increasing GA2ox activity. This interaction is suggested as a mechanism that could help refine the boundary between cells with SAM or leaf identity — a process which is expected to be particularly important when the molecules mediating cell fate are mobile.

Gibberellin and cytokinin have antagonistic effects in a number of processes — suggested to reflect convergence of cytokinin and gibberellin signals on the SPY protein [13] or incompatibility in the effects of cytokinin on cell division and gibberellin on cell expansion [8]. Such antagonism could further discourage specification of cells with intermediate identities at the SAM-leaf boundary. One of the many questions raised by these findings is how a high concentration of cytokinin, which can affect leaf development [14], is itself restricted to the SAM.

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Evolution: A Study in Bad Taste?

Bitter tastes are among the most salient of life's experiences — who can forget one's first encounter with dandelion milk or a stout beer? Studies of the genes underlying these tastes are providing new perspectives on human origins and health.

Stephen Wooding

Bitter-taste sensitivity, of course, begins on the tongue. Concentrated at the back of the tongue, on disc-like structures called circumvallate papillae, specialized bitter-taste receptor cells await contact with potentially bitter compounds. Upon exposure to an appropriate ligand, these receptor cells depolarize, generating a signal that is conveyed via the facial and glossopharyngeal nerves to the brain (Figure 1A). In principle, any mechanism that stimulates this neural pathway will lead to the sensation of bitter taste; however, recent studies have highlighted the importance of a small group of G-protein-coupled receptors encoded by the *TAS2R* (also called *T2R*) gene family [1,2].

In humans, this family includes roughly 25 functional genes and eight pseudogenes, each roughly a kilobase in length, found in three clusters on chromosomes 5, 7 and 12. The protein products of these genes are concentrated at the apex of bitter-taste receptor cells, near the taste pore, where they are positioned to bind bitter ligands as they wash past, dissolved in saliva (Figure 1A). Upon ligand binding, these receptors catalyze a series of reactions leading to the efflux of intracellular calcium, and the cascade of events leading to taste perception begins (Figure 1B).

Considerable effort has been directed at identifying ligands for these receptors, and a range of compounds have been identified that are capable of activating TAS2R10, TAS2R14, TAS2R16, TAS2R38, TAS2R43, and TAS2R44 and TAS2R61 [3–7]. These studies have produced a variety of interesting surprises. The artificial sweetener saccharin, for instance, activates TAS2R43 [6]. More striking, however, is the observation that an inordinate fraction of the compounds that activate the TAS2Rs are secondary compounds produced by plants. Further, many of these compounds are toxic, used by plants as means of defense against herbivores. TAS2R10, for instance, binds strychnine [3], the well-known toxin found in plants in the genus *Strychnos*, and TAS2R14 binds α-thujone,

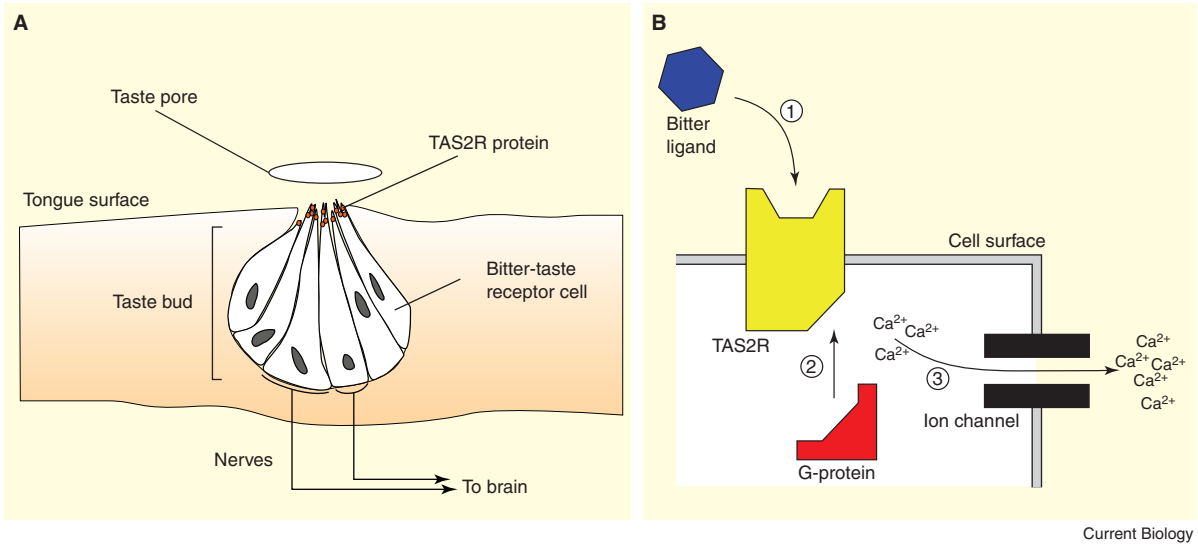

Figure 1. Bitter-taste receptor cells and proteins.

(A) Organization of bitter-taste receptor cells on the tongue. Many bitter-taste receptor cells, bundled together in a taste bud, are embedded in the surface of the tongue. The apices of these cells present bitter-taste receptor proteins to the interior of the mouth via the taste pore. (B) Basic series of events leading to bitter-taste receptor cell depolarization. 1: The bitter-taste receptor binds an appropriate ligand. 2: The TAS2R receptor couples with a G protein. 3: A series of catalyzed reactions release calcium ions from the cell, depolarizing it.

the principal neurotoxic component of absinthe, extracted from bitter wormwood [4]. Plants are a major component of primate diets, so the ability to taste these compounds could provide fitness advantages by enabling the individual to monitor, and thus regulate, toxin consumption.

