The highlight for April is by Dr. Federico Bermudez-Rattoni from the Institute of Cellular Physiology, National University of Mexico in Mexico City, Mexico. Dr. Bermudez-Rattoni’s work over the past 25 years has addressed the neuroanatomical and neurochemical mediation of taste aversion learning. Following his initial work with John Garcia on sensory gating in aversion learning in which he demonstrated a role for the basolateral amygdala in the gating of odors by tastes, he and his lab began exploring the neural mediation and integration of taste memory in aversion learning. Based on an impressive combination of behavioral, physiological, neurochemical and electrophysiological assessments, Dr. Bermudez-Rattoni and his colleagues have demonstrated that novel tastes cause the release of acetylcholine (in the insular cortex) that in turn establishes the memory trace of the gustatory event. Pre-training cholinergic deafferentation of the insular cortex disrupts aversion learning, indicating a role of ACH in its acquisition. When the animal is subsequently injected with an aversion-inducing agent, e.g., LiCl, glutamate is released (in the amygdala and insular cortex) that may be responsible for the formation of the memory for the aversive event. Through sophisticated work combining neurochemical and electrical activity in the amygdala and insular cortex, e.g., long-term potentiation induction and microinjections of glutamate, Dr. Bermudez-Rattoni demonstrated a convergence of activity in these two neural substrates (via acetylcholine and glutamate release) that results in the consolidation of taste aversion memory formation. Dr. Bermudez-Rattoni’s work highlights an impressive integration of neuroscience techniques and approaches that illustrates the importance of a broad-based investigation into any area of interest. His work on taste aversion learning reveals the relative complexity of what appears to be a simple behavioral issue, i.e., the avoidance of a previously-poisoned solution. This integration and its importance are highlighted in this month’s summary.

**Insular Cortex-Amygdala Dialogue During Taste Recognition Memory Formation**

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Imagine having a very good meal or a good glass of wine, suddenly realizing that it is the best you ever had. To reach that conclusion, you should have had a lot of experience eating and drinking a variety of meals and wines in your life. Clearly, our ability to judge food and drinking in this way is rather sophisticated. In this regard, the *Wall Street Journal* recently reported a large scale smuggling of Coke bottles from Mexico to the US.¹ A possible reason for that smuggling is that many people prefer the flavor of Mexican Coke to its US counterpart. Similar situations occur in nature in which it is not pleasure at stake, but survival. Taste recognition memory is the ability to identify a taste and relate it to past
consequences of its ingestion, an important survival skill that animals have developed throughout evolution\(^2\).

I have been involved in taste recognition memory for about 26 years, since I met John Garcia when he received the *Distinguish Scientific Contribution Award* from the American Psychological Association in 1979. Later, I continued my doctoral studies under his supervision at the University of California, Los Angeles. I remember that during my first interview with him, he proposed to me to find a brain “and-gate” that switched odors into aversive cues when they were presented together with tastes during taste aversion learning. Taste is an excellent conditioning stimulus for delayed illness, but odor is not. However, when odor and taste are combined into a compound stimulus and is followed by delayed illness, the odor becomes as aversive as taste when each is tested separately in extinction trials. Under these conditions odor aversions appear to be potentiated by taste, as if a sensory *and-gate* controlled by taste entered odors into food-related memory mechanisms\(^3-5\). In a way, we succeeded in finding the *and-gate* in the basolateral amygdala (BLA), since the BLA, but not other brain areas, was responsible for switching odors into aversive cues when they were presented together with taste\(^6-9\).

These results have been corroborated by a number of laboratories around the world\(^10-15\), and, furthermore, it was demonstrated that N-methyl-D-aspartate (NMDA) receptors in the BLA might play an important role in taste-potentiated odor aversions. That is, BLA injections of the NMDA receptor antagonist, d-2-amino-5-phosphonovalerate (APV), selectively blocked taste-potentiated odor aversions\(^11,13,15\). It has also been found that depletion of dopaminergic terminals in the BLA produce a strong disruption of taste-potentiated odor aversions but leaves taste aversions unaltered\(^16,17\). Therefore, it seems that the role of the amygdala in toxiphobia learning could be in the association of taste cues with visceral consequences and enabling potentiated odor conditioning by taste aversion learning. Later, I continued investigating the neural integration of taste memory. Together with students and colleagues, we were able to determine the cholinergic activity of the insular cortex (IC) and BLA during conditioned taste aversion (CTA) learning. Many of the studies at the time assessed the effects of different drugs on CTA by giving them before or after the presentation of the conditioned and the unconditioned stimuli (CS-US) pairing. Instead, we determined acetylcholine (ACh) release by using *in vivo* microdialysis to a CS separate from the US. Thus, we demonstrated that novel taste produced a strong release of ACh in the IC, whereas after several presentations of the stimulus the cortical ACh release decreased to the same levels as those produced by the familiar ones, indicating an inverse relationship between familiarity and cortical ACh release\(^18,19\). Similarly, specific cholinergic deafferentation restricted to the cortex produced a significant disruption in the acquisition of CTA. However, the same animals were able to recall the taste aversion when the conditioning trial was established before deafferentation\(^20,21\). These results suggest that cortical
cholinergic activity is involved in the recognition of taste novelty, but it is no longer necessary to recall aversive stimuli.

