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Challenges for taking primary and stem cells into clinical neurotransplantation trials for neurodegenerative disease ☆

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ABSTRACT

We review the first generations of clinical trials of novel cell therapies applied to a range of neurodegenerative diseases in the context of mechanisms of functional efficacy. This in turn helps to determine the best strategies to be adopted and the potential chances for success in developing new cell therapies to clinical application in different conditions. We then consider the scientific, technical, ethical, regulatory and logistic issues to be resolved in translating effective laboratory cell-based protocols to patients in clinical trials. We draw optimistic conclusions about the likelihood of success in developing radical new approaches to a range of devastating, and currently untreatable, neurodegenerative conditions, but caution that the problems are complex and the solutions are likely to be slow and costly to achieve in order to overcome significant ethical and regulatory as well as scientific challenges.

Neurotransplantation: basic science background

Cell survival, integration, and growth

Cell loss or damage in the central nervous system underlies all the most debilitating neurodegenerative diseases, and constitutes major – and ever increasing – costs in health, welfare and economics for patients, their families and the health care systems in all ageing societies. For more than 120 years there has been a dream of a 'cure' for neurodegenerative disease, not just by alleviating the symptoms but by replacing lost cells, repairing the damage and reconstructing neural circuits through cell transplantation (Thompson, 1890). However, it is only in the last 4 decades that the dream is turning to reality, with an emerging technology for reliable cell replacement and functional recovery in brain damage and disease (Dunnett, 2010). The key to achieving effective repair in the adult brain was the realisation that the transplanted nerve cells must be fully and accurately differentiated into the precise neuronal subtype lost to the disease process (for example, midbrain dopamine nerve cells for Parkinson's disease: PD). To achieve this it is necessary that the donor cells are transplanted at a time when their fate is already specified, as the adult brain does not express the developmental signals present in the foetal brain that are required to direct the cell's differentiation. Equally, the donor cells mustn't be too developmentally advanced, as this would compromise their chance of surviving the transplantation process. Thus, donor cells need to be within a particular developmental window. In many circumstances this requires harvesting donor cells from the developing foetal brain or spinal cord (Dunnett and Björklund, 1992; Olson et al., 1983), although now there are increasing opportunities for directed differentiation and/or re-specification of cells from embryonic or adult stem cell niches to specific development fates (see below). Once suitable sources of viable cells had been identified, the techniques for effective cell transplantation, first of tissue pieces (Gash et al., 1980; Olson and Malmfors, 1970; Stenevi et al., 1976) and subsequently of dissociated cell suspensions (Schmidt et al., 1981), developed rapidly during the 1970s and 1980s, resulting in a proliferation of studies of cell survival, development and

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function following transplantation in a wide variety of model systems, principally in rat brain (Dunnett, 2010).

A remarkable feature of primary embryonic/foetal cells following transplantation into the adult brain, is that they continue to express their specific developmental growth programmes, successively expressing the relevant developmental genes required to specify and direct their fate, maturating into fully differentiated functional neurons, appropriately embedded in their glial environment, giving rise to active neurite outgrowth, seeking out and establishing synaptic connections with appropriate targets (Björklund and Stenevi, 1985; Sladek and Gash, 1984). Perhaps the even more remarkable observation – in the light of presuppositions about the limited plasticity of adult mammalian brain that dominated since the time of Cajal – is that host axons retain into adulthood the capacity to sprout and innervate the grafts in response to local signals associated with developing foetal targets, allowing (at least in some situations) reciprocal connections to form between graft and host neurons, and thereby integrating the grafted cells into the local host neuron network (Bolam et al., 1987; Clarke and Dunnett, 1993; Xu et al., 1991).

Functional recovery

It is hoped that cell replacement will be accompanied by functional recovery. Dopamine producing foetal nigral cells can alleviate motor symptoms in dopamine-depleted Parkinsonian rats (Björklund et al., 1980; Perlow et al., 1979). Foetal striatal neurons can restore an animal's ability to learn fronto-striatal dependent spatial maze tasks following striatal lesions (Isacson et al., 1986). Diverse dopamine and cholinergic-rich transplants can alleviate motor and cognitive deficits of ageing (Gage et al., 1983, 1984). Hypothalamic tissues can restore vasopressin-dependent control of normal drinking in diabetic rats (Gash et al., 1980) and GnRH-dependent sexual maturation and function in hypogonadal mice (Gibson et al., 1984). Perhaps the most challenging is the achievement in at least some experimental models of recovery of locomotion in spinal injured animals (Cheng et al., 1996). Thus, speculation commenced from the earliest such studies in rats and mice that perhaps neural transplantation offered a new prospect for therapeutic repair in human neurodegenerative disease (Perlow et al., 1979).

