

shading indicative of the underlying molecular substructure. The stereo-pair, black-and-white image of the virus (Fig. 2b) was generated in the computer, taking the same direction of view as in Fig. 2a, by adding together separate projections of the low-resolution subunit model related by the screw symmetry of the virus helix (10). The RNA chain coiling over the top turn of the helix of protein subunits (Fig. 2b) was imaged at about a 4-Å resolution. Bumps on this chain, oriented sequentially (one extending out radially and two extending up), represent the three nucleotides associated with each protein subunit. The interlocked arrangement of the protein subunits and RNA chain (Fig. 2b) can be related to more detailed images of the parts to explain the higher levels of organization in terms of the specific molecular interactions.

We have aimed for a display that keeps the scale of detail in proportion to the overall dimensions of the image: in the distant view, only essential outlines of the parts are conserved; by zooming into the structure, the molecular detail can be augmented as the scale is magnified. This goal could not be realized by constructing an atomic model of the entire virus assembly and then displaying it at low resolution, since this would blur the subunit boundaries. Controlled reduction of image information will facilitate the display and interpretation of the structures of complex macromolecular assemblies.

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8. The graphics programs are written in Fortran and can be used to display structures of any type for which three-dimensional density maps can be calculated (for example, from atomic model coordinates or from x-ray diffraction, electron microscope, or optical data).
9. The Fortran graphics programs use the library subroutines supplied with the AED512 terminal to transfer the image from the core to the raster graphics screen. The programs could easily be adapted for any raster graphics terminal with corresponding subroutines. An integer between 0 and 255 is assigned to each pixel for the screen of the AED512 terminal, and each integer corresponds to a particular brightness and color taken from a table. For example, the color table could include 10 different colors (combinations of red, green, and blue) each with 25 levels of brightness, or 16 colors with 16 levels of brightness, and so on. For black and white images, 256 levels of brightness are available.
10. A two-dimensional image of each subunit in a six-turn helix segment was constructed in the computer. Because there are 49 symmetrically related subunits in the three-turn repeat of the helix, there are 49 different subunit orientations, some of which are hidden in the combined projected image. The individual subunit images, related by the helical symmetry, were combined by comparing *z* coordinates of corresponding pixels in the overlapping images to remove all hidden parts (that is, at each sampled point only the image pixel with the smallest *z* coordinate was displayed). The subunit boundaries are marked clearly in the virus image since it is built by adding together the images of the separate, and sharply outlined, subunits.
11. The definition of directions in the subunit and the terminology for the four major α -helical segments follows the nomenclature adopted to describe the protein structure in the two-turn disk aggregate [J. N. Champness, A. C. Bloomer, G. Bricogne, P. J. G. Butler, A. Klug, *Nature (London)* **259**, 20 (1976)]. The ordered part of the protein in the disk aggregate corresponds to the subunit structure in the virus helix at radii greater than 40 Å.
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13. We thank D. Hellerstein for help with the computer graphics programming. Supported by U.S. Public Health Service grants GM 25236 and GM 33265 (G.J.S.) and CA 15468 (D.L.D.C.). The VAX 11/780 computer was provided from Shared Instrument grant GM 21189-09S1 awarded to D. J. DeRosier.

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6 August 1984; accepted 23 October 1984

Two Gustatory Systems: Facial and Vagal Gustatory Nuclei Have Different Brainstem Connections

Abstract. *The gustatory sense in catfish consists of two dissociable components, a facial nerve system used for food selection and a vagal nerve system involved in swallowing. Neural tracing experiments demonstrate that the primary sensory nucleus for the facial gustatory system is connected to the reticular formation and trigeminal nuclei. In contrast, the primary sensory nucleus for the vagal gustatory system is connected to the motoneurons that mediate swallowing. These results provide anatomical evidence for parallel gustatory systems within the vertebrate central nervous system.*

Of all the sensory systems, the gustatory sense occupies a unique position at the portal to the alimentary canal. The sense of taste, however, is not limited to reflexive control of swallowing (1, 2), but plays an important role in food selection and determination of palatability. The location of taste buds within the oral cavity appears related to their function; taste buds located most externally are implicated in food selection and appreciation, while taste buds lying closest to the alimentary canal are concerned with ingestive and protective reflexes (2, 3). This dichotomy of function parallels the pattern of innervation of the different groups of taste buds. Taste buds lying closest to the esophagus—for example, on the palate and larynx—are innervated by branches of the vagus nerve; those lying most externally—for example, on the anterior part of the tongue or across the lips and barbels of certain fishes—are innervated by the facial nerve (4, 5). The glossopharyngeal nerve innervates taste buds lying between these extremes.