From these results, we were interested in determining if ACh activity was involved in signaling CS novelty. Although, it was known that cortical administration of scopolamine, a muscarinic receptor antagonist, blocked CTA, it remained unknown if that blockade would interfere with the taste or novelty perception. To better understand the role of ACh activity on gustatory memory trace, we used a learning protocol described in the 70s. When an animal drinks a novel taste solution, it innately hesitates to drink it, reducing its consumption (neophobia) until its post-digestive consequence has been assessed. Unlike CTA, when taste is followed by absence of malaise, an incremental increase in its consumption is observed, which is called attenuation of neophobia. Therefore, we assessed the effects of muscarinic receptor antagonists by giving microinjections of scopolamine or the NMDA-receptor antagonist APV into the IC on neophobia and attenuation of neophobia. The neophobic response was induced by presentation of a strong novel saccharin solution (0.5%) that was not affected by the injection of either drug given before the taste presentation, ruling out any impairment in taste perception or novelty discrimination. However, attenuation of neophobia was impaired by microinjection of scopolamine given before or after the first taste experience. However, APV did not affect the attenuation of neophobia, suggesting that this learning is independent of NMDA receptor activity. These results indicate that ACh activity is involved in the formation of the gustatory trace and that post-digestive consequences will produce safe (attenuation of neophobia) or aversive (CTA) gustatory memory formation.

Thus, we were interested in how a dialogue is established between the BLA and the IC during taste aversion memory formation. In this regard, we demonstrated that there is a functional communication between the BLA and IC by means of induced Long-Term Potentiation (LTP). We were able to demonstrate a significant increase of synaptic responses in the IC to low frequency stimulation after in vivo tetanic stimulation of the BLA in adult rats and that intracortical administration of APV disrupted the inducted LTP. In addition, induction of LTP in the BLA-IC projection before CTA training enhanced the retention of this task. These results point to a cortical functional facilitation induced by BLA stimulation that seems to be regulated by glutamate (Glu) through activation of NMDA receptors. In conclusion, the induced LTP in the BLA-IC projection is a possible mechanism for memory-related functions performed by the cortex. Thus, we decided to further investigate the possible role of Glu and if the US and the CS could differentially activate it. We demonstrated by in vivo microdialysis that the US (i.p. injection of LiCl), but not the saccharin presentation, induced a dramatic increase in Glu release in the BLA and a modest but significant release in the
IC\textsuperscript{29}. In addition, reliable and robust taste aversions could be elicited by intra-amygdala microinjections of Glu. That is, when Glu is administered in the BLA just before the presentation of a weak US (low LiCl dose), a clear CTA is produced\textsuperscript{29,30}. Furthermore, blockade of cortical NMDA receptors by APV applied before the novel taste impaired the induced taste aversions by the combined Glu injections and low doses of LiCl in the BLA\textsuperscript{30}. Altogether, these results suggest that cholinergic activity is involved in the taste memory trace, whereas glutamatergic activity participates in aversive memory formation. In addition, they indicate that Glu has the function of signaling at least partly the visceral input (US) that would eventually converge with the CS signal during consolidation of taste aversion memory formation. Altogether, these experiments demonstrate that behavioral enhancement of taste aversion could be induced by electrical (high frequency stimulation, LTP) or pharmacological (applications of Glu) stimulation of the BLA. The facilitations of CTA induced by BLA-LTP or by BLA-Glu infusions can be reversed by cortical infusions of NMDA receptor antagonists.

In summary, we are proposing that there are at least two forms of taste memory, i.e., safe and aversive, and that an active functional communication between the BLA and IC is established during taste memory formation. Thus, safe taste memory seems to involve cholinergic neurotransmission and its downstream signaling pathways. However, if a taste cue is followed by induction of visceral malaise it produces a release of Glu in the BLA and in the IC activating the NMDA receptors. If the US signal activation takes place during the few hours following consumption of the new taste, it interferes with the safe taste consolidation transforming the taste representation from "safe" to "aversive" trace and produces a long-lasting aversive memory trace\textsuperscript{31}.

References


