

Review Article

Unsuspected Plasticity of Single Neurons after Connection of the Corticospinal Tract with Peripheral Nerves in Spinal Cord Lesions

Giorgio Brunelli, M.D.,¹ Klaus von Wild M.D.²

¹Foundation for research on Spinal Cord Lesions, Brescia, Italy

²Medical School of the University of Münster, Germany

Objective : To report an unsuspected adaptive plasticity of single upper motor neurons and of primary motor cortex found after microsurgical connection of the spinal cord with peripheral nerve via grafts in paraplegics and focussed discussion of the reviewed literature.

Methods : The research aimed at making paraplegics walk again, after 20 years of experimental surgery in animals. Amongst other things, animal experiments demonstrated the alteration of the motor endplates receptors from cholinergic to glutamatergic induced by connection with upper motor neurons. The same paradigm was successfully performed in paraplegic humans. The nerve grafts were put into the ventral-lateral spinal tract randomly, without possibility of choosing the axons coming from different areas of the motor cortex.

Results : The patient became able to selectively activate the re-innervated muscles she wanted without concurrent activities of other muscles connected with the same cortical areas.

Conclusion : Authors believe that unlike in nerve or tendon transfers, where the whole cortical area corresponding to the transfer changes its function a phenomenon that we call "brain plasticity by areas", in our paradigm due to the direct connection of upper motor neurons with different peripheral nerves and muscles via nerve grafts motor learning occurs based on adaptive neuronal plasticity so that simultaneous contractions of other muscles are prevented. We propose to call it adaptive functional "plasticity by single neurons". We speculate that this phenomenon is due to the simultaneous activation of neurons spread in different cortical areas for a given specific movement, whilst the other neurons of the same areas connected with peripheral nerves of different muscles are not activated at the same time. Why different neurons of the same area fire at different times according to different voluntary demands remains to be discovered. We are committed to solve this enigma hereafter.

KEY WORDS : Paraplegia · Adaptive neuronal plasticity · Compensation · Nerve graft · Neural connection · Restoration of ambulation.

INTRODUCTION

In our long lasting research for finding a solution to paraplegia we connected (in rats and monkeys) the ventral-lateral corticospinal tract with muscular nerves of selected muscles^{1-7,11-13,22}. Research allowed us to demonstrate that the receptors of the motor end-plates shifted from cholinergic to glutamatergic⁸. After having obtained the permission of the Ethical Committee of the Italian Health Service we have also operated on three fully informed human volunteers. Unexpectedly, the reinnervated muscles were

able to contract individually without simultaneous contractions of the other muscles even if they were connected with axons coming from the same areas of the primary motor cortex²⁴.

MATERIALS AND METHODS

Connection of the central with the peripheral nervous system, in order to reinnervate the muscles disconnected from the cord, was done in hundreds of rats and in 4 groups of macaca fascicularis for a total of 50 monkeys and in three fully informed complete paraplegic human volunteers.

Our research was aimed at solving the problem of paraplegia and of the "matter of impossibility of performance" of the spinal cord for the advancement of the axons regrowing from the upper motoneurons (presynaptic)^{2,4}.

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• Address for reprints : Giorgio Brunelli, M.D.
Fondazione Midollo, Via Galvani 26, 25123 Brescia, Italy
Tel : 00390303385131, Fax : 00390303387595
E-mail : giorgio.brunelli@midollospinale.com

One operating protocol that we used among the others (in rats and monkeys) was the connection of the intact corticospinal tract above-the-lesion (that contains sound regrowing axons), with muscular nerves of selected muscles. This protocol led us to discover and demonstrate that the neuromuscular junctions changed their receptors from cholinergic to glutamatergic^{8,21)}.

The operation in humans (duration 12 hours) was done by 2 surgeons teams* simultaneously on both sides by taking the peroneal nerves (each 36-to 38 cm long, split from the tibial components of the sciatic nerves up to the upper third of the thigh) and inserting their central stumps 4-5 mm into the ventral-lateral bundles of the thoracic cord (the corticospinal tract, corticospinal tract that is the pyramidal tract encompassing Tractus reticulo-spinalis, vestibulo-spinalis, tecto-spinalis, olivo-spinalis et alii) just above the complete cord lesion. The distal stump of the grafts were divided into three artificial branches to connecting these branches each to the three different muscles of the lower limb : gluteus maximus, gluteus medius and quadriceps. These muscles have been demonstrated to allow the individual to stand up and to walk¹⁵⁾.

