less and require less than 50 μl of serum. It is expected that cell lines expressing the surface antigens of infectious agents can be configured into simple chlorometric tests that could be performed in a physician's office or a clinical laboratory. In principle, the strategy we have described could be applied to any situation where the expression of a membrane protein is desired.

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Electrical Brain Stimulation and the Localization of Cardiopulmonary Function

Eldridge *et al.* (*1*) found that electrical stimulation of a subthalamic locus could induce locomotion and augment respiration simultaneously. This finding does not support their conclusion that: "Hypothalamic command signals are thus primarily responsible for the proportional driving of locomotion and respiration during exercise." Two erroneous assumptions were made in drawing this conclusion. The first is that the responses elicited by focal stimulation could be assumed to be generated by the region stimulated. The second is that the stimulus-induced hyperpnea was functionally related to the locomotion because both responses could be evoked from the same site.

The subthalamic locomotor region is located within a large, ill-defined area, encompassing much of the hypothalamus, medial thalamus, and brainstem reticular core, all of which can give pressor responses (*2*, *3*) and hyperpnea (*3–5*) when stimulated. Cohen and Hugelin (*5*), among others, found that ventilation rate increased in proportion to the strength of electrical stimulation at many widespread sites in this area. It thus seems unlikely that the hyperpnea can be ascribed specifically to stimulation of the subthalamic locomotor region.

The second issue is whether a causal or functional relation between evoked hyperpnea and evoked locomotion can be assumed. One or both responses could have arisen from stimulation of fibers of passage, from an overlapping spatial distribution of structures subserving locomotion with those subserving cardiac and respiratory functions (*6*), or from current spread to adjacent structures (*7*). For instance, urination can be evoked in conjunction with locomotion by brainstem stimulation (*8*), even though these two responses are not functionally related. A functional relation may be suggested under the circumstances if, regardless of stimulus strength and location, the locomotion

and hyperpnea could not be dissociated. However, hyperpnea was induced in the complete absence of locomotion in the experiment of Eldridge *et al.*, leaving the question of the physiological role of the evoked hyperpnea entirely open.

By way of illustration, during the course of experiments in which electrical stimulation of the mesencephalic reticular formation was sometimes used (*9*), I obtained the usual pressor response, pupillary dilation, and neocortical desynchronization during the passage of current. In addition to these arousal-related responses, eye movements were elicited when the stimulating electrodes passed near the oculomotor nucleus. The intensity of the eye movements and pressor response was often proportional to stimulus intensity. In this case, it was unlikely that the pressor response was part of a feed-forward mechanism that anticipated the rise in oxygen consumption from activity in the extrinsic muscles of the eye. However, Eldridge *et al.* made this exact argument for an analogous finding obtained during electrical stimulation of the subthalamic locomotor region.

Eldridge *et al.* mentioned that one decorticate paralyzed animal had spontaneous intermittent locomotion and an associated hyperpnea. In contrast to the findings on evoked hyperpnea, this observation does support the idea that locomotion-related hyperpnea is not due to peripheral feedback. However, this was an isolated observation for which no data were presented. Even if this finding was consistent and well documented in a series of animals, thus demonstrating a central mechanism requiring no feedback, such observations would still not support the localization of the hyperpnea to the hypothalamus. That the hypothalamus may be unnecessary for exercise hyperpnea was shown by Shik *et al.* (*8*). They obtained hyperpnea during locomotion by stimulating the midbrain of cats whose hypothalamus had been removed.

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In his first sentence, Glenn states one of the findings of our report (*1*). He then states our conclusion and attacks that conclusion as though it were based solely on the experiments in which electrical stimulation was used.

Glenn has misquoted several of the references that he uses to show that hyperpnea can be evoked from a wider area of the brain than the subthalamic locomotor region (*2*). Nevertheless, we recognize the validity of the question and we are not in disagreement about the first issue he raises, that of the general problems of interpretation of electrically evoked responses.

However, we feel that the point is moot in relation to the second issue, the functional relation between locomotion and hyperpnea. In his criticism, Glenn has neglected to point out that many of our animals developed locomotion spontaneously and that respiration and arterial pressure increased when this occurred. He also failed to note that the speed of locomotion, respiratory responses, and pressor responses increased proportionately in an animal that walked spontaneously at two different treadmill speeds. These findings, which were essentially the same as those with electrical stimulation, indicate that Glenn's concern about fortuitous stimulation of unrelated structures subserving locomotion and respiration is not warranted.

Glenn does agree that the development of spontaneous fictive locomotion and hyperpnea in a paralyzed animal (a finding that we have observed in several experiments subsequent to our report) supports our conclusion. He then attempts to dismiss this finding by quoting Shik *et al.* (*3*) to the effect that the "hypothalamus may not be necessary for exercise hyperpnea." However, Glenn is apparently unaware, although we noted it in our report, that the hypothalamus is essential for the development of spontaneous locomotion (*4*) and that Shik *et al.* (*3*) made their observations in mesencephalic cats requiring electrical stimulation to induce locomotion.

Glenn makes the additional point that hyperpnea occurred in the absence of locomotion [see figure 2 of our report (*1*)]. We believe that this does not negate our conclusion, for at rest one of the effector systems (respiration) is above threshold and one (locomotion) is below threshold. A small activation of a common driving mechanism would thus have a demonstrable effect on the above-threshold system but might fail to bring the other to threshold. Our study shows that once both systems are above threshold, further increases of drive affect both in a demonstrably similar manner.

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7 March 1983