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Short communication

It hurts so good: oral irritation by spices and carbonated drinks and the underlying neural mechanisms

E. Carstens[a,*], Mirela Iodi Carstens[a], Jean-Marc Dessirier[b], Michael O'Mahony[c], Christopher T. Simons[c], Makoto Sudo[d], Satoko Sudo[d]

[a]*Section of Neurobiology, Physiology and Behavior, University of California, 1 Shields Avenue, Davis, CA 95616, USA*
[b]*Unilever Research US, 45 River Road, Edgewater, NJ 07020, USA*
[c]*Department of Food Science and Technology, University of California, 1 Shields Avenue, Davis, CA 95616, USA*
[d]*Department of Anesthesiology and Rescuscitology, Ehime University School of Medicine, Ehime, Matsuyama, Japan*

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Abstract

This paper reviews neurophysiological and psychological studies of oral irritation elicited by chemicals in spicy foods and carbonated drinks. Oral irritant, thermal and textural sensations are conveyed to the brain by the trigeminal pathway, which is separate from the gustatory and olfactory systems. In humans, repetitive application of capsaicin, citric acid, or concentrated NaCl elicits oral irritation that grows in intensity across trials ("sensitization"). After a rest period, reapplication elicits less irritation ("self-desensitization"), but if given recurrently will eventually evoke a progressive rise in irritation ("stimulus-induced recovery" = SIR). In neurophysiological recordings from neurons in the trigeminal subnucleus caudalis (Vc), the first relay in the pathway for oral somatosensation, these irritants elicit a similar pattern of progressively increasing firing, followed after a rest by self-desensitization and SIR. In contrast, nicotine, menthol or mustard oil elicit irritation that decreases across trials ("desensitization"), a pattern also observed in Vc neuronal responses to these irritants. Carbonated water elicits an oral tingling sensation and excites Vc neurons largely through its conversion to carbonic acid. The good correspondence in temporal profiles for perception and neuronal activity supports a role for Vc neurons in the mediation of oral irritation. Finally, the development of preference for foods containing aversive chemicals is addressed. This may involve mere exposure, social reinforcement, the "thrill" of the strong sensation, or physiological reinforcement associated with satiety or release of endorphins by the painful stimulus.

Keywords: Oral irritation; Trigeminal; Neuron; Sensitization; Desensitization; Stimulus-induced recovery; Capsaicin; Citric acid; Nicotine; Mustard oil; Menthol; Salt

1. Introduction

In addition to taste and smell, a variety of other sensory qualities contribute to the overall flavor of foods and drinks including texture, temperature and tingling or outright irritation. Textural, thermal and irritant qualities are conveyed via the trigeminal system in parallel with gustatory and olfactory pathways. Trigeminal sensory qualities can be very important to the acceptability of food. For example, many people prefer to sip coffee at a temperature that is actually above the threshold to elicit pain and tissue damage. Other examples are the crunchiness of chips, the tingling of carbonated drinks, and the spiciness associated with certain cuisines. In particular, carbonated drinks and spicy foods have both gained enormously in popularity in recent years. Total sales of carbonated products in the US in 1997 were estimated at $55 billion (Beverage Digest, 1998). While considerable effort has been made toward understanding the gustatory and olfactory aspects of food flavor, relatively less is known about trigeminal sensory qualities and their interaction with taste and smell. This paper reviews recent advances in our knowledge of oral irritant sensations based on human psychophysical studies, as well as animal studies aimed at understanding the underlying neural mechanisms.

* Corresponding author. Tel.: +1-530-752-6640; fax: +1-530-752-5582.
E-mail address: eecarstens@ucdavis.edu (E. Carstens).

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2. Irritation

Parker (1912) originally coined the term "common chemical sense" as a unitary sensory modality to detect irritant chemicals in the environment. More recently the term "chemesthesis" was introduced to describe a set of distinct sensory qualities (e.g., stinging, burning) elicited by irritant chemical stimulation of the skin or mucosa (Green, Mason, & Kare, 1990). Probably the most common term used to describe sensations elicited by pungent chemicals is "irritation". While lexical definitions of irritation usually include discomfort, soreness and even inflammation, irritation is not necessarily associated with inflammation or pain, and is often even desired. It is not certain if irritation lies along the same sensory continuum as pain, and few studies have assessed possible differences in sensory qualities according to the intensity of irritant chemical stimulation. Instillation of histamine into the eye was reported to elicit irritation at low concentrations and sticking pain at higher concentrations (Heubner, 1925; Keele & Armstrong, 1964). As discussed in detail below, there is evidence that application of irritant chemicals to the oral or ocular mucosa excites pain-transmitting trigeminal neurons (Carstens, Kuenzler, & Handwerker, 1998). That a common population of neurons is excited by noxious (overtly or potentially damaging) physical as well as irritant chemical stimuli suggests that irritation and pain are linked. However, it is not yet known if separate sensory pathways exist that can be activated only by irritant chemical stimuli.

3. Trigeminal pathway for oral irritation

Pain, temperature and touch sensations in the oral cavity are signaled by the trigeminal system. The mandibular branch of the trigeminal nerve gives rise to nerves innervating the oral cavity. In particular, the lingual nerve innervates the tongue. Individual lingual nerve fibers have specialized sensory receptors in the tongue tissue that are activated by thermal (warm or cold), mechanical (touch-pressure) and noxious stimuli. The latter are called nociceptors and generally have thinly myelinated or unmyelinated axons. Although our understanding of lingual nociceptors is incomplete, some of them respond to irritant chemicals applied to the tongue (Bryant & Moore, 1995; Hellekant, 1965; Komai & Brant, 1993; Wang, Erickson, & Simon, 1993; reviewed in Schults, 1992b). Lingual sensory fibers project into the brain stem trigeminal complex where they synaptically contact neurons, particularly in subnucleus caudalis (Vc) (for reviews, see Carstens et al., 1998; Schults, 1992a). Trigeminal neurons, in turn, send information to the somatosensory thalamus and cortex.

Although it is known that Vc neurons convey nociceptive information from the oral cavity (e.g. Amano, Hu, & Sessle, 1986), there was very little information regarding the signaling of oral chemical irritation. We wished to determine if Vc neurons respond to irritant chemical stimulation of the tongue. To first target the location of such neurons, we employed the method of c-fos immunohistochemistry (Carstens, Saxe, & Ralph, 1995). This method is based on the observation that the proto-oncogene, c-fos, is expressed by some neurons following strong sensory stimulation (Harris, 1998; Hunt, Pini, & Evans, 1987). Because the c-fos can be immunohistochemically tagged with an antibody, this method provides a static anatomical view of entire populations of neurons in the central nervous system that are activated by the stimulus. We found that application of a variety of different irritant chemicals (capsaicin, piperine, histamine, nicotine, etc.) elicited similar distributions of c-fos-positive neurons in the superficial layers of the dorsomedial aspect of Vc in rats. However, because only one irritant can be tested per animal, this method does not allow one to conclude whether there are separate populations of Vc neurons with overlapping distributions, each one selectively responsive to only one irritant, or if individual Vc neurons respond to multiple irritant chemicals. To answer this question, we used electrophysiological single-unit recording methods to assess the dynamic responses of individual Vc neurons to application of a variety of different stimuli. A typical example is shown in Fig. 1. This neuron, located in dorsomedial Vc, responded to noxious thermal stimulation of the tongue (upper left PSTH), as well as to application of a wide variety of different irritant chemicals. The other PSTHs show this neuron's responses to several irritant chemicals. In general, Vc neurons' responses to a given irritant were concentration-dependent, and they did not respond to any irritant at a concentration below threshold for irritation when self-applied to the tongue (Fig. 1, lower right PSTH; Carstens et al., 1998).

These studies have therefore identified a population of Vc neurons that are likely to be involved in signaling oral irritation. However, the fact that most Vc neurons respond non-selectively to a variety of chemicals poses a problem in terms of the ability to make qualitative distinctions among different irritant sensations. Clearly, one can distinguish between highly salty and acidic stimuli using taste cues. However, it is unclear to what extent humans can distinguish between irritants that are considered tasteless, such as capsaicin and piperine. Future psychophysical studies are needed to assess irritant discriminability. Moreover, if humans can make qualitative distinctions based solely on trigeminal input, additional neurobiological studies are needed to determine if chemically-specific neurons exist that could signal such distinctions.

4. Correlative psychophysical and neurobiological studies of oral irritation

4.1. Capsaicin-induced sensitization, self-desensitization and stimulus-induced recovery

Having identified Vc neurons that may be involved in brain processing of oral irritation, we wished to determine if their response patterns are similar to the temporal changes in the intensity of irritation elicited by repetitive application of chemicals in human psychophysical studies.

