their protocol is so similar to that of other investigators? Their previous experience with nonhuman primate ES cell derivation was certainly critical for this success. Thomson *et al.* (*11*) report that of 14 inner cell masses placed in culture, five ES cell lines were established. This result is excellent, but could it be better? Are there ways of assaying blastocysts for their potential of yielding ES cells? As in the mouse, are there predisposing genes for this property? Are there other extrinsic or intrinsic factors that may lead to a greater success rate?

The conditions for directed, lineage-restricted differentiation of ES cells must be defined. Studies to date on ES cell differentiation in vitro rely primarily on the selection and enrichment of specific lineages from the many that may be present when cell differentiation is induced. Also, strategies must be developed to obtain the large numbers of pure populations of cells that would be required for engraftments. In the short term, feeder cell-independent lines will have to be derived and methods for complete cell disaggregation developed. It must also be determined if the cells are amenable to transfections, enabling selection and gene-targeting strategies.

Reports on the isolation of human pluripotent stem cells will no doubt catch the public eye, and there will be expressions of concern, rekindling the debate on human embryo research. The debate will encompass the source of the cells, human cloning potential, and the possibilities of germ line modifications. Four years ago, the Human Embryo Research Panel's report to the director of the National Institutes of Health (NIH) concluded that research deriving ES cells is acceptable as long as embryos are not created expressly for research purposes. Several issues will have to be resolved to permit the appropriate exploitation of the uniqueness and potential of these cells. Currently, as broadly written, U.S. federal law bans the use of federal funds for the derivation of these cells [Public Law 105-78, Section 513(a)]. To date, research in this area has been sponsored through private and corporate funding, with hospital and academic institutional internal review board approval and informed patient consent. It is not clear whether NIH funding necessary to realize the biomedical potential of the cells will be available to support studies using the derived ES cells. Federal legislation and funding policies should be reexamined in light of the biomedical potential of human ES cells, now made more imminent by the Thomson *et al.* report. Federal guidelines must be established so that ES cell research can be funded after appropriate peer review and oversight.

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PERSPECTIVES: NEUROSCIENCE

Long-Term Change of Mind

Michael Merzenich

The removal of an arm or leg causes the grand-scale functional reorganization of maps on the surface of the brain representing body sensations and movements (see the figure) (*1–3*). In human amputees, many square centimeters of the cortex that were formerly engaged by the use of the missing limb come to respond to elaborated inputs from the intact limb stump, trunk, and face (*2, 3*). In parallel, the sensory sensitivity and ability to discriminate different sensations progressively improves on the limb stump; sensations evoked from the phantom hand or foot come to be topographically represented in somewhat unstable form on the amputation stump and, in many patients, on the face; and the perceived body form is distorted, with "phantom" fingers or toes moving progressively toward the arm or leg stump, often ultimately being perceived as being located near, on, or even within the stump (*4*). Enduring pain often parallels these changes, replaying the impact of the crushing machinery or the shark's bite that caused the amputation. The magnitude of this phantom limb pain is directly correlated with the extent of representational remodeling recorded within the cerebral cortex: The greater the changes in the cortex, the greater the pain experienced (*5*).

What mechanisms account for this massive representational cortical remodeling? Can this powerful capacity for representational translocation and functional revision be harnessed to prevent the genesis of phantom limb pain, or to improve rehabilitation after major peripheral or central nervous system injuries? Two reports in this issue (*6, 7*) shed new light on these questions, describing provocative large-scale changes in the morphology of the thalamus and in cortical network connectivity in these monkeys with long-term injuries.

Jones and Pons (page 1121) have examined the "Silver Spring monkeys," macaque monkeys that many years in their past suffered limb deafferentation by the cutting of sensory nerve roots as they enter the spinal cord (*1, 8*). These monkeys show degeneration of sectors of brainstem and thalamic nuclei formerly excited by sensory inputs from their long-insensate limb. In the reorganized thalamus, the representation of the face directly abuts that of the body trunk, just as in the remodeled cerebral cortex (*1, 2*).

How did this dramatic functional reorganization arise? The authors argue that thalamic shrinkage resulting from extensive cell death and subsequent physical rearrangement, combined with a probable sprouting of inputs from the face to neurons formerly representing the missing hand and arm, are the causes. From one perspective, sprouting would not appear to be on a very large scale, because the thalamic representations of the body and original face zones are both substantially shrunken, indicating that the thalamic organization recorded so many years after injury may primarily reflect a simple physical rearrangement of this deep brain nucleus. Alternatively, it is possible that this "rearrangement" has been achieved specifically through sprouting, by which a reinnervation of neurons formerly representing

