NEUROPATHOLOGY OF CARDIAC TRANSPLANTATION

Survey of 31 Cases

RALF SCHOBER  MARY M. HERMAN
Department of Pathology (Neuropathology), Stanford University School of Medicine, Stanford, California 94305, U.S.A.

Summary

The neuropathology of 31 cases of cardiac transplantation is reported. Lesions were found in the central nervous system of 18 cases, and were infectious, vascular, or neoplastic in nature. Fungal meningoencephalitis or abscess formation was found in 5 patients as part of a disseminated fungal infection. *Aspergillus* was the organism in 3 instances, and *Candida* or *Mucormyces* in 1 each. In 10 cases, disseminated microglial nodules were found in the brain in association with intranuclear inclusions suggestive of herpesvirus in the lungs. In two instances, large amphophilic intranuclear inclusions were found in neurons, and an antemortem rise in cytomegalovirus serology titre was demonstrated in two cases. The presence of widespread microglial nodules was interpreted as suggestive of viral encephalitis, with cytomegalovirus as the probable prevailing agent. In 1 case, *Toxoplasma* organisms were also demonstrated. Vascular lesions were seen in 5 cases, which included thrombotic embolisation in the middle cerebral artery in 1 case and multiple cerebral fat emboli in another. 1 patient had a reticulum-cell sarcoma-microglioma in the cerebellum, associated with a similar neoplastic lesion in one lung eight months after cardiac transplantation.

Introduction

In human transplantation the attention of pathologists has concentrated on the transplanted organ. However, in the light of the complications that usually determine the clinical course, tissue changes elsewhere in the body seem to be equally important. In this context the pathology of the central nervous system is significant, both to the clinician and to the pathologist. A frequent complication of immunosuppression is supervening infection, usually by fungi or viruses.1 Lately, the possibility of brain tumours arising de novo has been suggested.2 Detailed neuropathological reports have so far been limited to cases of renal transplantation.3,4 We describe here the neuropathological changes seen post mortem in patients given heart transplants at the Stanford University Medical Center.

Patients

The series consists of 31 cases of cardiac transplantation. All but one of 28 males and 3 females were middle-aged. None had had a major illness before transplantation, apart from cardiac failure. Postoperatively, they were under continuing immunosuppressive management.2

All brains were examined grossly and microscopically...
after formalin fixation for 21 days. Sections were stained with haematoxylin and eosin, and special stains were used when applicable. The earlier cases were sampled sparsely, since neuropathological changes were not anticipated at the beginning of the study. Spinal cords and peripheral nerves were included only if abnormalities were suspected on clinical grounds.

Results

13 cases had no significant neuropathological changes or had lesions that could be attributed to preoperative disease. The positive findings in the remaining 18 cases are summarised in the table. Most of the lesions were infectious or vascular in origin. Obvious manifestations of fungal infection were present in 5 cases. The infecting organisms were identified microscopically as Aspergillus (3 cases), Mucormyces, and Candida albicans. The gross picture in the cases of aspergillus meningoencephalitis was of

Fig. 3—Case 35: viral encephalitis.
A neuron with a large oval intranuclear inclusion is surrounded by reactive microglial cells. (Haematoxylin and eosin; ×900.)

Fig. 4—Case 42: toxoplasmosis.
Toxoplasma pseudocyst packed with organisms. (Haematoxylin and eosin; ×900.)

Fig. 5—Case 43: reticulum-cell sarcoma-microglioma.
Coronal section showing necrotic tumour nodule in the folia and adjacent white matter of the left lateral cerebellar hemisphere; hemorrhage and oedema in the left dentate nucleus and middle cerebellar peduncle.

Fig. 6—Case 43: reticulum-cell sarcoma-microglioma.
(Haematoxylin and eosin; ×400.)
multiple abscesses throughout the brain. They showed a predilection for the white matter, measured up to 3 cm in diameter, and were grey-brown and poorly defined (fig. 1). Microscopically, there was hemorrhage and intense necrosis of the neural tissue with relatively little inflammatory or astrocytic cellular response. Organisms were easily identified, especially in the leptomeninges, where they were found in the lumens and walls of numerous subarachnoid blood-vessels.

