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Immunological aspects of grafting in the mammalian central nervous system. A review and speculative synthesis

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1. INTRODUCTION

The successful grafting of homotopic tissue (see Table I for terminology), i.e. neuronal tissue, to the brain has recently increased the interest in the brain as an immunologically privileged transplantation site. Evidence accumulated over the past decade has demonstrated that under optimal conditions neuronal grafts seem to be able to survive indefinitely. It appears that they become integrated with the host brain to the extent that they form synaptic connections, and normalize behavioral and biochemical changes due to prior experimental damage of the host brain connectivity (for reviews see refs. 19 and 21). Most attempts at neuronal grafting have been performed with inbred strains of rodents or in other donor–host combinations with few transplantation antigen differences, thus avoiding complex immunological reactions. Therefore, most studies with intracerebral neuronal grafts have not really addressed the issue of immunological rejection in the brain.

The possibility of reversing many behavioral deficits in animal models of neurological diseases such as Parkinson's disease (PD), has focused attention on using neuronal grafts to ameliorate the clinical manifestations of the neuronal degeneration[21,34,182]. A major neuropathological feature of PD is the death of dopamine (DA)-containing neurons in the sub-

TABLE I

Immunology: terminology and definitions

Strong (major) transplantation antigens:	Major histocompatibility complex class I and II cell surface antigens. Induce graft rejection within 14 days.
Weak (minor) transplantation antigens:	Cell surface molecules that express inter-individual variability and which induce rejection, sometimes after processing and association with host major histocompatibility complex antigens.
Autologous graft: (Autograft)	The grafting of a tissue within one individual.
Syngeneic graft: (Syngraft, isologous graft, isograft)	Transplantation between genetically identical individuals (most often inbred strains or identical twins).
Allogeneic graft: (allograft, homologous graft, homograft)	Transplantation between genetically different individuals within a species.
Xenogeneic graft: (xenograft, heterologous graft, heterograft)	Transplantation between different species.
Homotopic graft: (orthotopic graft)	Transplantation of a tissue to its normal location, e.g. skin to skin.
Heterotopic graft:	Transplantation of a tissue to an abnormal location, e.g. skin to brain.

stantia nigra[4]. Indeed, results from some clinical trials involving transplantation to the brains of PD patients have already been published[8,144,151] with several more studies underway. In these trials the graft tissue was not neuronal in origin, but consisted of endocrine catecholamine-producing cells of the adrenal medulla from the patients themselves (autologous grafts, see Table I). In addition to the published cases, to our knowledge at least 150 more patients have undergone surgical procedures with autologous adrenal medullary tissue. Whereas the autografting procedure avoids the problem of immunological rejection, studies in experimental animals have indicated that the adrenal medulla grafts do not possess the same functional capacity as neuronal grafts containing DA neurons[79,145,182,236]. Therefore, it is of interest to seek a source for neuronal donor tissue for grafting to PD patients and also for other neurological diseases where there are no other tissues available that can substitute for the neurotransmitters lost in the disease process.

The intraparenchymal grafting of DA neurons to the adult brain seems to require that the donor tissue is fetal or immature for good survival to occur[16,30,35]. The most suitable donor tissue for grafting in PD patients may thus consist of immature mesencephalic DA neurons from aborted human fetuses. Inevitably, such tissue will be immunologically incompatible with the PD patients, and therefore it is clear that an understanding of the immunology of the brain will become increasingly important in the future.

The object of this paper is to very briefly review technical approaches to successful transplantation of neuronal tissue and to focus on the immunological aspects of grafting incompatible tissue to the brain, with special reference to the immunological problems facing a future clinical application of the technique in patients with PD. Although several studies have shown that immunologically incompatible tissue can survive grafting to the brain, little is known about the laws governing graft survival under these conditions. We will discuss the possible underlying reasons for the immunological privilege of the brain, the limitations of the privileged site, the likely sequence of cellular events after the entry of allo/xenogeneic tissue into the brain, the use of immunosuppression to prevent graft rejection, and finally present a hypothesis on how the immune reactions are regulated within the brain tissue.

2. NEURONAL GRAFTING TECHNIQUES

One can propose a few simple basic neurobiological conditions that need be fulfilled to obtain optimal survival and function of neuronal grafts in experimental animals (c.f. Table IX part I). Generally, the donor tissue must be immature (fetal or neonatal) and the transplantation surgery should be conducted under aseptic conditions. Furthermore, good intraparenchymal graft survival requires that the transplants should be rapidly vascularized by the host which limits the size of the grafted piece. Moreover, there are studies which clearly indicate that axonal outgrowth from transplanted tissue is most extensive when it is placed within reach of its natural anatomical target region in the brain (e.g. grafted DA neurons in the striatum[18]).

Several different methodological approaches have been used in experimental neuronal grafting research, i.e. transplantation of solid fetal or neonatal central nervous system (CNS) tissue to surgically prepared cavities; transplantation of solid fetal or neonatal tissue directly into brain parenchyma or into the ventricular spaces; and, finally, implantation of cell suspensions prepared from fetal tissue into brain parenchyma. There have been extensive studies of fetal neuronal grafts to the anterior chamber of the eye[181], but as the immunological status of this site is different to that of the brain[12], and since the anterior chamber of the eye is not a true CNS site, these studies are not included in this review.

The different methods and transplantation sites differ in aspects that probably are relevant to the immunological reactions against grafted tissues. There have been no systematic studies that directly compare the results when using different transplantation sites and techniques in the same donor–recipient combination with a given immunogenetic difference. For example, it can be reasoned that solid grafts placed into a prepared cavity in the cortex which is covered with a pial scar as a source of vascularization, are not identical from the point of view of origin of vessels and possibly also blood–brain barrier properties, to a graft of dissociated neuronal tissue implanted directly into the striatum. Moreover, the ventricular spaces may also present a somewhat dif-

ferent immunological situation compared to grafting into the brain parenchyma itself, as is mentioned in section 7.2.

2.1. *Brief description of the different grafting techniques currently used*

Small pieces of developing neuronal tissue can be inserted directly into the brain parenchyma as solid pieces by using a wide glass or metal cannula[54,55]. This approach gives access to most deep brain sites and also the ventricular spaces[78]. However, the technique is limited by yielding reproducibly good graft survival in the brain parenchyma only when the recipient is neonatal or young[53,54,269]. Thus it is not suitable for investigating immunological aspects of neuronal grafting since neonatal hosts (up to a few days of age) possess only an immature immune system and therefore immunological unresponsiveness (tolerance) might develop[102,104,159].

Larger solid pieces of CNS tissue can survive in adult hosts, with excellent growth, if they are placed in surgically prepared cavities lined by either a natural vascular bed or by well vascularized pial scar tissue[231]. With this technique, access is limited to brain structures at, or near, the surface of the brain. Grafting to deeper brain structures would require the removal of excessive amounts of host brain.

The cell suspension neuronal grafting technique[16] largely circumvents the problem of limited anatomical access and allows for graft tissue to be stereotaxically microinjected at almost any site in the CNS[31]. In a basic version of this method, the fetal CNS tissue is first incubated with trypsin and then rinsed repeatedly before being mechanically dissociated by repeated passing through the tip of a fire-polished Pasteur pipette. Due to the dissociation trauma, the cell suspension technique imposes greater constraints on the donor-age of the tissue for many regions in the CNS[31]. For example, when grafting rat mesencephalic DA neurons the donor fetus ought not be more than 16 days gestational age as otherwise little or no DA neuron survival is obtained[16,30,35]. Similarly, poor survival of grafted human DA neurons is obtained with human donor fetuses older than approximately 9 weeks postconception[33,36].

All the aforementioned grafting techniques cause damage to the host brain, with a concomitant rupture

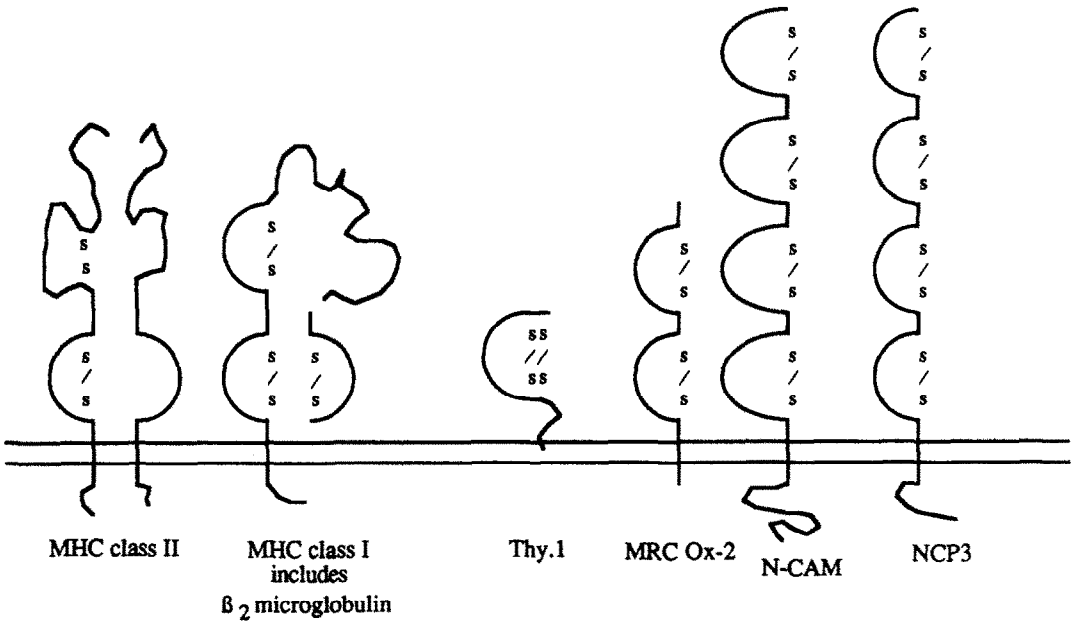


Fig. 1. Schematic drawings of possible targets for immunological reactions. The prime targets for an immune response in an allogeneic graft situation are the MHC class I and II molecules. Other membrane-bound glycoproteins that may display interindividual polymorphism are i.a. Thy-1 (distributed in at least man, rat, mouse), MRC Ox-2 (Medical Research Council monoclonal bank) (only described in the rat), N-CAM (neuron-cell adhesion molecule) (all species), NCP_3 (neurocytoplasmic protein 3) (so far only found in the rat). (Based on refs. 39, 46, 68, 111, 258.)

of the blood–brain barrier. Nonetheless, the extent of the inflammatory response elicited with the different techniques vary, due to the different degree of host brain damage.

3. TRANSPLANTATION IMMUNOLOGY

3.1. *Basic concepts in immunology*

In the following sections we outline a few basic principles of immunology that are relevant to transplantation in the brain. In order to limit the review we have disregarded the once common concept that the brain possesses a separate immune system of its own. It is now clear that lymphocytes within the brain tissue originate from outside the brain, either as precursors that might give rise to clones within the brain tissue, or as mature precomitted cells[249,260].

One important property of the immune system is the ability to distinguish between self and non-self structures and subsequently to inactivate those that are perceived as being non-self. The most important structures for the discrimination of self versus non-self are the membrane-bound glycoproteins encoded by the major histocompatibility gene complex (MHC) (see Fig. 1 and Table II). The immune system has the *potential* capacity to react to *any* foreign (antigenic, non-self) structure with the formation of antibodies (humoral response) and the generation of effector cells, such as helper T-cells and killer T-cells (cell-mediated responses, delayed type hypersensitivity (DTH) and cytotoxicity). B-lymphocytes can form antibodies against virtually any structure, even completely synthetic molecules. The repertoire of antigens that T-lymphocytes respond to is slightly more narrow. The clones of T-cells that ontogenetically are directed against the individual's own cells are selectively eliminated during development.

For an immune response (e.g. a graft rejection) to occur, the immune system has to be activated from the resting normal state. In general, for a T-cell to become activated foreign antigens must be associated with MHC molecules and bind to the T-cell receptor or, in the case of transplantation reactions, the T-cell receptor binds to the MHC molecules on the cell surface on grafted tissue directly. Activation leads to the production of effector mechanisms such as T cell help, T cell killing or antibody formation by B-cells. If such effector mechanisms develop, immunization has taken place.

Selected features and functions of antigen specific cells (lymphocytes) are summarized in Table IIIA. Several cells, without antigen-specific recognition capability, take part in the activation process and are called accessory cells of the immune system. Some functions of such accessory cells are summarized in Table IIIB.

Activated cells of the immune system produce lymphokines, which are substances that participate in the regulation of immune reactions. There are many such molecules that have been known for several years, including interleukin 1 (IL-1), IL-2 and γ-interferon (γ-IFN). Several new important lymphokines have recently been described and sequenced, including IL-3 (ref. 213), IL-4 (ref. 176), IL-5 (ref. 127), IL-6 (ref. 108), neuroleukin[96,97], α-lymphotoxin (α-LT)[48,205] and tumor necrosis factor-α (TNF-α)[14,171]. Proposed effects and sources of lymphokines are summarized in Table IV.

TABLE II

Major histocompatibility complex (MHC)

The major histocompatibility complex (MHC) comprises 3 gene loci, that give rise to class I, II and III antigens. Class I and II are cell surface glycoproteins, which display a high degree of polymorphism, i.e. interindividual differences. They serve as guidance and recognition molecules in the immune system. Class III molecules have a low degree of polymorphism and are found in the blood as complement factors. Strong transplantation antigens are the MHC class I and II gene products. Class I MHC molecules form complexes with the non-variable β_2 microglobulin. In the mouse MHC there are other class I structures with little or no polymorphism and which have hitherto been considered to play minor roles in the regulation of immune reactions. These molecules are encoded in the MHC regions called Tla (18 genes in the mouse) and Qa (8 genes in the mouse). The role of these molecules in the immune system is incompletely understood and the distribution of the homologues to these structures in other species are largely unknown.

Nomenclature	*Man*	*Rat*	*Mice*
Class I antigens	HLA, -A, -B, -C	RT1-A, -E	H-2; -D, -K
Class II antigens	HLA-DO, DP, DQ, DR	RT1-B, -D	H-2; I-E, I-A

Based on refs. 5, 129, 166, 167, 168.

3.2. *Lymphocyte activation and antigen presentation*

The most commonly occurring route of activation

of the normal immune response starts when an antigen is taken up by an accessory cell (e.g. macrophage). Subsequently the antigen is either directly associated with MHC class I or II molecules on the cell surface, or the antigens are broken down to smaller pieces in the cells (*antigen processing*)[252,253] before association with MHC class I or II molecules on the surface of the antigen-presenting cells[22]. In conjunction with presenting the antigen, the accessory cells are also stimulated to produce IL-1 (ref. 183). IL-1 is thought to be important for helper T-cell proliferation in the early stages of immune activation. However, it may well not be absolutely necessary for the proliferation to occur[62,95], nor is it likely to be the sole stimulating factor. Helper T-cells constitute one of the two main subclasses of T-cells that differ in their mode of activation. *Helper* T-cells usually become activated only when an antigen is presented together with self MHC class II structures, whereas *killer* T-cells are activated if the antigen is bound to self MHC class I on the target cell. This concept is called *MHC restriction*[270]. The helper T-cell is a key cell in providing necessary lymphokines to other cells in the immune system for regulation of differentiation and growth. Further subdivision of helper T-cells based on the lymphokines they secrete and their pre-

TABLE IIIA

Some features and functions of antigen-specific cells in the immune system

Phenotypes given are for mature resting cells, and functions after activation with either antigen alone or with help from T-cells. The subgrouping of helper T-cells is not ubiquitously accepted. The status of suppressor cells is also debated. The information below is given for clarification to the text only, and is not to be regarded as authoritative. Abbreviations used in Tables IIIA, B and IV: ADCC, antibody-dependent cellular cytotoxicity, killing of cells when immunoglobulins have bound to receptors on the killer cell and the target cells; +/−, cell expressing a surface marker is denoted + e.g. CD3+, a cell negative for that marker e.g. CD3−; CD, cluster of differentiation; structures with similar function and properties on cells of different species; CD3+, activation molecules of T-cells, used as a marker of mature T-cells able to respond to antigens; CD4+, CD4 molecules bind to MHC antigens class II; CD8+, CD8 molecules bind to MHC antigens class I; Ig-Ag, immune complexes; I-J, a proposed, but not generally accepted marker for a subtype of T-cells with an ability to down regulate immune reactions; LAK, lymphokine activated killer cells, NK cells with an expanded repertoire of cell-targets, after IL-2 treatment; LPS, lipopolysaccharide, a bacterial product, which can activate murine B-cells and macrophages; LTX, leukotrienes; NKLT, natural killer cell lymphotoxin; PG, prostaglandins; sIg, surface-bound immunoglobulin.

Name (ref.)	*Antigen recognition molecules – Functions*	*Markers*	*Secretion products after activation*
B-cells			
(48)	Immunoglobulin Antigen presentation Antibody production	sIg	IgM, IgA, IgE, IgG subclasses IL-1, α-LT
T-cells			
Helper-T cells (41, 42, 48, 96, 97, 108, 127, 161, 208, 225)	T-cell receptor Class II MHC restriction – assist B-cells in production and secretion of immunoglobulins – activate macrophages – induce proliferation of T-cells – induce cytotoxicity – assist B-cells to produce immunoglobulins; T_{H1} mediate delayed type hypersensitivity and T-cell help T_{H2} mediate B-cell help	CD4+	T_{H1}: IL-2, IL-3, γ-IFN T_{H2}: IL-4, IL-5 ? α-LT, TNF-α, IL-6, Neuroleukin?
Cytotoxic T-cells (38, 132, 161, 208)	T-cell receptor Class I MHC restricted – cytotoxic	CD8+	perforin, serine proteases a few may also produce γ-IFN
Suppressor T-cells (entity as a separate cell population is disputed) (161)	Receptor complex unknown Restriction pattern unknown – down regulates immune reactions in an antigen-specific manner	CD4+/CD8+ I-J+?, no unique marker yet.	Antigen-specific suppressor factors have been proposed, but none are sequenced.

cise function has recently been proposed[41,42].

In the initial steps of activation of the T-helper lymphocyte, foreign antigens associate with MHC class II molecules on the surface of antigen-presenting cells and the helper T-cell binds to this antigen-MHC class II molecule complex with its T-cell receptor (T-cell recognition structure for foreign antigens). This interaction leads to an unknown activation signal in the helper T-cell and the cell starts to produce lymphokines[259]. Precursor killer T-cells (non-activated killer T-cells) can bind to the antigen-presenting cell if the foreign antigen and MHC class I molecules also have formed a complex. IL-2 secreted by helper T-cells stimulates precursor killer T-cells to proliferate and mature into effector killer T-cells. Fig. 2 outlines a few steps in the normal activation of the immune system which results in the proliferation of helper T-cells and killer T-cells.

An alternative route for activation is possible if a foreign cell possesses an MHC molecule that is somewhat similar, yet different, to that of the host. This often occurs in transplantation situations. Under these conditions the T-cells may partially recognize the MHC molecules that have not bound any foreign antigen, but consider the MHC molecules to be associated with a foreign antigen as they are not identical to host MHC. This perception of *'altered-self'* leads to activation of the immune system, as outlined in Fig. 3.

3.3. *Transplantation immunology*

The general rule in transplantation is that whenever grafted cells have a surface structure (transplantation antigen) that the recipient cells do not share, the graft will be rejected.

In non-immunized adult hosts, the most rapid rejections are found when the graft is incompatible with regards to major transplantation antigens (MHC class I and class II antigens). Therefore these structures are sometimes called 'strong' transplantation antigens. In rodents, orthotopically grafted skin is typically rejected after 10–12 days if there are strong transplantation antigen differences between graft and recipient[130]. If the differences are of non-MHC structures (minor or weak transplantation antigens) the survival time of a skin graft may vary from 15 up to 250 days[130].