We have obtained a certification that included all applicable institutional and governmental regulations concerning the ethical use of human volunteers and animals that were followed during the course of these research projects.

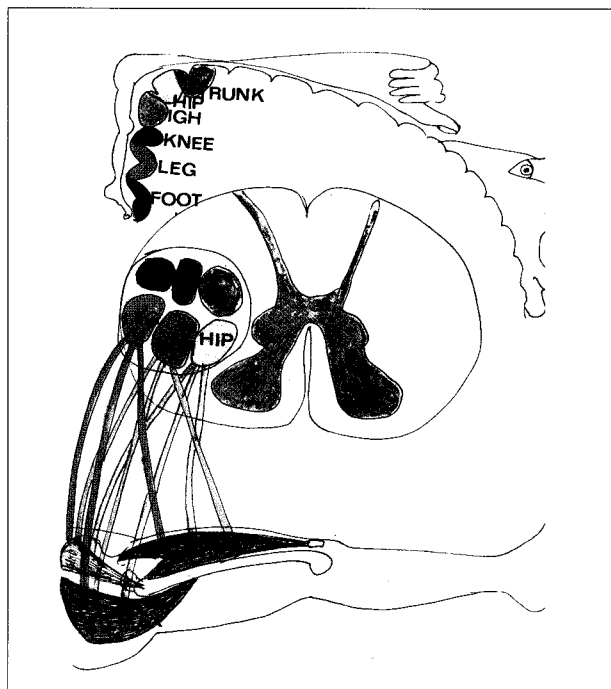


Fig. 1. Schematic drawing of the connection of the corticospinal tract with the muscles. Note that axons going to different muscles come from neurons scattered in different cortical areas.

RESULTS

This protocol for paraplegic volunteers showed another unsuspected plasticity of the brain that we would like to outline in this article.

As in a patient operated on by putting grafts from the corticospinal tract to peripheral nerves, the patients brain became able to command the function of selected muscles notwithstanding the connection of the nerve graft within the corticospinal tract for the outgrowth of the axons of upper motor (pre-synaptic) neurons had been performed only randomly. As a matter of fact, when we* put the graft into the corticospinal tract that contains motor-axons destined to many different muscles, we cannot chose the bundles of axons having the desired destiny and function and the connection is done blindly. Therefore, after graf-

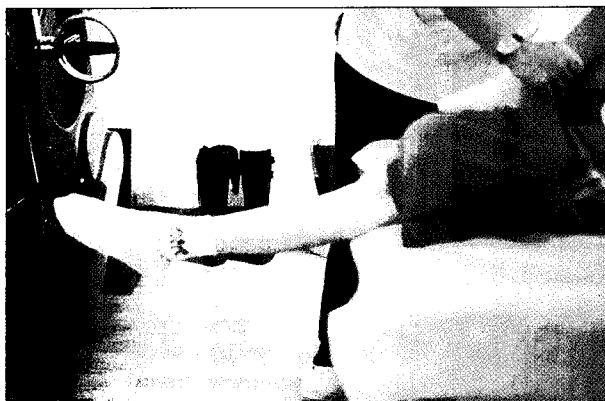


Fig. 2. Selective contraction of quadriceps without cocontractions.



Fig. 3. Selective contraction of the medius gluteus with abduction without disturbing co-contractions.

ting, the same cortical areas give origin to axons destined to different muscles (Fig. 1).

Neurons were placed in different areas and were connected to one muscle and fire together, even if remote from each other, for a selected contraction and movement. We call this phenomenon adaptive functional “brain plasticity by single neurons” in comparison with the well known normal brain plasticity that is called “brain plasticity by cortical areas”. We presume that those scattered presynaptic upper motor neurons connected with a new target learn and memorize their new capacity while forgetting their previous function and that also the neurons of the premotor and associative areas must learn the new function and forget the previous one.

As early as twelve months after surgery, some voluntary activation of the reinnervated muscles appeared and progressed in the following months. The voluntary function of the reinnervated muscles required 2 years to occur and 3 years to consolidate²⁴.