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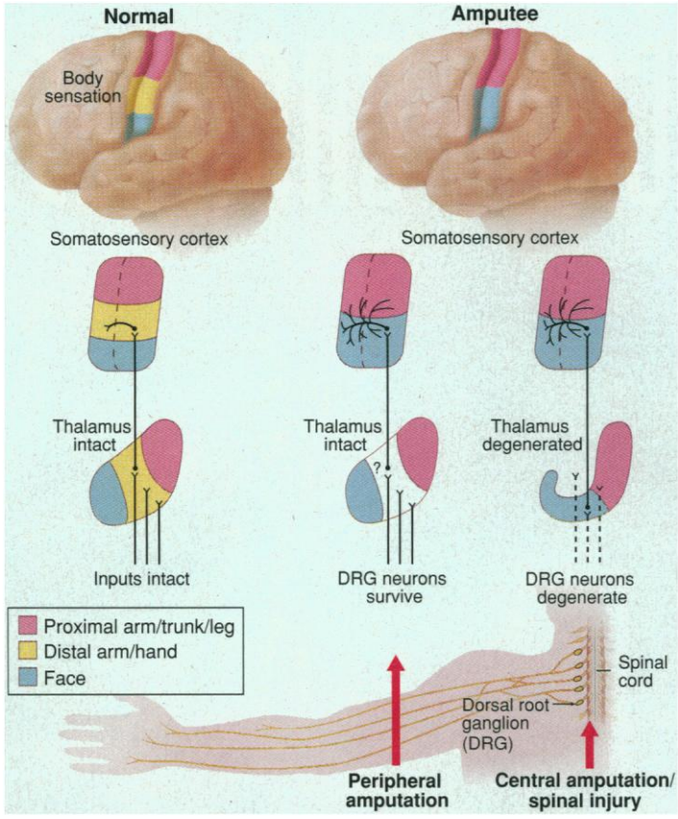
the limb by new inputs from the face also resulted in those inputs being withdrawn from neurons in the original face zone, resulting in cell death and shrinkage within both face and former-limb thalamic sectors. In any event, given this provocative picture (see the figure), further study of these questions will be of great general interest, because it is now reasonable to suppose that such remodeling eventually occurs after every perispinal and spinal injury that results in a substantial loss of sensory inputs.

Jones and Pons argue that these thalamic changes are on a scale that is sufficient to explain the cortical reorganization that has been recorded in the Silver Spring monkeys (*1*). They hypothesize that transneuronal degeneration of, and consequence representational rearrangement and connectional sprouting within, the thalamus may be the principal basis for representational remodeling after limb amputations.

Florence, Taub, and Kaas (page 1117) shed important additional light on this subject. They show that the physical status of the thalamus is essentially normal in monkeys that had suffered finger or arm amputation years in the past, contradicting the hypothesis of Jones and Pons that the changes that they recorded generalize to most or all cases of limb amputation. How could the thalamus remain intact after an upper arm amputation in the study of Florence *et al.*, while the thalamic hand/arm zones were massively deteriorated and rearranged in the Jones and Pons study? As Jones and Pons recognized might be the case, there appears to be a fundamental difference in the pattern of reorganization determined by the level of peripheral injury (see the figure). In most human amputations, the sensory nerves innervating the affected limb and their cell bodies in the dorsal root ganglia near the spinal cord remain largely intact. The Florence *et al.* study indicates that in that event, little or no neuronal degeneration is recorded at the thalamic level. When the injury leads to the death of dorsal root ganglion neurons (or of their nonregenerative central axons) because of spinal or perispinal injuries, slowly progressing transynaptic loss of neurons in the brainstem dorsal column nuclei and thalamus results in dramatic functional reorganization in the thalamus, as in the studies of Jones and Pons.



Massive changes. (**Top**) On the surface of the brain, large-scale reorganization of the representation of the body surface and of cortical outputs controlling body movements after limb deafferentation or amputation. (**Bottom**) Different outcomes likely result from different levels of injury. When spinal ganglion neurons are lost, transneuronal degeneration and possibly local sprouting result in a radically remodeled representation of the body in the thalamic body surface (*6*). When these neurons are spared a more distal injury (as in most amputations), the thalamus remains largely intact (*7*). Extensive sprouting of corticocortical connections (but no change in thalamocortical projections) also probably contributes to large-scale cortical remodeling in both types of lesion (*7*).

Florence *et al.* also discovered an unexpected and undoubtedly important additional contributor to representational remodeling in the cortex after either amputation or limb deafferentation. They showed that the intracortical projections within and between somatosensory cortical areas are far more extensive and elaborate in chronic amputees than in the normal case (see the figure). With these greatly exaggerated spreads in corticocortical axonal connections from the same and from other somatosensory cortical fields, activities from cortical face or arm stump sources are spread all across the large cortical territories that they come to representationally dominate. In striking contrast, thalamocortical projections into the somatosensory cortex appear to be normal.

The large-scale increases in the divergences of intrinsic and extrinsic corticocortical connections were, unexpectedly, as great in an adult macaque monkey with a normally innervated arm and hand that had suffered chronic hand disuse after a wrist injury as in monkeys with chronic arm amputations. This finding indicates that differences in the schedules of functional activation of these large cortical zones by little- or nonused dysfunctional or amputated hands and arms may underlie these large-scale adaptive changes in corticocortical projection anatomy.

Taken together, these studies indicate that there are at least two different brain plasticity scenarios that contribute separately—and, with especially severe or central injuries, together—to the large-scale functional reorganization of cortical representations seen after limb amputation or spinal deafferentation or damage. As our knowledge about the origins of these large-scale representational changes grows, possible strategies for manipulating reorganization outcomes are further elucidated. The control of phantom limb pain and of chronic somatic pain, and the controlled translocation of representations after peripheral and central injury, are eagerly anticipated products of this growing understanding.

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