The quantitatively dominating mode of activation of the immune system when grafting between strains of the same species (allogeneic grafts), and possibly also between species (xenogeneic grafting), is due to

TABLE IIIB

Some functions of cells in the immune system with unspecific antigen recognition

Non specific cells (ref.)	*Function*	*Stimulated by*	*Secretion*	*Features/markers*
Macrophages/ Monocytes (14, 142, 252, 253)	Antigen processing Antigen presentation ADCC Angiogenesis Tumoricidal	γ-IFN bacteria LPS Injury, Ig-Ag Activated T-cells UV light	IL-1 Arachidonate derivatives TNF-α Complement factor, proteases	Class II MHC expression
Microglia (105, 183)	Phagocytosis Antigen presentation	LPS, injury	IL-1	Class II MHC expression
Dendritic cells (183, 228, 252)	Antigen presentation			Class II MHC expression Stimulate T-cells
Natural killer cells (132)	Non-MHC restricted kills certain cell types ADCC mediator?	IL-2 induces LAK	NKLT (not cloned)	?
Mast cells (185)	Mediator of inflammat.	Complement factors sIgE complexes	Histamine PG and LTX	IgE Fc rec
Granulocytes	Phagocytosis	IL-3		

See Table IIIA for abbreviations.

direct stimulation of lymphocytes by graft MHC molecules, without major involvement of host accessory cells (Fig. 3). There are more precursor cells in the immune system that can react with antigens in an allograft than with other antigens[175]. The generally accepted interpretation of these findings is that the host immune system recognizes structures on the grafted cells as an *'altered-self'* structure, since the T-cell repertoire has evolved to react towards self-MHC molecules that carry a foreign antigen. The thymus deletes self-reactive clones. However, clones reactive against alloantigens are unaffected by the thymus processing. In addition, allogeneic MHC is assumed to resemble self-MHC that has bound several different antigens. This recognition of *'altered-self'* structures and alloantigens leads to strong immunological re-

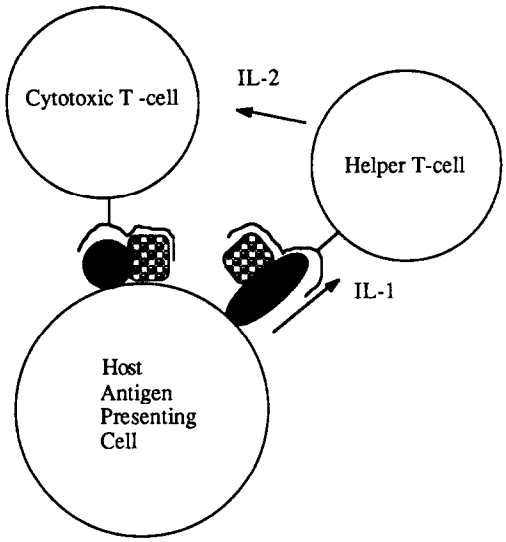
TABLE IV

Lymphokines, mentioned in text; names and some effects and features (of sequenced lymphokines)

Relevant references and other names in brackets.

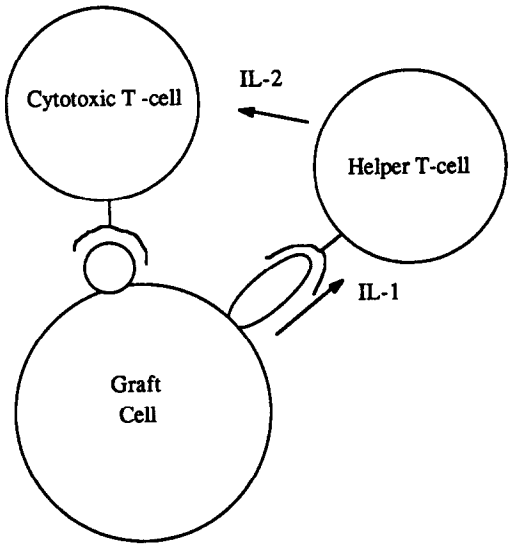
Name		*Effect*	*Cells producing the factor*
IL-1 (183)	Interleukin 1 (Lymphocyte Activat. Fact.) (Endogen, pyrogen)	Potentiation of T-cells Elevation of body temperature Fibroblast, endothelial and epithelial cell proliferation Chemotaxis and activation of neutrophils Induces acute phase protein synthesis in hepatocytes	Wide variety of cells; e.g. activated macrophages, epithelial cells, astrocytes
IL-2 (225)	Interleukin 2 (T-Cell Growth Fact.)	Stimulation of T-cell growth	Helper T-cells
IL-3 (81, 213, 261)	Interleukin 3 (Multi CSF) (Panspecific hemopoietin)	Stimulation of mast cell growth Multipotential hemopoietic cell growth factor	Helper T-cells Astrocytes
IL-4 (29, 176)	Interleukin 4 (B-Cell Stimulating Fact.-1) (B-Cell Growth Fact.-1) (T-Cell Growth Fact.-2) (Mast Cell Growth Fact.-2) (IgE/IgG Enhancing Fact.)	Co-stimulation of B-cell proliferation Limited T-cell growth stimulation Synergy with IL-3 on mast cell growth Enhancement of IgE and IgG1 production in B-cells Class II MHC expression on B-cells and macrophages Enhanced proliferation of hemopoietic progenitors	Helper T-cell Macrophages (certain cell lines) Mast cells
IL-5 (127)	Interleukin 5 (T-Cell Replacing Fact.) (IgA Enhancing Fact.) (B-Cell Growth Fact.-2)	Differentiation of eosinophiles Co-stimulation of B-cell growth Enhancement of IgA production by B-cells Stimulation of in vitro antibody responses	Helper T-cells
IL-6 (108)	B-Cell Stimulating Fact.-2 Interferon β 2	Maturation of B-cells to Ig-secretion T-cell help	T-cells Myxoma Bladder carcinoma
Neuroleukin (96, 97)		Neurotrophic for some embryonic spinal and sensory neurons Monocyte and T-cell dependent B-cell maturation	Activated T-cells Mouse salivary gland
TNF-α (14, 142, 171, 189, 205)	Tumor Necrosis Fact.-α (cachectin)	Cytotoxic to cells with receptors for TNF Activation of granulocytes and eosinophils Chemotaxis of granulocytes and phagocytes Proliferation of fibroblasts Angiogenesis Activation of macrophages Induces cachexia, suppresses lipoprotein lipase	Monocytes, macrophage Mast cell lines Activated lymphocytes
α-LT (48, 171, 205)	Lymphotoxin (TNF-β)	Induces inflammation Activation: granulocytes, eosinophils Stimulation of fibroblast proliferation Chemotaxis of granulocytes and phagocytes Cytotoxic for certain transformed cells	Activated lymphocytes B-lymphoblastoid cell line NK cells
IFN-γ (38, 163, 198, 208, 246)	Gamma-interferon (Immune interferon) (Macrophage Activating Fact.) (Migration Inhibition Fact.)	Cytotoxicity Induction of class I and II MHC Activation of macrophage Inhibits macrophage/monocyte migration Synergistic to α-LT in cytotoxicity	CD3+/– T lymphocytes (CD4+ and CD8+)

See Table IIIA for abbreviations.



Host class I MHC
Host class II MHC
Antigen
T-cell receptor

Fig. 2. Schematic drawing of T-cell activation via a host-derived antigen presenting cell (APC), which expresses high levels of MHC class I and II structures, and produces IL 1 and possibly other factors. The APC has picked up an antigen, and has associated this to the MHC class I and II molecules. Precursor (resting) helper T-cells bind to MHC class II and the associated antigen. IL-1 and other factors from the APC activate the helper T-cell, which then produces IL-2 and proliferates. Precursors of killer T-cells bind to MHC class I and antigen, and stimulated by IL-2 provided by the helper T-cells, start to proliferate. These activated cells expand in number (clonal expansion), and migrate to the peripheral tissues e.g. a wound.



Donor class I MHC
Donor class II MHC
T-cell receptor

Fig. 3. Same as Fig. 2, but the antigen presenting cell is graft-derived, and lacks an associated antigen. The illustration demonstrates the direct activation of host lymphocytes by donor MHC molecules. The donor APC provides IL-1 and other factors. Helper T-cells will proliferate in the same manner as described in Fig. 2.

sponses, in principle identical to the immune response against virus-infected cells[270].

The proliferation of host helper T-cells can be greatly enhanced if IL-1 is produced by donor-derived antigen presenting cells, so called 'passenger leukocytes'. These cells are not only capable of producing IL-1, but also possess high levels of MHC class I and II and are thought to be the strongest stimulus of a rejection process[133,153,221]. A likely mode of activation of the immune system in an allograft situation is summarized in Fig. 3.

Although the direct recognition of allogeneic MHC by T-cells is an important step in the activation of the immune response, from the previous section it should be clear that it is not the sole route by which grafted cells can cause an immune reaction. Host- or donor antigen-presenting cells can pick up foreign antigens that, once presented, can cause a graft rejection (Fig. 2). The antigen-presenting cells may be located in the graft-bearing tissue (resident antigen-presenting cells) or in lymphatic tissues that drain the grafted area. In order to reach lymphatic tissues, graft antigens either flow in a free fluid phase to the regional lymphatic tissue, or bind to resident antigen-presenting cells that in turn migrate to a regional lymphatic tissue. Here the antigens are associated with the host MHC class I and II antigens, possibly after antigen processing. They are also presented to lymphocytes. It has been proposed that some non-MHC transplantation antigens may not have to be processed in order to cause rejection, whereas other minor transplantation antigens do require processing[148]. In the context of fetal neuronal grafting, cell surface molecules that exhibit some degree of polymorphism such as Thy.1, N-CAM (nerve cell adhesion molecules) and Ng-CAM (neuroglia cell adhesion molecules) (Fig. 1), may possibly be immunoge-

nic as minor transplantation antigens and could conceivably cause rejection even in the absence of MHC differences between the graft and host.

A combination of the two mechanisms of immunity activation described above has been proposed by Sherwood et al.[216]. Graft-derived MHC molecules can be picked up by host accessory cells, which provide IL-1 and other factors and carry the graft MHC on their surface without associating them with host MHC. The host lymphocytes are then *directly* activated by the graft MHC on the host accessory cells in the presence of IL-1.

In a xenograft combination, the immunological reactions are not necessarily dominated by anti-MHC reactivity, although it is likely that it is an important antigenic stimulus. In support of MHC reactivity in xenografts Sachs et al.[206] have found antibodies reactive with species-specific MHC epitopes and also strain-specific MHC epitopes in rats immunized with mouse cells. However, it is also conceivable that the non-MHC antigens may play a more important role in xenografting than in allografting in determining graft survival. Empirical data have shown that the survival time of xenografts is often, but not always, relatively short.

Apart from strains with immunodeficiencies, all mammalian species have a complete graft rejection capacity. However, in certain host–recipient combinations there may be a variation in graft survival time depending on the actual combination made[107,130]. A graft between the strains A to B may survive a long time, whereas a graft from B grafted onto A may survive a shorter time. However, these cases are rare, and occur in inbred strains only. In outbred strains, this paradigm does not apply and the rejection capacity is usually intact, unless the animals have other compromising diseases. Transplantation laws are summarized in Table V.

TABLE V

Transplantation laws

Genetic rules

- The general rule in transplantation is that whenever the donor tissue possesses a cell surface structure (transplantation antigen, H = gene locus for transplantation antigen) that the recipient does not share, the graft will be rejected.
- *Syngeneic grafts* (grafts between individuals in an inbred strain) will survive.
- *Allogeneic grafts* (grafts between different inbred strains) will fail.
- *Xenogeneic grafts* (grafts between differen species) will fail.
- Grafts from males to females within an inbred strain where other transplantation antigens are identical may be rejected.
- Multiple histocompatibility differences are additive if of similar strength. This is most pronounced with minor transplantation antigen differences.
- Transplantation antigens are co-dominant, and a F1 hybrid expresses both alleles at each H locus.

Empirical rules

- Skin grafts incompatible for class I and/or II MHC structures are rejected within 10–12 days in the mouse.
- Skin grafts, differing at one or more non-MHC transplantation antigens between host and donor may be rejected between 15–250 days in the mouse.
- Allograft survival time is directly proportional to graft size. Some exceptions are known.
- The weaker the histocompatibility difference, the longer the graft survival of large grafts.
- With weaker histocompatibility differences, the onset of rejection is delayed and the duration longer, i.e. a prolonged time of rejection.
- Immunological memory is stronger, but of short-term duration with weaker histocompatibility differences.

Based on refs. 107, 130, 148, 222.

3.4. *Mechanisms of rejection*

In spite of decades of interest in the rejection mechanism of grafts, there is no real consensus on exactly which cells are necessary for the rejection process. Several recent reviews discuss this topic in great detail[113,155,230]. Knowledge of the cellular reactions underlying immunological rejections might provide a means of developing specific and less toxic ways of interfering with rejection processes.

Analysis of the events leading to rejection of a grafted tissue has for example been done by transferring different cell populations to animals bearing grafts. However, even in these studies controversy continues concerning the minimum required cell population that it is necessary to transfer in order to induce graft rejection[155]. If activated killer T-cells (from immunized animals) are transferred to animals bearing grafted tissue, they will cause rejection[155]. If helper T-cells are transferred to a host with a graft, the graft will be promptly rejected, possibly through the recruitment of host killer T-cells that mediate the actual graft rejection[230]. If a combination of helper and killer T-cells are transferred, the graft rejection is usually also prompt[232].

In addition to the specific cell-mediated responses discussed above, a rather unspecific inflammatory component probably also participates in the rejection process[113]. The so-called delayed-type hypersensitivity[224] is mediated by activated helper T-cells, which produce the lymphokines γ-IFN, IL-2 and α-LT[41]. These lymphokines can cause intense inflammation, monocytic/leukocytic aggregation and a direct cellular cytotoxicity[42,246]. This may result in killing of the grafted tissue, but also normal healthy cells may be killed ('bystander-kill'), and thus DTH is regarded as harmful for the organism. Claims have been made that privileged sites are protected against DTH reactions by an unknown mechanism, and if DTH reactions occur in such places they are believed to be particularly harmful[173,235]. Apart from toxins released from lymphocytes and macrophages, oligodendrocyte cytotoxic factors released from activated astrocytes have been described in vitro[200]. It is of importance to stress that even in DTH reactions, the initial eliciting event involves a specific recognition of the graft. There exists no completely unspecific mechanism of graft destruction.

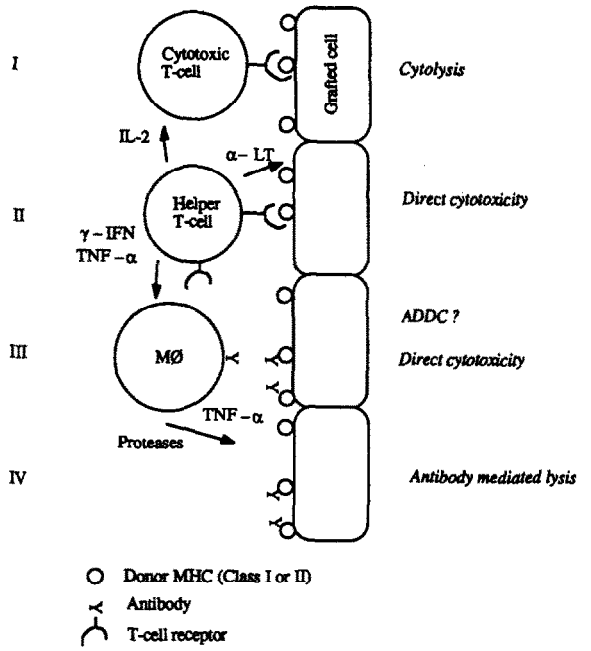


Fig. 4. Different mechanisms of graft rejection. I. Cytolysis mediated by killer T-cell directed i.a. against graft MHC. II. Helper T-cell provides IL-2 for itself and the killer T-cell. Helper T-cells may produce lymphokines that can be directly cytotoxic to some target cells. III. Helper T-cells, possible via γ-IFN and other lymphokines, may be chemotactic for inflammatory cells such as macrophages (MØ), which after activation by γ-IFN and other stimuli can be cytotoxic. Immunoglobulins may bind to macrophages and target cells and an antibody dependent cellular cytolysis (ADCC) may take place. IV. Antibodies may cause cytotoxicity after complement binding.

The role of antibodies in graft rejection remains uncertain[155]. Antibodies that bind to the surface of cells with foreign antigens can, under certain conditions, bind circulating complement factors that will cause cellular lysis of the target cell. Macrophages, monocytes, possibly natural killer cells and so-called null cells have receptors for the constant part of the immunoglobulin (F_c), and can attach to antibodies bound to grafted cells. The graft cells are subsequently killed in a process called antibody-dependent cellular cytotoxicity, ADCC[165]. The phenomenon is well described in vitro, but the role of ADCC in vivo remains to be determined. The hyperacute rejection (graft rejection within 24 h in a pre-immunized animal) is mediated mainly by antibodies and possibly ADCC[155]. In certain cases of xenografting, in the absence of immunization, preformed antibodies directed against the grafted tissue are present in the host before the grafting and are thought to lower the survival rate of the xenogeneic grafts[130].

Some important events in a generalized graft rejection are outlined in Fig. 4.

3.5. *Mechanisms of action of immunosuppressive drugs*

Currently, 3 immunosuppressive drugs are widely used for transplantation in clinical situations, chosen because of their relatively selective action on the immune system. These drugs can also be used as research tools to dissect the mechanisms that regulate immune responses.

Firstly, azathioprine is widely used for immunosuppression. It is an anti-mitotic substance that blocks the clonal expansion of activated lymphocytes, but it can also affect other rapidly dividing cells and is therefore bone marrow toxic.

Secondly, another commonly used immunosuppressive drug is cyclosporin A, which is not toxic to bone marrow. Although it has been used widely with success, the exact mechanism of action of cyclosporin A is not known[217]. A major immunosuppressive effect

is probably blockade of expression of high affinity IL-2 receptors and IL-2 production[122]. The production of γ-IFN is also directly inhibited[217]. If high affinity IL-2 receptors are already expressed on helper T-cells or killer T-cells, cyclosporin A is not effective[122,217]. This explains why cyclosporin A does not interfere with effector cell function. Cyclosporin A has also been reported to suppress the induction of MHC class I and II expression in grafted tissues, by inhibiting γ-IFN synthesis[162].

Thirdly, steroids may be used, particularly in combination either with cyclosporin A or azathioprine. The steroids have several effects, that are beyond the scope of this review, such as metabolic, electrolyte balance regulation, direct hormonal effects, actions on membranes and vasculature. A major role of steroids in immunosuppression is the anti-inflammatory effect. The exact mechanisms for these anti-inflammatory effects are not fully known, but some factors are of particular interest: the blocking IL-1 production[47]; and the activation of phospholipase A2, a regulatory enzyme step in the formation of all prostaglandins, thromboxanes and leukotrienes from arachidonic acid. The immunosuppressive action of steroids is further increased by the direct killing of immature T-cells resident in the thymus[130], the down-regulation of mainly MHC class II antigens[143,252] and the suppression of immunoglobulin production by B-cells[47].

3.6. *Lymphocyte circulation*

The circulation of lymphocytes is under strict control. Precursors of T-cells are formed in the bone marrow and then migrate to the thymus. Mature B-cells from the bone marrow or mature T-cells released from the thymus eventually migrate to lymphatic tissues such as lymph nodes, Peyer's patches in the gut, and to the spleen. Some B- and T-cells will be retained in these tissues by surface structures called homing receptors[83,117,118]. After activation of foreign antigens in the lymphatic tissue, lymphocytes lose their affinity for homing structures and enter the blood. Activated cells are known to accumulate in brains affected by inflammation[249]. A special kind of homing receptor for active lymphocytes, an ubiquinated glycoprotein, is induced by γ-IFN in inflamed tissues, for example the synovial membrane in an arthritic joint[65,83,119]. Cells in inflamed tissue and cells exposed to IL-1 or γ-IFN have been seen to express another molecule (ICAM-I), which increases the adherence of cells of the immune system[65]. The expression of MHC class II on endothelial cells and other tissues where MHC class II normally is not expressed has also been proposed to act as a stimulus for homing. The possible existence of similar homing signals in the brain is unknown.

4. THE BRAIN AS A PRIVILEGED SITE FOR TRANSPLANTS

4.1. *Definition*

The concept of the immunologically privileged status of the brain refers to the empirical finding that allogeneic or xenogeneic tissues (see Table I for definitions) exhibit a *prolonged survival,* or a *higher rate of survival after a given time,* when grafted into the brain parenchyma in comparison to when grafted to peripheral sites. This does not mean that immune reactions cannot take place in the brain, nor does it imply a permanent graft survival of immunogenetically incompatible tissues. In a classic review Barker and Billingham[11] stated: "the grafts (are) in some way partially or fully exempted from the normal rigors imposed by their histoincompatible status". Thus, grafting to a privileged site constitutes an exception to the normal transplantation laws.

Table VI lists a selection of experiments that show a prolonged graft survival in the brain as compared to graft survival in a peripheral site.