In catfish, the distinction between the facial and vagal gustatory systems is especially clear. Each nerve terminates

in a separate lobe protruding from the dorsal surface of the rostral medulla. Electrophysiological experiments show that the facial and vagal gustatory lobes receive similar chemosensory input from their respective gustatory fields (6). Nonetheless, selective lesion experiments demonstrate a behavioral dissociation of function for the facial and vagal gustatory lobes (2). A bilateral lesion of the facial lobe results in an animal that is unable to locate food pellets in the surrounding water but that will swallow food placed in its mouth. In contrast, a bilateral lesion of the vagal lobe produces an animal that can locate food in its environment and take it into its mouth, but appears unable to initiate swallowing and spits out any food that has accumulated in its mouth. The glossopharyngeal nerve seems to play a lesser role in gustatory-mediated behaviors in catfish. We have attempted to delineate the anatomical bases for the differences in behavior mediated by the facial and vagal gustatory subsystems.

Horseradish peroxidase (HRP) was used as an anterograde and retrograde tracer of neuronal connectivity in bull-

head catfish, *Ictalurus nebulosus* (7). In experiment 1, HRP was placed on the cut central ends of those branches of the vagus nerve that innervate the oropharyngeal muscles involved in swallowing. This experiment showed the location and morphology of the oropharyngeal motoneurons. In experiment 2, the HRP tracer was injected into either the facial or the vagal lobe to demonstrate the central connections of these primary gustatory nuclei.

The motoneurons that innervate the oropharyngeal musculature are located in a longitudinally oriented column situated just lateral to the midline at the floor of the fourth ventricle. Because these neurons are functionally and topologically homologous (8) to the nucleus ambiguus described in other vertebrates, they have been referred to by this name (9). The heavy retrograde filling of the nucleus ambiguus neurons shows that the dendrites extend ventrolaterally into the medullary tegmentum (Fig. 1A). The dendritic ramifications are densest near the lateral margin of the medulla, just ventral to the spinal trigeminal nucleus and tract.

Injections of HRP into the vagal lobe reveal a substantial projection from the vagal gustatory nucleus to the lateral medullary tegmentum exactly overlapping the distribution of the nucleus ambiguus dendrites (Fig. 1B) (10). There is virtually no output from the vagal lobe to the spinal trigeminal structures or to the medial reticular formation.

In contrast, the facial lobe does not project to the area of the nucleus ambiguus, but is connected to the medial reticular formation, funicular nuclei, and spinal trigeminal nuclei (Fig. 1C). These structures may play a role in spinal motor control and in correlation of somatosensory information with gustatory inputs from the face and barbels.

Thus, the facial and vagal gustatory inputs are processed in different structures within the central nervous system, which have different connections within the brainstem. The connections of the vagal gustatory lobe to the dendrites of the oropharyngeal motoneurons corresponds well to its role in initiating swallowing (Fig. 2). Electrophysiological studies show that despite the differences in connectivity of the facial and vagal gustatory systems, the two systems have largely similar chemical receptivities (6). Thus similarities in peripheral responsiveness do not imply similarities in central processing. Similar receptor responses may be utilized differently by different neural systems.

Some evidence exists that separate

facial and vagal gustatory subsystems may exist in mammals as well as in fish. Instead of the distinct gustatory lobes of some fish, the medullary gustatory nucleus in mammals and birds constitutes a portion of the nucleus solitarius complex. Hamilton and Norgren (5) reported that the gustatory nerves innervating the tongue project to the rostral portion of nucleus solitarius, whereas the nerves innervating taste buds on the palate and

pharynx terminate in the caudal portion of the same nucleus. The reported connections of the rostral and caudal portions of nucleus solitarius differ and may reflect underlying differences in connectivity of the facial and vagal gustatory subsystems in mammals. These results in mammals, however, are difficult to interpret since the general visceral portions of nucleus solitarius are immediately adjacent to the gustatory portions.