The pioneering studies of Lawless (e.g. Lawless & Stevens, 1988; Stevens & Lawless, 1987) and Green (e.g. Green, 1988, 1989; Green & Lawless, 1991) have established psychophysical methods to track the intensity of oral irritation. Capsacinoids such as capsaicin or piperine elicit irritation, the intensity of which usually increases progressively across trials of repeated application at regular intervals (e.g. 1 min) to the tongue (Dessirier, Nguyen, O'Mahoney, Siffermann, & Carstens, 1999; Dessirier, O'Mahoney, & Carstens, 1997; Green, 1989; Karrer & Bartoshuk, 1991) although there is considerable individual variability (Prescott, 1999). This phenomenon is called "sensitization". Following a rest period, reapplication of capsaicin elicits a much-reduced level of irritation; this is called "self-desensitization". However, if capsaicin reapplication is continued, the irritation eventually increases in magnitude, a phenomenon called "stimulus-induced recovery" (SIR; Green, 1996).

We investigated if Vc neurons exhibit similar firing patterns to repeated or sustained application of capsaicin (Dessirier, Simons, Sudo, Sudo, & Carstens, 2000).

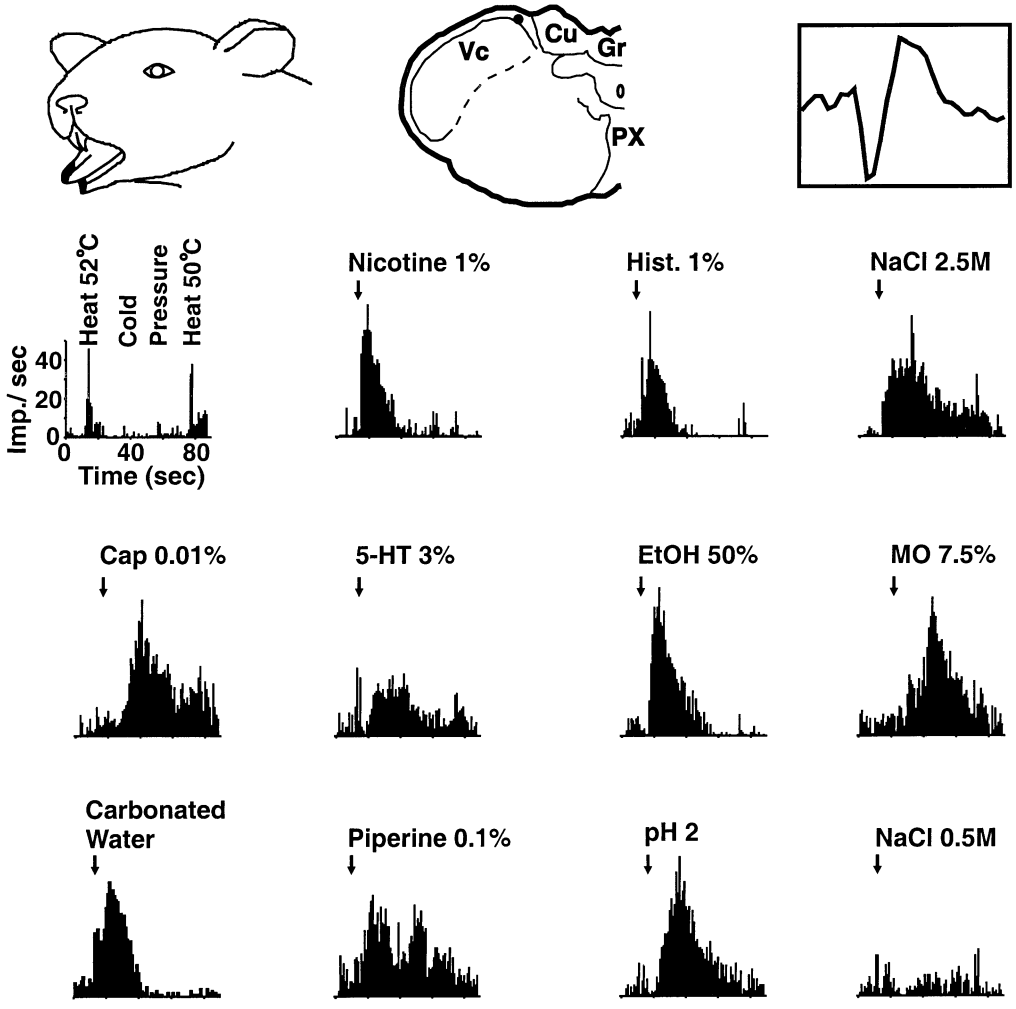

Fig. 1. Non-selective responses of Vc neuron to different oral irritants. The figurines in the upper row show, from left to right, a drawing of the rat's face with the receptive field of this neuron on the tongue and chin (black), a cross-section of the medulla showing the recording site (dot) in the superficial part of dorsomedial Vc, and the action potential waveform. The lower three rows show peristimulus-time histograms (PSTHs, bin width: 1 s) of responses to various stimuli. Calibrations given in left-hand PSTH, second row. Abbreviations: hist., histamine; Cap, capsaicin; 5-HT, serotonin; EtOH, ethanol; MO, mustard oil; pH2, HCl at pH of 2. Abbreviations for upper middle inset: Vc, trigeminal subnucleus caudalis; Cu, cunate n.; Gr, gracile n.; PX, pyramidal decussation. Adapted from Carstens et al. (1998).

When capsaicin (100 ppm) was applied repetitively at 1-min intervals, there was a progressive rise in Vc neuronal firing over the initial 8–10 trials (Fig. 2A) consistent with sensitization. After a rest period of 30–60 min, capsaicin was reapplied and initially elicited no response (self-desensitization), followed eventually by a rise in firing similar to a partial SIR (Fig. 2B). A similar sensitizing pattern, followed after a rest period by self-desensitization and SIR, was observed with constant-flow application of capsaicin; because repetitive and continuous application elicited similar firing patterns, we have used flow in subsequent studies. A single application of capsaicin induced a significant increase in Vc neuronal firing, but over a much slower time course. Furthermore, when capsaicin was singularly reapplied after a 30–60 min rest period, the Vc neurons no longer responded at all. This indicates that repetitive or continual application of capsaicin is necessary to induce sensitization and SIR. It is interesting that a capsaicin concentration of 100 ppm, but not 10 ppm, was necessary to excite Vc neurons while capsaicin concentrations well below 10 ppm are perceived as irritating by humans. In behavioral studies in our laboratory using a two-bottle paired-preference model, rats and mice reject capsaicin at concentrations of less than 10 ppm (Simons, Dessirier, Jinks, & Carstens, 2001). These findings suggest that higher capsaicin concentrations are necessary to excite Vc neurons because of the depressant effect of the anesthetic used in the neurophysiological experiments.

Sensitization might be explained by an increase in the sensitivity of peripheral receptors (peripheral "sensitization", a physiological change in receptor sensitivity not to be confused with the psychophysical term) or of Vc neurons (central sensitization) to subsequent application of the irritant. Sensitization might also be explained by spatial recruitment of trigeminal receptors by diffusion of capsaicin. Thus, immediately upon application of capsaicin, trigeminal receptors at the application site are excited. However, some of the capsaicin will diffuse into the adjacent lingual epithelium. As the capsaicin is replenished by repeated or continual application, it will diffuse to a border at which the diffusion rate is balanced by clearance rate. During the initial period of diffusion, additional trigeminal receptors are activated that may contribute to an increase in Vc neuronal firing and perceived intensity of irritation. Once the diffusion border is reached, the neuronal firing and irritant intensity reach a plateau (Fig. 2A).

Self-desensitization might also be explained by a peripheral or central depression of trigeminal activity. A peripheral site seems more likely and is supported by recent advances in cell biology. A molecular vanilloid receptor (VR-1) that is activated by noxious heat and capsaicin has recently been identified (Caterina, Schmacher, Tominage, Rosen, Levine, & Julius, 1997). Using patch-clamp methods it is possible to record inward depolarizing currents elicited by capsaicin when it binds to VR-1 receptors expressed in spinal dorsal root ganglion neurons or trigeminal ganglion neurons (a surrogate for trigeminal nerve endings). Interestingly, there is a progressive reduction (tachyphylaxis) in the size of currents when capsaicin is applied repeatedly (Liu & Simon, 1996b). More recently, it was discovered that capsaicin can interfere with the generation of action potentials, apparently by blocking voltage-sensitive Na^+ channels (Liu, Oortgiesen, Li, & Simon, 2001; Su, Wachtel, & Gebhart, 1999). This latter effect may well explain not only self-desensitization by capsaicin, but also why capsaicin readily cross-desensitizes irritation elicited by subsequent application of many other irritant chemicals (Dessirier et al., 1997, 1999; Green, 1991).