4.2. *Early history of the brain as a transplantation site*

Prior to the development of in vitro culturing techniques, there was a need to find hospitable sites for experimental grafts in order to study the development of embryonic tissue and tumors. The brain was identified as one such potentially interesting site, but it was not immediately classified as a particularly good transplantation site[20]. In 1907, Del Conte[58] grafted a variety of *non-neuronal fetal tissues* to the cerebral cortex of adult dogs with some minor success. However, at best he found that there was only partial survival of certain types of tissue and he concluded that the brain was an unfavorable site for im-

TABLE VI

Examples recorded in the literature of a prolonged survival time and/or increased rate of graft survival of intracerebrally grafted tissue where a comparison has been made between the survival rate of tissue from the same donor in a peripheral reference site and the brain

Ref.	*Tissue grafted*	*Survival rate in CNS*	*Survival rate in periphery*	*Observation time, median (range)*	*Host/graft combination*	*Notes*
164	Carcinoma	95%	70%	tumor take	allogeneic	a
	Carcinoma	89%	21%	tumor take	allogeneic	a, b, c
266	Embryonic	80%	50%	56 (21–77)	allogeneic	a, d
158	Skin	33%	0%	10	allogeneic	a, e
218	Tumor	10–35%	0%	84	allogeneic	f
199	Skin	100%	0%	21	allogeneic	a, c
194	Skin	71%	0%	50	allogeneic	g
106	Skin	100%	0%	20 (15–40)	allogeneic	h
	Skin	88%	0%	35 (15–50)	allogeneic	i
	Parathyroid	54%	0%	38 (11-96)	allogeneic	i
154	Neural	54%	0%	30	allogeneic	j
263	Neural/skin	100%	0%	49 (42–90)	allogeneic	k

a, No inbred strains used.
b, Host immunized by donor blood injection 10 days earlier.
c, Simultaneous grafting to the groin and the brain of the tumor.
d, Grafts placed in the brain and subcutanously in the neck at the same time.
e, Hosts immunized by a donor skin graft prior to grafting.
f, Inbred mouse strains used.
g, BN and Lewis inbred rat strains used.
h, Inbred strains Lewis and FI rat used.
i, Inbred strains DA and FI rats used.
j, Inbred strains PVG and AO used, graft placed in 3rd ventricle and under kidney capsule.
k, Inbred strains of mice used, differing at class I, II, I and II MHC antigens, non-MHC and MHC and non-MHC. Comparison with a skin graft in the periphery.

plantation of most types of fetal tissue.

In 1913 came, to our knowledge, the first report of successful intracerebral grafting of an allogeneic *tumor*, thus identifying the brain as a suitable site for the study of transplants (study mentioned in ref. 67). Since then, many reports have been published reporting that allogeneic and xenogeneic tumors can grow in the brain (e.g. refs. 67, 92, 93, 94, 103, 146, 164, 209, 210, 219). Very few studies directly compare the grafting of identical tissue within and outside the brain and, thus, the issue of the brain being a particularly good transplantation site was not directly addressed. Nevertheless it was often remarked that the tumors only grew when grafted to the brain.

Thompson's early claims in 1890 of survival of xenografted *adult neuronal tissue*[245] can be disputed, as more recent work clearly indicates that the grafted adult neurons are very unlikely to have survived. In fact, the first report reliably showing that *neuronal* tissue could survive grafting to the brain by Dunn involved *immature* cortical tissue grafted between rat siblings[61].

The first report of successfully grafted *fetal tissue* into the brain was published by Willis in 1935[266]. In this study, the grafting of various, mostly minced, whole body parts was conducted in an allograft combination in rats. The age of the fetal tissue was deemed important and it was concluded that the brain is: "a favorable site for (the) differentiation and development of embryonic tissue to mature tissue, without host resistance reactions"[266]. Differentiation and development of the fetal tissue did not occur after grafting to a subcutaneous site. Concerning grafts of *neuronal tissue*, the importance of using immature donors has repeatedly been confirmed following the early findings of Dunn[61]. Both Le Gros Clark[141] and Glees[88] found good survival of fetal cortical graft implanted intracerebrally in rodents and more recent work has demonstrated that the donor age is a limiting and crucial factor when grafting neuronal tissue to the brain[16,35,214].

Furthermore, several types of *non-neuronal adult tissue*, such as skin[85,86,106,158,194,199,210], parathyroid gland[23,106,136], thyroid gland[137,220], adrenal medulla[79,182,185,236], endocrine pancreas[251], and pituitary gland[250], have since also been successfully grafted to

the brain. Recently there have also been several reports of tumor cell lines and neuroblastoma cell lines being grafted to the brain[84,116]. Some of these cell lines completely lack expression of MHC molecules, thus making a classification into allo- and xenogeneic grafting somewhat difficult.

In summary, several different types of tissues can survive grafting to the brain. Neuronal tissue displays optimal survival only if immature, whereas the development stage of non-neuronal tissue seems less critical.

4.3. *Reasons for caution in interpretation of early graft studies*

The grafting of tumors and non-neuronal tissue may not be an ideal approach to study the immunologically privileged status of the brain and its direct relevance in a neuronal graft context is questionable. For example, unlike neuronal grafts non-neuronal graft tissues do not form a blood–brain barrier[233,241]. Moreover, it is conceivable that malignant cells may outgrow the host immune response despite the existence of a true immunological response, perhaps via an in vivo selection of tumor cells that express little or low levels of transplantation antigens[128,191]. In addition, tumors may be *less* vascularized than normal tissues, and therefore be relatively inaccessible for immune reactions[199]. Alternatively, tumors may thrive in the brain, and achieve a high survival rate, because of a *relatively better* blood supply and a better access to growth factors in the brain as compared to a peripheral site[146].

Tissues can differ in their antigenic properties, and can arbitrarily be grouped into tissues of high or low immunogenicity[106]. Their antigenicity affects the outcome of the transplantation (Table VI). However, it is important to note that a transformation from low to high immunogenicity can occur by exposure to MHC class I and class II antigen induction factors (see section 6). Even tissues that at the time of grafting may express very low levels of donor MHC[218], may later express high levels of MHC antigens after grafting in vivo[154]. The relative abundance of antigen-presenting cells in the grafted tissue may also be an important factor in governing immunogenicity. Tissues such as cartilage and acellular bone can be defined as an 'immunologically privileged tissue' (tissue of exceptionally low immunogenicity) which does not appear to evoke an immune response at all when transplanted[12].

In summary, when evaluating graft survival in a privileged site it is important not only to consider the genetic origin of the donor tissues, but also to take into account properties of the grafted tissue that are inherent to its particular cellular composition.

4.4. *Attempts at defining the immunogenetic basis for transplantation in the brain*

Several attempts have been made to define an immunogenetic basis for the survival of intracerebral grafts in order to determine the number of MHC and non-MHC structures that may differ between the donor and host, while still allowing good graft survival. For heterotopic grafts there are examples of good graft survival for prolonged periods with xenogeneic adrenal medulla grafted into brain cavities or parenchyma[187]. Other heterografted allogeneic tissues of high and low immunogenicity have survived for prolonged times[106,194,199]. Geyer and Gill indicated that heterologous, allogeneic grafts placed in the brain parenchyma can survive for prolonged times. However, MHC class I structures were still found to immunize the host and there were signs of rejection in grafts when there were MHC class I differences between the donor and host[85,86].

Xenogeneic neuronal grafts implanted into the brain parenchyma and the ventricles generally do not survive transplantation permanently. Although rejection occurs in a large proportion of animals[32,37,72,114], there are still reports of good xenograft survival for long time periods in a subset of recipients[17,32,51,52].

The neuronal graft combination of interest in a clinical context is a combined MHC and non-MHC antigen difference between donor and host. Such grafts have been seen to survive up to 15 weeks without signs of rejection when grafted as a cell suspension into the parenchyma[263]. Neuronal grafts that differ at only MHC class I antigens have survived for a minimum of 4–7 weeks in the parenchyma[150,263]. However, similar intraventricular grafts have been found to undergo rejection within 4–7 weeks in a majority of cases[150,154,172,218]. Grafts differing at both MHC class I and II have been shown to survive for up to 60 days in the ventricle[154] and the parenchyma[263].

Similarly grafts differing at non-MHC antigens only have shown good survival both in the parenchyma and the ventricles[154,263].

In summary, there does not seem to be a single certain immunogenetical combination between donor and host that is exempt from immunological rejection. Nor is there as yet strong evidence for a certain immunogenetical difference between donor and hosts which allows indefinite, permanent graft survival, completely without immunological reactions against grafts implanted in the brain. The contrast in survival between neural and non-neural grafts is possibly related to the degree of MHC antigen expression (see section 6). There is then a mere quantitative difference between the graft survival in the brain compared to other sites when it comes to the length of graft survival and the relative strength of the antigenic difference between donor and host, as has been proposed by Hasek[103].

5. ALLEGED COMPONENTS IN THE IMMUNOLOGICALLY PRIVILEGED STATUS OF THE BRAIN IN A GRAFT SITUATION

5.1. *Components of the afferent arc of the immune system*

The afferent arc of the immune system comprises the events leading up to the initiation of an immunological reaction. This includes the drainage of antigens, either free or cell-bound, from a transplantation site to a lymphatic tissue, and the subsequent activation of the immune system.

5.1.1. *Lack of lymphatic drainage from the brain*

The important role of regional lymphatic tissue in graft rejection has been repeatedly stressed in transplantation immunology[11,153,155,247]. The removal of the regional lymph nodes close to the site of grafting may allow for a *prolonged* graft survival[60] although rejection still can occur[11,82]. The most common explanation given for the prolonged survival time of grafts in the brain is the lack of lymphatic drainage[12,106,158]. Indeed, there are no endothelium-lined lymphatic vessels in normal neuronal tissue. However, the perivascular spaces present along the larger vessels in the brain, have been suggested to serve as an equivalent of lymphatics in the brain[192].

The lack of classical lymphatic vessels in the normal brain does not mean that there are no routes by which antigens in the brain can reach a regional lymphatic tissue. Tracer substances injected into the brain parenchyma can pass to the deep cervical lymph nodes in all species tested so far[25,27,45,147,243,261,264]. The fraction of tracer substances recovered in the lymph nodes is partly dependent on the molecular weight of the tracer substance[25,27], the charge of the tracer particles[265] and the species used. After an intracerebral injection of antigens, specific antibodies are formed[100,264] and cells directed against the antigen proliferate in the deep cervical lymph nodes, peaking on day 5 after the injection[262,264]. Up to 50% of particles are estimated to be cleared via the lymphatics when injected into the caudate nucleus in rats[243]. This passage is extracellular and is found in the perivascular spaces draining into the subarachnoid space, or along white matter fiber bundles towards the olfactory bulb. The particles finally pass through the cribriform lamina into the lymphatic vessels of the nasal mucosa[27,69]. The existence and potential role of this route of passage in humans remains to be determined. In addition to these routes there are extensive networks of lymphatic vessels in the meningial tissue and dura mater[7]. In animal models of chronic inflammatory conditions, such as chronic relapsing experimental encephalomyelitis and multiple sclerosis in humans, an organization of the regions of the perivascular spaces into tissue resembling actual lymphatic nodes has been observed[192]. These formations have been implicated in the drainage of the interstitial fluid and to present antigens[192,193]. Finally, it should be mentioned that a direct activation of lymphocytes circulating in blood vessels through a graft has been suggested to be important during late rejection of alymphatic skinflaps[247]. However, due to the complex pattern of cellular interaction in the normal immune system this route of immunization is today regarded to be of lesser or no importance.

5.1.2. *Lack of dendritic cells in the brain parenchyma; antigen presentation capacity in brain tissue*

One important component of the afferent arc of the immune system is antigen presentation: the process of bringing the antigen and cells of the immune system together, resulting in an activation of the lymphocytes.

Several *accessory cells* in the immune system have this capacity as summarized in Table IIIB. The importance of such cells in transplantation immunology has been stressed repeatedly and they are considered to be the potentially strongest inducers of allograft rejection[133,155,221]. One such accessory cell is the dendritic cell, which is a non-phagocytic cell of bone marrow origin that can present antigens to lymphocytes[228]. The dendritic cell also expresses high levels of MHC class I and II antigens. Its presence in the brain has been debated[101]. However, Head and Griffin have found MHC class II expressing cells that might be dendritic cells lining the ependyma, and localized along major vessels and in the white matter in the rat[106].

Another cell population of interest is the alleged 'resident macrophage' of the brain, the microglia, which is most probably also of bone marrow origin[188]. Microglia have phagocytotic capacity, and resemble the macrophages of other tissues since they can produce IL-1 after activation[182,253]. These cells are the major cell component expressing MHC class II antigens within the brain[105] and normally constitute 13% of the glial cells in the white matter of humans[105].

Furthermore, cultured astrocytes can produce IL-1 after exposure to endotoxins such as lipopolysaccharide in vitro. A subset of astrocytes may express high levels of MHC class I and II antigens after exposure to γ-IFN[70,73,75]. Such stimulated astrocytes have been found to be capable of presenting antigens to T-cell lines and to promote T-cell clonal growth in vitro, and thus may function as facultative antigen-presenting cells in vivo[74,160,212,260].

Cerebral endothelial cells have also been proposed to act as antigen-presenting cells in certain situations. These cells have been shown to express MHC class II antigens in human cerebral vessels from active inflammation sites in cases of multiple sclerosis and systemic lupus erythematosus (SLE)[248]. In animal experiments, antigens injected into the brain parenchyma have been found to traverse the endothelium and reside on the luminal side of the endothelium, suggesting that endothelial cells may have bound the antigens on their surface[255]. Cultured cerebral endothelial cells have also been shown to present brain-specific antigens, to associate them with MHC class I and II molecules and to promote clonal growth of specific T-cell clones reactive with the brain-specific antigen[157].

In summary, there are several cells present in the brain which have the potential to act as antigen-presenting cells in a transplantation situation. The exclusion of cells with antigen-presenting capacity from the graft tissue itself prior to transplantation may provide a means to reduce the immune response and prolong graft survival.

5.1.3. *Immunological unresponsiveness induced by a preferentially vascular route of antigen presentation*

As mentioned earlier, a common belief was previously that the brain completely lacked lymphatic drainage. It was also taken for granted that the interstitial fluid of the brain drained completely into the cerebrospinal fluid (CSF) and that this in turn drains directly into the blood. It was therefore reasoned that antigens introduced into the brain were released into the blood stream. A vascular route of intracerebral graft antigen presentation has been suggested to induce a state of unresponsiveness. An accidental injection of grafted tissue into blood stream during the actual implantation, can probably be regarded to behave in a similar way. An incompatible graft implanted in the anterior chamber of the eye has been reported to be promptly rejected if the host spleen is removed prior to graft surgery, whereas a prolonged graft survival is seen if the spleen is left intact[125]. Two different concepts have been suggested to cause this 'immune deviation' as the phenomenon has been called; enhancing antibodies[125] and T-suppressor cells[235]. In theory, enhancing antibodies are immunoglobulins that bind to transplantation antigens and 'hide' them from effectors of cell-mediated graft rejection and from further antigen presentation[226,229], thereby prolonging the graft survival.

Streilein et al.[235] suggest that suppressor T-cells resident in the spleen can down-regulate the immune responses against grafts placed in a privileged site. However, as pointed out earlier, we now know that the brain in rodents is not totally lacking a passage into the lymphatics and moreover, recent studies indicate that antigens in the CSF can be picked up and presented by cells in the choroid plexus[170]. Therefore the emphasis on the spleen and a preferential vascular route of antigen presentation in immunological privilege may be overrated.

5.1.4. *Evidence for immunization of the host by the graft*

Activation of the immune system is the final outcome of the afferent arc. Thus immunization of graft recipient indicates that the afferent arc is operant.

It is essential to determine that the grafted tissue can be immunogenic. Fetal neuronal tissue does not normally express MHC antigens at the time of grafting[218], but if grafted to a non-privileged site a vigorous rejection can be observed, indicating that fetal neuronal tissue can be immunogenic[154].

A sensitive, but technically difficult way of determining if an animal is immunized by a graft is to measure the survival time of a subsequent homotopic skin graft from the same donor strain. If the host has become sensitized, the skin graft is rejected more rapidly, in a second set fashion, in contrast to the slower first set reaction. To make this analysis with xenografts is particularly difficult, since the primary graft survival often is poor, with absence of formation of a regular blood supply. A primary xenogeneic skin graft thus rarely survives for 7 days. Using the technique of homotopic skin grafting, Geyer and Gill[85] have monitored immunization due to a single intracerebral allogeneic skin graft and they emphasize the importance of MHC class I antigen differences in evoking immunization. Head et al.[106] report a survival time of 7 days of a second skin graft after an intracerebral grafting. However, it is claimed that this short time is a normal first set reaction in that particular strain combination, and the authors do not consider this to be a proof of host immunization[106]. Lund et al.[150] report a second set reaction in rats that have received a neuronal intracerebral xenograft in the *neonatal* period and have then received a skin graft when adult.

Another test that measures the cellular response following immunization is the primed lymphocyte test. This is based on the appearance of a swifter proliferative response of previously activated lymphocytes when these are re-exposed to an antigenic stimulus. Raju and Grogan have found such reactivity in hosts receiving intracerebral allogeneic skin grafts[194].

Simonsen's test of alloimmunization is an alternative and sensitive method of measuring host sensitization of cells in vivo[222]. The test determines the proliferation capacity of lymphocytes from animals that have been exposed to a given transplantation antigen combination in vivo. If a host has been immunized by a graft, lymphocytes transferred to neonatal animals of the donor–host recipient combination will proliferate vigorously if the cells are re-exposed to the antigens. The test is among the most sensitive immunization assays available, but requires the use of highly inbred strains. With this test we have monitored immunization in several, but not all, combinations of allogeneic fetal neuronal grafts[263].

Even though antibodies do not play a major role in actual graft rejection, a common technique of monitoring immunization by grafts is to determine the titer of antibodies formed against a grafted tissue. This method is advantageous as it can be used in immunosuppressed animals. This is a purely qualitative test, and it is very important to emphasize that there is no correlation between the amount of antibodies formed and the state of anti-graft reactivity[155]. Intraparenchymal allogeneic skin grafts[194] and intraventricular allogeneic neuronal grafts[150] have both been found to cause antibody production in the recipients. Recently we have found graft-specific antibodies in immunosuppressed rats receiving intrastriatal implants of fetal human or mouse neuronal tissue, using an indirect fluorescent antibody detection of bound rat immunoglobulins directed against graft antigens[36,37].

To summarize, there are several studies indicating that grafts placed in the brain immunize the host. Thus, some form of the afferent arc from the brain to the immune system must be present.

5.2. *Factors of the efferent arc of the immune system*

The efferent arc of the immune system comprises the effector mechanism for an immunological reaction, resulting in the elimination of grafted cells.

5.2.1. *The blood–brain barrier*

The maintenance of homeostasis in the internal milieu of the brain is essential for normal brain function and is partially dependent on the blood–brain barrier complex. The most important component in this complex is the cerebral capillary endothelium which has a surface area within the brain 5000 times larger than all other intracerebral vessels[184]. Notably, the barrier complex also comprises an interstitial barrier in the so-called 'blood–brain barrier-free regions': the pia–vessel adventitial barrier; the choroid

plexus epithelium–ventricular system barrier; and finally, the blood–meningeal barrier. Thus, all components contribute to effectively shield the brain tissue from direct contact with the blood (for reviews see refs. 24 and 195). The factors that contribute to the restricted passage across the endothelium in the cerebral microvessels are: (1) zonula occludens component of tight junctions[197], (2) transcellular potential difference[44], (3) enzymatic set-up that degrades e.g. many potent plasma-derived putative neurotransmitter substances[99], (4) low pinocytotic capacity[267] and (5) reduced response to inflammatory mediators that normally increase permeability[26,44,195]. In general, substances that normally *readily* pass the barrier system are highly lipophilic, uncharged and relatively small[24,195]. The cerebral endothelium promotes transport of desired substances such as glucose[43] and certain amino acids[13,178] against chemical gradients. Some of the transport and barrier functions mentioned earlier are partially energy-dependent. Rat brain microvessel endothelial cells contain a higher number of mitochondria than other endothelial cells, reflecting a high energy demand[179].

Thus, the proteins or cells of the immune system cannot pass the barrier under normal conditions, but may do so under abnormal conditions: viruses may be transported into the brain in conjunction with lymphocytes[238]. In fact, activated helper T-cells have recently been reported to cross the intact blood–brain barrier[260]. The same study also reports that long-lived, antigen-specific lymphocytes, that are not in an activated state do not pass the barrier, even if administered in high numbers[260]. The passage is thought to be mainly mediated by lymphocyte properties, such as degrading enzymes produced by activated lymphocytes[169]. However, the passage of activated lymphocytes into the brain seems to be confined to the postcapillary venules[260] and therefore other factors, such as vessel properties or homing receptors, may well be involved.

In multiple sclerosis patients, T-cells labeled by fluorescent antibodies that are injected into the blood, can be recovered from the CSF[98]. B-cells can also pass the barrier into the CSF in MS patients. This is illustrated by the fact that after systemic immunization with tetanus toxoid, B-cells that produce anti-tetanus immunoglobulin can be recovered from the CSF[207]. However, it is important to stress that MS patients do not possess a normal barrier complex and they exhibit an abnormal MHC class II expression on cerebral vessels[249].

When considering the contribution of the barrier to the immunologically privileged status of the brain, it is important to remember that highly immunogenic tissues such as skin, which do not form a barrier complex[234], still exhibit a prolonged survival when grafted to the brain[106]. This indicates that the blood–brain barrier cannot be the sole contributor to the immunologically privileged status of the brain.

In summary, whereas the blood–brain barrier was previously thought to constitute a complete barrier to components of the immune system, there is today evidence for a limited passage of immunocompetent cells under certain conditions.

