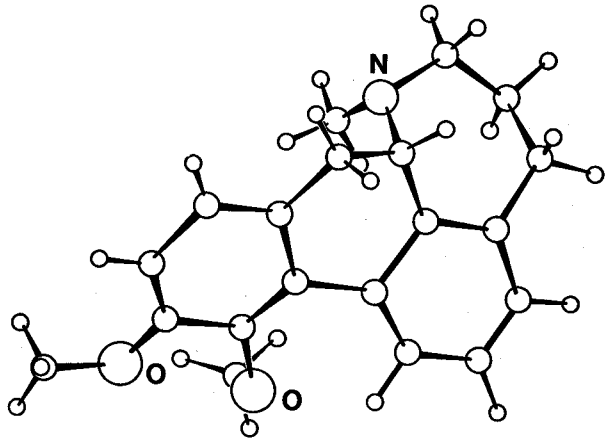
The result of the crystal structure analysis is shown in the Figure. The nitrogen atom is axial to the B-ring, and the N-methyl group is also, unusually, axial to the seven-membered ring. This conformation is quite different from that of apomorphine and is sufficient explanation for the biological inactivity of **2**. The torsion angles of interest, τ_1 and τ_2, are -146 and $+86°$, respectively. Interestingly, for molecules with the same absolute configuration at the asymmetric centre, the sense of the twist between the two benzene rings is opposite in apomorphine and in this homologue **2**.



0,0-Dimethyl derivative of apomorphine homologue (**3**): perspective drawing of the structure as observed in crystals of the base.

Summary. 11,12-Dihydroxy-7-methyl-4,5,6,7,7a,8-hexahydrophenanthro[10,1-b,c]-azepine (**2**), a homologue of apomorphine (**1**), has been found to be devoid of dopaminergic effects. The biological differences between apomorphine and this homologue are explained in terms of differences in conformation of the two molecules.

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The Process of Survival of Denervated and Freely Autotransplantated Skeletal Muscle

Survival of free autotransplants of entire skeletal muscles was first demonstrated by STUDITZKY[1,2], but the importance of previous denervation was emphasized by THOMPSON[3]. Successful autotransplantations have later been reported by several authors[4–10]. In a series of investigations on denervated cat muscle autotransplants[7,11–13], we have analyzed the various phases of early survival, revascularization and reinnervation of the graft. Those studies resulted in the finding that the muscle fibres undergo differential changes in the superficial and deep regions of the graft within the first week after the transplantation[7]. In particular, three concentric zones could be clearly distinguished where differential histochemical changes were demonstrated. However, the nature of these changes could not be clearly established by light microscopy. An ultrastructural study was therefore undertaken in order to define more precisely the process of survival of the graft.

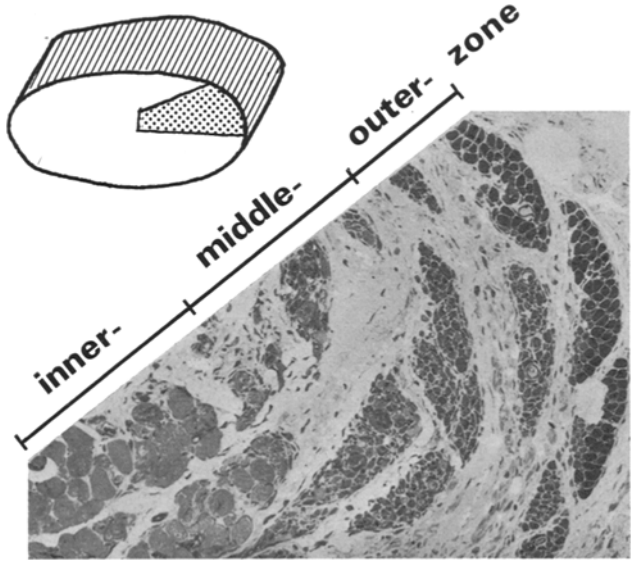

Fig. 1. Survey of muscle graft 8 days after transplantation. 3 zones are distinguished (outer, middle and inner zone). The boundaries between the different zones are not clear-cut and their relative extension varies in different parts of the transplant. × 60.