Sensitization and self-desensitization may reflect competing cellular mechanisms, whereby sensitization dominates during the maintained presence of capsaicin, while desensitization predominates after cessation of

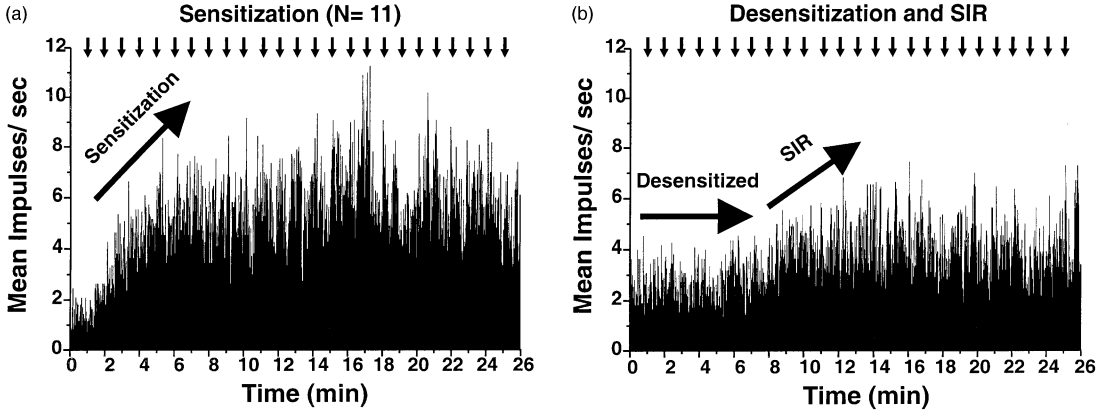

Fig. 2. Capsaicin sensitization, desensitization and stimulus-induced recovery (SIR) in Vc neuronal responses. (a) Averaged PSTH (bin width 1 s) of the population response of 11 Vc neurons to repeated application of 100 ppm capsaicin at 1-min intervals (arrows). (b) Averaged PSTH of the same neurons shown in A to reapplication of capsaicin 30–60 min after the end of the first series. From Dessirier, Simons, Sudo et al. (2000) with permission.

capsaicin. SIR evoked by recurrent capsaicin application may represent a sensitization process that eventually overcomes desensitization (Green, 1996).

4.2. Sensitization by acids

Acidic stimuli taste sour and elicit irritation at higher concentrations. The magnitude of citric acid-induced irritation is proportional to concentration, and exhibits sensitization (Fig. 3A), self-desensitization, and cross-desensitization by capsaicin (Dessirier, O'Mahoney, Iodi Carstens, & Carstens, 2000; Gilmore & Green, 1993). Following capsaicin-induced cross-desensitization, recurrent application of citric acid induces an increase in irritant intensity: cross-SIR (Dessirier, O'Mahoney et al., 2000). Similarly, Vc neurons give increasing responses to HCl over a pH range of 4 to 1 (Carstens et al., 1998) and respond to repeated application of pentanoic acid (5-min intervals) although the responses do not exhibit tachyphylaxis (Dessirier, Simons, Sudo et al., 2000). Responses of Vc neurons to continual application of 250 mM citric acid exhibit a progressive increase in firing similar to sensitization (Fig. 3B). Thus, as with capsaicin, there is a marked similarity in the sensitizing pattern of irritant sensation and Vc neuronal firing (Fig. 3A, B).

There are at least two mechanisms by which acids might excite trigeminal receptors. The recent discovery of acid-sensitive ion channels (ASICs), which are members of the amiloride-sensitive Na^+ channel/degenerin family (Waldman, Champigny, Bassilana, Heurteaux, & Lazdunski, 1997; Waldman, Champigny, Lingueglia, De Weille, Heurteaux, & Lazdunski, 1999), provides one means for extracellular protons to depolarize trigeminal nerve endings. To date, three sub-types of ASIC [ASIC-1, ASIC-2 (formerly MDEG = mammalian degenerin) and ASIC-3 (formerly DRASIC = dorsal root ASIC)] have been identified in sensory neurons (Waldman & Lazdunski, 1998). Patch-clamp studies of trigeminal ganglion neurons revealed several patterns of acid-evoked inward currents, some of which are blocked by amiloride (Liu & Simon, 2000). In this regard, we found that amiloride reduced the magnitude of citric acid-evoked irritation (Dessirier, O'Mahoney et al., 2000), consistent with a role for ASIC. A second mechanism involves VR-1. Capsaicin-evoked inward currents in dorsal root and trigeminal ganglion cells are enhanced by acidification (e.g. Liu & Simon, 2000), and it was recently reported that cutaneous nociceptors from knockout mice lacking the VR-1 receptor show a significantly lower incidence of responsiveness to acidic stimuli compared to nociceptors in wildtype mice (Caterina et al., 2000). Consistent with this, acid-evoked inward currents in some trigeminal ganglion cells were blocked by the VR-1 antagonist, capsazepine (Liu & Simon, 2000).

4.3. NaCl sensitization

High molar concentrations of NaCl elicit oral irritation that exhibits sensitization upon repeated application and self-desensitization following a hiatus (Dessirier, O'Mahoney, Iodi Carstens, Yao, & Carstens, 2001; Gilmore & Green, 1993; Green & Gelhard, 1989) (Fig. 4A). Similarly, Vc neurons exhibit a sensitizing pattern of firing in response to continual lingual application of 5 M NaCl (Fig. 4B). Interestingly, NaCl induces cross-sensitization of capsaicin-evoked irritation (Dessirier, O'Mahoney, Iodi Carstens, Yao, & Carstens, 2001). One explanation is that the capsaicin-evoked depolarization of trigeminal nerve endings is enhanced if there is a higher extracellular concentration of Na^+ ions available to pass through the VR-1-gated cation channel. Finally, we observed that NaCl-evoked

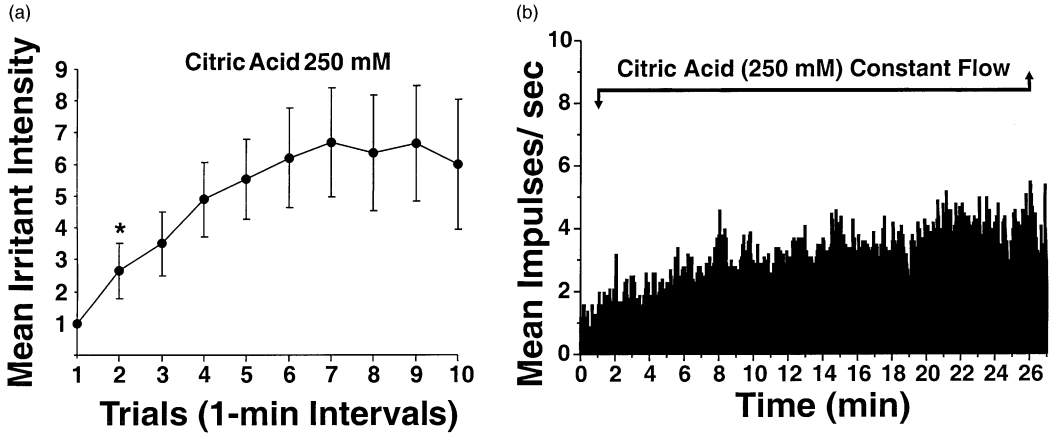

Fig. 3. Sensitization by citric acid. (a) Graph plots mean ratings of irritant intensity elicited by application of 250 mM citric acid by filter paper onto one side of the tongue 10 times in succession at 1-min intervals (from Dessirier, O'Mahoney, Iodi Carstens, & Carstens, 2000, with permission). (b) Averaged PSTH of population response of 10 Vc neurons to constant-flow (0.32 ml/min) application of 250 mM citric acid to the tongue in rats anesthetized with thiopental and halothane. Flow began at downward arrow and ended at upward arrow.

oral irritation was reduced by amiloride, suggesting involvement of amiloride-sensitive Na^+ channels.

4.4. Nicotine desensitization

In contrast to the sensitization observed with repeated application of capsaicin, citric acid and NaCl, we found that nicotine, a bioactive agent in tobacco, evokes irritation that declines in intensity across trials (Dessirier et al., 1997, 1999). This is called desensitization (Fig. 5A). Similarly, Vc neurons are initially excited by lingual application of nicotine, followed by a decline in firing to baseline levels despite the maintained presence of nicotine (Fig. 5B). Nicotine-evoked irritation exhibits self-desensitization following a rest period. Likewise, responses of Vc neurons to nicotine repeated at 5-min intervals exhibited significant tachyphylaxis (Carstens et al., 1998). Moreover, nicotine at sufficiently high concentration (300 mM) cross-desensitized capsaicin-evoked irritation (Dessirier, Chang, O'Mahoney, & Carstens, 2000). Nicotine (600 mM) also cross-desensitized responses of Vc neurons to pentanoic acid (authors' unpublished observations).