5.2.2. *Local immunosuppressive factors/agents. Systemic effects of lesions in the brain*

As early as in 1923 Murphy and Sturm[164] proposed that the brain could produce substances that suppress immune reactions. However, there still exists very little experimental evidence in support of this hypothesis. Glucocorticosteroids, α-fetoprotein and prostaglandin E_2 are well established down regulators of MHC class I and II antigen expression[252], and they could theoretically act as local immunosuppressive factors in the brain. Interestingly astrocytes have been shown in vitro to produce prostaglandin E_2 (ref. 73), but whether this plays a significant physiological role is unknown. Observations have indicated that active lymphoid cells may be confined to the perivascular spaces of postcapillary venules in the brain, without migrating further into the parenchyma[180,260]. This could suggest that the brain parenchyma produces a substance which hinders local lymphocyte migration.

It should be kept in mind that under certain circumstances some lesions in the brain can affect systemic immunity by interrupting the hypothalamic-pituitary axis which participates in the regulation of hormonal levels in the systemic circulation. This is most pronounced if the lesions are placed in the pituitary, hypothalamus and the anterior hypothalamus[204]. However, for the large majority of grafting experiments to the brain such lesions are not of relevance.

6. REGULATION OF MAJOR HISTOCOMPATIBILITY COMPLEX (MHC) MOLECULES OF NEURONAL TISSUES

The MHC expression of neuronal tissues is under strict regulation. As the MHC plays a paramount role in the induction of immunological reactions as well as in the execution of immune effector function, the regulation of these structures is discussed separately. Normally in the adult mouse, rat and human, MHC class I and class II molecules are not expressed on neurons, astrocytes and oligodendrocytes[49,70,109,126,134,135,215,218,240,248,268]. However, both in vivo and in vitro studies have shown that murine and human neurons, astrocytes and oligodendrocytes can express MHC I antigens during viral infection[156,218], after exposure to γ-IFN[70,126,134,135,268], or in other inflammatory situations[240,248]. Isolated mouse astrocytes have also been shown to produce α- and β-like IFN activity in response to standard IFN-inducing agents in vitro. The α/β-like IFN produced has been shown to induce high levels of MHC class I antigen expression on a subset of mouse astrocytes in vitro[244]. Under similar conditions astrocytes and cerebral endothelium have also been shown to be able to express high levels of MHC class II antigens[70,126,248,268]. In recent papers, Mason et al.[154] and Nicholas et al.[172] have found that fetal neuronal tissues grafted to ventricular system express high levels of MHC antigens when undergoing rejection. An increased cell surface MHC expression has also been observed on grafted neuronal tissue after syngeneic transplantation[154]. This suggests that the inflammatory response caused merely by surgical trauma is sufficient to induce an increase of MHC antigen expression in fetal neuronal tissue. The exact mediators of this MHC induction during classical inflammation are not known but could include α-TNF[139]. As mentioned previously the presence of MHC molecules is of prime importance for immunological reactions to occur[270]. Nicholas et al.[172] report of preferential neuronal cell survival in rejecting grafts, and suggest that these cells express lower levels of MHC than the surrounding non-neuronal cells within the graft parenchyma. The consequences of low MHC expression is illustrated by the transfer of activated killer T-cells directed against lymphochoriomeningitis (LCM) virus-infected cells to an LCM virus-tolerant animal (neonatally infected with the LCM virus). All the cells in the body that expressed the LCM viral antigens in association with the MHC were killed within two weeks. The LCM-infected brain cells did not express MHC, and therefore still remained alive after 120 days[180].

As MHC antigens appear on brain cells only after induction, the turnover rate of MHC molecules is of particular interest. Unfortunately, little is known about the stability of MHC antigens on brain cells once they have been expressed and after the original inductive stimulus has subsided. Studies on other cell types suggest that the half-life of MHC molecules after viral induction may be relatively short, in the range of 15 h[6]. If cerebral endothelial cells are induced to express MHC class II molecules in vivo and the endothelium is subsequently isolated from the inductive agent, the MHC class II antigens cannot be found on the cell surface 72 h later in an in vitro culture[157]. Table VII summarizes brain tissue cells that express MHC antigens in and the effect of some inductive agents on MHC antigen expression.

7. IMMUNOLOGICAL EXPERIENCE FROM RECENT EXPERIMENTS WITH ALLO- AND XENOGRAFTS OF NEURONAL AND SOME RELATED TISSUES

7.1. *Intraparenchymal grafts*

The first recorded trials involving grafting of neuronal tissue across species barriers stem from almost a century ago. In 1890 Thompson reported that cortical tissue grafted from a cat to a dog, and vice versa, survived in all the 3 xenograft cases studied[245]. Two of the animals were studied only after a maximum of 4 days graft survival and therefore these results are difficult to interpret. In view of present knowledge of the general conditions required for survival of neuronal grafts it is likely that a third animal, that was studied 7 weeks after transplantation, only contained the remnants of resorbed graft tissue. Although several successful intracerebral graft studies followed during the first half of this century (see section 4.2), it is only over the past 6 years that the survivability of allo- and xenografted neuronal tissue has specifically been addressed. These studies have shown that the survival of intracerebral neuronal allo- and xenografts is reduced compared to syngeneic transplants. Nevertheless it is, in certain circumstances, possible

TABLE VII

Expression of MHC molecules in CNS tissue cell components.

Tissue	*Normal*		*Inductive agent present*		*Inductive agent*
MHC class:	*I*	*II*	*I*	*II*	
Neurons	–	–	+	–	γ-IFN, TNF-α, rejection
Astrocytes	–	–	+	+	γ-IFN, virus infection, rejection, TNF-α
	–	–	+	(+)	αβ-IFN
Oligodendrocytes	–	–	+	–	γ-IFN, virus infection
Endothelial cells	+	–	+	+	Inflammation (MS, SLE), γ-IFN, rejection
Microglia	+	+			
Dendritic cells	+	+	++	++	γ-IFN
Choroid plexus	–	–	+	+	virus infection
Ependyma	–	–	?	?	

–, absent; (+), low amounts; +, present; ++, high levels.
Based on refs. 74, 112, 126, 139, 154, 156, 170, 172, 239, 240, 244, 248, 268.

for neuronal allo- and xenografts to survive and function for prolonged periods without immunosuppression.

In the adult rodent brain parenchyma systematic experiments with histoincompatible neuronal grafts have been conducted mainly in two anatomical sites: the striatum and the hippocampus (see sections 7.1.1. and 7.1.2.). Extraparenchymal intraventricular grafts are dealt with separately in section 7.2 as the immunological reactions in this site may be quite different to other intracerebral sites.

7.1.1. *Striatum*

DA neurons have been found to survive well when grafted from rat fetuses to the adult rat striatum, in a model of direct relevance to PD[15,186]. In this model the graft recipients are first given a unilateral lesion of the mesostriatal DA pathway using the neurotoxin 6-hydroxydopamine (6-OHDA), resulting in an easily quantifiable motor asymmetry[211,254].

In the first xenograft study, solid pieces of fetal mouse mesencephalic tissue were placed in cortical cavities overlying the caudate-putamen[17]. In 10 out of 18 (55.5%) cases, surviving DA neurons were found either as graft tissue in the cortical cavity or, in most cases, as scattered cells that had migrated into the caudate-putamen proper. In a subsequent study which used the cell suspension technique, xenografted mouse DA neurons survived in 3 out of 7 (43%) rats for up to 42 days[32]. However, using the same technique there were no surviving grafts in 15 rats after 6 months[37], which may indicate that there occurs a time-related continuous loss of xenografts. Similarly, xenografted rabbit fetal mesencephalic cells have shown very poor survival without immunosuppression[66,80]. Recent experiments have shown that fetal allografts containing DA neurons can survive intraparenchymal grafting to the striatum of mice[263] and adult monkeys[9,10,223] without the use of immunosuppression. Prior to grafting, the primate recipients were injected with the DA neurotoxin 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) which results in severe parkinsonian symptoms. In the best cases, these neurological deficits were clearly reduced after the transplantation surgery[76,196].

Human fetal mesencephalic tissue of different ages has also been grafted to 6-OHDA-denervated rats which were immunosuppressed with cyclosporin A[33,36,237]. In studies utilizing the cell suspension technique, good survival of DA neurons was obtained when the graft tissue was taken from 9-postconception-weeks old, or younger, fetuses, whereas older tissue yielded poor or no DA neuron survival[33,36]. This is presumably related to the similar donor-age restrictions found when using rat fetal donor tissue[16,30,35]. In contrast to the good survival in cyclosporin A-treated rats there was no graft survival after 20 weeks when human DA neurons were implanted in non-immunosuppressed rats. However, Kamo et al. have reported that cultured fetal human spinal cord cells[124], and cells from fetal human sympathetic ganglia[123], survive grafting into the striatum or a cortical cavity for several weeks, even in non-immunosuppressed rats.

TABLE VIII

Survival of fetal neural tissue homotopically grafted into adult hosts in settings with well-defined immunogenetical differences

A. Mesencephalic grafts into the striatum

Donor species and strain	*Donor age E (days, w) CRL (mm)*	*Grafting technique*	*Host species and strain*	*Immunosuppression*	*Survival rate (%)*	*Observation time (days)*	*Host immunization*	*Graft combination*	*Ref.*
Mouse, NMRI	E 16–17	Sol	Rat, S.D.	None	55.5	180	–	xeno	17
Mouse, A.SW	E 13–15	Susp	Rat, S.D.	None	43	42	–	xeno	32
Mouse, A.SW	E 13–15	Susp	Rat, S.D.	CyA	100	42 (21–140)	–	xeno	32
Monkey	CRL 170	Dis	Monkey	None	100	69	–	allo	223
Monkey	CRL 13	Dis	Monkey	None	100	56	–	allo	223
Rabbit	E 21	Susp	Rat	None	<10	42	–	xeno	66
Rabbit	E 14–17	Susp	Rat, S.D.	CyA	57	63	–	xeno	80
Rabbit	E 14–17	Susp	Rat, S.D.	None	few cells	63	–	xeno	80
Monkey	CRL 15–16	Dis	Monkey	None	100	63	No	allo	10
Monkey	CRL 18–45	Dis	Monkey	None	100	120–180	No	allo	76
Mouse, C3H/He	E 13	Susp	Mouse, C57Bl	None	100	60	–	allo	218
Human	E 6.5–8 w	Susp	Rat, S.D.	CyA	100	140	Yes, Ig	xeno	36
Human	E 11.5 w	Susp	Rat, S.D.	CyA	20	140	Yes, Ig	xeno	36
Human	E 8 w	Susp	Rat, S.D.	None	0	140	Yes, Ig	xeno	36
Human	E 9–12 w	Sol	Rat, S.D.	CyA	100	70–80	–	xeno	237
Mouse, A.SW	E 13–14	Susp	Rat, S.D.	None	0	175	Yes, Ig	xeno	37
Mouse, A.SW	E 13–14	Susp	Rat, S.D.	CyA, 3 w	33	175	Yes, Ig	xeno	37
Mouse, A.SW	E 13–14	Susp	Rat, S.D.	CyA, 6 w	21	175	Yes, Ig	xeno	37
Mouse, C57Bl	E 13–15	Susp	Mouse, A.SW	None	100	42–49	Yes, cells	allo	263
Mouse, A.	E 13–15	Susp	Mouse, A.SW	None	100	42–49	Yes, cells	allo	263

B. Septal or hippocampal grafts into the hippocampus

Rat, S.D.	E 15–17	Sol	Rat, Wistar	None	80	90	–	allo	149
Mouse, C57Bl	E 15–17	Susp	Rat, S.D.	None	80	up to 120	–	xeno	51
Mouse, C57Bl	E 15–17	Susp	Rat, S.D.	None	100	63	–	xeno	51
Rat, Wistar	E 16–17	Sol	Rat, Wistar	None	82	120–180	–	allo	256
Rat, Wistar	E 16–17	Sol	Rabbit	None	67	90–180	–	xeno	256
Mouse, C57Bl	E 16–17	Sol	Rat, Wistar	CyA	65	35	–	xeno	72
Mouse, C57Bl	E 16–17	Sol	Rat, Wistar	CyA	43	56	–	xeno	72
Mouse, C57Bl	E 16–17	Sol	Rat, Wistar	None	36	35	–	xeno	72
Mouse, C57Bl	E 16–17	Sol	Rat, Wistar	None	18	56	–	xeno	72
Human	E 9–10 w	Susp	Rat, S.D.	CyA	60	30	Yes, Ig	xeno	174

C. Intraventricular grafts

Donor species and strain	*Donor age*	*Tissue grafted*	*Grafting technique*	*Host species and strain*	*Immunosuppression*	*Site of graft*	*Survival rate (%)*	*Observation time*	*Host immunization*	*Graft combination*	*Ref.*
Bovine	Adult, cult	Adrenal	Susp	Rat, S.D.	None	Lat. vent	100	60	–	xeno	187
Rat, BN	E 18–19	S.Nig.	Sol	Rat, F344	None	Lat. vent	81	210	Not found	allo (MHC+)	77
Mouse, BALB/c	P 0	Cort	Sol	Rat, Wistar	CyA, 2 w	3rd vent	100	28	–	xeno	114
Mouse, BALB/c	P 0	Cort	Sol	Rat, Wistar	None	3rd vent	0	28	–	xeno	114
Mouse, BALB/c	E 17–19	Cort	Sol	Rat, PVG	None	3rd vent	0	30	–	xeno	154
Rat, PVG	E 17–19	Cort	Sol	Rat, AO	None	3rd vent	54	30	–	allo (MHC+)	154
Rat, PVG	E 17–19	Cort	Sol	Rat, AO	None	3rd vent	25	60	–	allo (MHC+)	154
Rat, AO	E 17–19	Cort	Sol	Rat, PVG	None	3rd vent	80	30	–	allo (MHC+)	154
Rat, PVG(AO)	E 17–19	Cort	Sol	Rat, AO	None	3rd vent	86	60	–	allo (non MHC)	154
Rat, PVG	E 17–19	Cort	Sol	Rat, PVG/AO	None	3rd vent	100	60	–	allo (MHC)	154
Rat, BN	E 14	Cort	Susp	Rat, DA	None	Ventricle	0	30	Yes, Ig	allo	150
Mouse, AKR	E 16–19	Sup op	Sol	Mouse, HPG	None	3rd vent	100	1–90	–	allo	28

E, embryonic day; E w, embryonic week; CRL, crown rump length (mm); Sol, solid grafts; S.D., Sprague–Dawley; CyA, cyclosporin A; Susp, suspension grafts; Dis, dissociated graft; xeno, xenogeneic; allo, allogeneic; A., A.SW, C3H/He, C57Bl, inbred mouse strains; NMRI, outbred mouse strain; P 0, postnatal day 0; Lat. vent, lateral ventricles; 3rd vent, third ventricle; Cort, cortex; Sup op, supra optic area; MHC+, grafts differing at MHC and non-MHC antigens; non-MHC, grafts differing at non-MHC antigens; MHC, grafts differing at MHC antigens only; AO, BN, F344, DA, PVG, PVG(AO): inbred rat strains; AKR, BALB/c, HPG: inbred mouse strains; S.Nig., substantia nigra.

Experiments involving incompatible grafts of fetal mesencephalic tissue into the striatum are summarized in Table VIIIA.

7.1.2. *Hippocampus*

Solid grafts of rat hippocampus and septum have shown a 67% survival rate when placed in a surgical cavity overlying the hippocampus in rabbits without immunosuppressive treatment[256]. A slightly lower survival rate (36%) has been found after 5 weeks with solid intraparenchymal xenografts of fetal mouse hippocampal tissue implanted into the hippocampus of adult rats[72]. With a longer observation period of 8 weeks merely 18% of the grafts were still viable[72] suggesting a continuous loss of grafted cells due to rejection.

Using the cell suspension technique, mouse septal tissue, rich in cholinergic neurons, has successfully been xenografted to the rat hippocampus. At 17 weeks postgrafting survival, the rate was estimated to approximately 80%. In the rats with surviving xenografts there is a restoration of the normal hippocampal acetylcholinesterase lamination pattern[52], choline acetyltransferase levels[50] and a partial reversal of lesion-induced deficits in conditioned learning[51]. Interestingly, the cell suspension implants were difficult or impossible to discern as discrete tissue masses, and surviving grafts were identified only as scattered acetylcholinesterase-positive cells or by the existence of acetylcholinesterase-positive stained fibers in the host hippocampus. In an allogeneic setting grafts of solid basal forebrain tissue displayed approximately 80% survival rate when placed in a cavity adjacent to the hippocampus[149]. Experiments of allogeneic and xenogeneic grafts into the hippocampus are summarized in Table VIIIB.

7.2. *Intraventricular grafts*

The survival of grafts in the ventricular system might be somewhat different from intraparenchymal grafts. Therefore the results with grafts placed in the ventricular system are treated separately. Table VIIIC lists a few examples of allogeneic and xenogeneic grafts placed in the ventricular system. The frequency of surviving xenografts in rats varies between 0% for fetal mouse cortical tissue[114] to 100% for adult bovine adrenal medulla[187]. The variation in allogeneic graft survival is likewise large: from 0% in one particular graft combination of fetal cortical grafts in suspension[150] to 25% for solid grafts differing at MHC and non-MHC antigens or 100% for grafts with incomplete histoincompatibility, at 60 days postgrafting[154].

7.3. *Immunosuppression with cyclosporin A*

The immunosuppressive drug cyclosporin A (10 mg/kg daily i.p.) markedly increases the survival of cell suspension grafts of mouse DA neurons implanted in rat striatum. In an initial study, all of the immunosuppressed rats exhibited behaviorally functional grafts and large numbers of surviving DA neurons 6 weeks after transplantation, whereas only 3 out of the 7 non-immunosuppressed control rats possessed small surviving grafts[32]. Cyclosporin A has also been found to increase the survival of intraparenchymal xenografts of rabbit DA neurons[80] and mouse hippocampal tissue[72], and cortical mouse grafts placed in the ventricular system of rats[114,115].

In our initial study, two rats in the cyclosporin A-treated group were studied for a further 4 and 14 weeks, respectively, after termination of the 6-week immunosuppression period. Both these rats retained large grafts, rich in DA neurons, that remained functional[32]. This finding prompted us to investigate if short-term cyclosporin A treatment could support long-term survival of intracerebral DA grafts. Using the same model we grafted fetal mouse mesencephalic tissue to rats that received no immunosuppression, or were allocated to either 10-, 21- or 42-day cyclosporin A treatment schemes. Three to 6 weeks after transplantation several rats in the cyclosporin A-treated groups showed, as expected, behavioral evidence of surviving functional transplants. However, 3 weeks or more after the cessation of the respective cyclosporin A treatment the majority, but not all, of the rats exhibited evidence of rejection (loss of behavioral graft effect)[37]. Thus, at morphological analysis, 6 months after transplantation, the large majority of immunosuppressed rats lacked surviving grafts, suggesting that short-term immunosuppressive treatment cannot reliably support long-term survival of intrastriatal fetal DA cell suspension grafts between mice and rats.

8. OTHER EXPERIENCES WITH IMPLICATIONS ON THE IMMUNOLOGICAL REACTION AGAINST GRAFTED NEURONAL TISSUES

8.1. *Grafted cells, characteristics and development at the graft site*

The antigenicity of fetal neuronal tissue is related to the developmental stage and the mode of tissue preparation. The cellular components in a graft and the cell surface structures of fetal neuronal cells used for transplantation have not been systematically studied. Careful separation of the neuronal tissue from the overlying mesenchymal covering will largely remove pleuripotent mesenchymal cells, hematopoietic cells and stem cells all of which have the potential to develop into non-neuronal tissues and express MHC antigens. Thereby the graft tissue consists mainly of maturing neurons, glial cells, endothelial cells and their respective precursors. Following dissection the tissue can be implanted either as a solid piece or injected as a cell suspension.

Normally, there are very low or no levels of MHC class I and II molecules expressed on the neuro- and glio-blasts in the graft at the developmental stage when they are grafted[90,109,218]. The trypsin treatment sometimes used to prepare cell suspensions is thus unlikely to affect the MHC expression. Human MHC molecules are not trypsin-sensitive when membrane-bound[131], whereas mouse MHC (H-2) can be degraded by trypsin if sufficiently high concentrations of the enzyme are used. In contrast, the vascular endothelium can express class I antigens, and under certain conditions may also express class II MHC antigens[113]. It is likely that fetal neuroblasts express other potentially polymorphic molecules such as Thy.1 (refs. 39, 40, 91), cell adhesion molecules[46, 64,68] and species-specific cell surface glycoproteins[258]. Notably, solid grafts differing only on Thy molecules (a minor transplantation antigen), have survived implantation into the mouse cerebellum for over 180 days without histological signs of rejection[56]. Moreover, it is important to remember that cells in the grafted tissue can be stimulated to express transplantation antigens after graft surgery, as discussed earlier. Fetal neuronal tissue has also been shown to be capable of immunizing a host after intracerebral grafting (see section 5.1.4.) and cause rejection of a prior intracerebral graft if grafted under the kidney capsule[154].