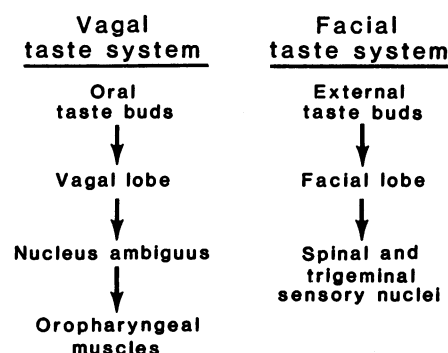
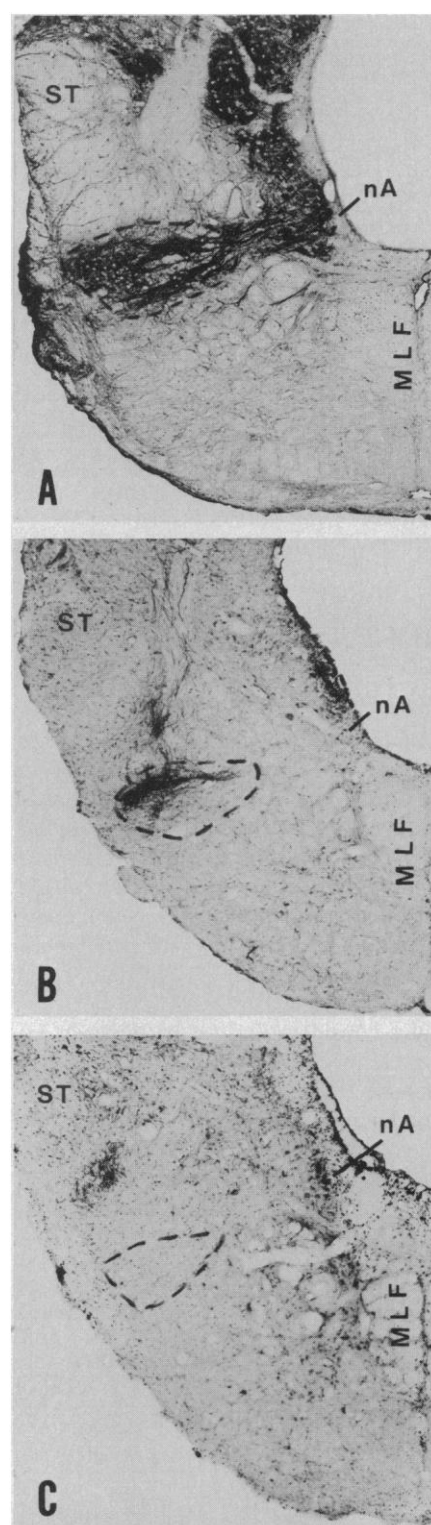


Fig. 1 (left). Photomicrographs of sections through the region of the nucleus ambiguus on the left side of the brainstem of three different experimental animals. All figures are approximately $\times 20$. (A) The oropharyngeal motoneurons are retrogradely filled by an injection of HRP into the vagus nerve. The dendrites of the motoneurons of nucleus ambiguus (nA) extend laterally into the area ventral of the spinal trigeminal tract and nucleus (ST). This area of dendritic ramification is outlined by dashes in parts (B) and (C). The medial longitudinal fasciculus (MLF) is also indicated for purposes of orientation. (B) The vagal lobe was injected with HRP to determine its output. Labeled axons can be followed from the injection site to their terminus in the area containing the lateral dendrites of the nucleus ambiguus neurons. (C) The facial lobe was injected with HRP. No terminal labeling occurred in the outlined region, which contains the lateral dendrites of the nucleus ambiguus. Labeled fibers and terminals can be found, however, in the spinal trigeminal nucleus (ST), which is located dorsal of the outlined area. Fig. 2 (right). Diagram highlighting the differences of reflex connectivity of the vagal and facial gustatory systems. The vagal, but not the facial, taste system has relatively direct access to those motoneurons involved in swallowing. This is related to the behavioral observation that an intact vagal taste system is necessary for the animal to initiate swallowing (2). Not illustrated are the ascending connections arising from each of the taste systems. These connections also differ for the vagal and facial taste systems. The facial system has direct connections to both pontine and thalamic taste nuclei, whereas the vagal system is directly connected only to the pontine area.

Differences in connections could be attributed to the differing general visceral functions of the nucleus. This source of confusion is absent in catfish, in which the general visceral nucleus is anatomically distinct and separated from the gustatory lobes.

The gustatory system of vertebrates, including perhaps mammals and humans, can thus be viewed as consisting of two (or more) subsystems that mediate different behaviors. Although differential parallel processing of inputs in other sensory modalities is well known (11), this is, to our knowledge, the first anatomical evidence for parallel subsystems of the gustatory sense.