Nicotine-evoked Vc neuronal responses (Carstens et al., 1998) and c-fos expression (Carstens, Simons, Dessirier, Iodi Carstens & Jinks, 2000), as well as nicotine-evoked irritant sensation (Dessirier, O'Mahoney, Sieffermann, & Carstens, 1998), are blocked by mecamylamine, a neuronal nicotinic acetylcholine receptor (nAChR) antagonist. This implicates the involvement of nAChRs expressed in trigeminal nerve endings in nicotine-evoked irritation and pain. nAChRs consist of a pentameric combination of α- and β-nAChR subunits surrounding a central ion channel (for reviews, see

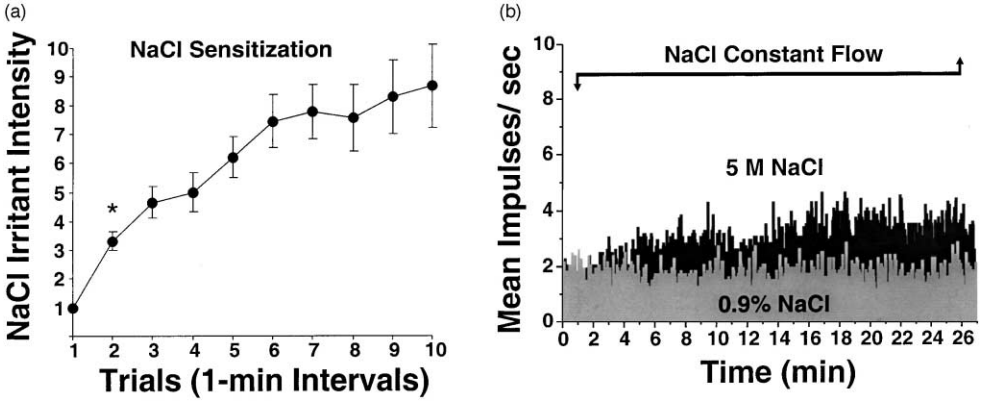

Fig. 4. Sensitization by NaCl. (a) Graph as in Fig. 3a showing mean ratings of irritation elicited by successive application of 5 M NaCl to the tongue 10 times at 1-min intervals. Asterisk denotes significant difference from trial 1 (from Dessirier, O'Mahoney, Iodi Carstens, & Carstens, 2001, with permission). (b) Averaged PSTH of response of nine Vc neurons to constant-flow application of NaCl to the tongue in rats anesthetized with thiopental and halothane (format as in Fig. 3b). Black PSTH: response to 5 M NaCl; gray PSTH: absence of response of nine other Vc neurons to isotonic (0.9%) saline.

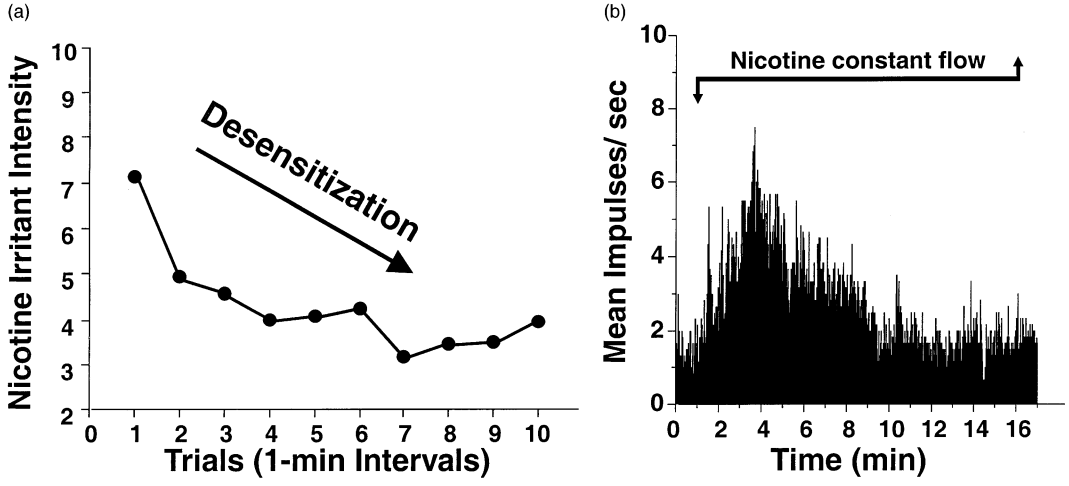

Fig. 5. Desensitization by nicotine. (a) Graph as in Fig. 3a of mean ratings of irritation elicited by sequential application of 7.2 mM nicotine by filter paper to the tongue 10 times at 1-min intervals. From Dessirier et al. (1997) with permission. (b) Averaged PSTH of responses of Vc neurons to constant-flow application of 600 mM nicotine to the tongue of thiopental-anesthetized rats (from Dessirier, Simons, Sudo et al., 2000, with permission).

Cordero-Erausquin, Marubio, Klink, & Changeaux, 2000; Gotti, Fornasari, & Clementi, 1997; Paterson & Nordberg, 2000); α4β2 and α2β4 heteromers and α7 homomers have been identified in sensory neurons. An interesting feature of nAChRs is tachyphylaxis to repeated application of nicotine (Liu & Simon, 1996a), which might underlie desensitization observed psychophysically and in Vc neuronal responses (Fig. 5).

4.5. Menthol desensitization

Repeated application of menthol to the oral cavity at 1-min intervals elicits irritation (and cooling) that desensitizes across trials (Fig. 6A; Cliff & Green, 1994, 1996; Dessirier, O'Mahoney, & Carstens, 2001). Menthol-responsive Vc units found to date exhibit a response profile similar to that of nicotine; i.e. excitation followed by a decline, consistent with desensitization. An example is shown in Fig. 6B. Menthol irritation exhibits self-desensitization (independent of its cooling action) as well as cross-desensitization of nicotine-evoked irritation (Dessirier, O'Mahoney, & Carstens, 2001), a finding that may be of relevance to the acceptance of mentholated tobacco products. Menthol also reduced capsaicin-evoked irritation when presented shortly before or in mixture (Green & McAuliffe, 2000), but cross-sensitized capsaicin irritation when presented 15 min earlier (Cliff & Green, 1996). Interestingly, repeated delivery of menthol at more rapid intervals (20 s) elicited irritation that rose significantly over the first few trials, followed by significant desensitization (Dessirier, O'Mahoney, & Carstens, 2001). After a rest period, recurrent reapplication of menthol elicited progressively increasing irritation indicative of SIR. These findings underscore the importance of interstimulus interval in determining the temporal profile of irritation, and raise the question as to whether other desensitizing agents such as zingerone (Prescott & Stevenson, 1996a, 1996b) might exhibit an initial phase of sensitization when delivered rapidly.

Ethanol also elicits irritation that desensitizes across trials repeated at 1-min intervals (Prescott & Swain-Campbell, 2000), and excites Vc neurons (Carstens et al., 1998). It is currently not known if menthol and ethanol excite trigeminal receptors via specific molecular receptors or by some general effect on the membrane of fiber endings.

4.6. Mustard oil desensitization

Like nicotine and menthol, repeated application of mustard oil (allyl-isothiocyanate; 0.125%) to the tongue elicits irritation that desensitizes across trials (Fig. 7A, authors' unpublished observations). Mustard oil induces self-desensitization and exhibits reciprocal cross-desensitization with capsaicin. Mustard oil also readily excites Vc neurons with a moderate decline in firing over time (Fig. 7B), consistent with the psychophysical data.

4.7. Carbonated water

The fizzy sensation of carbonated drinks is due to bubbles containing CO_2 gas. Several lines of evidence indicate that the sensation is not due solely to the mechanical bursting of bubbles against the oral mucosa. The tingling sensation persists after the carbonated water is expectorated (Green, 1992). Moreover, ratings of several qualitative descriptors (e.g. pricking, tingling, mouth-burn, etc.) of carbonated water were the same whether subjects drank it under normal atmospheric conditions or in a hyperbaric chamber that prevented the formation of bubbles (S. McEvoy, personal

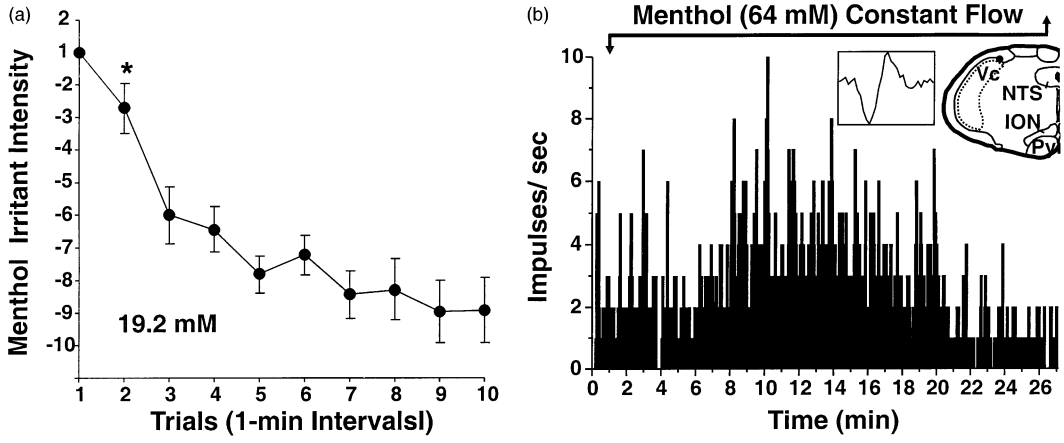

Fig. 6. Desensitization by menthol. (a) Graph as in Fig. 3a showing progressive decline in ratings of oral irritation elicited by repeated application of 19.2 mM menthol to the tongue at 1-min intervals (from Dessirier, O'Mahoney, & Carstens, 2001). (b) Example of a Vc neuron's response to constant-flow application of 64 mM menthol to the tongue of a rat anesthetized with thiopental and halothane. Insets show action potential waveform (left) and Vc recording site (dot) on a cross-section of the brain stem (right). Abbreviations: NTS, n. of the solitary tract; ION, inferior olivary n.; Py, pyramid.

