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A COMPARATIVE EXPERIMENTAL STUDY ON TRANSPLANTATION OF AUTOGENOUS AND HOMOGENOUS TENDON TISSUE

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Regeneration and transplantation of tendon tissue have been subject for numerous investigations. Nevertheless it is still much discussed in literature whether a true regeneration occurs or the tendon defect is replaced by ordinary scar being in its structure similar to tendon tissue. The supporters of the first opinion consider the regeneration to be effected by a tenoblast proliferation from the cut ends of the tendon itself (*Seggel* 1903, *Garlock* 1927, *Stewart* 1936, *Voronin* 1954, *Amosova* 1958). Other authors consider the tendon tissue to be high differentiated and consequently unable for a true regeneration. According to them the regeneration arises mainly from the sheats of the tendon stumps by cicatrisation (*Adams* 1860, *Beltzow* 1883, *Schwarz* 1922, *Tomilova* 1927, *Krivorotov* 1936, *Nikolaev* 1937, *Rozov* 1952, *Skoog & Persson* 1954, *Potenza* 1962).

Mason & Shearon (1932), *Flynn and coll.* (1962) and others are of the opinion that union is first effected by proliferation of the sheaths tissues and after the 4th or 5th day—by proliferation of stumps tenoblasts, which penetrate into the "callus".

Not less is the controversy about the fate of the tendon transplant. According to the opinion of some authors the autogenous tendon transplant remains alive with the possible exception of the central areas in ticker transplants and showes tenoblastic proliferation (*Garlock* 1927, *Mason & Shearon* 1932, *Peer* 1955, *Lindsay & McDougall* 1961, *Cordrey and coll.* 1963). *Lewis & Davis* (1911) considered the fresh homogenous tendon transplant taken as a whole to remain alive.

Other group of authors holds that the tendon transplant—either autogenous or homogenous—undergo necrosis and act solely as a strut for ingrowing cells from the host tissues (*Neuhof* 1923, *Teneff & Fonda*

1953, *Skoog & Persson* 1954, *Flynn and coll.* 1960, 1962, *Dimoff and coll.* 1962, *Degtiareva & Lavrishtcheva* 1962, *Herzog* 1963, *Andreeff & Dimoff* 1965).

With the present work we had the task to investigate the possible difference in healing of fresh autogenous tendon transplants and homogenous tendon transplants, preserved by means of different methods.

MATERIAL AND METHODS

For our experiments we used calcaneus tendon of rabbit because in its anatomical arrangement it resembles the true synovial tendons in man (*Nisbet* 1960).

Thirty-seven rabbits were operated. For evaluation of the obtained results we took 31 out from that number. The experiments were divided into 4 series. In the first series in seven animals a segment of soleus tendon was excised, turned round and resutured end to end with black silk "Sutagor" 000 placed in figure of eight fashion.

After the operation the leg was immobilised for two weeks in a plaster extending to the groin.

In the other series were used three types of homogenous tendon transplants, which were sutured under moderate tension in the same way into the gap, produced by excising a segment of soleus tendon of two-three centimeters. In 12 rabbits were used homogenous tendon transplants preserved by means of the method we have already reported with proteine hydrolizate at +4° C in the course of 30–90 days. In 9 rabbits were used homogenous tendon transplants preserved in the course of 30–84 days by refrigeration at —25° C in 20 per cent Glycerin-Ringer's solution. In 9 rabbits were used freeze-dried homogenous tendon transplants. The residual moisture of these transplants was defined to be 2 per cent.

Successively after the 1th, 2nd, 3rd, 4th, 5th, 6th and 7th week after the operation soleus tendon and its transplant were revealed. After local examination the whole transplant together with the adjacent ends of soleus tendon and the sheaths were removed and fixed in 10 per cent neutral formaline for morphological examination. Classical and histochemical methods were applied as follows: for collagen contents (Haematoxylin-eosine and Picrofuxine), for elastic fibres (Orseine), for mucopolysaccharides (Tolouidin blau and Periodic Acid-Schiff reaction) and for nucleine acids (Acridine orange).

RESULTS

Finally in all experiments the continuity of tendon and transplant was restored and since the 5th week on the animals were able to use their leg freely hopping and rising themselves to it.

The reparative processes which have been observed were almost identical and therefore we shall consider all of them together, noting the difference in the corresponding places.

