an inverse effect of leptin might enhance sweet taste sensitivity. Endocannabinoids such as anandamide [*N*-arachidonoylethanolamine (AEA)] and 2-arachidonoyl glycerol (2-AG) are known as orexigenic mediators that act *via* cannabinoid receptors (CB_1) in the hypothalamus and limbic forebrain to induce appetite[80,81] and stimulate food intake.[82] Circulating endocannabinoid levels inversely correlate with plasma levels of leptin.[83] Therefore endocannabinoids are potential candidates to enhance sweet taste sensitivity.[84] The administration of AEA or 2-AG into wild-type mice increases gustatory nerve responses and behavioral responses to sweeteners without affecting responses to salty, sour, bitter, and umami substances. Sweet enhancing effects of endocannabinoids were not observed in CB_1 knockout mice, suggesting that administration of endocannabinoids selectively enhances sweet taste responses and the endocannabinoid effect is mediated by their receptor, CB_1. In taste cells, about 60% of T1R3-expressing cells coexpressed with CB_1 and responses of sweet sensitive cells were enhanced by the administration of AEA or 2-AG. Effects of endocannabinoids are diminished by the administration of AM251, a CB_1 receptor antagonist. Thus endocannabinoids enhance sweet taste information by acting on CB_1 receptors in sweet-sensitive taste cells.

Leptin and endocannabinoids are well known to regulate food intake reciprocally *via* central mechanisms. In addition, peripheral taste organs are another important target of these orexigenic and anorexigenic mediators. Leptin reduces the palatability of sweet foods, whereas endocannabinoids enhance the palatability of sweet foods. Such reciprocal regulation of peripheral sweet taste reception by leptin and endocannabinoids may contribute to their opposing actions on food intake and play an important role in regulating energy homeostasis.

5. CONCLUSION

We summarized data on molecular mechanisms for detection and transduction of taste in taste bud cells, connections between taste cells and gustatory fibers, and regulatory mechanisms of taste information at peripheral taste organs.

Our understanding is that taste information discriminated among taste bud cells is straightforwardly relayed onto gustatory nerve fibers and then the central nervous system. Not only these straight lines but also processing of taste information among the taste bud exist and may be important for the coding of taste information. Modulation of taste information at the peripheral taste organs is evident for sweet taste but not for other tastes. We believe that similar modulatory systems may also function in other tastes to help maintain homeostasis.

Acknowledgments This work was supported by Japan Society for the Promotion of Science, Grant-in-Aid for Scientific Research (KAKENHI) 18109013 and 18077004 (YN).