communication). Finally, mountain climbers taking Diamox (acetazolamide) for mountain sickness reported that carbonated drinks were no longer fizzy and that beer tasted like "dishwater"; this was termed the "champagne blues" (Graber & Kelleher, 1988). Acetazolamide and related drugs (dorzolamide) block the enzyme, carbonic anhydrase, that catalyzes the conversion of CO_2 gas to carbonic acid. Acetazolamide blocks responses of lingual nerve sensory fibers to carbonated water (Komai & Bryant, 1993). Taken together, these data suggest that formation of carbonic acid is critical to the oral sensation of carbonated drinks.

Since carbonated water excites nociceptive Vc neurons (Fig. 1, lower left PSTH), we hypothesized that the oral sensation of carbonation is mediated by the diffusion of CO_2 gas from bursting bubbles into the oral epithelium, where it is converted into carbonic acid that excites trigeminal receptors which, in turn, activate nociceptive Vc neurons. We predicted that blockers of carbonic anhydrase would reduce the intensity of the oral sensation of carbonation, as well as the responses of Vc neurons to carbonated water. In psychophysical studies (Dessirier, Simons, O'Mahoney, & Carstens, 2000; Simons, Dessirier, Iodi Carstens, O'Mahoney, & Carstens, 1999) we pretreated one side of the tongue with acetazolamide or dorzolamide, and the other side with a control solution matched for taste and viscosity. The subjects' tongues were then exposed to carbonated water bilaterally, and in a 2-alternative forced-choice paradigm the subject stated which side of the tongue had a stronger sensation of carbonation. Subjects also rated the intensity of the sensation independently on both sides. With both blockers of carbonic anhydrase, a significant majority of subjects chose the control side as having the stronger sensation and assigned significantly higher intensity ratings to that side. To rule out a non-specific effect of the blockers on acid-evoked irritation, the experiment was repeated except that citric

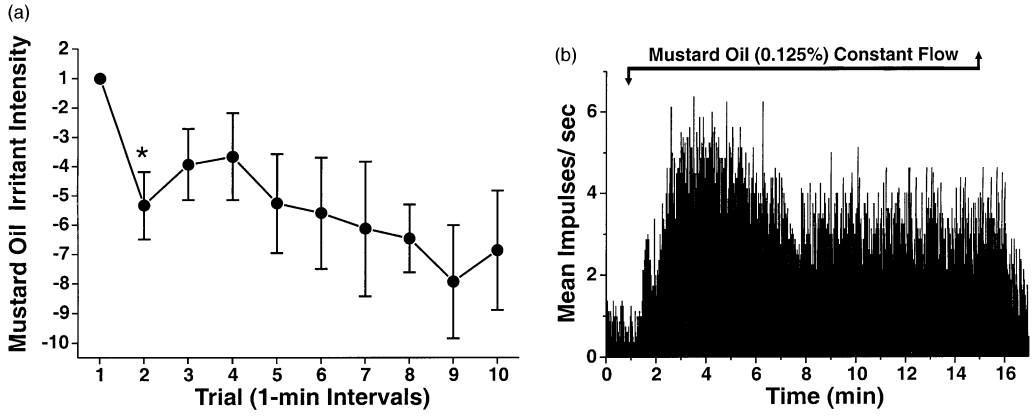

Fig. 7. Desensitization by mustard oil. (a) Graph as in Fig. 3a showing decline in successive ratings of irritation elicited by application of 0.125% mustard oil (allyl-isothiocynate) to the tongue. (b) Averaged PSTH of response of eight Vc neurons to constant-flow application of 0.125% mustard oil to the tongue in rats anesthetized with thiopental and halothane.

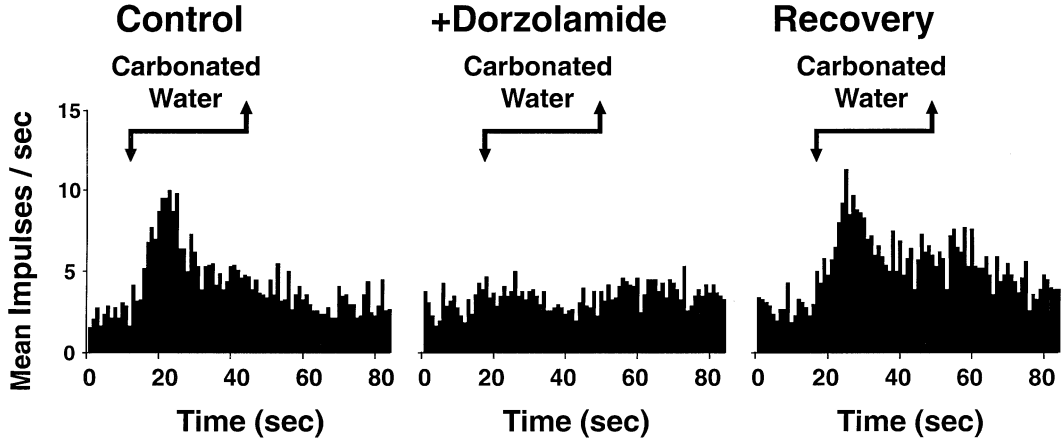

Fig. 8. Dorzolamide blockade of response of Vc neurons to carbonated water. Shown are averaged PSTHs of 10 Vc neurons. The left-hand PSTH shows the response to carbonated water flowed over the tongue for 60 s (on- and offset of flow indicated by downward and upward arrows, respectively). The middle PSTH shows absence of response when the carbonated water stimulus was preceded by application of 2% dorzolamide to the tongue. The right-hand PSTH shows recovery of the response 30 min later (from Simons et al., 1999, with permission).

or pentanoic acid was applied bilaterally instead of carbonated water. Pretreatment with the carbonic anhydrase blockers had no effect, as subjects chose the control and pretreated sides of the tongue in equal proportions and gave comparable intensity ratings of acidic irritation for the two sides of the tongue. A variety of control experiments ruled out possible anesthetic, taste and pH effects. We conclude that the oral sensation of carbonated water, but not acids in general, requires the conversion of CO_2 gas into carbonic acid in a carbonic anhydrase-dependent reaction.

In parallel animal experiments, we also showed that acetazolamide and dorzolamide significantly reduced the activation of Vc neurons as assessed by both c-fos expression and electrophysiological single-unit recordings (Dessirier, Simons, O'Mahoney, & Carstens, 2000; Simons et al., 1999). Dorzolamide blockade of carbonated water-evoked excitation of Vc neurons is illustrated in Fig. 8.

Carbonic acid might excite trigeminal receptors in the oral epithelium via ASIC or VR-1 receptor mechanisms. If ASIC is involved, amiloride would be predicted to reduce the sensation of carbonation. However, we recently found that amiloride enhances, rather than reduces, the sensation elicited by carbonated water (Dessirier, Simons, O'Mahoney, & Carstens, 2001). This is consistent with a previous study showing that amiloride enhanced the responses of cutaneous nociceptors to CO_2 (Steen, Wegner, & Reeh, 1999). The latter finding was interpreted in relation to amiloride's ability to block a cellular Na^+/H^+ exchange pump that regulates cell volume and pH (Lang et al., 1998; Madshus, 1988). However, amiloride was also recently shown to enhance certain acid-evoked inward currents recorded in trigeminal ganglion cells (Liu & Simon, 2000), consistent with our psychophysical data. The exact site and transduction mechanism by which carbonic acid activates trigeminal nerve endings remains to be elucidated.