8.2. *Vascularization and origin of vessels*

The injection of 1.5 μl volume of cell suspension into the rat brain causes a transient breakage of the blood–brain barrier[37] followed by local edema. Initially, the grafted cells are probably dependent on diffusion from distant blood vessels for their nutrition. Solid tissue pieces that are grafted, either of heterotopic origin (e.g. skin graft) or of homotopic origin, are likely to differ from cell suspension grafts with regard to the origin of the vessels in the graft and the time course of revascularization. Skin grafts (which have been prepared in such a way that they do not contain any intact vessels when grafted) are vascularized on day 4–5 after transplantation to the brain[234]. Primordial vessels in the grafted tissue develop into capillaries and larger vessels within the grafted skin tissue and these vessels seemingly hook up to host vessels at the graft–host interface[234]. The same process has been found to be operative for solid embryonic neuronal grafts placed in the third ventricle[28]. The origin of the vessels seems to vary in neuronal cell suspension grafts with some cells in the vessels coming from the host and others being derived from the grafted tissue[28]. The origin of the vasculature in the graft may be of great importance, since the endothelial lining of blood vessels can express high levels of donor MHC and may be the first site of attack of an immune response against the graft. Inflammatory mediators have angiogenic properties and participate in the revascularization process[189]. For example, IL-1 released by activated astrocytes and macrophages can induce proliferation of endothelial cells[183] and several other specific endothelial growth factors have been isolated from brain tissue[2,121].

8.3. *Blood–brain barrier development in intracerebral grafts*

The establishment of a blood–brain barrier and the time course of its development in the graft may be of some importance for the survival of the graft. Although lymphocytes in a certain activation stage can pass the barrier, the passage of lymphocytes may still remain restricted. Interestingly, vessels in peripheral

tissue grafted to the brain do not form an intact blood–brain barrier[233,241], whereas CNS tissue grafted to the periphery can attain a functional blood–brain barrier[233]. It has been hypothesized that the glial cells surrounding the vessels induce the blood–brain barrier properties[57]. This idea is supported by studies where purified astrocytes have been injected into the anterior chamber of the eye. The vessels from the iris invading the aggregated astrocytes form blood–brain barrier properties with typical tight junctions and these vessels exclude macromolecular tracers[120].

Slightly conflicting results have been obtained when studying the development of a blood–brain barrier in intracerebral neuronal grafts. Rosenstein[202] has described incomplete barriers that leak macromolecular tracers, horseradish peroxidase and endogenous proteins, for several weeks after the grafting of solid fetal or neonatal neuronal tissue into the cortex or the fourth ventricle. In contrast, Broadwell et al.[28] have demonstrated an intact blood–brain barrier in solid fetal neuronal grafts placed in the third ventricle in mice after 7 days, when revascularization occurred. Moreover, we have recently found that the vessels in an intrastriatal cell suspension graft in rats will attain a low permeability to Evans blue-labeled macromolecules within 12 days after grafting[37]. Similar results of a tight blood–brain barrier to macromolecules have been found in solid fetal grafts in the hippocampus[242], in cell suspension grafts in the thalamus[63] and finally in intracortical solid grafts in neonatal rats[140]. In summary, it would seem that fetal neuronal tissue grafts can form a blood–brain barrier to macromolecules in most situations. However, it remains unclear whether the development of an intact blood–brain barrier plays an important role in hindering or limiting immune responses to intracerebral grafts.

8.4. *Inflammatory response*

Inevitably surgical trauma evokes an inflammatory response in the brain around the implantation site. Host vessels are likely to be disrupted, which will unleash the coagulation cascade and the aggregation of thrombocytes. Debris of disrupted and dead cells in the graft are probably cleared from the area by phagocytosing cells such as resident microglia and blood-derived macrophages and monocytes. In this process a multitude of different inflammatory mediators are likely to be released[138]. The effect of these inflammatory mediators on the permeability across the cerebral endothelium, or their effects on the blood flow are not known in detail, although their effects are likely to be less marked than in the peripheral circulation since the cerebral vessels lack receptors for several of the permeability and vasoactive mediators[26,44,195].

Inflammatory mediators may be released from the endothelial cell itself[138], resident astrocytes[73] and microglia[188], or invading cells such as platelets, monocytes and leukocytes[138]. In the transplantation situation, inflammatory mediators can affect the expression of MHC antigens. The endothelial cells in orthotopic skin grafts undergoing rejection express higher levels of MHC class I antigens and are stimulated to express class II antigens[59]. As mentioned previously the normally low level of MHC expression is slightly elevated in grafted CNS tissue even in syngeneic grafting combinations[154]. One possible candidate for this inflammatory induction signal is platelet-derived growth factor, PDGF, which has been shown to induce MHC antigen expression on B-cells in vitro[3], and is released by aggregated platelets and from activated monocytes[203]. Other candidates are the astrocyte-derived IFN-like substance recently described[244] or α-TNF[139].

Another important step in the inflammatory response may be the release of IL-1, produced either by resident astrocytes, microglia or invading monocytes[62]. IL-1 is strongly lymphotactic[163] and will induce migration and proliferation of astrocytes[87] (see Fig. 5).

8.5. *Mechanisms of rejection of intracerebral neuronal grafts*

Converging data point to the importance of classical T-cell infiltration in intracerebral grafts undergoing rejection. Mason et al. have found approximately equal proportions of helper T-cells, killer T-lymphocytes, and macrophages in neuronal grafts that are undergoing rejection in the ventricular system[154]. Similar results have recently been published for solid cortical allografts in the lateral ventricle[172] and intraparenchymal solid xenogeneic grafts in the hippocampus[71]. The accumulation of inflammatory

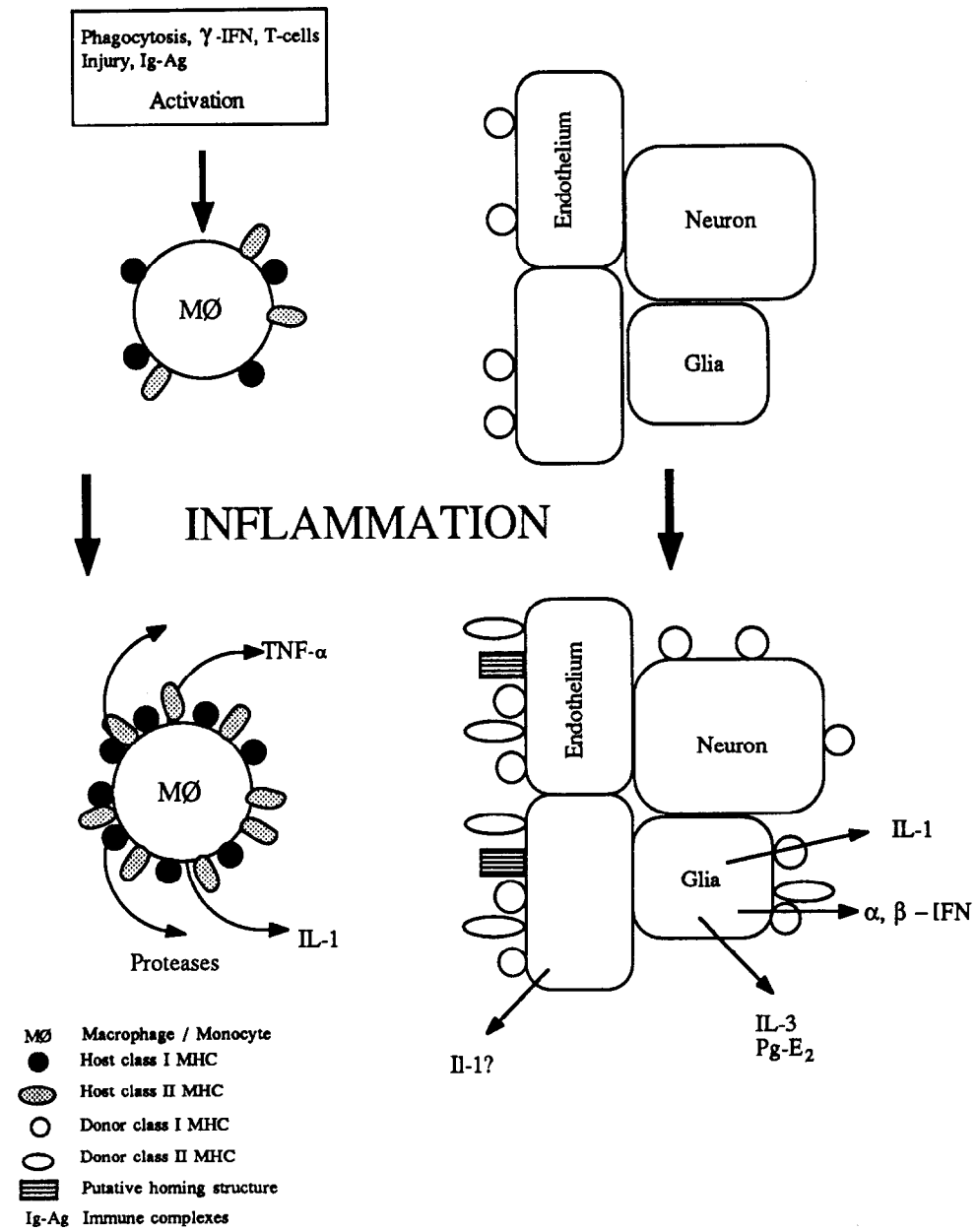


Fig. 5. Schematic drawing of inflammatory events leading to enhanced MHC class I and II expression on the grafted tissue. The exact source of the mediators are not known in a transplantation situation, but monocytes and macrophages (MØ), platelet-derived inflammatory mediators, the coagulation and complement cascades may produce mediators after activation. Stimulus for macrophage activation is i.a. phagocytosis, injury and immunological factors such as immune complexes and γ-IFN. Endothelium and glia cells are also able to produce mediators that can take part in inflammatory events, e.g. lymphotaxis.

cells and lymphocytes peaked in the graft area on the third week after a xenogeneic solid neuronal graft placed in the hippocampus[71]. Lymphocytes were also present in the meninges, choroid plexus and in the perivascular spaces. Following a rejection process induced by an orthotopic skin graft, Houston et al.[110] have observed MHC expression and cellular infiltrates in mouse retinal xenografts implanted in neonatal rats. Lymphocytic infiltrates peaked when graft degeneration was evident on day 8, and were particularly prominent in the perivascular spaces and in the surrounding host brain.

The observation that the T-cell-specific immunosuppressive drug cyclosporin A, as discussed in section 7.3, increases the survival of xenogeneic grafts also supports the notion that the T-cell mediated immunological reactions comprise a crucial component in graft rejection also in the brain.

8.6. *Late rejection*

The absence of lymphocytes in or around a graft does not necessarily imply that an immunological reaction has not taken place. Nor does it exclude a slow ongoing chronic rejection. Even with intracerebral fetal neuronal grafts the necessary stimuli for rejection seem to be present for long time periods in rats which have received intrastriatal xenografts. For example, a xenograft is in general rejected within 3–6 weeks after the withdrawal of therapeutic immunosuppression[37]. This late rejection may be initiated due to the presence of foreign endothelium in the vasculature, which could provide a chronic stimulus for the host immune system. This would lead to a high number of circulating graft-specific and activated lymphocytes. Similarly, well integrated allografts that have survived for prolonged periods may be at risk of rejection if there is an inflammation or unrelated activation of the immune system, which would subsequently increase the MHC expression on vessels and the grafted tissue.

8.7. *Risk of autoimmune reactions against the normal host nervous system and potential harmful effects of a rejection in the central nervous system*

The immune system is implicated in several neurological diseases, including multiple sclerosis and acute polyradiculoneuritis (inflammatory polyneuritis)[1]. The brain contains several antigenic structures that often are regarded as potent autoantigens, most notably myelin basic protein, apoprotein and some glycolipids[89]. In the experimental model known as experimental allergic encephalitis (EAE), immunization with a small octapeptide derived from myelin basic protein is sufficient to cause a demyelinating disease in specific strains that are prone to develop the disease[193,249,260]. To date no reports of similar lesions against normal host nervous tissue due to neural transplantation have been published. Notably, tissue used for cell suspension grafts are taken at a developmental stage when they do not contain any myelin components[227] which limits the risk of EAE-like conditions. Release of the putative autoantigens after normal tissue damage or neurosurgery have not been found to be a major cause of demyelinating or aberrant autoimmune reactions. There is a theoretical possibility that the rejection of an established neuronal graft could cause perturbations in the adjacent host vasculature such that parts of the normal host brain are afflicted. However, studies with rejection of functional xenografts in rats do not indicate that the host brain becomes significantly involved[37].

8.8. *Tolerance induction*

The topic of immunological tolerance is a complex subject beyond the scope of this review[104,177,201]. However, specific immunological unresponsiveness against grafted tissue is a possible explanation for why intracerebral neuronal allo- and xenografts can function up to 7 months after grafting[17,196]. If complete tolerance has evolved, a second skin graft from the same donor should then be permanently accepted. This was not the case in a study by Nicholas et al. who observed a swifter rejection of fetal neuronal allogeneic intraventricular grafts if an orthotopic skin graft was transplanted two weeks after intracerebral grafting in adult mice[172]. Tolerance is usually only accomplished if grafting is performed when the host is 1–2 days of age[159]. A recent study indicates that good survival of xenogeneic neuronal tissue implanted in the parenchyma is possible in hosts up to day 12 postnatally[150]. The xenograft is then accepted for up to 60 days, without any signs of rejection until challenged with a skin graft. Rejection then occurs. This may be an example of partial tolerance, a poorly understood immunological phenomenon[102]. One possible explanation for partial tolerance is that the lymphocytes that can react strongly with the first grafted tissue are eliminated during the neonatal period, but clones of lymphocytes with lesser avidity may be still present in the host. These clones do not harm the grafted tissue until an adequate second stimulus, such as an orthotopic skin graft, activates sufficiently many host lymphocytes that can react with the grafted tissue and cause a full rejection of both grafts.

8.9. *Influence of age on the immune system*

The age of the graft recipient may affect the immune response towards the grafted tissue in various ways. If the host is neonatal or very immature, permanent immunological tolerance is likely to occur (see section 8.8). At the other end of the scale, it has also been proposed that if the recipient of the graft is very old the immune response may be different from that of a younger individual. Indeed, there are reports in the literature indicating that there can occur an age-dependent decline in certain immunological parameters[152] and that the decline is partly species- and strain-specific[190]. The influence of age on immunological reactions, and the putative role of immune reactions in the aging process is a vast subject beyond this review[152,257]. However, there are no reports indicating a reduced graft rejection capacity in elderly, nor does clinical experience of transplantation in elderly support that other regimes for immunosuppression be used in aged individuals.

9. CONCLUSIONS FROM RECENT NEURONAL GRAFTING EXPERIMENTS

9.1. *Defining graft survival and monitoring graft rejection*

When studying incompatible intracerebral neuronal grafts, it is essential to avoid overestimation or underestimation of the extent of immunological rejection. The identification of a few graft-derived cells in the brain may lead to the belief that there is no graft rejection. Also when discussing grafts with a low survival rate one must directly compare the survival rate with that which a similar technique would yield in a syngeneic combination or in immunosuppressed hosts, in order to be able to define the exact role of immunological rejection.

A major problem in monitoring grafted fetal neuronal tissue is the lack of specific methods to demonstrate an ongoing immunological reaction in vivo. There are no tests available that measure the status of the immune system on a day-to-day basis and which could then reveal an increased risk of imminent rejection. When other organs are grafted there are usually simple measures of the function of the grafted organ available, which can be monitored. With signs of poor graft function measures against rejection episodes can be taken. In addition, graft biopsies can confirm an ongoing rejection and support the adoption of special measures against rejection. There are no such functional tests available in neuronal tissue grafting before the grafted cells give rise to functional effects. In general, the long latency between the graft surgery and the development of functional effects renders the neuronal transplant investigator blind for an extended time period during which immunological attacks may take place against the grafted tissue.

9.2. *Possible site-dependent differences in graft survival*

There is a tendency towards better survival of incompatible grafts in the hippocampus as compared to the striatum, Table VIIIA and B. A similar suggestion of differential graft survival was made by Head et al. who proposed that histoincompatible skin grafts placed in the occipital lobe survive better than those implanted in the forebrain, but the data are unconvincing[106]. Since most of the experiments have been performed in rats, the finding that interstitial fluid from the forebrain is preferentially drained into the lymphatics and fluid from the hindbrain drains into CSF may bear some relevance[243].

When comparing the fate of grafts in the ventricular system with the fate of intraparenchymal grafts, no definite conclusion can be made regarding which site is most hospitable for grafts. One can propose that intraventricular grafts can interact with the hosts in at least two principally different manners: firstly, small pieces of graft tissue can probably receive full nutritional support from the cerebrospinal fluid. In the absence of blood vessels they are almost completely out of reach of the immune system and this might allow complete survival across major histocompatibility barriers[187]. Secondly, solid grafts in the ventricles have been seen to receive a blood supply from neighboring brain tissue. The vessels within solid grafts are likely to be completely derived from the donor, and appear to hook up to host vessels at the interface between graft and host[28]. The grafted tissues will then be at greater risk for rejection than the non-vascularized grafts in the ventricles, or may also be rejected more quickly than intraparenchymal

grafts that might contain vessels of a mixture of both graft and host origin.

To summarize, both the mode of tissue preparation and the site of grafting affects not only the neurobiological properties of the grafts but also their immunological status.

9.3. *Regrafting*

In a clinical setting it is conceivable that regrafting to the brain would be necessary, either in order to improve poor function of an existing graft or to permit reinnervation of additional target areas, for example contralateral to an initial graft. Although it is well known that intracerebral grafts placed into a sensitized host do not survive[106,154,158,194], there are no published reports concerning intracerebral regrafting of allogeneic or xenogeneic neuronal tissue. Studies with repeated intracerebral grafting of skin from an incompatible donor have given some indication that a second graft may jeopardize the survival of a successful first graft[85,86].

9.4. *Differences in survival between intracerebral allo- and xenografts*

It seems as if allografts (even with complete histoincompatibility) generally survive significantly better than xenografts in the brain (see e.g. Table VIII). These empirical findings do not conform to the findings obtained in peripheral sites and are not easily explained theoretically. In the xenogeneic setting the host is likely to possess a higher number of clones that recognize antigenic determinants on both MHC and non-MHC structures than in the allogeneic setting. This difference between xeno- and allografts is not necessarily reflected in graft survival time in the periphery where the immune response is working under unrestricted conditions. However, the difference in host capacity to recognize graft antigens may be a decisive factor for intracerebral graft survival.

10. PROPOSED SEQUENCE OF EVENTS FOLLOWING INTRACEREBRAL IMPLANTATION OF FETAL NEURONAL CELL SUSPENSION GRAFTS

A possible sequence of events of cellular and tissue responses after intracerebral grafting of immunogenetically different fetal neuronal tissue in cell suspension into a healthy adult rodent brain is briefly described.

The donor tissue is prepared from newly aborted fetuses: the meninges are carefully dissected away, and the desired neuronal region is mechanically disrupted to form a suspension of free cells and cells in small aggregates. When stereotaxically injected into the brain, the implantation causes transient local edema, and a localized rupture of the blood–brain barrier. Rupture of small vessels will evoke a coagulation process and the concomitant tissue damage will give rise to an inflammatory process. This leads to activation of astrocytes to start migration and proliferation. Activated astrocytes are capable of producing angiogenic factors and possibly IL-1 and IFN-like substances. The activated astrocytes produce increased amounts of glial fibrillary protein. Aggregated monocytes and macrophages, some of which might be microglia, are likely to be attracted to the graft area. As the graft-derived cells are exposed to several inflammatory mediators, their MHC expression is likely to be increased. Grafted cells with high MHC level expressed are probably brought to a local lymphatic tissue, either as whole migrating cells or in pieces carried by host-derived defense cells.

Initially the graft survives on diffusion of nutrients and oxygen. Endothelial cells from the donor integrate with the vessels that grow into the graft from the ends of the ruptured host vessels. Revascularization is well underway within 3–4 days and a week after the implantation the blood–brain barrier is reformed. Cells that die in the grafts are scavenged by invading macrophages, resident microglia and activated astrocytes.