REFERENCES

1) Chandrashekar J., Hoon M. A., Ryba N. J., Zuker C. S., *Nature* (London), **444**, 88—94 (2006).
2) Ishimaru Y., *Odontology*, **97**, 1—7 (2009).
3) Yoshida R., Yasumatsu K., Shigemura N., Ninomiya Y., *Arch. Histol. Cytol.*, **69**, 233—242 (2006).
4) Yoshida R., Ninomiya Y., *Int. Rev. Cell. Mol. Biol.*, **279**, 101—134 (2010).
5) Jyotaki M., Shigemura N., Ninomiya Y., *Endocr. J.*, **57**, 467—475 (2010).
6) Hoon M. A., Adler E., Lindemeier J., Battey J. F., Ryba N. J., Zuker C. S., *Cell*, **96**, 541—551 (1999).
7) Bachmanov A. A., Li X., Reed D. R., Ohmen J. D., Li S., Chen Z., Tordoff M. G., de Jong P. J., Wu C., West D. B., Chatterjee A., Ross D. A., Beauchamp G. K., *Chem. Senses*, **26**, 925—933 (2001).
8) Kitagawa M., Kusakabe Y., Miura H., Ninomiya Y., Hino A., *Biochem. Biophys. Res. Commun.*, **283**, 236—242 (2001).
9) Max M., Shanker Y. G., Huang L., Rong M., Liu Z., Campagne F., Weinstein H., Damak S., Margolskee R. F., *Nat. Genet.*, **28**, 58—63 (2001).
10) Montmayeur J. P., Liberles S. D., Matsunami H., Buck L. B., *Nat. Neurosci.*, **4**, 492—498 (2001).
11) Nelson G., Hoon M. A., Chandrashekar J., Zhang Y., Ryba N. J., Zuker C. S., *Cell*, **106**, 381—390 (2001).
12) Nelson G., Chandrashekar J., Hoon M. A., Feng L., Zhao G., Ryba N. J., Zuker C. S., *Nature* (London), **416**, 199—202 (2002).
13) Sainz E., Korley J. N., Battey J. F., Sullivan S. L., *J. Neurochem.*, **77**, 896—903 (2001).
14) Li X., Staszewski L., Xu H., Durick K., Zoller M., Adler E., *Proc. Natl. Acad. Sci. U.S.A.*, **99**, 4692—4696 (2002).
15) Zhao G. Q., Zhang Y., Hoon M. A., Chandrashekar J., Erlenbach I., Ryba N. J., Zuker C. S., *Cell*, **115**, 255—266 (2003).
16) Yamaguchi S., *J. Food. Sci.*, **32**, 473—478 (1967).
17) Yamaguchi S., *Physiol. Behav.*, **49**, 833—841 (1991).
18) Zhang F., Klebansky B., Fine R. M., Xu H., Pronin A., Liu H., Tachdjian C., Li X., *Proc. Natl. Acad. Sci. U.S.A.*, **105**, 20930—20934 (2008).
19) Shigemura N., Shirosaki S., Sanematsu K., Yoshida R., Ninomiya Y., *PLoS ONE*, **4**, e6717 (2009).
20) Damak S., Rong M., Yasumatsu K., Kokrashvili Z., Varadarajan V., Zou S., Jiang P., Ninomiya Y., Margolskee R. F., *Science*, **301**, 850—853 (2003).
21) Chaudhari N., Landin A. M., Roper S. D., *Nat. Neurosci.*, **3**, 113—119 (2000).
22) San Gabriel A., Uneyama H., Yoshie Y., Torii K., *Chem. Senses*, **30**, i25—i26 (2005).
23) Yasuo T., Kusuhara Y., Yasumatsu K., Ninomiya Y., *Biol. Pharm. Bull.*, **31**, 1833—1837 (2008).
24) Adler E., Hoon M. A., Mueller K. L., Chandrashekar J., Ryba N. J., Zuker C. S., *Cell*, **100**, 693—702 (2000).
25) Chandrashekar J., Mueller K. L., Hoon M. A., Adler E., Feng L., Guo W., Zuker C. S., Ryba N. J., *Cell*, **100**, 703—711 (2000).
26) Matsunami H., Montmayeur J. P., Buck L. B., *Nature* (London), **404**, 601—604 (2000).
27) Meyerhof W., Batram C., Kuhn C., Brockhoff A., Chudoba E., Bufe B., Appendino G., Behrens M., *Chem. Senses*, **35**, 157—170 (2010).
28) Behrens M., Foerster S., Staehler F., Raguse J. D., Meyerhof W., *J. Neurosci.*, **27**, 12630—12640 (2007).
29) Kuhn C., Bufe B., Batram C., Meyerhof W., *Chem. Senses*, **35**, 395—406 (2010).
30) Fox A. L., *Sci. News Lett.*, **9**, 249 (1931).
31) Bufe B., Breslin P. A., Kuhn C., Reed D. R., Tharp C. D., Slack J. P., Kim U. K., Drayna D., Meyerhof W., *Curr. Biol.*, **15**, 322—327 (2005).
32) Wong G. T., Gannon K. S., Margolskee R. F., *Nature* (London), **381**, 796—800 (1996).