Materials and methods. The transplantations were made in two stages[7]. Primarily, a denervation of the m. peroneus longus of adult cats was made. Secondarily, 3 weeks after denervation, the muscles (diameter 8–9 mm, length 4 cm) were transplanted. By blunt dissection a tunnel was created under the fascia of one of the intercostal muscles. In this way the fibres of the transplant ran at approximately right angles to the intercostals. Both ends of the grafts were then sutured to the intercostal fascia under slight tension. The muscles were removed 2, 8 or 15 days after transplantation and processed for electron microscopy, i.e. treated with 2.5% glutaraldehyde for 16 h, cut into slices, postfixed in 1% osmium tetroxide and plastic embedded. For simplicity, only the findings after 8 days of transplantation will be described here.

Results and discussion. Three zones could be distinguished in the graft (Figure 1): an outer zone composed of a few layers of muscle fibres with slightly reduced diameter, a middle zone comprizing several fascicles of very small muscle fibres interspersed with numerous other cells, and an inner zone where large pale muscle fibres were observed at sites surrounded and penetrated by small cells.

Outer zone (Figure 2). The surface of the muscle fibres displayed small papillary projections of the basement membrane which contained cytoplasmic evaginations. The contractile material showed a tendency to become confluent into large fields. This was apparently related to an altered distribution of sarcotubular system elements. Triads were frequently seen transversely oriented. Honeycomb tubular structures derived from the T-systems were also seen. At variance with the control,

muscle mitochondria were predominantly longitudinally oriented. Myelin bodies and autophagic vacuoles were seen with variable frequence. In conclusion, the outer zone of the transplant consisted of surviving atrophic fibres which displayed pathological changes similar to those known to occur in denervated muscle fibres[14–16]. There was no evidence of necrosis or muscle regeneration in this zone.

Middle zone (Figures 3 and 4). The cell composition of this zone was markedly heterogenous. The main component was represented by extremely atrophic muscle fibres. They were contained within a loosely fitting, pleated basal lamina and showed often central nuclei with dispersed chromatin and prominent nucleoli. The contractile material was reduced and signs of myofibrillar disintegration were apparent. Large cytoplasmic areas were occupied by autophagic vacuoles and residual bodies. The effect of ischemia presumably summated with the effect of denervation in triggering the activation of the lysosomal system and atrophy. The transverse tubular system showed focal dilatations and honeycomb structures. On the contrary, mitochondria were generally well preserved. Large mononuclear cells were frequently seen inside the sarcolemmal tubes. Some of these cells could be identified as macrophages. However, more often these cells had features of undifferentiated cells with abundant free and membrane-bound ribosomes. Many of these cells had a characteristically clear cytoplasm with sparse filaments, often of an intermediate type, and glycogen accumulations. Frequently cells containing Z-bodies and clearly recognizable myofilaments were also seen, occurring either with atrophic muscle fibres or with undifferentiated cells and macrophages inside the same basal lamina. In conclusion, the middle zone was characterized both by extreme muscle atrophy, and by fibre regeneration.

Inner zone (Figure 5). The muscle fibres were here all clearly necrotic. Macrophages often appeared within these fibres. Further, undifferentiated mononuclear cells or clearly recognizable myoblasts were seen, generally adherent to the inner surface of the sarcolemmal tubes. Infiltration by macrophages and concomitant regenerative processes appeared to occur starting at the junction of the middle and inner zone, whereas in the more central areas no viable cell was seen. This demolition-reconstitution process seemed to occur as a progressive concentric wave penetrating deeper into the graft as shown in other models of muscle regeneration[17, 18]. Ischemic necrosis in