Stunningly, when one muscle contracts due to voluntary command the other muscles do not co-contraction with it. In Fig. 2 and 3, the isolated function of medius gluteus and of quadriceps muscles are shown without cocontractions of the other muscles. In Fig. 4, the lady walking with quadripode sticks is shown.

The operation was done after many successful operations performed by the senior author on rats and monkeys and the human patient was a fully informed volunteer.

Notwithstanding the success in animals we were concerned about the possibility that in an human being cocontraction



Fig. 4. Photogram from a movie showing the ambulating patient, who suffered from “guillottione” severance of the cord at T 8 level (by dislocation of the vertebra) and was operated by connecting her corticospinal tract (bilaterally) with the motor nerves of quadriceps, gluteus medius and gluteus maximus. The patient is walking with tetrapode sticks, without cocontractions (same as in Fig. 2 and 3).

of the reinnervated muscles could compromise the result of the operation. Luckily no simultaneous concurrent contraction of other muscles occurred!

The success of this operations led us, the senior author with a team of researchers of the Medical School of the University of Brescia (Brunelli et al.⁷) and Pizzi et al.²¹) to a series of sophisticated analysis⁸) (i.e. the compound action potentials, the ChAT immunoreactivity, the VGluT1 & VGluT2 expression in control and grafted nerves, the immunoblot analysis of ChAT and of Glu receptors, the T.E.M. exam of N.M.J. in control and reinnervated muscles, the A.M.P.A. receptors in reinnervated muscles, the immunoprecipitation analysis of proteins interacting with GluRs and the C.T.B. retrograde tracing of upper neurons) which allowed us to state that the neuromuscular junctions of the reinnervated muscles had changed their receptors from cholinergic to glutamatergic excitation¹².

DISCUSSION

How the brain was able to command one single movement without co-contraction of the other muscles connected with the same C.S.T. remained an open scientific question to be explained. To say the truth we have not been able to find any explanation. We only were able to infer a hypothesis.

The plasticity of the brain cortex is known for many years and has been first guessed (and then demonstrated by means of fMRI) after amputations and after transfers of neuromuscular units or of nerves in case of palsies. Plasticity has manifold ways of expressing itself in both the intact and the lesioned nervous system. In this respect we would like to refer for more details to the numerous specific literatures on neuroprotection, neural recovery and neural repair, that cannot be discussed here^{8-10,14,16-20,23}). We distinguish today between negative (e.g. with increase in apoptotic cell death²⁰) and positive plasticity (regarding functional and adaptive plasticity²³). Underlying mechanisms thought to be involved in adaptive plasticity include unmasking of existing connections, long-term potentiation, long-term depression, axonal sprouting, dendritic sprouting, synaptogenesis, angiogenesis, and neurogenesis. Such reorganization may demand a high price, since the mature central nervous system has a certain number of redundant or backup systems that participate in the takeover of restored functioning¹⁸).

It has been demonstrated that intensive physical training that is targeted to a very specific functional task and daily

*The two teams of surgeons operating on the left and right side were headed by Prof. Brunelli (orthopaedic surgeon) and by Prof. von Wild (neurosurgeon).

exercises in an enriched environment can modify and improve significantly impaired higher cortical functioning. But, in those cases plasticity regarded the switch of the function of one cortical area to a different function : that of the area corresponding to the palsied muscles to compensate its loss of function.

In our paradigm, both experimental and human cases, on the contrary, there was not the change of function of an area in its entirety but a multitude of single neurons scattered in various areas of the cortex changed their target and function firing together for a new selected voluntary movement whereas the other neurons scattered in the same areas, although connected with different muscles, did not fire (Fig. 2, 3, 4)

This functional plasticity was never demonstrated nor even presumed before. The explanation of this phenomenon is though and daunting as by means of the surgical connection only upper motor neuron fibres were connected with the muscles and, in principle, no feedback from sensory corpuscles could be possible.

We thought that the selection of the single movements could be possible by visual control but the patient was able to do the single required movements also with her eyes closed.

How the scattered neurons can chose the moment to fire together for a voluntary movement without coactivation of the adjacent neurons remain a mystery that we will try to solve hereafter.

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