4.8. Summary

The data summarized above indicate that a variety of irritant chemicals in food spices, as well as carbonation, activate trigeminal pain pathways. It is interesting that for a given chemical, the temporal pattern of irritant sensation, and firing of Vc neurons, is similar. There appear to be two distinct temporal patterns. Sensitization (i.e. a progressive increase in irritant intensity and Vc firing) is observed with capsaicin, piperine, citric acid and concentrated NaCl. In contrast, desensitization (i.e. irritation and Vc firing that progressively declines) is seen with nicotine, menthol, mustard oil, zingerone and ethanol. The pattern may also depend on stimulus frequency, as shown for menthol-evoked irritation which desensitizes at long interstimulus intervals but which exhibits sensitization followed by desensitization at shorter intervals. Some irritants appear to excite trigeminal receptors via a distinct molecular receptor: VR-1 for capsaicin, ASIC for acids, and nAChRs for nicotine. Transduction mechanisms for other irritant chemicals have not yet been elucidated. Our combined approach reveals striking similarities in psychophysical and neural measures of irritation, and supports a role for Vc neurons in the mediation of oral irritation.

5. Acquired preference for spicy food

Given that irritants like capsaicin activate trigeminal pain pathways, it should be expected that they are aversive to animals and humans and would be rejected. It is thus interesting to consider factors that lead many people to develop a preference for spicy food and tingly drinks. An excellent discussion of this topic may be found in a review by Rozin (1990).

It is generally assumed that irritant spices are aversive when first encountered, as suggested by the negative reactions normally exhibited by young children or pets. Before discussing possible factors in the acquisition of a preference for spicy food in humans, it is interesting to consider whether animals exhibit innate or learned preferences for spicy food. In virtually all previous studies in which animals had a choice between normal dry food and food spiced with pepper, the latter was rejected (Hilker, Hee, Higashi, Ikehara, & Paulsen, 1967; Rozin, Gruss, & Berk, 1979). However, there are cases in which rats or chimpanzees developed a preference for the spicy food after social interactions (Dib, 1990; Galef, 1989; Rozin & Kennel, 1983). To establish a rodent model of oral irritation by aqueous solutions, we employed a two-bottle paired preference method (Simons et al., 2001). One bottle contained water and the other a given concentration of capsaicin in water, and the mouse or rat had free access to both bottles. To avoid positional preferences, bottle position was switched daily during the 7-day period in which the animal was exposed to each capsaicin concentration. The amount of fluid consumed from each bottle was measured. The relative consumption of capsaicin compared to water decreased in a concentration-dependent manner above a threshold concentration of approx. 2 μM, and in no case did we observe a preference for the capsaicin solution.

The preference for spicy food by humans is likely to be acquired in most cases, often between the ages of 5 and 9 (Rozin, 1990). Clearly, exposure to the spice is necessary in order to develop a liking, but is exposure alone sufficient? Furthermore, some individuals may show an innate liking of the burning sensation evoked by spices, while others find it neutral or aversive. In the latter two instances, what factors govern the development of a preference for an initially neutral or aversive sensation?

There are few studies addressing whether the initial contact with spices is indeed aversive. In interviews of college students, fully 43% reported never having disliked chili (Rozin & Schiller, 1980) and 15% reported liking chili upon the initial exposure (Rozin, 1990). These data suggest that for many people the initial experience with spicy food is not necessarily aversive. This idea was pursued by Stevenson and Yeomans (1995), who reasoned that very infrequent consumers of spicy foods (chili "non-likers") may be representative of novices at the initial stage of exposure to spices. Compared to non-likers, chili-likers (frequent consumers) consistently gave lower intensity ratings and higher pleasantness ratings of the burn elicited by oral capsaicin. As an aside, female panelists gave higher sensory ratings than males, consistent with current data indicating that females are more sensitive to pain (Fillingim & Ness, 2000). Interestingly, the lower capsaicin concentrations (1, 2 ppm) were rated as hedonically neutral by non-likers and positive by likers, while the highest capsaicin concentration (4 ppm) was rated negatively by both groups. Furthermore, pleasantness ratings were higher for likers than non-likers even when compared at the same rated burn intensity. The data appear to better support a model in which novices are initially neutral toward the burning sensation evoked by a moderate level of spice.

Is mere exposure sufficient for the development of a preference for spicy food? This question was addressed by Stevenson and Yeomans (1995). They selected panelists who were infrequent consumers of spicy food yet were willing to experience it (i.e. they neither liked nor disliked it). At weekly intervals they consumed a two-part experimental meal consisting of an initial unspiced portion followed by an identical portion except that capsaicin (either 2.5 or 5 ppm) was added. There was a linear increase in the rated liking of the spiced (but not unspiced) food across weekly sessions. However, this effect did not transfer to the rated degree of liking of a capsicin-spiced tomato juice that was tested before and after the series of spicy meals. The results support the idea that mere exposure can contribute to the acquired liking of spicy food; factors such as novelty, rated burn intensity and arousal were considered to be unlikely contributors (Stevenson & Yeomans, 1995).

As discussed above, while there may be rare cases of liking upon initial exposure to spices, most novices are likely to judge the irritant sensation as affectively neutral or negative. Furthermore, mere exposure appears to be an important factor in acquisition of preference for spicy food. What other factors are likely to contribute to this acquired preference? A variety of adaptive, psychological and physiological factors have been suggested (Rozin, 1990). Potential adaptive qualities of spices include:

- promotion of cooling (by sweating) or salivation in dry climates
- nutritional (e.g. source of vitamins A, C)
- antimocrobial

The latter antimicrobial effect is supported by an interesting recent survey of cuisines in a variety of different cultures (Billing & Sherman, 1998).

Besides innate liking and exposure, additional psychological factors are likely to contribute to the acquisition of preference for spicy food. Reported desirable attributes of spicy food include "good taste", enjoyment of the burning or tingling sensation, and enhancement of the flavor of food (Rozin, 1990). In the latter case, "flavor enhancement" presumably means the overall impact including taste, smell and irritant sensations, since capsaicin actually reduces the intensity of certain taste qualities such as sweetness (Lawless & Stevens, 1988). With fizzy drinks, it is easy to imagine that the initial aversiveness and surprise could be compensated by curiosity and liking of the unusual sensation, particularly when it is recognized that the experience is tolerable. Attempts are currently underway to develop and market carbonated foods, such as fruits and vegetables, one rationale being that the carbonation makes the food more interesting for children.

Social reinforcement is a psychological factor that almost certainly contributes importantly to the acquisition of preference for spicy food in humans as well as in the rare cases with animals. Another uniquely human psychological factor may be the "enjoyment of constrained risks" (Rozin, 1990), whereby the "thrill" of the strong sensation of spicy food may be likened to the sensory and emotional experience associated with danger or perceived danger, such as a roller coaster ride. In interviews with college students, Rozin (1990) reports weak correlations between the liking of chili pepper and amusement rides ($r = 0.22$) or gambling ($r = 0.19$).

Finally, physiological factors possibly contributing to the development of preference for spices have been put forward. Numerous studies in recent years have consistently shown that frequent consumers of chili have a higher threshold for detection of capsaicin (or zingerone), and give lower intensity ratings, compared to infrequent consumers (Cowart, 1987; Lawless, Hartono, & Hernandez, 2000; Lawless, Rozin, & Schenker, 1985; Prescott & Stevenson, 1995, 1996a, 1996b; Rozin, 1990; Stevenson & Prescott, 1994; Stevenson & Yeomans, 1995). This might reflect a chronic state of desensitization due to regular consumption of high capsaicin levels by the chili likers. It might also reflect a context effect, in which the experimental capsaicin stimulus is much more intense than anything normally encountered by an infrequent user and thus rated highly, while it is given a lower rating by frequent consumers who often experience strong oral irritation. A reduced sensitivity to the

oral burning sensation of spices might diminish their innate aversiveness and contribute to increased consumption, but this does not account for why the consumer originally developed a preference for spicy food.

Conceivably, the oral burning sensation paired with meal consumption might lead to a learned association with the pleasant experience of satiety. Another possible case of associative learning is the pairing of the oral burn with a pleasant emotional state caused by release of endogenous opioids (endorphins) in the brain. This suggestion has merit in that irritant chemicals activate trigeminal pain pathways, which in turn could conceivably lead to endorphin release. This idea predicts that interfering with the action of endorphins, for example by pharmacologically blocking their effect with the opiate antagonist naloxone, should reduce the liking of spicy food. Rozin (1990) again reports tantalizing preliminary data that weakly support this notion. This idea is consistent with a role for endogenous opioids in general food palatability (for reviews, see: Cooper, Jackson, Kirkham, & Turkish, 1988; de Zwaan & Mitchell, 1992; Reid, 1985; Yeomans, 1998). It has been shown that opiate antagonists such as naloxone reduce hedonic ratings of certain foods in human studies (e.g. Yeomans & Gray, 1997; Yeomans & Wright, 1991) and are anorectic in animal studies (e.g. Yeomans & Clifton, 1997). In the case of nicotine, it is easy to understand how the initial aversiveness of irritation from cigarette smoke or chewing tobacco could be overcome by positive hedonic factors associated with addiction.

Ultimately, it is likely that many or all of the factors mentioned above contribute in some way to the development of preference for products containing irritant chemicals.