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7. The HRP was prepared as a thick paste in 1 percent lysolecithin and applied to the nerves or superficial brain structures by means of repeated stabs with an HRP-coated insect pin. After surviving for 2 to 8 days, the animals were fixed by means of perfusion with 4 percent glutaraldehyde in 0.1M phosphate buffer of pH 7.2. The brains were removed from the skull, embedded in gelatin and fixed for 2 to 6 hours before being cryoprotected in buffer containing sucrose. After being sectioned on a freezing sliding microtome, the brain was processed for peroxidase activity according to a modified Hanks-Yates procedure (Y. Morita and T. E. Finger, *J. Comp. Neurol.*, in press).
8. In mammals, the nucleus ambiguus is a migrated cell group; that is, during embryogenesis the neuroblasts leave the proliferative zone at the ventricular surface and move radially outward into the developing medullary tegmentum. In frogs and some fishes, the nucleus ambiguus does not travel far from the ventricular surface, and so the cell bodies come to lie dorsomedial to the position of the same cell group in mammals.
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10. The vagal lobe is also interconnected with the commissural nucleus of Cajal, which is the general visceral sensory nucleus of the caudal medulla. In catfish, the general visceral sensory nucleus is entirely separate from the more rostrally placed gustatory nuclei. This contrasts sharply with the situation in the nucleus solitarius, the homologous structure in mammals and birds.
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14 September 1984; accepted 24 September 1984

Courtship Disruption Modifies Mate Choice in a Lek-Breeding Bird

Abstract. *The aggregation of displaying males in lek-breeding birds is often associated with disruption of courtship and mating but effects of this disruption on mate choice have not been shown. In a 4-year study of *Rupicola rupicola* in Suriname, interference among territorial adult males disrupted 31 percent of all female courtship visits and terminated 32 percent of all matings at a lek where there were an average of 55 territorial males. Disruption in this rain forest species caused females to modify their courtship and mating patterns, and males that used intense and persistent disruption received a disproportionate share of this redirected mate choice.*

Mate choice is a fundamental aspect of sexual selection and has emerged as a focus of evolutionary theory (1). Of particular interest is the degree to which members of the limiting sex (usually females) are able to control with whom they mate. The lek-mating system is gen-

erally considered to be particularly favorable for the operation of mate choice because females of lek species receive no resources from males, initiate mating by solicitation, and are not subject to forced copulation (2).

The one direct means of thwarting

female choice that is available to the males of lek species is courtship disruption. This can be defined as any interference in courtship or mating that decreases the number of complete copulations performed by a male or that increases the time and energy required to perform them (3). Interruption of mating by competing males has been reported for all well-studied lek species, at levels ranging from 2 to 68 percent (4). However, because no direct benefits resulting from this behavior have been shown, the adaptive significance of courtship disruption is controversial (2, 3).

I present evidence that courtship disruption in a lek-breeding bird, the Guianan cock of the rock (Cotingidae: *Rupicola rupicola*), can provide direct benefits to disrupters by modifying female choice. These data were collected during a 4-year study of a color-banded population of *Rupicola* at the Raleigh Falls-Voltzberg Nature Reserve in Suriname, South America. The study lek, located in dense rain forest, had an average of 55 territorial males during the 1980 through 1983 breeding seasons (range, 51 to 61 males). Each *Rupicola* display territory is defended by a single male and consists of a cleared ground court (approximately 1 m in diameter) and the surrounding perches, to a height of 1 to 2 m above the court. The nearest-neighbor distance between court edges on this lek averaged 1.4 ± 1.26 m (mean $\pm$ standard deviation; $n = 96$), providing many opportunities for males to observe and disrupt each other's courtship.

Females come to the lek for courtship and mating, typically visiting the territories of a number of males over several days before copulating one or two times. Such a period is called a mating bout. Females initially observe the males from perches in the subcanopy and then enter individual display territories for courtship interactions (5). I limit the term "courtship disruption" to harassment of females actually visiting a male's territory.

During the study period 385 copulations and 3169 courtship visits by females to male territories were observed. As in other lek species, male *Rupicola* mating success is highly skewed, with the most successful male performing an average of 30 percent of all matings per year (range, 23 to 39 percent), and 67 percent of territorial males failing to mate at all each year (5). On average, adult males disrupted 31 percent of courtship visits (range, 28 to 36 percent) and 32 percent of matings (range, 21 to 41 percent) annually. Approximately half the territorial males, representing the full

Table 1. Courtship and mating of female *Rupicola* in relation to disruption during the first copulation of the mating bout. Disruption significantly increased the amount of courtship, number of males sampled, and degree of polygamy (all comparisons at $P < 0.05$ level, arcsin test for degree of polygamy; one-way analysis of variance for other measures). Means and standard deviations are shown.

First mating	Mating bouts	Courtship visits	Males visited	Polygamous females (%)
Disrupted	17	15.3 ± 10.74	4.2 ± 3.32	47
Complete	55	6.7 ± 5.82	2.8 ± 1.58	18