33) Zhang Y., Hoon M. A., Chandrashekar J., Mueller K. L., Cook B., Wu D., Zuker C. S., Ryba N. J., *Cell*, **112**, 293—301 (2003).
34) Hisatsune C., Yasumatsu K., Takahashi-Iwanaga H., Ogawa N., Kuroda Y., Yoshida R., Ninomiya Y., Mikoshiba K., *J. Biol. Chem.*, **282**, 37225—37231 (2007).
35) Zhang Z., Zhao Z., Margolskee R. F., Liman E., *J. Neurosci.*, **27**, 5777—5786 (2007).
36) Ugawa S., Yamamoto T., Ueda T., Ishida Y., Inagaki A., Nishigaki M., Shimada S., *J. Neurosci.*, **23**, 3616—3622 (2003).
37) Stevens D. R., Seifert R., Bufe B., Muller F., Kremmer E., Gauss R., Meyerhof W., Kaupp U. B., Lindemann B., *Nature* (London), **413**, 631—635 (2001).
38) Lin W., Burks C. A., Hansen D. R., Kinnamon S. C., Gilbertson T. A., *J. Neurophysiol.*, **92**, 2909—2919 (2004).
39) Richter T. A., Dvoryanchikov G. A., Chaudhari N., Roper S. D., *J. Neurophysiol.*, **92**, 1928—1936 (2004).
40) Miyamoto T., Fujiyama R., Okada Y., Sato T., *J. Neurophysiol.*, **80**, 1852—1859 (1998).
41) Huang A. L., Chen X., Hoon M. A., Chandrashekar J., Guo W., Trankner D., Ryba N. J., Zuker C. S., *Nature* (London), **442**, 934—938 (2006).
42) Ishimaru Y., Inada H., Kubota M., Zhuang H., Tominaga M., Matsunami H., *Proc. Natl. Acad. Sci. U.S.A.*, **103**, 12569—12574 (2006).
43) Huque T., Cowart B. J., Dankulich-Nagrudny L., Pribitkin E. A., Bayley D. L., Spielman A. I., Feldman R. S., Mackler S. A., Brand J. G., *PLoS ONE*, **4**, e7347 (2009).
44) Heck G. L., Mierson S., DeSimone J. A., *Science*, **223**, 403—405 (1984).
45) Ninomiya Y., Funakoshi M., *Brain Res.*, **451**, 319—325 (1988).
46) Spector A. C., Guagliardo N. A., St John S. J., *J. Neurosci.*, **16**, 8115—8122 (1996).
47) Yoshida R., Horio N., Murata Y., Yasumatsu K., Shigemura N., Ninomiya Y., *Neuroscience*, **159**, 795—803 (2009).
48) Chandrashekar J., Kuhn C., Oka Y., Yarmolinsky D. A., Hummler E., Ryba N. J., Zuker C. S., *Nature* (London), **464**, 297—301 (2010).
49) Lyall V., Heck G. L., Vinnikova A. K., Ghosh S., Phan T. H., Alam R. I., Russell O. F., Malik S. A., Bigbee J. W., DeSimone J. A., *J. Physiol.*, **558**, 147—159 (2004).
50) Ruiz C., Gutknecht S., Delay E., Kinnamon S., *Chem. Senses*, **31**, 813—820 (2006).
51) Caicedo A., Kim K. N., Roper S. D., *J. Physiol.*, **544**, 501—509 (2002).
52) Yoshida R., Shigemura N., Sanematsu K., Yasumatsu K., Ishizuka S., Ninomiya Y., *J. Neurophysiol.*, **96**, 3088—3095 (2006).
53) Yoshida R., Miyauchi A., Yasuo T., Jyotaki M., Murata Y., Yasumatsu K., Shigemura N., Yanagawa Y., Obata K., Ueno H., Margolskee R. F., Ninomiya Y., *J. Physiol.*, **587**, 4425—4439 (2009).
54) Ninomiya Y., Tanimukai T., Yoshida S., Funakoshi M., *Physiol. Behav.*, **49**, 913—918 (1991).
55) Ninomiya Y., Imoto T., Yatabe A., Kawamura S., Nakashima K., Katsukawa H., *Am. J. Physiol.*, **274**, R1324—R1330 (1998).
56) Yasumatsu K., Katsukawa H., Sasamoto K., Ninomiya Y., *J. Neurosci.*, **23**, 4362—4368 (2003).
57) Yasumatsu K., Kusuhara Y., Shigemura N., Ninomiya Y., *Eur. J. Neurosci.*, **26**, 1521—1529 (2007).
58) Ninomiya Y., Imoto T., Sugimura T., *J. Neurophysiol.*, **81**, 3087—3091 (1999).
59) Seta Y., Toyoshima K., *Anat. Embryol.*, **191**, 83—88 (1995).
60) Clapp T. R., Yang R., Stoick C. L., Kinnamon S. C., Kinnamon J. C., *J. Comp. Neurol.*, **468**, 311—321 (2004).
61) Bo X., Alavi A., Xiang Z., Oglesby I., Ford A., Burnstock G., *Neuroreport*, **10**, 1107—1111 (1999).