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References

Amano, N., Hu, J. W., & Sessle, B. J. (1986). Responses of neurons in feline trigeminal subnucleus caudalis (medullary dorsal horn) to cutaneous, intraoral and muscle afferent stimuli. *Journal of Neurophysiology*, *55*, 227–243.

Beverage Digest. (1998). 1997 retail value of the carbonated soft drink business in the US only: $54.7 billion. February 20:1.

Billing, J., & Sherman, P. W. (1998). Antimicrobial functions of spices: why some like it hot. *Quart. Rev. Biol.*, *73*, 3–49.

Bryant, B. P., & Moore, P. A. (1995). Factors affecting the sensitivity of the lingual trigeminal nerve to acids. *American Journal of Physiology*, *268*, R58–R65.

Carstens, E., Kuenzler, N., & Handwerker, H. O. (1998). Activation of neurons in rat trigeminal subnucleus caudalis by different irritant chemicals applied to oral or ocular mucosa. *Journal of Neurophysiology*, *80*, 465–492.

Carstens, E., Saxe, I., & Ralph, R. (1995). Brainstem neurons expressing c-Fos immunoreactivity following irritant chemical stimulation of the rat's tongue. *Neuroscience*, *69*, 939–953.

Carstens, E., Simons, C. T., Dessirier, J.-M., Iodi Carstens, M., & Jinks, S. L. (2000). Role of neuronal nicotinic acetylcholine receptors in the activation of neurons in trigeminal subnucleus caudalis by nicotine delivered to the oral mucosa. *Experimental Brain Research*, *132*, 375–383.

Caterina, M. J., Leffler, A., Malmberg, A. B., Martin, W. J., Trafton, J., Petersen-Zeitz, K. R., Koltzenburg, M., Basbaum, A. I., & Julius, D. (2000). Impaired nociception and pain sensation in mice lacking the capsaicin receptor. *Science*, *288*, 306–313.

Caterina, M. J., Schmacher, M. A., Tominage, M., Rosen, T. A., Levine, J. D., & Julius, D. (1997). The capsaicin receptor: a heat-activated ion channel in the pain pathway. *Nature*, *389*, 816–824.

Cliff, M. A., & Green, B. G. (1994). Sensory irritation and coolness produced by menthol: evidence for selective desensitization of irritation. *Physiology and Behavior*, *56*, 1021–1029.

Cliff, M. A., & Green, B. G. (1996). Sensitization and desensitization to capsaicin and menthol in the oral cavity—interactions and individual differences. *Physiology and Behavior*, *59*, 487–494.

Cooper, S. J., Jackson, A., Kirkham, T. C., & Turkish, S. (1988). Endorphins, opiates and food intake. In R. J. Rodgers, & S. J. Cooper (Eds.), *Endorphins, opiates and behavioral processes* (pp. 143–186). John Wiley: Chichester.

Cordero-Erausquin, M., Marubio, L. M., Klink, R., & Changeaux, J.-P. (2000). Nicotinic receptor function: new perspectives from knockout mice. *Trends in Pharmacological Sciences*, *21*, 211–217.

Cowart, B. J. (1987). Oral chemical irritation: does it reduce perceived taste intensity? *Chemical Senses*, *12*, 467–479.

de Zwaan, M., & Mitchell, J. E. (1992). Opiate antagonists and eating behavior in humans: a review. *Journal of Clinical Pharmacology*, *32*, 1060–1072.

Dessirier, J.-M., Chang, H.-K., O'Mahony, M., & Carstens, E. (2000). Cross-desensitization of capsaicin-evoked oral irritation by high but not low concentrations of nicotine in human subjects. *Neuroscience Letters*, *290*, 133–136.

Dessirier, J.-M., Nguyen, N., O'Mahony, M., Sieffermann, J.-M., & Carstens, E. (1999). Oral irritant properties of piperine and nicotine: psychophysical evidence for asymmetrical desenstization effects. *Chemical Senses*, *24*(N4), 405–413.

Dessirier, J.-M., O'Mahony, M., & Carstens, E. (1997). Oral irritant effects of nicotine: psychophysical evidence for decreased sensation following repeated application and lack of cross-desensitization with capsaicin. *Chemical Senses*, *22*, 483–492.

Dessirier, J.-M., O'Mahony, M., & Carstens, E. (2001). Oral irritant properties of menthol: sensitizing and desensitizing effects of repeated application and cross-desensitization to nicotine. *Physiology and Behavior*, *73*(1–2), 25–36.

Dessirier, J.-M., O'Mahony, M., Iodi-Carstens, M., & Carstens, E. (2000). Sensory properties of citric acid: psychophysical evidence for sensitization, self-desensitization, cross-desensitization and cross-stimulus-induced recovery following capsaicin. *Chemical Senses*, *25*, 769–780.

Dessirier, J.-M., O'Mahony, M., Iodi Carstens, M., Yao, E., & Carstens, E. (2001). Oral irritation by sodium chloride: sensitization, self-desensitization and cross-sensitization to capsaicin. *Physiology and Behavior*, *72*, 317–324.

Dessirier, J.-M., O'Mahony, M., Sieffermann, J.-M., & Carstens, E. (1998). Mecamylamine inhibits nicotine but not capsaicin irritation on the tongue: psychophysical evidence that nicotine and capsaicin activate separate molecular receptors. *Neuroscience Letters*, *240*, 65–68.

Dessirier, J.-M., Simons, C. T., O'Mahony, M., & Carstens, E. (2000). Psychophysical and neurobiological evidence that oral irritation from carbonated water is of chemogenic origin. *Chemical Senses*, *25*, 277–284.

Dessirier, J.-M., Simons, C.T., O'Mahony, M. and Carstens, E. (2001) The oral sensation of carbonated water: cross-desensitization by capsaicin and potentiation by amiloride. *Chemical Senses*, 26, 639–643.

Dessirier, J.-M., Simons, C. T., Sudo, M., Sudo, S., & Carstens, E. (2000). Sensitization, desensitization and stimulus-induced recovery (SIR) of responses of rat trigeminal caudalis neurons to repeated oral application of capsaicin and nicotine. *Journal of Neurophysiology*, *84*, 1851–1862.

Dib, B. (1990). After 2 weeks habituation to capsaicinized food, rats prefer this to plain food. *Pharmacology Biochemistry and Behavior*, *37*(4), 649–653.

Fillingim, R. B., & Ness, T. J. (2000). Sex-related hormonal influences on pain and analgesic responses. *Neuroscience and Biobehavioral Reviews*, *24*, 485–501.

Galef, B. G. Jr. (1989). Enduring social enhancement of rats' preferences for the palatable and the piquant. *Appetite*, *13*(2), 81–92.

Gilmore, M. M., & Green, B. G. (1993). Sensory irritation and taste produced by NaCl and citric acid: effects of capsaicin desensitization. *Chemical Senses*, *18*, 257–272.

Graber, M., & Kelleher, S. (1988). Side effects of acetazolamide: the champagne blues. *American Journal of Medicine*, *84*, 979–980.

Green, B. G. (1988). Spatial and temporal factors in the perception of ethanol irritation on the tongue. *Perception Psychophysics*, *44*, 108–116.

Green, B. G. (1989). Capsaicin sensitization and desensitization on the tongue produced by brief exposures to a low concentration. *Neuroscience Letters*, *107*, 173–178.

Green, B. G. (1991). Capsaicin cross-desensitization on the tongue: psychophysical evidence that oral chemical irritation is mediated by more than one sensory pathway. *Chemical Senses*, *16*, 675–689.

Green, B. G. (1992). The effects of temperature and concentration on the perceived intensity and quality of carbonation. *Chemical Senses*, *17*, 435–450.

Green, B. G. (1996). Rapid recovery from capsaicin desensitization during recurrent stimulation. *Pain*, *68*, 245–253.

Green, B. G., & Gelhard, B. (1989). Salt as an oral irritant. *Chemical Senses*, *14*, 259–271.

Green, B. G., & Lawless, H. T. (1991). The psychophysics of somatosensory chemoreception in the nose and mouth. In T. V. Getchell, R. L. Doty, L. M. Bartoshuk, & J. B. Snow (Eds.), *Smell and taste in health and disease* (pp. 235–253). New York: Raven.

Green, B. G., Mason, J. R., & Kare, M. R. (1990). *Chemical senses, irritation,* (Vol. 2). New York: Dekker.

Green, B. G., & McAuliffe, B. L. (2000). Menthol desensitization of capsaicin irritation: evidence of a short-term anti-nociceptive effect. *Physiology and Behavior*, *68*, 631–639.

Gotti, C., Fornasari, D., & Clementi, F. (1997). Human neuronal nicotinic receptors. *Progress in Neurobiology*, *53*, 199–237.

Harris, J. A. (1998). Using c-fos as a neural marker of pain. *Brain Research Bulletin*, *45*, 1–8.