Once antigens from the graft reach the most proximal lymphatic tissue there is activation of the immune system. Proliferation of lymphocytes is initiated and activated lymphocytes are released into the blood stream within a few days. The cells circulate until they are homed into the inflammatory region around the wound and graft. Further proliferation of lymphocytes takes place in and around the graft if the antigen against which they are primed is present. The active lymphocytes produce increased amounts of lymphokines which increase the intensity of the inflammation, causing enhanced expression of MHC antigens on host and graft cells. Killer T-cells

TABLE IX

Donor, host and technical factors that influence the outcome of intracerebral neural and non-neural tissue grafting

Transplantation combination	*Factors that affect survival*
I. General aspects	Aseptic conditions
Homotopic tissue (neural tissue to brain)	Donor age, dependent on technique used Ability of graft tissue to survive ischemia
Heterotopic tissue (non-neural tissue to brain)	Ability to survive the immediate postoperative period Duration before revascularization Trophic factors-target and non-target specific Virally infected donor tissue Intra- and postoperative complications, e.g. bleeding and bacterial infections Purity of dissection (e.g. amount of meninges in the graft preparation) Blood–brain barrier formation
Autograft	All factors above (few tissues applicable, adrenal medulla, carotic body)
Syngeneic	All factors above (limited to inbred strains)
II. Immunological aspects	
Allogeneic	All factors above, and in addition the following: Host age Immunological reactions against MHC and non-MHC antigens Content of antigen presenting cells in grafted tissue Content of preformed vessels or precursors of vessels Degree of angiogenesis caused by inflammation Revascularization; source of vessels (host- or donor-derived) Duration of blood–brain barrier rupture and malfunction Degree of inflammation caused by the surgery Response to inflammatory stimuli, amount of MHC induced on vessels and brain tissue Duration of antigenic stimulus from the grafted tissue Duration of activated lymphocytes in circulation Induction of homing structures on vessels Tolerance induction Site of graft in the brain
Xenogeneic	All factors above, and in addition the following: Non-MHC antigens tending to give rise to strong immunological reactions resembling MHC reactivity. Preformed antibodies Matching between trophic factors and receptors?
Regrafting	All factors above and in addition: Specific host immunization present prior to the second graft

can then proceed to lyse cells, possibly starting with graft-derived endothelial cells. Moreover, lymphotoxins released from lymphocytes and macrophages can kill cells which have receptors for the toxins. Finally, the grafted tissue will be more or less disintegrated and engulfed by macrophages. Table IX summarizes donor, host and technical factors that influence the survival of intracerebral grafts. Detailed knowledge of the mechanisms and events may provide means of interfering with immunological reactions, to further prolong the graft survival.

11. A MECHANISTIC HYPOTHESIS FOR GRAFT SURVIVAL: THE BRAIN AS AN IMMUNOLOGICALLY PRIVILEGED SITE FOR TRANSPLANTS

Partly based on the above sequence of events we present a model for graft survival, which may provide a tentative explanation as to why the brain is an immunologically privileged site.

Most of the factors that once were thought to fully explain the privileged status of the brain such as the absence of lymphatic drainage; the absence of anti-

gen-presenting cells; the absence of passage across the blood–brain barrier of lymphocytes and the absence of MHC class I and II antigens on nervous tissue cells have now been shown not to apply within brain tissue under certain conditions. In spite of this, the brain is beyond doubt a privileged site for grafts, when privilege is defined as a prolonged graft survival compared to the survival time in another locus. The reason for this is still unclear.

One hypothetical explanation for prolonged survival is that — although all the components necessary for a normal immune response (graft rejection) are present — the kinetics and degree of the *regulation* of the individual steps in an immune response, following intracerebral grafting, may differ from those in the periphery. Thereby, the simple *delay* in the immune response may result in *prolonged* graft survival.

Further, one can postulate that if certain crucial events, such as MHC expression on grafted brain cells declines before the execution of the next step in the immune reaction, such as the passage of activated lymphocytes across a reformed barrier complex, the normal chain of events in the immune reaction is irreversibly broken, resulting in *permanent* graft survival.

One could therefore regard the privileged status of the brain as a state of quantitative and temporally balanced *regulated* events, which is under control of a large number of cellular mediators, among them lymphokines and other intercellular communication molecules. With an increased understanding of the factors underlying the regulation of the immunological response in grafting in the brain across immunological barriers it would perhaps be possible to interfere with these steps and in a relatively mild way ensure prolonged graft survival, ultimately also in a clinical setting.

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REFERENCES

1 Aarli, J.A., The immune system and the nervous system, *J. Neurol.*, 203 (1983) 137–154.
2 Abraham, J.A., Mergia, A., Whang, J.L., Tumolo, A., Friedman, J., Hjerrild, K.A., Gospodarowicz, D. and Fiddes, J.C., Nucleotide sequence of a bovine clone encoding the angiogenic protein, basic fibroblast growth factor, *Science*, 233 (1986) 545–548.
3 Acres, R.B., Lamb, J.R. and Feldman, M., Effects of platelet-derived growth factor and epidermal growth factor on antigen-induced proliferation of human T-cell lines, *Immunology*, 54 (1985) 9–16.
4 Agid, Y., Javoy-Agid, F. and Ruberg, M., Biochemistry of neurotransmitters in Parkinson's disease. In C.D. Marsden and S. Fahn (Eds.), *Movement Disorders 2*, Butterworths, London, 1987, pp. 166–230.
5 Aizawa, M. and Natori, T., Transplantation immunology in the rat. In Y. Yamamura and T. Tada (Eds.), *Progress in Immunology V*, Academic, Tokyo, 1983, pp. 1439–1448.
6 Andersson, M., Pääbo, S., Nilsson, T. and Peterson, P.A., Impaired intracellular transport of class I MHC antigens as a possible means for adenoviruses to evade immune surveillance, *Cell*, 43 (1985) 215–222.
7 Andres, K.H., von Düring, M., Muszynski, K. and Schmidt, R.F., Nerve fibers and their terminals of the dura mater encephali of the rat, *Anat. Embryol.*, 175 (1987) 289–301.
8 Backlund, E.O., Granberg, P.O., Hamberger, B., Knutsson, E., Mårtensson, A., Sedvall, G., Seiger, Å. and Olson, L., Transplantation of adrenal medullary tissue to striatum in parkinsonism. First clinical trials, *J. Neurosurg.*, 62 (1985) 169–173.
9 Bakay, R.A., Fiandaca, M.S., Barrow, D.L., Schiff, A. and Collins, D.C., Preliminary report on the use of fetal tissue transplantation to correct MPTP-induced Parkinson-like syndrome in primates, *Appl. Neurophysiol.*, 48 (1985) 358–361.
10 Bakay, R.A., Barrel, D.L., Fiandaca, M.S., Iuvone, P.M., Schiff, A. and Collins, D.C., Biochemical and behavioral correction of MPTP Parkinson-like syndrome by fetal cell transplantation, *Ann. N.Y. Acad. Sci.*, 495 (1987) 623–640.
11 Barker, C.F. and Billingham, R.E., The role of afferent lymphatics in the rejection of skin homografts, *J. Exp. Med.*, 128 (1968) 197–220.
12 Barker, C.F. and Billingham, R.E., Immunologically privileged sites, *Adv. Immunol.*, 25 (1977) 1–54.
13 Betz, A.L. and Goldstein, G.W., Polarity of the blood–

brain barrier: neutral amino acid transport into isolated brain capillaries, *Science*, 202 (1978) 225–227.
14 Beutler, B., Greenwald, D., Hulmes, J.D., Chang, M., Pan, Y.-C.E., Mathison, J., Ulevitch, R. and Cerami, A., Identity of tumour necrosis factor and the macrophage-secreted factor cachectin, *Nature (Lond.)*, 316 (1985) 552–554.
15 Björklund, A. and Stenevi, U., Reconstruction of the nigrostriatal pathway by intracerebral nigral transplants, *Brain Res.*, 177 (1979) 555–560.
16 Björklund, A., Schmidt, R.H. and Stenevi, U., Functional reinnervation of the neostriatum in the adult rat by use of intraparenchymal grafting of dissociated cell suspensions from the substantia nigra, *Cell Tissue Res.*, 212 (1980) 39–45.
17 Björklund, A., Stenevi, U., Dunnett, S.B. and Gage, F.H., Cross-species grafting in a rat model of Parkinson's disease, *Nature (Lond.)*, 298 (1982) 652–654.
18 Björklund, A., Stenevi, U., Schmidt, R.H., Dunnett, S.B. and Gage, F.H., Intracerebral grafting of neuronal cell suspensions. II. Survival and growth of nigral cell suspensions implanted in different brain sites, *Acta Physiol. Scand.*, Suppl. 522 (1983) 9–18.
19 Björklund, A. and Stenevi, U., Intracerebral neural implants: neuronal replacement and reconstruction of damaged circuits, *Annu. Rev. Neurosci.*, 7 (1984) 279–308.
20 Björklund, A. and Stenevi, U., Intracerebral neural grafting: a historical perspective. In A. Björklund and U. Stenevi (Eds.), *Neural Grafting in the Mammalian CNS*, Elsevier, Amsterdam, 1985, pp. 3–14.
21 Björklund, A., Lindvall, O., Isacson, O., Brundin, P., Wictorin, K., Strecker, R.E., Clarke, D.J. and Dunnett, S.B., Mechanism of action of intracerebral neural implants: studies on nigral and striatal grafts to the lesioned striatum, *Trends Neurosci.*, 10 (1987) 509–516.
22 Bjorkman, P.J., Spaer, M.A., Samraoui, B., Bennet, W.S., Strominger, J.L. and Wiley, D.C., Structure of the human class I histocompatibility antigen HLA-A2, *Nature (Lond.)*, 329 (1987) 506–518.
23 Block, M.A., Tworek, E.J. and Miller, J.M., Parathyroid homografts in brain tissue, *Arch. Surg.*, 92 (1966) 778–784.
24 Bradbury, M.W., *The Concept of a Blood–Brain Barrier*, Wiley, Chichester, 1979.
25 Bradbury, M.W. and Westrop, R.J., Factors influencing exit of substances from cerebrospinal fluid into deep cervical lymph of the rabbit, *J. Physiol. (Lond.)*, 339 (1983) 519–534.
26 Bradbury, M.W., The structure and function of the blood–brain barrier, *Fed. Proc.*, 43 (1984) 186–190.
27 Bradbury, M.W. and Westrop, R.J., Lymphatics and the drainage of cerebrospinal fluid. In K. Sharpio, A. Marmarou and H. Portnoy (Eds.), *Hydrocephalus*, Raven, New York, 1984, pp. 69–81.
28 Broadwell, R.D., Charlton, H.M., Balin, B.J. and Salcman, M., Angioarchitecture of the CNS, pituitary gland, and intracerebral grafts revealed with peroxidase cytochemistry, *J. Comp. Neurol.*, 260 (1987) 47–62.
29 Brown, M.A., Pierce, J.H., Watson, C.J., Falco, J., Ihle, J.N. and Paul, W.E., B cell stimulatory factor-1/interleukin-4 mRNA is expressed by normal and transformed mast cells, *Cell*, 50 (1987) 809–818.
30 Brundin, P., Isacson, O. and Björklund, A., Monitoring of cell viability in suspension of embryonic CNS tissue and its use as a criterion for intracerebral graft survival, *Brain Res.*, 331 (1985) 251–259.
31 Brundin, P., Isacson, O., Gage, F.H., Stenevi, U. and Björklund, A., Intracerebral grafts of neuronal cell suspensions. In A. Björklund and U. Stenevi (Eds.), *Neural Grafting in the Mammalian CNS*, Elsevier, Amsterdam, 1985, pp. 51–59.
32 Brundin, P., Nilsson, O.G., Gage, F.H. and Björklund, A., Cyclosporin A increases survival of cross-species intrastriatal grafts of embryonic dopamine-containing neurons, *Exp. Brain Res.*, 60 (1985) 204–208.
33 Brundin, P., Nilsson, O.G., Strecker, R.E., Lindvall, O., Åstedt, B. and Björklund, A., Behavioural effects of human fetal dopamine neurons grafted in a rat model of Parkinson's disease, *Exp. Brain Res.*, 65 (1986) 235–240.
34 Brundin, P., Strecker, R.E., Lindvall, O., Isacson, O., Nilsson, O.G., Barbin, G., Prochiantz, A., Forni, C., Nieoullon, A., Widner, H., Gage, F.H. and Björklund, A, Intracerebral grafting of dopamine neurons: experimental basis for clinical trials in patients with Parkinson's disease, *Ann. N.Y. Acad. Sci.*, 495 (1987) 473–496.
35 Brundin, P., Barbin, G., Strecker, R.E., Isacson, O., Prochiantz, A. and Björklund, A., Survival and function of dissociated dopamine neurones grafted at different development stages or after being cultured in vitro, *Dev. Brain Res.*, 39 (1988) 233–243.
36 Brundin, P., Strecker, R.E., Widner, H., Clarke, D.J., Nilsson, O.G., Åstedt, B., Lindvall, O. and Björklund, A., Human fetal dopamine neurons grafted in a rat model of Parkinson's disease: immunological aspects, spontaneous and drug-induced behaviour, and dopamine release, *Exp. Brain Res.*, 70 (1988) 192–208.
37 Brundin, P., Widner, H., Nilsson, O.G., Strecker, R.E. and Björklund, A., Intracerebral xenografts of dopamine neurons: the role of immunosuppression and the blood–brain barrier, *Exp. Brain Res.*, in press.
38 Budd, R.C., Cerottini, J.-C. and MacDonald, H.R., Selectively increased production of interferon-γ by subsets of Lyt-2+ and L3T4+T cells identified by expression of Pgp-1, *J. Immunol.*, 138 (1987) 3583–3586.
39 Cambell, S.I., Thy-1, *Biochem. J.*, 195 (1981) 15–30.
40 Charlton, H.M., Barclay, A.N. and Williams, A.F., Detection of neuronal tissue from brain grafts with anti-Thy-1.1 antibody, *Nature (Lond.)*, 305 (1983) 825–827.
41 Cher, D.J. and Mosman, T.R., Two types of murine helper T cell clone. 2. Delayed type-hypersensitivity is mediated by Th1 clones, *J. Immunol.*, 138 (1986) 3688–3696.
42 Cherwinski, H.M., Shumacher, J.D., Brown, K.D. and Mosman, T.R., Two types of mouse helper T cell clone. III. Further differences in lymphokine synthesis between Th1 and Th2 clones revealed by RNA hybridization, functionally monospecific bioassays, and monoclonal antibodies, *J. Exp. Med.*, 166 (1987) 1229–1244.
43 Crone, C., Facilitated transport of glucose from blood into brain, *J. Physiol. (Lond.)*, 181 (1965) 103–113.
44 Crone, C., Modulation of solute permeability in microvascular endothelium, *Fed. Proc.*, 45 (1986) 77–83.
45 Cserr, H.F., The convection of brain interstitial fluid. In K. Sharpio, A. Marmarou and H. Portnoy (Eds.), *Hydrocephalus*, Raven, New York, 1984, pp. 59–67.
46 Cunningham, B.A., Cell adhesion molecules: a new perspective on molecular embryology, *Trend. Biol. Sci.*, 11 (1986) 423–426.
47 Cupps, T.R. and Fauci, A.C., Corticosteroid-mediated

immunoregulation in man, *Immunol. Rev.*, 65 (1982) 133–155.
48 Cuturi, M.C., Murphy, M., Costa-Giomi, M.P., Weinmann, R., Perussia, B. and Trinchieri, G., Independent regulation of tumor necrosis factor and lymphotoxin production by human peripheral blood lymphocytes, *J. Exp. Med.*, 165 (1987) 1581–1594.
49 Daar, A.S., Fuggle, S.V. and Fabre, J.W., The detailed distribution of HLA-A, B, C antigens in normal human organs, *Transplantation*, 38 (1984) 287–292.
50 Daniloff, J.K., Wells, J. and Ellis, J., Cross-species septal transplants: recovery of choline acetyl transferase activity, *Brain Res.*, 324 (1984) 151–154.
51 Daniloff, J.K., Bodony, R.P., Low, W.C. and Wells, J., Cross-species embryonic septal transplants: restoration of a conditioned learning behavior, *Brain Res.*, 346 (1985) 176–180.
52 Daniloff, J.K., Low, W.C., Bodony, R.P. and Wells, J., Cross-species neural transplants: embryonic septal nuclei to the hippocampal formation of adult rats, *Exp. Brain Res.*, 59 (1985) 73–82.
53 Das, G.D. and Altman, J., Transplanted precursors of nerve cells: their fate in the cerebellums of young rats, *Science*, 173 (1971) 637–638.
54 Das, G.D., Hallas, B.H. and Das, K.G., Transplantation of neural tissue in the brains of laboratory mammals: technical details and comments, *Experientia*, 35 (1979) 143–153.
55 Das, G.D., Intraparenchymal transplantation. In A. Björklund and U. Stenevi (Eds.), *Neural Grafting in the Mammalian CNS*, Elsevier, Amsterdam, 1985, pp. 23–31.
56 Date, I., Kawamura, K. and Nakashima, H., Histological signs of immune reactions against allogeneic solid neural grafts in the mouse cerebellum depend on the MHC locus, *Exp. Brain Res.*, in press.
57 deBault, L.E. and Cantilla, P.A., γ-Glutamyl transpeptidase in isolated brain endothelial cells: induction by glial cells in vitro, *Science*, 207 (1980) 653–655.
58 Del Conte, G., Einpflanzungen von embryonalem Gewebe ins Gehirn, *Pathol. Anat., Beitr.*, 42 (1907) 193–202.
59 deWaal, R.M.W., Bogman, M.J.J., Maass, C.N., Cornelissen, L.M.H., Tax, W.J.M. and Koene, R.A.P., Variable expression of Ia antigens on the vascular endothelium of mouse skin allografts, *Nature (Lond.)*, 303 (1983) 426–429.
60 Dorsch, S., Wotherspoon, J., Keith, I. and Roser, B., The role of regional lymph node in the response to secondarily vascularized grafts, *Transplantation*, 35 (1983) 231–239.
61 Dunn, E.H., Primary and secondary findings in a series of attempts to transplant cerebral cortex to the albino rat, *J. Comp. Neurol.*, 27 (1917) 565–582.
62 Durum, S.K., Schmidt, J.A. and Oppenheim, J.J., Interleukin 1: an immunological perspective, *Annu. Rev. Immunol.*, 3 (1985) 263–287.
63 Dusart, I., Nothias, F., Roudier, F. and Peschanski, M., Vascularization of the neonucleus constructed by embryonic neurons grafted in the adult CNS as a cell suspension, *Soc. Neurosci. Abstr.*, (1987) 81.17.
64 Dustin, M.L., Rothlein, R., Bhan, A.K., Dinarello, C.A. and Springer, T.A., Induction by IL-1 and interferon-γ: tissue distribution and function of a natural adherence molecule (ICAM-1), *J. Immunol.*, 137 (1986) 245–254.
65 Duijvestijn, A.M., Schreiber, A.B. and Butcher, E.C., Interferon-γ regulates an antigen specific for endothelial cells involved in lymphocyte traffic, *Proc. Natl. Acad. Sci. U.S.A.*, 83 (1986) 9114–9118.
66 Dymecki, J., Jedrzejewska, A., Poltorak, M., Pucilowski, O., Bidzinski, A. and Wosko, W., Effects of transplanting rabbit substantia nigra into the striatum of rats with experimental hemiparkinsonism, *Ann. N.Y. Acad. Sci.*, 495 (1987) 696–698.
67 Ebeling, E., Experimentelle Gehirntumoren bei Mäusen, *Z. Krebsforsch.*, 14 (1914) 151–156.
68 Edelman, G.M., Cell-adhesion molecules in neural histogenesis, *Annu. Rev. Physiol.*, 48 (1986) 417–430.
69 Erlich, S.S., McComb, J.G., Hyman, S. and Weiss, M.H., Ultrastructural morphology of the olfactory pathway for cerebrospinal fluid drainage in the rabbit, *J. Neurosurg.*, 64 (1986) 466–473.
70 Fierz, W., Endler, B., Reske, K., Wekerle, H. and Fontana, A., Astrocytes as antigen-presenting cells. I. Induction of Ia antigen expression on astrocytes by T cells via immune interferon and its effect on antigen presentation, *J. Immunol.*, 134 (1985) 3785–3793.
71 Finsen, B., Oteruelo, F. and Zimmer, J., Immunological reactions to brain grafts. Characterization of lymphocytic and astroglial reactions, *Neurosci. Lett.*, Suppl. 258 (1987) 774P.
72 Finsen, B., Poulsen, P.H. and Zimmer, J., Xenografting of fetal mouse hippocampal tissue to the brain of adult rats. Effect of cyclosporin A treatment, *Exp. Brain Res.*, 70 (1988) 117–133.
73 Fontana, A., Kristensen, F., Dubs, R., Gemsa, D. and Weber, E., Production of prostaglandin E and interleukin-1 like factor by cultured astrocytes and C_6 glioma cells, *J. Immunol.*, 129 (1982) 2413–2419.
74 Fontana, A., Fierz, W. and Wekerle, H., Astrocytes present myelin basic protein to encephalolitogenic T-cell lines, *Nature (Lond.)*, 307 (1984) 273–276.
75 Fontana, A., Erb, P., Pircher, H., Zinkernagel, R., Weber, E. and Fierz, W., Astrocytes as antigen-presenting cells. Part II. Unlike H-2K-dependent cytotoxic cells, H-2 Ia-restricted T cells are only stimulated in the presence of interferon-γ, *J. Neuroimmunol.*, 12 (1986) 15–28.
76 Freed, C.R., Richard, J.B., Hutt, C., Whalen, J., Peterson, R. and Reite, M., Behavioral effects of fetal dopamine cell transplantation in bonnet monkeys with MPTP-induced parkinsonism, *Soc. Neurosci. Abstr.*, (1987) 219.5.
77 Freed, W.J., Functional brain tissue transplantation: reversal of lesion induced rotation by intraventricular substantia nigra and adrenal medullary grafts, with a note on intracranial retinal grafts, *Biol. Psychiatry*, 18 (1983) 1205–1267.
78 Freed, W.J., Transplantation of tissues to the cerebral ventricles: methodological details and rate of graft survival. In A. Björklund and U. Stenevi (Eds.), *Neural Grafting in the Mammalian CNS*, Elsevier, Amsterdam, 1985, pp. 31–41.
79 Freed, W.J., Cannon-Spoor, H.E. and Krauthamer, E., Intrastriatal adrenal medulla grafts in rats, long-term survival and behavioral effects, *J. Neurosurg.*, 65 (1986) 664–670.
80 Freeman, T.B., Brandies, L., Pearson, J. and Flamm, E.S., Cross-species grafts of embryonic rabbit mesencephalic tissue survives and cause behavioral recovery in the presence of chronic immunosuppression, *Ann. N.Y.*