At the end of the first week the stumps sheaths and the tissues round the transplant seemed to be hyperemic. Round the joints there were

Figure 1. Homogenous tendon transplant preserved in proteine hydrolizate at the end of the 3rd week after transplantation into the gap of soleus tendon. The transplant is completely integrated with the stumps. There is a disordered capillary net round the stumps and the transplant.

slight adhesions which were easily separated from them. Such adhesions were not to be found in the freeze-dried homogenous tendon transplants. The mesotenonium was healed to the transplant. The transplant appeared shiny and slightly oedematous like the stumps as well but well defined from them. At the second week there was moderate hyperemia and adhesions round the stumps and transplants and slight adhesions appeared round the freeze-dried homogenous tendon transplant. The joints seemed to be well healed. At the end of the third week the adhesions increased and there was a disordered capillary net. The transplant was completely integrated with the stumps, the latter being generally thickened (Figure 1). At the fourth week the capillary net round the transplant and the stumps decreases. The next week the capillary net had almost a normal appearance and there was longitudinal orientation of the vessels. The adhesions were slighter and they did not disturb the normal function of the whole tendon. The transplant was defined only by the silk sutures. At the end of the 7th week round the stumps and transplant there were found only slight adhesions.

Morphological changes: The morphological changes were fully identical in all the 4 series of experiments. The difference as much as it existed was exceptionally of quantitative character.

For better distinctness and clearness of our description the evaluation of the essential changes is reflected in Table 1.

During the first week necrosis appears in all tendon transplants. The necrotic changes are demonstrated by kariopicnosis, kariolysis and

Table 1. 1. Fresh Autogenous Tendon Transplants. 2. Homogenous Tendon Transplants Preserved in Proteine Hydrolizate at +4° C (by the Method of the Authors). 3. Homogenous Tendon Transplants Preserved by Refrigeration at —25° C in 20 per cent Glycerin-Ringer's Solution. 4. Homogenous Lyophilised Tendon Transplants.

Morphological Changes	Experimental Series	Time Elapsed from Transplantation (in Weeks) 1	2	3	4	5	6	7
Ischemic Necrosis of Transplant	1	+++++	+++++	++++	+++	++	+	—
	2	+++++	+++++	++++	+++	++	++	—
	3	+++++	+++++	++++	++++	+++	+++	+
	4	+++++	+++++	+++++	++++	+++	+++	++
Paratenon and Stumps Capillary Proliferation	1	++++	++++	++++	++++	++	+	—
	2	+++	++++	++++	++++	++	++	+
	3	++	+++	++++	++++	+++	++	++
	4	+	++	+++	++++	+++	+++	++
Paratenon and Stumps Fibroblastic Proliferation	1	+++	++++	+++	+++	++	+	—
	2	+++	++++	+++	+++	+++	+	—
	3	++	++++	+++	++++	++	++	—
	4	++	+++	+++	++++	+++	++	—
Capillary and Fibroblastic Proliferation in Transplant	1	++	+++	+++	++++	++++	+++	++
	2	+	++	++++	++++	++++	+++	+++
	3	+	+	++	+++	++++	++++	+++
	4	±	+	+	+++	++++	+++	+++
Organisation of Transplant	1	+	++	+++	+++	++++	+++±	+++
	2	—	+	++	+++	++++	++++	++++
	3	—	—	+	++	+++	++++	+++
	4	—	—	—	++	+++	++++	+++
Capillary Reduction and Collagenisation of Transplant	1	—	—	—	+	++	+++	++++
	2	—	—	—	—	+	++	+++
	3	—	—	—	—	—	++	++
	4	—	--	—	±	+	++	++

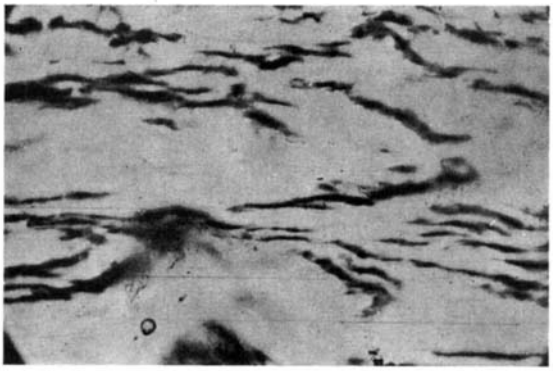

Figure 2. Proliferation of capillaries in the necrotic homogenous lyophilised tendon transplant on the 5th day after transplantation. Reaction for sour phosphatasis. (× 100).

by changes in the colorific properties of the collagenous fibres of the ground substance.