Hellekant, G. (1965). The effect of ethyl alcohol on non-gustatory receptors of the tongue. *Acta Physiologica Scandinavica*, *65*, 243–250.

Heubner, W. (1925). Zur Pharmakologie der Reizstoffe. *Arch Exp. Path. Pharmak.*, *107*, 129–154.

Hilker, D. M., Hee, J., Higashi, J., Ikehara, S., & Paulsen, E. (1967). Free choice consumption of spiced diets by rats. *Journal of Nutrition*, *91*(1), 129–131.

Hunt, S. P., Pini, A., & Evan, G. (1987). Induction of c-fos-like protein in spinal cord neurons following sensory stimulation. *Nature*, *328*, 632–634.

Karrer, T., & Bartoshuk, L. (1991). Capsaicin desensitization and recovery on the human tongue. *Physiology and Behavior*, *49*, 757–764.

Keele, C. A., & Armstrong, D. (1964). *Substances producing pain and itch*. London: Arnold.

Komai, M., & Bryant, B. P. (1993). Acetazolamide specifically inhibits lingual trigeminal nerve responses to carbon dioxide. *Brain Research*, *612*, 122–129.

Lang, F., Busch, G. L., Ritter, M., Volkl, H., Waldegger, S., Gulbins, E., & Haussinger, D. (1998). Functional significance of cell volume regulatory mechanisms. *Pharmacology Reviews*, *78*(1), 247–306.

Lawless, H. T., Hartono, C., & Hernandez, S. (2000). Thresholds and suprathreshold intensity functions for capsaicin in oil and aqueous based carriers. *Journal of Sensory Studies*, *15*, 437–447.

Lawless, H. T., Rozin, P., & Schenker, J. (1985). Differences in perception of capsaicin between people who frequently consume and like chili pepper and others who rarely consume and dislike it. *Chemical Senses*, *10*, 579–589.

Lawless, H. T., & Stevens, D. A. (1988). Responses by humans to oral chemical irritants as a function of locus of stimulation. *Perception Psychophysics*, *43*, 72–78.

Liu, L., Oortgiesen, M., Li, L., & Simon, S. A. (2001). Capsaicin Inhibits Activation of Voltage-Gated Sodium Currents in Capsaicin-Sensitive Trigeminal Ganglion Neurons. *Journal of Neurophysiology*, *85*, 745–758.

Liu, L., & Simon, S. A. (1996a). Capsaicin and nicotine both activate a subset of rat trigeminal ganglion neurons. *American Journal of Physiology*, *270*, C1807–C1814.

Liu, L., & Simon, S. A. (1996b). Capsaicin-induced currents with distinct desensitization and Ca^{2+} dependence in rat trigeminal ganglion neurons. *Journal of Neurophysiology*, *75*, 1503–1514.

Liu, L., & Simon, S. A. (2000). Capsaicin, acid and heat-evoked currents in rat trigeminal ganglion neurons: relationship to functional VR1 receptors. *Physiology and Behavior*, *69*, 363–378.

Madshus, I. H. (1988). Regulation of intracellular pH in eukaryotic cells. *Biochemical Journal*, *250*(1), 1–8.

Parker, G. H. (1912). The relation of smell, taste and the common chemical sense in vertebrates. *J. Acad. Natl. Sci. Ser. 2, 15*, 221–234.

Paterson, D., & Nordberg, A. (2000). Neuronal nicotinic receptors in the human brain. *Progress in Neurobiology*, *61*, 75–111.

Prescott, J. (1999). The generalizability of capsaicin sensitization and desensitization. *Physiology and Behavior*, *66*(5), 741–749.

Prescott, J., & Stevenson, R. J. (1995). Effects of oral chemical irritation on tastes and flavors in frequent and infrequent users of chili. *Physiology and Behavior*, *58*, 1117–1127.

Prescott, J., & Stevenson, R. J. (1996a). Desensitization to oral zingerone irritation: effects of stimulus parameters. *Physiology and Behavior*, *60*, 1473–1480.

Prescott, J., & Stevenson, R. J. (1996b). Psychophysical responses to single and multiple presentations of the oral irritant zingerone: relationship to frequency of chili consumption. *Physiology and Behavior*, *60*, 617–624.

Prescott, J., & Swain-Campbell, N. (2000). Responses to repeated oral irritation by capsaicin, cinnamaldehyde and ethanol in PROP tasters and non-tasters. *Chemical Senses*, *25*(3), 239–246.

Reid, L. D. (1985). Endogenous opioid peptides and regulation of feeding and drinking. *American Journal of Clinical Nutrition*, *42*, 1099–1132.

Rozin, P. (1990). Getting to like the burn of chili pepper.. Biological, psychological and cultural perspectives. In B. G. Green, J. R. Mason, & M. R. Kare (Eds.), *Chemical senses, irritation,* (Vol. 2) (pp. 231–269). New York: Dekker.

Rozin, P., Gruss, L., & Berk, G. (1979). Reversal of innate aversions: attempts to induce a preference for chili peppers in rats. *J. Comp. Physiol. Psychol.*, *93*(6), 1001–1014.

Rozin, P., & Kennel, K. (1983). Acquired preferences for piquant foods by chimpanzees. *Appetite*, *4*(2), 69–77.

Rozin, P., & Schiller, D. (1980). The nature and acquisition of a preference for chili pepper by humans. *Motivat. Emotion*, *4*, 77–101.

Schults, R. C. (1992a). Nociceptive neural organization in the trigeminal nuclei. In A. R. Light (Ed.), *The initial processing of pain and its descending control: spinal and trigeminal systems* (pp. 178–202). New York: Karger.

Schults, R. C. (1992b). Trigeminal primary afferent receptors. In A. R. Light (Ed.), *The initial processing of pain and its descending control: spinal and trigeminal systems* (pp. 50–74). New York: Karger.

Simons, C. T., Dessirier, J.-M., Iodi Carstens, M., O'Mahony, M., & Carstens, E. (1999). Neurobiological and psychophysical mechanisms underlying the oral sensation produced by carbonated water. *Journal of Neuroscience*, *19*(N18), 8134–8144.

Simons, C. T., Dessirier, J.-M., Jinks, S. L., & Carstens, E. (2001). An animal model to assess aversion to intra-oral capsaicin: increased threshold in mice lacking substance P. *Chemical Senses*, *26*, 491–497.

Steen, K. H., Wegner, H., & Reeh, P. W. (1999). The pH response of rat cutaneous nociceptors correlates with extracellular [Na+] and is increased under amiloride. *European Journal of Neuroscience*, *11*(8), 2783–2792.

Stevens, D. A., & Lawless, H. T. (1987). Enhancement of responses to sequential presentation of oral chemical irritants. *Physiology and Behavior*, *39*, 63–65.

Stevenson, R. J., & Prescott, J. (1994). The effects of prior experience with capsaicin on ratings of its burn. *Chemical Senses*, *19*, 651–656.

Stevenson, R. J., & Yeomans, M. R. (1995). Does exposure enhance liking for the chilli burn? *Appetite*, 107–120.

Su, X., Wachtel, R. E., & Gebhart, G. F. (1999). Capsaicin sensitivity and voltage-gated sodium currents in colon sensory neurons from rat dorsal root ganglia. *American Journal of Physiology—Gastrointestinal and Liver Physiology*, *277*, G1180–G1188.

Waldmann, R., Champigny, G., Bassilana, F., Heurteaux, C., & Lazdunski, M. (1997). A proton-gated cation channel involved in acid-sensing. *Nature*, *386*, 173–177.

Waldmann, R., Champigny, G., Lingueglia, E., De Weille, J. R., Heurteaux, C., & Lazdunski, M. (1999). H(+)-gated cation channels. *Annals of the New York Academy of Scence*, *868*, 67–76.

Waldmann, R., & Lazdunski, M. (1998). H(+)-gated cation channels: neuronal acid sensors in the NaC/DEG family of ion channels. *Current Opinion in Neurobiology*, *8*, 418–424.

Wang, Y., Erickson, R. P., & Simon, S. A. (1993). Selectivity of lingual nerve fibers to chemical stimuli. *J. Gen. Physiol.*, *101*, 843–866.

Yeomans, M. R. (1998). Taste, palatability and the control of appetite. *Proc. Nutrition Soc.*, *57*, 609–615.

Yeomans, M. R., & Clifton, P. G. (1997). Exposure to sweetened solutions enhances the anorectic effect of naloxone but not d-fenfluramine. *Physiology and Behavior*, *62*, 255–262.

Yeomans, M. R., & Gray, R. W. (1997). Effects of naltrexone on food intake and changes in subjective appetite during eating: evidence for opioid involvement in the appetizer effect. *Physiology and Behavior*, *62*, 15–21.

Yeomans, M. R., & Wright, P. (1991). Lower pleasantness of palatable foods in nalmefene-treated human volunteers. *Appetite*, *16*, 249–259.