The case of mucormycosis showed similar microscopical features (fig. 2). It differed in that the infectious process involved the orbitofrontal regions and adjacent olfactory structures and overlying leptomeninges. Extensive bilateral panophthalmitis was also present, with mycotic involvement of the orbits and gangrenous cellulitis of the central part of the face. There was a recent infarct in the right temporal lobe, but it seemed to be anatomically unrelated to the distribution of mucormycosis. In the case of cerebral candidiasis there were multiple small red annular foci with pale centres, predominantly situated in the parietal and occipital white matter. Microscopically, these foci consisted in areas of necrosis and microglial proliferation which sometimes surrounded fungal hyphae. The leptomeninges were free of organisms but were infiltrated by lymphocytes and plasma cells. Microglial nodules were found in 10 cases. They were composed of collections of cells with darkly staining elongated nuclei and scanty cytoplasm, overlaying a background of usually loosely textured or fragmented neural tissue. In all cases they were widespread, but showed a predilection for the thalamus, basal ganglia, and pons. Associated Cowdry type-A intranuclear inclusion bodies (fig. 3) were found in ganglion cells in 2 instances—namely, in a spinal ganglion in case 23, and, after extensive sampling, in the hippocampus, caudate nucleus, putamen, and thalamus in case 35. The inclusions were round or oval and measured up to 8 µ in diameter. They demonstrated amphiphilic staining properties and were surrounded by a clear halo.

The microscopical examination of the brain in case 42, where numerous microglial nodules were found,
also revealed multiple diffusely scattered toxoplasma organisms closely packed in pseudocysts (fig. 4). Most of them were unassociated with cellular infiltrates. In a few instances, however, they were found in or near a microglial nodule. Mature toxoplasma organisms or calcifications were not seen.

Vascular lesions were present in 5 instances. Patient 2, who died three days after cardiac transplantation, had multiple small recent pericapillary hemorrhagic white matter infarcts, with oil-red-O positive material in central capillary blood-vessels. The lesions were interpreted as the result of fat embolisation and were associated with pulmonary fat emboli which also contained bone-marrow cells. Case 8 showed a large hemorrhagic infarct in the left frontoparietal region associated with a recent embolus in the left middle cerebral artery, the source of which was a thrombus on an ulcerated atherosclerotic plaque in the left common carotid artery. In the remaining 3 cases, the vascular lesions were small recent anemic superficial cortical infarcts (1 parietal, 1 parieto-occipital, and 1 temporal), which were presumably due to ischemic anoxia resulting from severe circulatory failure.

In the last case of this series a neoplastic lesion of the reticuloendothelial system was demonstrated both intracerebrally and extracerebrally. This case deserves special attention:

The patient was a twenty-year-old White male with a cardiomyopathy of unknown aetiology. After cardiac transplantation on May 18, 1972, the immediate clinical course was favourable, with only one episode of late rejection two months later. Maintenance immunosuppressive therapy consisted of azathioprine, 150 mg. daily, and prednisone, 50 mg. daily. Seven months after transplantation, the patient was admitted complaining of a sore throat and fever. Chest X-rays showed diffuse infiltrates in both lungs which on biopsy were found to be caused by Pneumocystis. On the second day of admission the patient developed headaches and then became progressively confused and agitated. Twelve days later he was obtunded and had a stiff neck. A lumbar puncture showed 28 red blood-cells, 1 white blood-cell, 248 mg. protein per 100 ml., and a normal glucose content. A brain scan revealed nothing significant. The patient had a sudden respiratory arrest. He remained deeply comatose for several hours until he was pronounced dead, on the 227th postoperative day.

At necropsy, the lungs showed extensive Pneumocystis pneumonia and abscesses caused by Nocardia. Microscopic examination revealed also one small focus of neoplastic cells interpreted as reticulum-cell sarcoma (histiocytic lymphoma). Abnormal neuropathological findings were confined to the left cerebellar hemisphere. This con-