The potential involvement of TAS2R receptors in sensing plant toxins has spurred a number of recent studies of the effects of natural selection on the *TAS2R* genes. Go *et al*. [8] and Parry *et al*. [9] investigated rates of pseudogenization, which are potentially indicative of major changes in evolutionary constraint. These studies found that rates of pseudogenization in the primates are higher than in other taxa; thus the range of bitter compounds perceived by primates is likely reduced relative to other groups.

Studies of amino-acid substitution rates have led to a related finding: rates of amino acid substitution are high in the TAS2Rs [10–14]. This finding could indicate a relaxation of evolutionary constraints, though it is also consistent with the hypothesis that *TAS2Rs* in primates are undergoing rapid adaptive evolution. For example, in a study of genetic variation in *TAS2R38*, which controls sensitivity to phenylthiocarbamide (PTC), my colleagues and I [10] found five nucleotide substitutions, all of which caused amino acid changes. The presence of unusually high numbers of intermediate-frequency alleles led to the conclusion that the PTC 'taster' and 'non-taster' alleles have been maintained by balancing natural selection favoring heterozygotes, who may taste a slightly broader range of bitter compounds than do homozygotes. Most recently, Kim *et al*. [13] found evidence that human populations differ more with respect to *TAS2R* genes than with respect to most other regions of the genome — consistent with the argument that local adaptation has been a pervasive force in the evolution of these genes.

Missing from all of these studies has been the exploration of connections between sensitivity to specific bitter compounds and adaptive processes. This gap has finally been filled by Soranzo *et al*. [15] in an investigation of population genetic variation in *TAS2R16*, reported very recently in *Current Biology*. The functional properties of TAS2R16 were first explored by Bufe *et al*., [3] who found that this receptor responds to β-glucopyranosides, a family of compounds that includes salicin, a natural analgesic found in bark of the willow (*Salix* spp.). Given the response of this receptor to a common, widespread family of plant toxins, Soranzo *et al*. [15] wanted to know the answers to several questions. How has natural selection shaped patterns of variation in this receptor? Is functional variation present? And how is this variation distributed among human populations. To find out, they resequenced *TAS2R16* in a sample of nearly 1,000 people worldwide, representing more than 52 populations from Africa, Asia, Europe, and North and South America.

Population genetic tests for natural selection in these populations revealed three evolutionarily derived nucleotide variants at very high frequencies. One of these caused an amino acid substitution, K172N. Further, levels of linkage disequilibrium (LD) around *TAS2R16* were found to extend for approximately

100 kb, an exceptionally long genomic distance. The presence of derived variants on a high LD background is consistent with the presence of a selective sweep, the classic selective event in which a new mutant is strongly favored and rises rapidly to high frequency.

To determine whether the selected K172N polymorphism was associated with phenotypic variance, Soranzo *et al*. [15] extended the earlier functional analyses of Bufe *et al*. [3]. These experiments revealed that the K172N variant is indeed correlated with phenotypic variance *in vitro*, with the evolutionarily derived N172 allele conferring two-fold greater sensitivity to salicin, arbutin (found in bearberries), and amygdalin (found in bitter almonds). Soranzo *et al*. [15] did not test any living humans for sensitivity to these bitter plant toxins, but previous studies have found strong agreement between *in vitro* and *in vivo* analyses of *TAS2R16* [3]. Thus, the *TAS2R16* variant that seems to have been rapidly driven to high frequency is likely associated with greater sensitivity to β-glucopyranosides. Greater sensitivity to β-glucopyranosides might have provided a fitness advantages by allowing the regulation of β-glucopyranoside intake.

Evidence that natural selection has favored a new, high-sensitivity N172 mutant at *TAS2R16* makes sense, but it also raises a question: why has the allele not become completely, rather than just nearly, fixed in human populations? Here, Soranzo *et al*. [15] present an intriguing, albeit more speculative, explanation. Among the populations harboring the low-sensitivity *TAS2R16* allele (K172), Soranzo *et al*. [15] observed, most of these populations are found in areas of Africa harboring endemic malaria. A number of plant-derived compounds are recognized as therapeutic treatments for malaria, and Soranzo *et al*. [15] argue that individuals carrying the low-sensitivity K172 allele might be predisposed to consume more of these compounds than are individuals carrying the high-sensitivity allele. If these compounds really are protective against malaria, the disadvantage of eating these poisons might have been offset by the advantage of avoiding malaria. If correct, this hypothesis is consistent with more than a simple selective sweep; it is consistent with local adaptation.

The value of inferences about the origins and distribution of variation in bitter-taste sensitivity extends beyond their evolutionary interest. As demonstrated by Soranzo *et al*. [15], such studies not only generate specific, testable hypotheses useful in genotype–phenotype association studies, they provide insights into similarities and differences among populations that may be helpful in revealing important subtleties in genetic epidemiology. In the realm of bitter-taste sensitivity, studies based on phenotype–phenotype correlations have a long and distinguished history, uncovering many important relationships between bitter-taste sensititivity and health-related behaviors [16]. For example, variable aversions to bitter compounds have been found to correlate with rates of thyroid-deficiency disease (with PTC nontasters being more susceptible than PTC tasters) [16]. Similarly, bitter-taste sensitivity has been implicated as a factor affecting smoking habits [17]. The development of detailed connections between genotype, phenotype, and population history like those outlined by Soranzo *et al*. promise to refine and fortify these important biomedical studies by injecting a new evolutionary perspective that tells us not just about ancient human history, but about modern human biology.

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