Simultaneously with that the reparative processes develop as well. They begin with exudation and capillary and fibroblastic proliferation of the paratenoneum and stumps. The granulation tissue, consisting of fibroblasts, capillaries and the fluid and cells of the exudate surround the stumps and the transplant quickly and fill in the gaps between them. The newly formed granulation tissue penetrates into the necrotic transplant.

Capillary proliferation most manifestated was established in the autogenous tendon transplants during the first week. Vascular response of such a character was found in the homogenous tendon transplants of the 2nd series at the end of the second week, while in the others—at the end of the 3rd and 4th week. Since the 5th week on this superficial capillary proliferation begins to decrease in the autogenous tendon transplants as well as in the homogenous tendon transplants, preserved in hydrolizate solution while in the homogenous tendon transplants of the 3rd and 4th series—since the 6th or 7th week on.

Here we must note that capillary vessels, penetrating into the periphery of the transplant, were found also in the freeze-dried homogenous tendon transplants even during the first week (Figure 2).

A fibroblastic proliferation develops parallelly with the capillary proliferation. Some perivasal muffs of fibroblasts orientated longitudinally were found still during the first week in the granulation tissue round the stumps and autogenous tendon transplant, as well as in the homo-

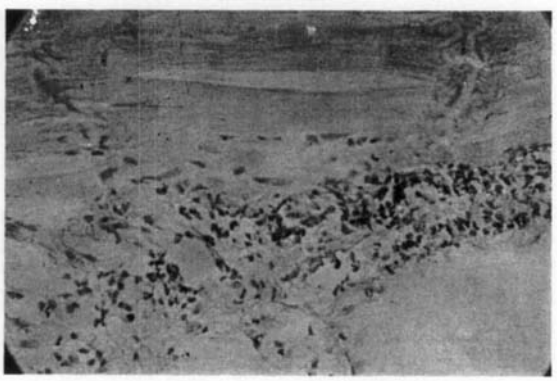

Figure 3. Necrotic region in a organized homogenous tendon transplant of the 5th week. Colouring—a combined reaction for sour mucopolysaccharides and for desoxyribonucleine acide. (× 100).

genous tendon transplants of the 2nd series. This reaction was less manifestated in the other types homogenous tendon transplants. It begins to decrease since the 3–5th week on.

Concerning the capillary and fibroblastic proliferation in the transplant it develops considerably slowlier. It is less manifestated at the end and superficial layers of the autogenous tendon transplant during the first week. In the homogenous tendon transplants preserved in hydrolizate solution and by means of freezing it begins at the end of the 1st–2nd week while in the freeze-dried homogenous tendon transplants —still later (since the 3rd week).

The process of organisation of the transplant begins earliest in the 1st and 2nd series and some later in the frozen and freeze-dried homogenous tendon transplants. In some transplants—autogenous and homogenous as well—acellular parts of the necrotic changed transplant different in their size remain among the organized tissue (Figure 3).

Reduction of the small vessels and differently expressed collagenous formation was observed in the granulation tissue between the 4th and 7th week. Bundles of fibrous tissue with collagenous fibres orientated longitudinally are formed during that period. The richness of cells elements decreases gradually and the transplant gets the appearance of a connective-tissue cicatrice. The quantity of collagenous fibres orientated longitudinally increases conversely proportionally to the reduction of the proliferated capillaries. Since the 6th–7th week on the autogenous tendon transplants receive the morphological appearance of tendon tissue which differs from the normal tendon only by its greater quan-

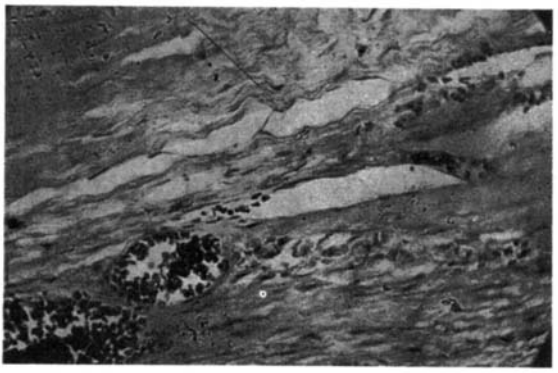

Figure 4. Vessels in a collagenized autogenous tendon transplant at the end of the 6th week. Colouring—Haematoxyline eosine. (× 100).

tity of vessels (Figure 4). This process in the homogenous tendon transplants develops slowlier. The slowliest one is in the frozen and freeze-dried homogenous tendon transplants.