Acad. Sci., 495 (1987) 699–702.
81 Frej, K., Bodmer, S., Schwerdel, C. and Fontana, A., Astrocyte-derived interleukin 3 as a growth factor for microglia cells and peritoneal macrophages, *J. Immunol.*, 137 (1986) 3521–3527.
82 Futrell, J.W. and Myers, G.H., Role of regional lymphatics in tumor allograft rejection, *Transplantation*, 13 (1972) 551–557.
83 Gallatin, M., St John, T.P., Siegelman, M., Reichert, R., Butcher, E.C. and Weissman, I.L., Lymphocyte homing receptors, *Cell*, 44 (1986) 673–680.
84 Gash, D.M., Notter, M.F.D., Okawara, S.H., Kraus, A.L. and Joynt, R.J., Amitotic neuroblastoma cells used for neural implants in monkeys, *Science*, 233 (1986) 1420–1422.
85 Geyer, S.J. and Gill, T.J., Immunogenetic aspects of intracerebral skin transplanted in inbred rats, *Am. J. Pathol.*, 94 (1979) 569–579.
86 Geyer, S.J., Gill, T.J., Kunz, H.W. and Moody, E., Immunogenetic aspects of transplantation in the rat brain, *Transplantation*, 39 (1985) 244–247.
87 Giullan, D. and Lachman, L.B., Interleukin-1 stimulation of astroglial proliferation after brain injury, *Science*, 228 (1985) 497–499.
88 Glees, P., Studies of cortical regeneration with special reference to cerebral implants. In W.F. Windle (Ed.), *Regeneration in the Central Nervous System*, Thomas, Springfield, 1955, pp. 94–111.
89 Gonatas, N.K., Greene, M.I. and Waksman, B.H., Genetic and molecular aspects of demyelination, *Immunol. Today*, 7 (1986) 121–126.
90 Goridis, C., Deagostini-Bazin, H., Hirn, M., Hirsch, M.R., Rougon, G., Sadoul, R., Langley, O.K., Gombos, G. and Finne, J., Neural surface antigens during nervous system development, *Cold Spring Harbour Symp. Q. Bull.*, 48 (1983) 527–537.
91 Granholm, A.C., Almqvist, P., Seiger, Å. and Olson, L., Thy-1-like immunoreactivity in human brain during development, *Brain Res. Bull.*, 17 (1986) 107–115.
92 Greene, H.S., Transplantation of tumors to the brains of heterologous species, *Cancer Res.*, 11 (1951) 529–534.
93 Greene, H.S., Transplantation of human brain tumors to brains of laboratory animals, *Cancer Res.*, 13 (1953) 422–426.
94 Greene, H.S., Heterotransplantation of tumors, *Ann. N.Y. Acad. Sci.*, 69 (1957) 818–829.
95 Gullberg, M. and Larsson, E.L., Selective inhibition of antigen-induced 'step-one' in cytotoxic T lymphocytes by anti-Lyt-2 antibodies, *Eur. J. Immunol.*, 12 (1982) 1006–1011.
96 Gurney, M.E., Heinrich, S.P., Lee, M.R. and Yin, H.-S., Molecular cloning and expression of neuroleukin, a neurotrophic factor for spinal and sensory neurons, *Science*, 234 (1987) 566–574.
97 Gurney, M.E., Apatoff, B.R., Spear, G.T., Baumel, M.J., Antel, J.P., Brown Bania, M. and Reder, A.T., Neuroleukin: a lymphokine product of lectin-stimulated T cells, *Science*, 234 (1987) 574–581.
98 Hafler, D.A., Fox, D.A., Manning, M.E., Schlossman, S.F., Reinherz, E.L. and Weiner, H.L., In vivo activated T lymphocytes in the peripheral blood and cerebrospinal fluid of patients with multiple sclerosis, *N. Engl. J. Med.*, 312 (1985) 1405–1411.
99 Hardebo, J.E. and Owman, C., Barrier mechanisms for neurotransmitter monoamines and their precursors at blood–brain interface, *Ann. Neurol.*, 8 (1980) 1–11.
100 Harling-Berg, C., Knopf, P., Pettigrew, K.D. and Cserr, H.F., Role of cervical lymphatics in the systemic humoral immune response to human serum albumin (HSA) microinfused into cerebrospinal fluid (CSF), *Soc. Neurosci. Abstr.*, (1987) 380.1.
101 Hart, D.N. and Fabre, J.W., Demonstration and characterization of Ia-positive dendritic cells in the interstitial connective tissue of rat heart and other tissues, but not brain, *J. Exp. Med.*, 154 (1981) 347–361.
102 Hasek, M., Lengerova, A. and Hraba, T., Transplantation immunity and tolerance, *Adv. Immunol.*, 1 (1961) 1–66.
103 Hasek, M., Chutna, J., Sladecek, M. and Lodin, Z., Immunological tolerance and tumour allografts in the brain, *Nature (Lond.)*, 268 (1977) 68–69.
104 Hasek, M. and Chutna, J., Complexity of the state of immunological tolerance, *Immunol. Rev.*, 46 (1979) 4–26.
105 Hayes, G.M., Woodroofe, M.N. and Cuzner, M.L., Microglia are the major cell type expressing MHC class II in human white matter, *J. Neurol. Sci.*, 80 (1987) 25–37.
106 Head, J.R. and Griffin, S.T., Functional capacity of solid tissue transplants in the brain: evidence for immunological privilege, *Proc. R. Soc. Lond. Ser. B.*, 224 (1985) 375–387.
107 Hildeman, W.H., Components and concepts of antigenic strength, *Transpl. Rev.*, 3 (1970) 5–22.
108 Hirano, T., Yasukawa, K., Harada, H., Taga, T., Watanabe, Y., Matsuda, T., Kashiwamura, S., Nakajima, K., Koyama, K., Iwamatsu, A., Tsunasawa, S., Sakiyama, F., Matsui, H., Takahara, Y., Taniguchi, T. and Kishimoto, T., Complementary DNA for a human interleukin (BSF-2) that induces B lymphocytes to produce immunoglobulin, *Nature (Lond.)*, 324 (1986) 73–76.
109 Hofman, F.M., Danilovs, J.A. and Taylor, C.R., HLA-DR (Ia)-positive dendritic-like cells in human fetal non-lymphoid tissues, *Transplantation*, 37 (1984) 590–594.
110 Houston, M.B., Kunz, H.W., Gill, T.J. and Lund, R.D., Course of transplant rejection within the CNS, *Soc. Neurosci. Abstr.*, (1987) 81.12.
111 Hunkapiller, T. and Hood, L., The growing immunoglobulin gene superfamily. News and views, *Nature (Lond.)*, 323 (1986) 15–16.
112 Hämmerling, G.J., Tissue distribution of Ia antigens and their expression of lymphocyte subpopulations, *Transpl. Rev.*, 30 (1976) 64–82.
113 Häyry, P., von Willebrand, E., Parthenais, E., Nemlander, A., Soots, A., Lautenschlager, I., Alfoldy, P. and Renkonen, R., The inflammatory mechanisms of allograft rejection, *Immunol. Rev.*, 77 (1984) 85–142.
114 Inoue, H., Kohsaka, S., Yoshida, K., Ohtani, M., Toya, S. and Tsukada, Y., Cyclosporin A enhances the survivability of mouse cerebral cortex grafted into the third ventricle of rat brain, *Neurosci. Lett.*, 54 (1985) 85–90.
115 Inoue, H., Kohsaka, S., Yoshida, K., Otani, M., Toya, S. and Tsukada, Y., Immunohistochemical studies on mouse cerebral cortex grafted into the third ventricle of rats treated with cyclosporin A, *Neurosci. Lett.*, 57 (1985) 289–294.
116 Jaeger, C.B., Immunocytochemical study of PC12 cells grafted to the brain of immature rats, *Exp. Brain Res.*, 59 (1985) 615–624.
117 Jalkanen, S.T., Bargatze, R.F., Herron, L.R. and Butch-

er, E.C., A lymphoid cell surface glycoprotein involved in endothelial cell recognition and lymphocyte homing in man, *Eur. J. Immunol.*, 16 (1986) 1195–1202.

118 Jalkanen, S., Reichert, R.A., Gallatin, W.M., Bargatze, R.F., Weissman, I.L. and Butcher, E.C., Homing receptors and the control of lymphocyte migration, *Immunol. Rev.*, 91 (1986) 40–60.

119 Jalkanen, S., Steere, A.C., Fox, R.I. and Butcher, E.C., A distinct endothelial cell recognition system that controls lymphocyte traffic into inflamed synovium, *Science*, 233 (1986) 556–558.

120 Janzer, R.C. and Raff, M.C., Astrocytes induce blood–brain barrier properties in endothelial cells, *Nature (Lond.)*, 325 (1987) 253–257.

121 Jaye, M., Howk, R., Burgess, W., Ricca, G.A., Chiu, I., Ravera, M.W., O'Brien, S.J., Modi, W.S., Maciag, T. and Drohan, W.N., Human endothelial cell growth factor: cloning, nucleotide sequence, and chromosome localization, *Science*, 233 (1986) 541–544.

122 Kahan, B.D., Cyclosporin: the agent and its actions, *Transpl. Proc.*, 17, Suppl. 1 (1985) 5–18.

123 Kamo, H., Kim, S.U., McGeer, P.L. and Shin, D.H., Functional recovery in a rat model of Parkinson's disease following transplantation of cultured human sympathetic neurons, *Brain Res.*, 397 (1986) 372–376.

124 Kamo, H., Kim, S.U., McGeer, P.L., Araki, M., Tomimoto, H. and Kimura, H., Transplantation of human spinal cord cells into the rat motor cortex: use of phaseolus vulgaris leucoagglutinin as a cell marker, *Neurosci. Lett.*, 76 (1987) 163–167.

125 Kaplan, H.J. and Streilein, J.W., Do immunologically privileged sites require a functional spleen?, *Nature (Lond.)*, 251 (1974) 553–554.

126 Kim, S.U., Antigen expression by glial cells grown in culture, *J. Neuroimmunol.*, 8 (1985) 255–282.

127 Kinashi, T., Harada, N., Sevrinson, E., Tanabe, T., Sideras, P., Konishi, M., Azuma, C., Tominaga, A., Bergstedt-Lindqvist, S., Takahashi, M., Matsuda, F., Yaoita, Y., Takatsu, K. and Honjo, T., Cloning of complementary DNA encoding T-cell replacing factor and identity with B-cell growth factor II, *Nature (Lond.)*, 324 (1986) 70–73.

128 Klein, G., Factors that interfere with or prevent effective destruction of tumors via immune mechanisms. In R.T. Smith and M. Laundy (Eds.), *Immunobiology of Tumor-Host Relationship*, Academic, London, 1975, pp. 203–276.

129 Klein, J., Juretic, A., Baxevanis, C.N. and Nagy, Z.A., The traditional new version of the mouse H-2 complex, *Nature (Lond.)*, 291 (1981) 455–460.

130 Klein, J., *Immunology, the Science of Self-Nonself Discrimination*, Wiley, 1982, pp. 445–507.

131 Klein, J., *Natural History of the Major Histocompatibility Complex*, Wiley, 1986, pp. 194–195 and 211.

132 Koren, H.S., Proposed classification of leukocyte associated cytolytic molecules, *Immunol. Today*, 8 (1987) 69–71.

133 Lafferty, K.J., Prowse, S.J., Simenonovic, C.J. and Warren, H.S., Immunobiology of tissue transplantation: a return to the passenger leukocyte concept, *Annu. Rev. Immunol.*, 1 (1983) 143–173.

134 Lampson, L.A. and Fischer, C.A., Weak HLA and β2-microglobulin expression of neuronal cell lines can be modulated by interferon, *Proc. Natl. Acad. Sci. U.S.A.*, 81 (1984) 6476–6480.

135 Lampson, L.A., Molecular bases of the immune system response to neural antigens, *Trend. Neurosci.*, 10 (1987) 211–216.

136 Lance, E.M., A functional and morphological study of intracranial parathyroid allografts in the dog, *Transplantation*, 5 (1967) 1471–1483.

137 Lance, E.M., A functional and morphological study of the intracranial thyroid allografts in the dog, *Surg. Gynecol. Obstet.*, 125 (1967) 529–539.

138 Larsen, G.L. and Henson, P.M., Mediators of inflammation, *Annu. Rev. Immunol.*, 1 (1983) 335–359.

139 Lavi, E., Suzumura, A., Zoltick, P.W., Murasko, D.M., Silberberg, D.H. and Weiss, S.R., Tumor necrosis factor induces MHC class I antigen expression on mouse astrocytes, *J. Neuroimmunol.*, 16 (1987) 102.

140 Lee, M.H., Lossinsky, A.S., Rabe, A. and Wisniewski, H.M., Intact blood–brain barrier in large neocortical transplants in the rat brain, *Soc. Neurosci. Abstr.*, (1987) 81.18.

141 Le Gros Clark, W.E., Neuronal differentiation in implanted fetal cortical tissue, *J. Neurol. Psychiatry*, 3 (1940) 263–284.

142 Leibovich, S.J., Polverini, P.J., Shepard, H.M., Wiseman, D.M., Shively, V. and Nuseir, N., Macrophage-induced angiogenesis is mediated by tumor necrosis factor-α, *Nature (Lond.)*, 329 (1987) 630–632.

143 Leszczynski, D., Ferry, B., Schellekens, H., v.d. Meide, P. and Häyrä, P., Antagonistic effects of γ-interferon and steroids on tissue antigenicity, *J. Exp. Med.*, 164 (1986) 1470–1477.

144 Lindvall, O., Backlund, E.-O., Farde, L., Sedvall, G., Freedman, R., Hoffer, B., Nobin, A., Seiger, Å. and Olson, L., Transplantation in Parkinson's disease: two cases of adrenal medullary grafts to the putamen, *Ann. Neurol.*, 22 (1987) 457–468.

145 Lindvall, O., Dunnett, S.B., Brundin, P. and Björklund, A., Transplantation of catecholamine-producing cells to the basal ganglia in Parkinson's disease: experimental and clinical studies. In C. Rose (Ed.), *Parkinson's Disease; Clinical and Experimental Advances*, Libbey, London, 1987, pp. 189–206.

146 Lodin, Z., Hasek, M., Chutna, J., Sladecek, M. and Holan, V., Transplantation immunity in the brain, *J. Neurol. Sci.*, 3 (1977) 275–280.

147 Love, J.A. and Leslie, R.A., The effects of raised ICP on lymph flow in the cervical lymphatic trunks in cats, *J. Neurosurg.*, 60 (1984) 577–581.

148 Loveland, B. and Simpson, E., The non-MHC transplantation antigens: neither weak nor minor, *Immunol. Today*, 7–8 (1987) 223–229.

149 Low, W.C., Lewis, P.R. and Bunch, S.T., Embryonic neuronal transplants across a major histocompatibility barrier: survival and specificity of innervation, *Brain Res.*, 262 (1983) 328–333.

150 Lund, R.D., Rao, K., Hankin, M.H., Kunz, H.W. and Gill III, T.J., Transplantation of retina and visual cortex to rat brains of different ages. Maturation, connection patterns, and immunological consequences, *Ann. N.Y. Acad. Sci.*, 495 (1987) 227–241.

151 Madrazo, I., Drucker-Colin, R., Diaz, V., Martinez-Mata, J., Torres, C. and Becerril, J.J., Open microsurgical autograft of adrenal medulla to the right caudate nucleus in two patients with intractable Parkinson's disease,

N. Engl. J. Med., 316 (1987) 831–834.
152 Makinodan, T. and Kay, M.B., Age influence on the immune system, *Adv. Immunol.*, 29 (1980) 287–330.
153 Mason, D.W., Charlton, H.M., Jones, A., Parry, D.M. and Simmonds, S.J., Immunology of allograft rejection in mammals. In A. Björklund and U. Stenevi (Eds.), *Neural Grafting in the Mammalian CNS*, Elsevier, Amsterdam, 1985, pp. 91–98.
154 Mason, D.W., Charlton, H.M., Jones, A.J., Lavy, C.B., Puklavec, M. and Simmonds, S.J., The fate of allogeneic and xenogeneic neuronal tissue transplantated into the third ventricle of rodents, *Neuroscience*, 19 (1986) 685–694.
155 Mason, D.W. and Morris, P.J., Effector mechanisms in allograft rejection, *Annu. Rev. Immunol.*, 4 (1986) 119–145.
156 Massa, P.T., Dörries, R. and ter Meulen, V., Viral particles induce Ia antigens expression on astrocytes, *Nature (Lond.)*, 320 (1986) 543–546.
157 McCarron, R.M., Kempski, O., Spatz, M. and McFarlin, D., Presentation of myelin basic protein by murine cerebral vascular endothelial cells, *J. Immunol.*, 134 (1985) 3100–3103.
158 Medawar, P.B., Immunity to homologous grafted skin. III. The fate of skin homografts transplanted to the brain, to subcutaneous tissue, and to the anterior chamber of the eye, *Br. J. Exp. Pathol.*, 29 (1948) 58–69.
159 Medawar, P.B., *Theories of Immunological Tolerance*, Ciba Foundation Symp., *Cellular Aspects of Immunity*, 1960, pp. 134–149.
160 Merrill, J.E., Miacroglia: neural cells reactive to lymphokines and growth factors, *Immunol. Today*, 8 (1987) 146–150.
161 Meuer, S.C., Acuto, O., Hercend, T., Schlossman, S.F. and Reinherz, E.L., The human T-cell receptor, *Annu. Rev. Immunol.*, 2 (1984) 23–50.
162 Milton, A., Spencer, S.C. and Fabre, J.W., The effects of cyclosporin on the induction of donor class I and II MHC antigens in the heart and kidney allografts in the rat, *Transplantation*, 42 (1986) 337–347.
163 Miossec, P., Yu, C. and Ziff, M., Lymphocyte chemotactic activity of human interleukin-1, *J. Immunol.*, 133 (1984) 2007–2011.
164 Murphy, J.B. and Sturm, E., Conditions determining the transplantability of tissues in the brain, *J. Exp. Med.*, 38 (1923) 183–197.
165 Möller, E., Contact induced cytotoxicity by lymphoid cells containing foreign isoantigens, *Science*, 147 (1965) 873–879.
166 Möller, G. (Ed.), Molecular genetics of class I and II MHC antigens, 1, *Immunol. Rev.*, 84 (1985) 7–143.
167 Möller, G. (Ed.), Molecular genetics of class I and II MHC antigens, 2, *Immunol. Rev.*, 85 (1985) 5–168.
168 Möller, G. (Ed.), Molecular genetics of class III MHC antigens, *Immunol. Rev.*, 87 (1985) 7–208.
169 Naparstek, Y., Cohen, I.R., Fuks, Z. and Vlodavsky, I., Activated T lymphocytes produce a matrix degrading heparin sulphate endoglycosidase, *Nature (Lond.)*, 310 (1984) 241–244.
170 Nathanson, J.A. and Chun, L.Y., Immunological defense of the brain: role of the choroid plexus, *Soc. Neurosci. Abstr.*, 12 (1986) 342.2.
171 Nedwin, G.E., Svedersky, L.P., Bringman, T.S., Palladino, M.A. and Goeddel, D.V., Effect of interleukin 2, interferon-γ and mitogens on the production of tumor necrosis factor α and β, *J. Immunol.*, 135 (1985) 2492–2496.
172 Nicholas, M.K., Antel, J.P., Stefansson, K. and Arnason, B.G.W., Rejection of fetal neocortical neural transplants by H-2 incompatible mice, *J. Immunol.*, 139 (1987) 2275–2283.
173 Niederkorn, J.Y. and Streilein, J.W., Alloantigens placed into the anterior chamber of the eye induce specific suppression of delayed-type hypersensitivity but normal cytotoxic T lymphocytes and helper T lymphocyte responses, *J. Immunol.*, 131 (1983) 2670–2674.
174 Nilsson, O.G., Brundin, P., Widner, H., Strecker, R.E. and Björklund, A., Human fetal basal forebrain neurons grafted to the denervated rat hippocampus produce an organotypic cholinergic reinnervation pattern, *Brain Res.*, 456 (1988) 193–198.
175 Nisbet, N.W., Simonsen, M. and Zaleski, M., The frequency of antigen-sensitive cells in tissue transplantation, *J. Exp. Med.*, 129 (1969) 459–467.
176 Noma, Y., Sideras, P., Naito, T., Bergstedt-Lindquist, S., Azuma, S., Severingson, E., Tanabe, T., Kinashi, T., Matsuda, F., Yaoita, Y. and Honjo, T., Cloning of cDNA encoding the murine IgG1 induction factor by a novel strategy using SP6 promoter, *Nature (Lond.)*, 319 (1986) 640–644.
177 Nossal, G.J., Cellular mechanisms of immunologic tolerance, *Annu. Rev. Immunol.*, 1 (1983) 33–62.
178 Oldendorf, W.H., Carrier-mediated blood–brain barrier transport of short-chain monocarboxylic organic acids, *Am. J. Physiol.*, 224 (1973) 1450–1453.
179 Oldendorf, W.H. and Brown, W.J., Greater number of capillary endothelial cell mitrochondria in brain than in muscle, *Proc. Soc. Exp. Biol. Med.*, 149 (1975) 736–738.
180 Oldstone, M.B., Blount, P., Southern, P.J. and Lampert, P.W., Cytoimmunotherapy for persistent virus infection reveals a unique clearance pattern from the central nervous system, *Nature (Lond.)*, 321 (1986) 239–243.
181 Olson, L., Seiger, Å. and Strömberg, I., Intraocular transplantations in rodents. A detailed account of the procedure and example of its use in neurobiology with special reference to brain tissue grafting. In S. Federoff and L. Hertz (Eds.), *Adv. Cellular Neurobiology, 4*, Academic, New York, 1983, pp. 407–442.
182 Olson, L., Backlund, E.-O., Freed, W.J., Herrera-Marschitz, M., Hoffer, B., Seiger, Å. and Strömberg, I., Transplantation of mono-amine producing cell systems in oculo and intracranially: experiments in search of a treatment for Parkinson's disease, *Ann. N.Y. Acad. Sci.*, 457 (1985) 105–126.
183 Oppenheim, J.J., Kovacs, E.J., Matsushima, K. and Durum, S.K., There is more than one interleukin-1, *Immunol. Today*, 7 (1986) 45–56.
184 Pardridge, M.W., UCLA conference, Blood–brain barrier: interface between internal medicine and the brain, *Ann. Intern. Med.*, 105 (1986) 82–95.
185 Parwaresch, M.R., Horny, H.-P. and Lennert, K., Tissue mast cell in health and disease, *Pathol. Res. Pract.*, 179 (1985) 439–461.
186 Perlow, M.J., Freed, W.J., Hoffer, B.J., Seiger, Å., Olson, L. and Wyatt, R.J., Brain grafts reduce motor abnormalities produced by destruction of nigrostriatal dopamine system, *Science*, 204 (1979) 643–647.
187 Perlow, M.J., Kumakura, K. and Guidotti, A., Prolonged survival of bovine adrenal chromaffin cells in rat cerebral