62) Finger T. E., Danilova V., Barrows J., Bartel D. L., Vigers A. J., Stone L., Hellekant G., Kinnamon S. C., *Science*, **310**, 1495—1499 (2005).
63) Romanov R. A., Rogachevskaja O. A., Bystrova M. F., Jiang P., Margolskee R. F., Kolesnikov S. S., *EMBO J.*, **26**, 657—667 (2007).
64) Huang Y. J., Maruyama Y., Dvoryanchikov G., Pereira E., Chaudhari N., Roper S. D., *Proc. Natl. Acad. Sci. U.S.A.*, **104**, 6436—6441 (2007).
65) Murata Y., Yasuo T., Yoshida R., Obata K., Yanagawa Y., Margolskee R. F., Ninomiya Y., *J. Neurophysiol.*, **104**, 896—901 (2010).
66) Kataoka S., Yang R., Ishimaru Y., Matsunami H., Sevigny J., Kinnamon J. C., Finger T. E., *Chem. Senses*, **33**, 243—254 (2008).
67) Huang Y. J., Maruyama Y., Lu K. S., Pereira E., Plonsky I., Baur J. E., Wu D., Roper S. D., *J. Neurosci.*, **25**, 843—847 (2005).
68) Friedman J. M., *Nat. Med.*, **10**, 563—569 (2004).
69) Lee G. H., Proenca R., Montez J. M., Carroll K. M., Darvishzadeh J. G., Lee J. I., Friedman J. M., *Nature* (London), **379**, 632—635 (1996).
70) Ninomiya Y., Sako N., Imai Y., *Am. J. Physiol.*, **269**, R930—R937 (1995).
71) Ninomiya Y., *Proc. Natl. Acad. Sci. U.S.A.*, **95**, 5347—5350 (1998).
72) Kawai K., Sugimoto K., Nakashima K., Miura H., Ninomiya Y., *Proc. Natl. Acad. Sci. U.S.A.*, **97**, 11044—11049 (2000).
73) Shigemura N., Ohta R., Kusakabe Y., Miura H., Hino A., Koyano K., Nakashima K., Ninomiya Y., *Endocrinology*, **145**, 839—847 (2004).
74) Martin B., Shin Y. K., White C. M., Ji S., Kim W., Carlson O. D., Napora J. K., Chadwick W., Chapter M., Waschek J. A., Mattson M. P., Maudsley S., Egan J. M., *Diabetes*, **59**, 1143—1152 (2010).
75) Saladin R., Vos D. P., Guerre-Millo M., Leturque A., Girard J., Staels B., Auwerx J., *Nature* (London), **377**, 527—529 (1995).
76) Ahima R. S., Prabakaran D., Mantzoros C., Qu D., Lowell B., Maratos-Flier E., Flier J. S., *Nature* (London), **382**, 250—252 (1996).
77) Sinha M. K., Sturis J., Ohannesian J., Magosin S., Stephens T., Heiman M. L., Polonsky K. S., Caro J. F., *Biochem. Biophys. Res. Commun.*, **228**, 733—738 (1996).
78) Nakamura Y., Sanematsu K., Ohta R., Shirosaki S., Koyano K., Nonaka K., Shigemura N., Ninomiya Y., *Diabetes*, **57**, 2661—2665 (2008).
79) Schoeller D. A., Cella L. K., Sinha M. K., Caro J. F., *J. Clin. Invest.*, **100**, 1882—1887 (1997).
80) Jamshidi N., Taylor D. A., *Br. J. Pharmacol.*, **134**, 1151—1154 (2001).
81) Cota D., Marsicano G., Tschop M., Grubler Y., Flachskamm C., Schubert M., Auer D., Yassouridis A., Thone-Reineke C., Ortmann S., Tomassoni F., Cervino C., Nisoli E., Linthorst A. C., Pasquali R., Lutz B., Stalla G. K., Pagotto U., *J. Clin. Invest.*, **112**, 423—431 (2003).
82) Kirkham T. C., Williams C. M., Fezza F., Di Marzo V., *Br. J. Pharmacol.*, **136**, 550—557 (2002).
83) Monteleone P., Matias I., Martiadis V., De Petrocellis L., Maj M., Di Marzo V., *Neuropsychopharmacology*, **30**, 1216—1221 (2005).
84) Yoshida R., Ohkuri T., Jyotaki M., Yasuo T., Horio N., Yasumatsu K., Sanematsu K., Shigemura N., Yamamoto T., Margolskee R. F., Ninomiya Y., *Proc. Natl. Acad. Sci. U.S.A.*, **107**, 935—939 (2010).