DISCUSSION

The results obtained from our comparative study on fresh autogenous tendon transplants and homogenous tendon transplants preserved by means of different methods confirm the opinion that their healing runs principally in one and the same way. Our former studies (1962, 1965) and the present ones, as well as those of *Skoog & Persson, Flynn and coll., Degtiareva & Lavrishtcheva, Herzog and others* prove that necrotic changes appear in the transplanted autogenous and homogenous tendon tissue, manifestated by necrobiotic and necrotic changes of the cell elements and of the ground substance of the tendon tissue. These changes advance almost in the same terms. Tenoblastic proliferation as mentioned by *Mason & Shearon, Peer, Lindsay and others* was not established in anyone of the transplants. That makes us to consider the autogenous as well as the homogenous tendon transplants to play the role only of a bridge through which ingrow the vessels and fibroblasts from the surrounding tissue and stumps of the host.

There is no essential difference between the reparative processes in the transplanted autogenous and homogenous tendon tissue. The existing difference refers to the terms of appearance and termination of the reparative processes. A more precise difference exists between autogenous and homogenous tendon transplants, preserved by means

of different methods. The homogenous tendon transplants preserved in hydrolizate solution stand closest to the autogenous tendon transplants.

SUMMARY

The authors have carried out comparative experimental studies on the calcaneus tendon in rabbits in which they have transplanted fresh autogenous tendon tissue and homogenous tendon tissue preserved by means of different methods: in hydrolizate solution at +4° C (a methed of the authors), in 20 per cent Glycerin-Ringer's solution at —20° C and by freeze-drying. The transplants were examined every week in the course of seven weeks. Ordinary and histochemical methods were used.

It has been established that as the autogenous also the homogenous tendon transplants heal with the stumps well and serve as a bridge through which ingrow the vessels and fibroblasts from the surrounding tissues and stumps. Finally the tendon transplant turns into a scar which receives the morphological and functional features of a mature tendon. Only some quantitative difference was established in the regenerative processes between the autogenous and homogenous tendon transplants. The homogenous tendon transplants preserved by means of the authors' method stand nearest to the autogenous tendon transplants.

RESUME

Les auteurs ont procédé à des études expérimentales comparatives sur le tendon d'Achille chez des lapins auxquels ils avaient transplanté du tissu tendineux autogène frais et du tissu tendineux homogène conservé au moyen de différentes méthodes: dans une solution hydrolysée à +4° C (méthode des auteurs), dans une solution de Ringer de 20 pour cent de glycérine à moins —20° C et par lyophilisation. Les greffes ont été examinées chaque semaine pendant sept semaines. Les méthodes ordinaires et histochimiques ont été utilisées.

Il a été établi que comme les autogreffes, les homogreffes des tendons réussient aussi bien et servent comme un pont à travers lequel poussent les vaisseaux et les fibroblastes des tissus environnants et même comme par des bouts de tendon sectionné. Finalement, le tendon transplanté se transforme en cicatrice qui a les aspects morphologiques et fonctionnels d'un tendon arrivé à maturité. Il n'a pu être établi qu'une différence quantitative dans le processus de régénération entre les trans-

plants de tendons autogènes et homogènes. Les homogreffes des tendons conservés par la méthode des auteurs se rapprochent le plus aux autogreffes des tendons.

ZUSAMMENFASSUNG

Die Verfasser haben vergleichende, experimentelle Studien an der Calcaneussehne von Kaninchen ausgeführt, bei denen sie frisches autologenes Sehnengewebe und mittels verschiedener Methoden konservirtes homologenes Sehnengewebe verpflantz haben: In Hydrolysatlösung bei +4° C (eine Methode der Verfasser), in 20 Prozent Glycerin-Ringerlösung bei —20° C und mittels Gefrierungs-Trocknung. Die Transplantate wurden jede Woche im Laufe von sieben Wochen untersucht. Gewöhnliche und histochemische Methoden wurden angewandt.

Es wurde festgestellt, dass sowohl die autologenen als auch die homologenen Sehnentransplantate mit dem Stümpfen gut zusammenheilen und als Brücke durch welche die Gefässe und Fibroblasten des umgebenden Gewebes und der Stümpfe durchwuchern dienen. Schliesslich verwandelt sich das Sehnentransplantat in eine Narbe, die die morphologischen und funktionellen Zeichen einer reifen Sehne erhält. Nur einige quantitative Verschiedenheiten hinsichtlich der regenerativen Prozesse zwischen autologenen und homologenen Sehnentransplantaten wurden festgestelll. Die mittels der Methode der Verfasser konservierten homologenen Sehnentransplantate nähern sich in ihrem Verhalten am meisten der autologenen Sehnentransplantaten.

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