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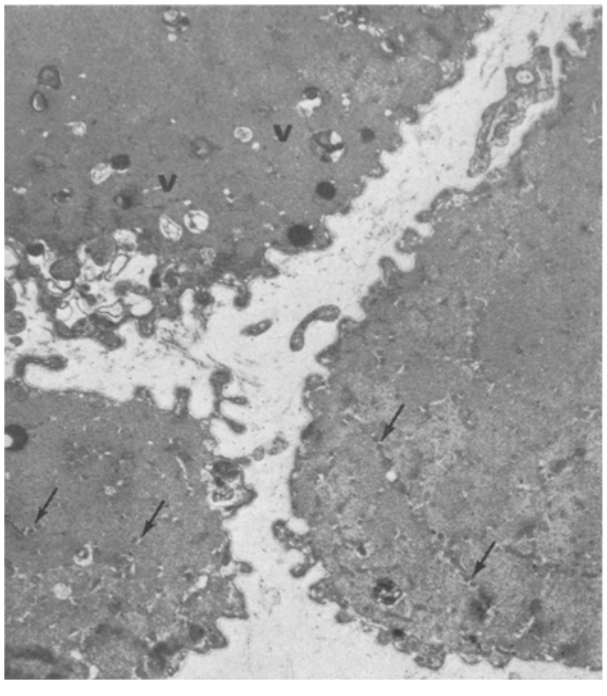

Fig. 2. Outer zone: Fibres from the periphery of the graft, slightly atrophic and with a surface displaying small papillary projections. The sarco-tubular system shows an altered organization with transversely running triads (arrows) and areas with autophagic vacuoles (v) are seen. ×9,000.

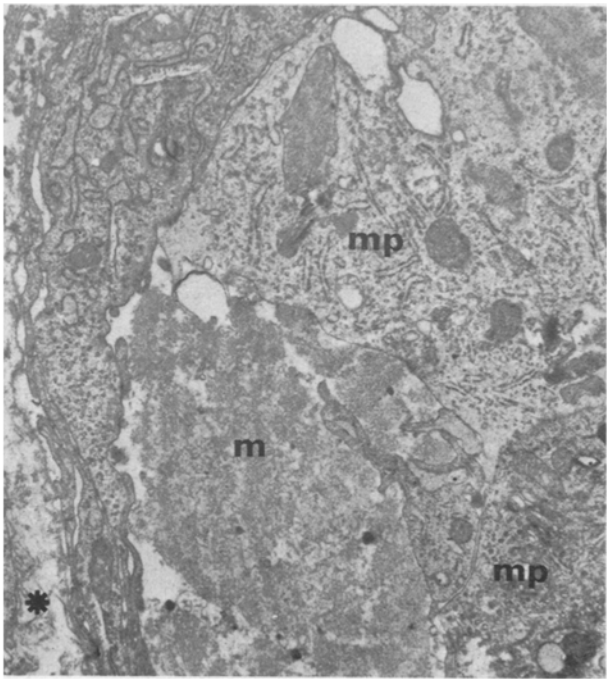

Fig. 5. Inner zone: A necrotic muscle fibre (m) infiltrated by macrophages (mp) whose long pseudopodia penetrate the disorganized myofibrillar material. Portions of undifferentiated cells lying close to the basement membrane of the necrotic fibre are also seen. Extracellular space (x). ×9,000.