### SUMMARY OF 18 CASES WITH NEUROPATHOLOGICAL ABNORMALITIES—continued

<table>
<thead>
<tr>
<th>Stanford series no.</th>
<th>Age</th>
<th>Sex</th>
<th>Days postop.</th>
<th>Neurological symptoms*</th>
<th>Neuropathology, gross</th>
<th>Neuropathology, microscopical</th>
<th>Related general pathology</th>
</tr>
</thead>
<tbody>
<tr>
<td>34</td>
<td>52</td>
<td>M</td>
<td>45</td>
<td>R hemiparesis and disorientation, 6 days</td>
<td>Large abscesses in L cerebral hemisphere, R frontal and occipital lobes, and L cerebellar hemisphere, with intraventricular hemorrhage</td>
<td>Necrotising Aspergillus meningoencephalitis; microglial nodules</td>
<td>Pulmonary aspergillosis and C.M.V. infection</td>
</tr>
<tr>
<td>10</td>
<td>47</td>
<td>F</td>
<td>820</td>
<td>Headache and nausea, 6 days</td>
<td>Yellow discoloration of 3 × 1.5 × 1 cm. in L pericallosal white matter</td>
<td>Focal leuencephalopathy of unknown aetiology; senile plaques</td>
<td>Chronic rejection</td>
</tr>
<tr>
<td>37</td>
<td>49</td>
<td>M</td>
<td>65; 15</td>
<td>Transient focal seizures after second transplant; terminal coma</td>
<td>Recent infarct, R temporal lobe; necrosis of olfactory bulbs and supraorbital gyri bilaterally</td>
<td>Recent infarct; necrotising mucormycoses meningoencephalitis and panophthalmitis; microglial nodules</td>
<td>Severe acute rejection; mucormycoses panencephalitis, pulmonary C.M.V. infection</td>
</tr>
<tr>
<td>41</td>
<td>51</td>
<td>M</td>
<td>66</td>
<td>Terminal seizures</td>
<td>Fresh hemorrhage in R frontal lobe with extension into lateral ventricle; secondary brainstem hemorrhages</td>
<td>Microglial nodules</td>
<td>Pulmonary aspergillosis and C.M.V. infection</td>
</tr>
<tr>
<td>42</td>
<td>45</td>
<td>M</td>
<td>65</td>
<td>...</td>
<td>Normal</td>
<td>Disseminated Toxoplasma pseudocysts; microglial nodules</td>
<td>Severe acute rejection; pulmonary aspergillosis, C.M.V. and Toxoplasma infection; positive C.M.V. titre</td>
</tr>
<tr>
<td>35</td>
<td>48</td>
<td>M</td>
<td>321</td>
<td>Coma, 2 days</td>
<td>Normal</td>
<td>Microglial nodules and 4 neural nuclear inclusions</td>
<td>Severe acute rejection; pulmonary C.M.V. infection; positive C.M.V. titre</td>
</tr>
<tr>
<td>49</td>
<td>56</td>
<td>M</td>
<td>63</td>
<td>...</td>
<td>Normal</td>
<td>Microglial nodules</td>
<td>Severe acute rejection; pulmonary aspergillosis and C.M.V. infection</td>
</tr>
<tr>
<td>43</td>
<td>20</td>
<td>M</td>
<td>227</td>
<td>Headache, stiff neck and otorrhoea, 10 days</td>
<td>Necrotic lesion of 3 cm. diameter and small hemorrhages, L cerebellar hemisphere</td>
<td>Reticulum-cell sarcoma (microglioma)</td>
<td>Small reticulum-cell sarcoma, lung; pulmonary Pneumocystis and Nocardia infection</td>
</tr>
</tbody>
</table>

* Figures refer to time before death.  
† Cardiac allograft pathology in ref. 11.  
‡ Complete necropsy report in ref. 12.
tained an ill-defined grey-brown round mass, measuring 3 cm. in diameter and involving the cortical folia and part of the underlying white matter (fig. 3). Several foci of recent hemorrhage were present in the ipsilateral dentate nucleus and the middle cerebellar peduncle. The mass was surrounded by edema and associated with striking tonsillar herniation, which had caused medullary compression. Microscopical examination revealed a reticulum-cell sarcoma (microglioma) (fig. 6) characterised by extensive necrosis and a typical perivascular distribution of the tumour cells. Special stains for fungi and bacteria were negative.

Discussion

The neuropathological findings of infectious nature in this series of cases of cardiac transplantation are comparable to those seen after renal transplantation. Schneck's series of 35 cases included 14 with microglial nodules, 3 of which showed inclusion bodies and reactive cellular changes compatible with cytomegalovirus infection. Dorfman has reported widespread glial nodule encephalitis presumably due to cytomegalovirus in the brains of 4 adults examined in this department, 3 of whom were renal-transplant recipients. Patients in Schneck's series had widespread cerebral mycotic infection. Collectively, therefore, the infectious lesions in Schneck's and Dorfman's series and in ours point to immunosuppressive therapy as the common underlying factor that determines supervening infection, regardless of the organ transplanted.

Generalised fungal infection seems to be a common cause of death in transplant recipients. In our series, all 5 cases with fungal lesions in the central nervous system had evidence of fungal infection elsewhere in the body. The extent of the lesions in the brain and the concomitant meningeal involvement make it highly likely that the infection was blood-born. The pattern of lesions in the case of mucormycosis, similar to that of the kidney-transplant case reported by Haim et al. and to other instances of mucormycosis unassociated with organ transplantation, is suggestive of rhino-cerebral spread. The case of toxoplasmosis showed neuropathological features that were similar to those in a renal transplantation case complicated by generalised toxoplasmosis reported by Reynolds et al. Of special interest was the finding of disseminated microglial nodules in 10 cases. Although we did not try to isolate a virus we believe, with Bailey et al., that circumstantial evidence favours a disseminated viral encephalitis.