The first caution to note is that, even with good surviving grafts, functional recovery is not invariable. Rather, reliable efficacy is dependent upon getting the technical conditions correct. Behavioural recovery may affect some aspects of a lesion syndrome but not others, the extent of recovery is critically dependent on cell survival, differentiation and integration of connections within the host brain, and the profile of recovery can be dependent upon exactly where the graft is placed, which afferent connections it attracts, and which targets it reinnervates. Thus, it was recognised from the earliest studies that transplantation parameters need to be designed and specified to address particular aspects of the functional syndrome of particular interest. Moreover, effective functional repair requires attention not just to the neurobiology of cell transplantation but also to understanding the functional organisation of the brain and how the pathogenic processes of disease translate into profiles of more or less debilitating symptoms.

The second important caution is that, although ineffective or inappropriate grafts are in most cases simply without functional impact, in some circumstances the grafts may produce unanticipated overt adverse side effects. The most obvious adverse effects are that tissues can overgrow or (depending on the cell type) form tumours, space-occupying lesions, or occlusions of ventricular circulation. Poor dissection of embryonic tissues can include non-neural tissues differentiating into cell types of other embryonic lineages. In some circumstances, the grafted cells can exert specific adverse functional influences on normal brain processes, the most widely cited of which are the draft-induced dyskinesias that are occasionally associated with nigral-rich grafts in Parkinson's disease (PD) patients. Even if the graft itself does not survive, any surgical implantation process is intrinsically associated with some non-negligible risk, for example vascular damage. The potential for adverse effects warrant particular attention when considering the safety case for clinical applications.

Mechanisms of recovery

The first reports of functional alleviation of genetic or lesion-induced behavioural deficits in transplanted animals involved replacement of the lost cells in appropriate areas of the brain, leading to the natural conclusion that the recovery was attributable to repair of the damage and restitution of normal processes within the host brain circuits. There are certainly circumstances where this 'reparative' hypothesis can still be sustained (Dunnett et al., 2000). However, it soon became apparent that in many other situations a variety of less specific mechanisms could still yield functional benefit in the absence of effective circuit reconstruction (Dunnett and Björklund, 1987). Grafts might simply provide tonic delivery of deficient neurotransmitters, neurotrophic and tropic factors or neuroprotective agents, exerting their function via pharmacological mechanisms, albeit with more effective transport across the blood–brain barrier, more effective targeting, and local delivery at physiological levels. Alternatively, the grafted cells might provide substrates for axon support and remyelination, thus promoting endogenous survival and regenerative capacity of host neurons, rather than actually replacing or repairing neuronal loss. Indeed, a variety of alternative mechanisms of functional influence have now been identified in a range of model systems (see Table 1), such that simply equating functional recovery with structural repair is not sustainable.

This does not lessen the impact of transplantation as a repair strategy, but does have two implications. First, we have a range of strategies among which to select in order to achieve reliable and sustainable therapeutic efficacy. An effective strategy in one condition is not automatically generalisable to other conditions; the most effective solution will not be the same for all conditions, nor even for all the symptoms of any particular condition. Secondly, when seeking to develop a novel treatment for disease X, more effective progress is likely if the therapeutic design is based upon a rational analysis of the relationship of the pathogenic process to patterns of neuronal dysfunction, cell loss, or circuit disruption, and of how these in turn underlie the symptoms of greatest clinical importance.

Features of both the donor cells and the host influence successful outcome

Over the last two decades, considerable preclinical work has been undertaken to explore the translation of alternative cell therapeutic approaches to a number of clinical applications (see Table 2). Several important issues need to be weighed in considering the relevance of cell therapeutic strategy in each particular consideration.

Mechanisms required for effective remediation

The realisation that diverse mechanisms of actions underlie the success of neural transplantation therapy (i.e., ranging from circuit repair to alterations to the course of disease or downstream consequences of injury) opens the number of ways in which cellular transplantation might offer important therapeutic benefit. The use of transplants to deliver large therapeutic molecules (whether endogenous growth, tropic or transcription factors, or exogenous neuroprotective agents) that do not by themselves cross the blood brain barrier offers new opportunities to deliver effective neuroprotection against the abnormal cellular processes involved in neuropathological progression and to stimulate endogenous neuroplasticity enhancing intrinsic circuit reorganisation and compensation. This approach has been most widely advanced to

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