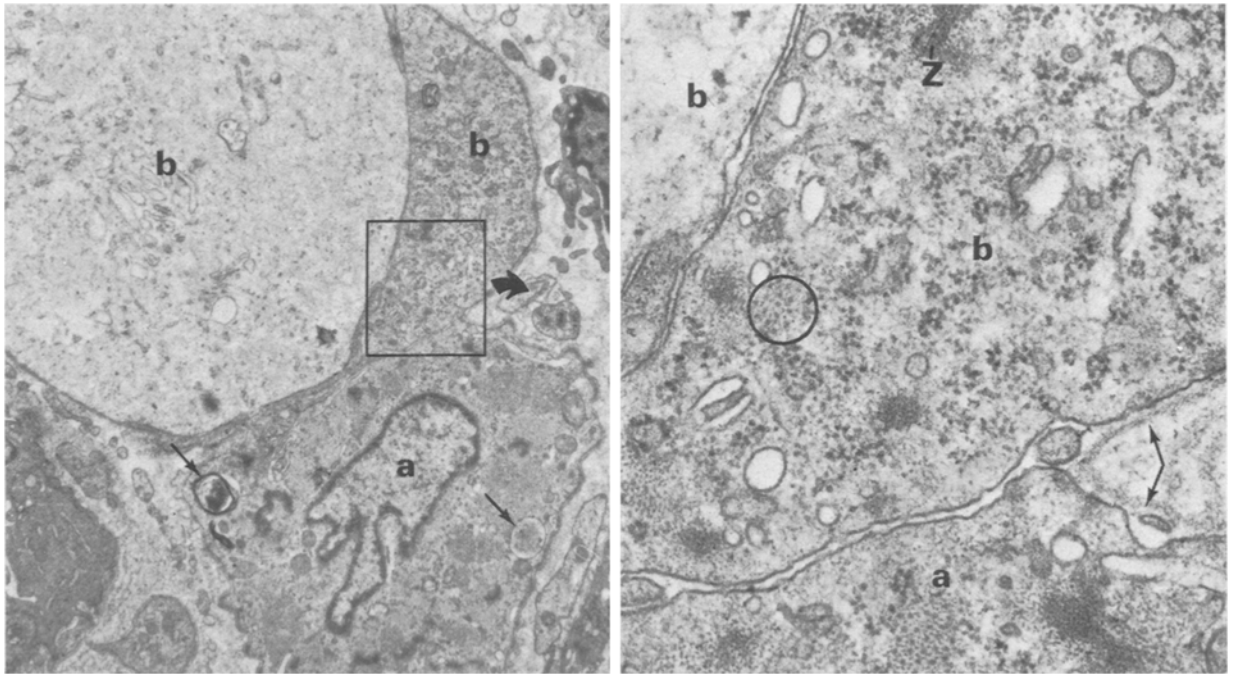

Fig. 3. Middle zone: The cell composition is markedly heterogenous. Both extremely atrophic fibres (a) and regenerating muscle cells (b) are seen, sometimes, as in this figure, even within the same basal lamina (c.f. Figure 4). Autophagic vacuoles and residual bodies are seen in the atrophic fibre (arrows). ×9,000.

Fig. 4. Higher magnification of the area indicated in Figure 3, showing a myoblast (b) containing abundant free and membrane-bound ribosomes, Z-bodies (Z) and thick and thin myofilaments organized in a regular hexagonal pattern (circle). The basal lamina (arrows). × 45,000.

the inner zone must have occurred shortly after transplantation before the muscle fibres could undergo any adaptive process. This is consistent with the early loss of histochemical staining for oxidative enzymes observed at these sites in contrast with the positive reaction persisting in the more peripheral zones.

This study shows that two distinct processes occur in autografts of denervated intact muscle during the early, critical phases of the transplantation: Survival of transplanted fibres at the periphery of the graft, and regeneration of new muscle fibres following breakdown of the originally transplanted fibres in the central areas. In this respect, transplantation of intact muscle differs basically from the transplantation of minced muscle which is exclusively dependent on regenerative processes[1, 2, 4, 19]. It seems important to establish the relative contribution of the two processes since the factors which may affect respectively the survival and regeneration of muscle fibres are not necessarily the same, or one factor could affect the two processes in different ways. Thus, it is probable that previous denervation makes it possible for the muscle fibres to survive as structural entities by decreasing the size of the fibres, making metabolic exchanges easier through a more favourable surface-to-volume ratio, and possibly also by reducing their energetic requirements[20]. On the other hand, denervation appears to affect muscle regeneration mainly through an increase in the number of satellite cells and thus of the population of myoblasts participating in the regenerative process[21–23]. The choice of the optimal denervation period in muscle autotransplantation is clearly dependent on a more precise analysis of these different aspects of the 'plastic state'[2] induced in the muscle by denervation.

Summary. The study shows that two distinct processes occur in free autografts of denervated intact muscle during the early critical phase of the transplantation: survival of transplanted fibres at the periphery of the graft, and regeneration of new muscle fibres following breakdown of the originally transplanted fibres in the central areas.

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