This interpretation is based first on the demonstration of characteristic intranuclear inclusions suggestive of herpesvirus infection in the neurons of 2 of our cases. The probability of finding inclusions appears, in our experience, to be proportional to the number of sections taken of the grey matter. Secondly, the significance of the microglial nodules as evidence of encephalitis is reinforced by the fact that all 10 cases had histological evidence of a concomitant pulmonary viral infection. The characteristic large intranuclear inclusions found in one or more visceral organs were interpreted as cytomegalovirus infection in 8 instances. The inclusion bodies in the other 2 were morphologically identical, but were interpreted as probably due to a herpes-simplex infection because of the clinical features: case 22 had a generalised herpes-simplex dermatitis, and case 23 had a severe oral herpes-simplex infection. The third line of evidence is provided by the serological tests. Serological studies for cytomegalovirus were done in 3 patients who were subsequently found to have microglial nodules. In 2 of these there had been a significant ante-mortem rise in titre—four months after transplantation (six months before death) in one and one month after transplantation (one month before death) in the other. In Dorfman's series of 4 cases with disseminated glial nodule encephalitis, intranuclear inclusions characteristic of cytomegalovirus infection were found in 2 brains, and the virus was cultured from body fluids in 3 instances.

In 10 of the 18 cases in this series, fungal infection was present outside the central nervous system, and in 7 of these it was associated with demonstrable pulmonary cytomegalovirus infection; microglial nodules were present in the brain in all 7 of these cases. Moreover, 3 of these 7 cases with microglial nodules had associated fungal infection in the brain. Whether this evident association between fungal and viral infection implies a facilitating effect favouring invasion by more than one opportunistic agent or a decrease of immune resistance in general cannot be answered on morphological grounds. 5 of the 10 patients with microglial nodules in the brain died during a severe rejection crisis, which would argue against the second hypothesis. Of further interest in this respect is the simultaneous occurrence of toxoplasmosis and viral infection in case 42. Double infections of this kind have been experimentally reproduced in human fibroblast cultures.

In the absence of distinct neurological symptoms it is difficult to estimate the impact that viral encephalitis may have on the clinical course. Neuropathological studies alone cannot provide information as to the time of onset. Furthermore, in most instances other potential causes of diffuse encephalopathy may be present (i.e., generalised bacterial or fungal infection, azotemia, electrolyte disturbances, and the effects of medication). Increasing awareness of opportunistic viral infections of the brain should, however, lead to the development of more specific clinical and laboratory techniques for their control.

The finding of a reticulum-cell sarcoma (microglioma) in the brain of 1 patient in this series seems important. The presumption that it is related to transplantation and the effects of immunosuppression is strong. Schneck and Penn have reported a 6% frequency for neoplasms after renal transplantation and drew attention to the special frequency of tumours of the reticuloendothelial system involving the brain. In their survey of 24 mesenchymal neoplasms reported to arise de novo in patients with organ homografts, 11 (46%) were in the brain, 8 of them affecting this organ exclusively. The case reported in this paper is analogous to their case 6 (also reported by Penn), in which simultaneous involvement of the brain and lungs by a reticulum-cell sarcoma was found in a woman of thirty-nine who died fourteen months after renal transplantation.

The possible mechanisms that may be brought into play in the genesis of tumours in homograft recipients or in patients and laboratory animals with diseases
associated with immunological deficiencies and the factors that determine the tendency of the neoplastic process to involve the brain more than other organs have been discussed by Schnick and Penn. 8 The hypotheses have included stimulation of the host's immunological apparatus by foreign antigens of the homograft, chromosomal breakage induced by immunosuppressive drugs, loss or impairment of a surveillance mechanism for neoplastic mutant cells, and proliferation of an oncogenic virus. The tendency to involve the brain more than other organs could be due to the relatively poor immunological response on the part of the central nervous system to stimulation by a foreign antigen, as contrasted to the relative preservation of the surveillance function of the extracerebral immunological apparatus in destroying or reducing the number of neoplastic cells.

On the basis of this hypothesis, our case would appear to demonstrate that a breakdown of this surveillance mechanism had occurred not only in the brain but in the extracerebral tissues as well. The pulmonary lesion in our case was smaller than that in the cerebellar hemisphere. Whether the neoplasm arose multicentrically or as a result of a metastatic process is open to speculation. Schnick and Penn 2 suggest that one of the reasons why, in such cases, the neoplasms in the brain have until now invariably been of the lymphoreticular system and not gliomas is that they could be derived from neoplastic mesenchymal cells that arose outside the brain and were subsequently implanted intracerebrally. Our case provides no additional information that might help to reinforce or exclude this hypothesis. In any event, the late onset of the clinical symptoms, the young age of the patient in this report, and the extremely focal nature of the neoplastic lesions leave little room to doubt that the tumour arose after cardiac transplantation and is to be related to the transplantation procedure and/or the associated immunosuppressive therapy.

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Requests for reprints should be addressed to R. S.

REFERENCES