ventricles, *Proc. Natl. Acad. Sci. U.S.A.*, 77 (1980) 5278–5281.
188 Perry, V.H., Hume, D.A. and Gordon, S., Immunohistochemical localization of macrophages and microglia in the adult and developing brain, *Neuroscience*, 15 (1985) 313–326.
189 Philip, R. and Epstein, L.B., Tumour necrosis factor as immunomodulator and mediator of monocyte cytotoxicity induced by itself, γ-interferon and interleukin-1, *Nature (Lond.)*, 323 (1986) 86–89.
190 Ponnappan, U., Cinander, B., Gerber, V. and Blaser, K., Antibody response and aquired tolerance of A/J mice: age and immunogen-related isotype differences, *Scand. J. Immunol.*, 27 (1988) 419–425.
191 Prehn, R.T., Do tumors grow because of the immune response of the host, *Transpl. Rev.*, 28 (1976) 33–42.
192 Prineas, J.W., Multiple sclerosis: presence of lymphatic capillaries and lymphoid tissue in the brain and spinal cord, *Science*, 203 (1979) 1123–1125.
193 Raine, C.S., Mokhtarian, F. and McFarlin, D.E, Adoptively transferred chronic relapsing experimental autoimmune encephalomyelitis in the mouse. Neuropathological analysis, *Lab. Invest.*, 51 (1984) 534–546.
194 Raju, S. and Grogan, J.B., Immunological study of the brain as a privileged site, *Transpl. Proc.*, 9 (1977) 1187–1191.
195 Rapoport, S.I., *Blood–Brain Barrier in Physiology and Medicine*, Raven, New York, 1976, pp. 1–315.
196 Redmond, D.E., Sladek, J.R., Roth, R.H., Collier, T.J., Elsworth, J.D., Deutch, A.Y. and Haber, S., Fetal neuronal grafts in monkeys given metylphenyltetrahydropyridine, *Lancet*, I 8490 (1986) 1125–1127.
197 Reese, T.S. and Karnovsky, M.J., Fine structural localization of a blood–brain barrier to exogenous peroxidase, *J. Cell Biol.*, 34 (1967) 207–217.
198 Revel, M. and Chebath, J., Interferon-activated genes, *Trend Biol. Sci.*, 11 (1986) 166–170.
199 Ridley, A. and Cavanagh, J.B., The cellular reactions to heterologous, homologous and autologous skin implanted into brain, *J. Pathol.*, 99 (1969) 193–203.
200 Robbins, D.S., Shiazi, Y., Drysdale, B.-E., Lieberman, E., Shin, H.S. and Shin, M.L., Production of cytotoxic factor for oligodendrocytes by stimulated astrocytes, *J. Immunol.*, 139 (1987) 2593–2597.
201 Roser, B. and Dorsch, S., The cellular basis of transplantation tolerance in the rat, *Immunol. Rev.*, 46 (1979) 55–85.
202 Rosenstein, J.M., Neocortical transplants in the mammalian brain lack a blood–brain barrier to macromolecules, *Science*, 235 (1987) 772–774.
203 Ross, R., Raines, E.W. and Bowen-Pope, D.F., The biology of platelet-derived growth factor, *Cell*, 46 (1986) 155–169.
204 Roszman, T.L. and Brooks, W.H., Neural modulation of immune function, *J. Neuroimmunol.*, 10 (1985) 59–69.
205 Ruddle, N.H., Tumor necrosis factor and related cytotoxins, *Immunol. Today*, 8 (1987) 129–130.
206 Sachs, D.H., Winn, H.J. and Russel, P.S., The immunologic response to xenografts. Recognition of mouse H-2 histocompatibility antigens by the rat, *J. Immunol.*, 107 (1971) 481–492.
207 Sandberg-Wollheim, M., Zweiman, B., Levinson, A.I. and Lisak, R.P., Humoral immune responses within the human central nervous system following systemic immunization, *J. Neuroimmunol.*, 11 (1986) 205–214.
208 Sandvig, S., Laskey, T., Andersson, J., De Ley, M. and Andersson, U., Gamma-interferon is produced by CD3+ and CD3– lymphocytes, *Immunol. Rev.*, 97 (1987) 51–65.
209 Scheinberg, L.C., Edelman, F.L. and Levy, A.W., Is the brain an immunologically privileged site? I. Studies based on intracerebral tumor transplantation and isotransplantation to sensitized hosts, *Arch. Neurol.*, 11 (1964) 248–264.
210 Scheinberg, L.C., Kotsilimbas, D.G., Karpf, R. and Mayer, M., Is the brain an immunologically privileged site? III., *Arch. Neurol.*, 15 (1966) 62–67.
211 Schmidt, R.H., Ingvar, M., Lindvall, O., Stenevi, U. and Björklund, A., Functional activity of substantia nigra grafts reinnervating the striatum: neurotransmitter metabolism and (^{14}C)-2-deoxy-D-glucose autoradiography, *J. Neurochem.*, 38 (1982) 737–748.
212 Schnyder, B., Weber, E. and Fontana, A., On the role of astrocytes in polyclonal T cell activation, *J. Neuroimmunol.*, 10 (1986) 209–218.
213 Schrader, J.W., The panspecific hemopoietin of activated T lymphocytes, *Annu. Rev. Immunol.*, 4 (1986) 205–230.
214 Seiger, Å. and Olson, L., Quantification of fiber growth in transplanted central monoamine neurons, *Cell Tissue Res.*, 179 (1977) 285–316.
215 Shen, A.L., Hu, C. and Hisa, R., Cellular and antigenic properties of cultured normal and fetal brain and glioma cells, *J. Neuroimmunol.*, 8 (1985) 141–151.
216 Sherwood, R.A., Brent, L. and Rayfield, L.S., Major histocompatibility complex antigens are presented by murine host accessory cells, *Transpl. Proc.*, 19 (1987) 239–241.
217 Shevach, E.M., The effects of cyclosporin A on the immune system, *Annu. Rev. Immunol.*, 3 (1985) 397–423.
218 Shimuzu, K., Tsuda, N., Matsui, Y., Okamoto, Y., Miyao, Y., Tamura, K., Nakatani, S. and Mogami, H., Surgical trials on treatment of Parkinson's disease in mice, *Soc. Neurosci. Abstr.*, (1987) 219.1.
219 Shirai, Y., Transplantation of the rat sarcoma in adult heterogenous animals, *Japn. Med. World*, 1 (1921) 14–15.
220 Siebert, W.J., Auto and homeotransplantation of thyroid gland into brain of guinea pigs, *Proc. Soc. Exp. Biol. Med.*, 26 (1928) 236–237.
221 Silvers, W.K., Bartlett, S.T., Chen, H., Fleming, H.L., Naji, A. and Barker, C.F., Major histocompatibility complex restriction and transplantation immunity. A possible solution to the allograft problem, *Transplantation*, 37 (1984) 28–32.
222 Simonsen, M., Graft versus host reactions. Their natural history, and applicability as tools of research, *Prog. Allerg.*, 6 (1962) 349–367.
223 Sladek, J.R., Collier, T.J., Haber, S.N., Roth, R.H. and Redmond, D.E., Survival and growth of fetal catecholamine neurons grafted into primate brain, *Brain Res. Bull.*, 17 (1986) 809–818.
224 Smith, F.I. and Miller, J.F.A.P., Delayed hypersensitivity to allogeneic cells in mice. III., *J. Exp. Med.*, 150 (1979) 965–976.
225 Smith, K.A., Interleukin 2, *Annu. Rev. Immunol.*, 2 (1984) 319–334.
226 Snell, G.D., The homograft reaction, *Annu. Rev. Microbiol.*, 11 (1957) 439–458.
227 Sorg, B.A., Smith, M.M. and Campagnoni, A.T., Developmental expression of the myelin proteolipid protein and basic protein mRNA in normal and dysmyelinating mutant mice, *J. Neurochem.*, 49 (1987) 1146–1154.

228 Steinman, R.M. and Nussenzweig, M.C., Dendritic cells: features and functions, *Immunol. Rev.*, 53 (1980) 127–147.

229 Steinmuller, D., Methods for induction of permanent graft survival, *Transpl. Proc.*, 11 (1979) 1198–1206.

230 Steinmuller, D., Which T cells mediate allograft rejection?, *Transplantation*, 40 (1985) 229–233.

231 Stenevi, U., Kromer, L.F., Gage, F.H. and Björklund, A., Solid neural grafts in intracerebral transplantation cavities. In A. Björklund and U. Stenevi (Eds.), *Neural Grafting in the Mammalian CNS*, Elsevier, Amsterdam, 1985, pp. 41–51.

232 Stepkowski, S. and Duncan, W.R., The role of T_{DTH} and T_C populations in organ graft rejection, *Transplantation*, 42 (1986) 406–412.

233 Stewart, P.A. and Wiley, M.J., Developing nervous tissue induces formation of blood–brain barrier characteristics in invading endothelial cells: a study using quail-chick transplantation chimeras, *Dev. Biol.*, 84 (1981) 183–192.

234 Stewart, P.A., Clements, L.G. and Wiley, M.J., Revascularization of skin transplanted into the brain: source of the graft endothelium, *Microvasc. Res.*, 28 (1984) 113–124.

235 Streilein, J.W. and Niederkorn, J.Y., Characterization of the suppressor cell(s) responsible for anterior chamber-associated immune deviation (ACAID) induced in BALB/c mice by P815 cells, *J. Immunol.*, 134 (1985) 1381–1387.

236 Strömberg, I., Herrera-Marschitz, M., Ungerstedt, U., Ebendal, T. and Olson, L., Chronic implants of chromaffin tissue into the dopamine-denervated striatum. Effects of NGF on graft survival, fiber growth and rotational behavior, *Exp. Brain Res.*, 60 (1985) 335–349.

237 Strömberg, I., Bygdeman, M., Goldstein, M., Seiger, Å. and Olson, L., Human fetal substantia nigra grafted to dopamine-denervated striatum of immunosuppressed rats: evidence for functional reinnervation, *Neurosci. Lett.*, 71 (1986) 271–276.

238 Summers, B.A., Greisen, H.A. and Appel, M.J.G., Possible initiation of viral encephalomyelitis in dogs by migrating lymphocytes infected with distemper virus, *Lancet*, II 8082 (1978) 187–189.

239 Suzumura, A., Lavi, E., Weiss, S.R. and Silberberg, D.H., Coronavirus infection induces H-2 antigen expression on oligodendrocytes and astrocytes, *Science*, 232 (1986) 991–993.

240 Suzumura, A., Silberberg, D.H. and Lisak, R.P., The expression of MHC antigens on oligodendrocytes: induction of polymorphic H-2 expression by lymphokines, *J. Neuroimmunol.*, 11 (1986) 179–190.

241 Svendgaard, N.A., Björklund, A., Hardebo, J.E. and Stenevi, U., Axonal degeneration associated with a defective blood–brain barrier in cerebral implants, *Nature (Lond.)*, 255 (1975) 334–336.

242 Swensen, R.B., Sörensen, J.C., Zimmer, J. and Castro, A.J., Neocortical transplants grafted into the newborn rat brain demonstrates a blood–brain barrier to macromolecules, *Soc. Neurosci. Abstr.*, (1987) 81.16.

243 Szentistvanyi, I., Patlak, C.S., Ellis, R.A. and Cserr, H.F., Drainage of interstitial fluid from different regions of rat brain, *Am. J. Physiol.*, 246 (Renal Fluid Electrolyte Physiol. 15) (1984) F835–F844.

244 Tedeschi, B., Barrett, J.N. and Keane, R.W., Astrocytes produce interferon that enhances the expression of H-2 antigens on a subpopulation of brain cells, *J. Cell Biol.*, 102 (1986) 2244–2253.

245 Thompson, W.G., Successful brain grafting, *N.Y. Med. J.*, 51 (1890) 701–702.

246 Thurman, G.B., Braude, I.A., Gray, P.W., Oldham, R.K. and Stevenson, H.C., MIF-like activity of natural and recombinant human interferon-γ and their neutralization by monoclonal antibody, *J. Immunol.*, 134 (1985) 305–309.

247 Tilney, N.L. and Gowans, J.L., The sensitization of rats by allografts transplanted to alymphatic pedicles of skin, *J. Exp. Med.*, 133 (1970) 951–962.

248 Traugott, U., Scheinberg, L. and Raine, C., On the presence of Ia-positive endothelial cells and astrocytes in multiple sclerosis lesions and its relevance to antigen presentation, *J. Neuroimmunol.*, 8 (1985) 1–14.

249 Trotter, J. and Steinman, L., Homing of Lyt-2^+ and Lyt-2^- T-cell subsets and B lymphocytes to the central nervous system of mice with acute experimental allergic encephalomyelitis, *J. Immunol.*, 132 (1984) 2919–2923.

250 Tulipan, N.B., Huang, S. and Allen, G.S., Pituitary transplantation: cyclosporin enables transplantation across a minor histocompatibility barrier, *Neurosurgery*, 18 (1986) 316–322.

251 Tze, W.H. and Tai, J., Intracerebral allotransplantation of purified pancreatic endocrine cells and pancreatic islets in diabetic rats, *Transplantation*, 38 (1984) 107–111.

252 Unanue, E.R., Antigen-presenting function of the macrophage, *Annu. Rev. Immunol.*, 2 (1984) 395–428.

253 Unanue, E.R. and Allen, P.M., The basis for the immunoregulatory role of macrophages and other accessory cells, *Science*, 236 (1987) 551–557.

254 Ungerstedt, U. and Arbuthnott, G.W., Quantitative recording of rotational behavior in rats after 6-hydroxydopamine lesions of the nigrostriatal dopamine system, *Brain Res.*, 24 (1970) 485–493.

255 Vass, K., Lassmann, H., Wiesniewski, H.M. and Iqbal, K., Ultrachemical distribution of myelin basic protein after injection into the cerebrospinal fluid. Evidence for transport through the blood–brain barrier and binding to the luminal surface of cerebral veins, *J. Neurol. Sci.*, 63 (1984) 423–433.

256 Vinogradova, O.S., Bragin, A.G. and Kitchigina, V.F., Spontaneous and evoked activity of neurons in intrabrain allo- and xenografts of the hippocampus and septum. In A. Björklund and U. Stenevi (Eds.), *Neural Grafting in the Mammalian CNS*, Elsevier, Amsterdam, 1985, pp. 409–421.

257 Walford, R.L., *Maximum Lifespan*, Avon, New York, 1983, pp. 1–256.

258 Webb, M. and Barclay, A.N., Localization of the MRC Ox-2 glucoprotein on the surface of neurones, *J. Neurochem.*, 43 (1984) 1061–1067.

259 Weiss, A., Imboden, J., Hardy, K., Manger, B., Terhorst, C. and Stobo, J., The role of the T_3/antigen receptor complex in T-cell activation, *Annu. Rev. Immunol.*, 4 (1986) 539–620.

260 Wekerle, H., Linington, C., Lassmann, H. and Meyerman, R., Cellular immune reactivity within the CNS, *Trends Neurosci.*, 6 (1986) 271–277.

261 Whetton, A.D. and Dexter, T.M., Haemopoietic growth factors, *Trends Biol. Sci.*, 11 (1986) 207–211.

262 Widner, H., Johansson, B.B. and Möller, G., Qualitative demonstration of a link between brain parenchyma and the lymphatic system after intracerebral antigen deposition, *J. Cerebr. Blood Flow Metab.*, 5 (1985) 88–89.

263 Widner, H., Brundin, P., Björklund, A. and Möller, E., Survival and host immunization of allogeneic fetal neural dopamine cell suspension grafts implanted into the brains of adult mice, *Exp. Brain Res.*, in press.
264 Widner, H., Johansson, B.B. and Möller, G., Immune response in deep cervical lymph nodes and spleen in the mouse after intracerebral antigen deposition, *Scand. J. Immunol.*, in press.
265 Widner, H., Jönsson, B.-A., Hallstadius, L., Wingårdh, K., Strand, S.E. and Johansson, B.B., Scintigraphic method to quantify the passage from brain parenchyma to the deep cervical lymph nodes in the rat, *Eur. J. Nucl. Med.*, 13 (1988) 456–461.
266 Willis, R.A., Experiments on the intracerebral implantation of embryo tissues in rats, *Proc. R. Soc. Lond. Ser. B.*, 117 (1935) 400–412.
267 Wolff, J., Elektronmikroskopische Untersuchungen über die Vesikulationen in Kapillarendotel, *Z. Zellforsch.*, 73 (1966) 143–164.
268 Wong, G.H., Bartlett, P.F., Clark-Lewis, I., Battye, F. and Schrader, J.W., Inducible expression of H-2 and Ia antigens on brain cells, *Nature (Lond.)*, 310 (1984) 688–691.
269 Zimmer, J., Lesion-induced reorganization of central nervous connections, with a note on central nervous transplants. In M.W. van Hof and G. Mohn (Eds.), *Functional Recovery from Brain Damage*, Elsevier/North-Holland, Amsterdam, 1981, pp. 289–304.
270 Zinkernagel, R.M. and Doherty, P., MHC-restricted cytotoxic T cells: studies on the biological role of polymorphic major transplantation antigens determining T-cell restriction-specificity, function, and responsiveness, *Adv. Immunol.*, 27 (1979) 51–177.