ELECTRICALLY STIMULATED BIOLOGICAL SKELETAL MUSCLE VENTRICLE: PRELIMINARY RESULTS OF CHRONIC EXPERIMENTS IN SHEEP AND GOAT

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Abstract - *A new type of skeletal muscle ventricle (SMV) constructed of biological materials only was designed as an aortic counterpulsation device: In a series of acute experiments in sheep the hemodynamic efficacy was demonstrated and a standardized surgical procedure was established.*
Following this a series of chronic experiments was scheduled. In six sheep the latissimus dorsi muscle (LDM) was preconditioned by means of a newly developed fully implantable ECG-triggered multi-channel-stimulator and the SMV was constructed. In one goat an unconditioned LDM was used for SMV construction.
The implanted stimulators worked without any technical problems. Two sheep died after SMV-construction due to surgical reasons. Four sheep could not be weaned from mechanical ventilation and had to be sacrificed two to three days after the procedure. The goat survived the procedure without any complications. Three months after surgery the LDM was stimulated chronically at a rate of 1:3 compared with the heart rate and the SMV was patent.
The newly developed fully implantable stimulator was tested successfully. The configuration produced distinct hemodynamic changes required for aortic counterpulsation. Goat turned out to be more appropriate for this kind of experiments than sheep. After ninety days in circulation the observed results are promising and the experiments will be continued.

Keywords: Animal experiment, Cardiac assistance, Functional Electrical Stimulation, Skeletal Muscle Ventricle

1. Introduction

The chronic shortage of donor organs for cardiac transplantation and the high costs for mechanical assist devices demand the development of alternative cardiac assist devices for the treatment of severe heart failure. Cardiac assistance by stimulated skeletal muscles is currently investigated as such a possible alternative.

A skeletal muscle ventricle (SMV) consisting of biological materials only was designed as an aortic counterpulsation device and was tested in sheep. During a series of acute experiments a standardized surgical procedure suitable for long-term studies was established. Under stable hemodynamic conditions the stimulation of the biological SMV induced a significant increase of mean diastolic pressure by 26% and reduced maximum left ventricular pressure by 5%. During pharmacologically induced periods of cardiac failure the stimulation of the neo-ventricle and increased mean diastolic pressure by 19% and reduced maximum left ventricular pressure by 8%. In all cases the diastolic peak pressure was raised to supra-systolic values during stimulation. Thus the configuration produced distinct hemodynamic changes required for aortic counterpulsation [3].

Following this a first series of chronic experiments was scheduled in order to evaluate the

reliability and hemodynamic efficacy of this newly designed stimulated SMV.

2. Material and Methods

6 adult female sheep and one adult female goat were used in this experiment. Surgical procedures were performed under general anesthesia. Animals received care in accordance with the Austrian federal law for the care and use of laboratory animals.

In all sheep four ring-shaped stimulation electrodes were implanted to the epineurium of the thoracodorsal nerve and a newly developed fully implantable stimulation unit was placed in a subcutaneous pocket near the thoracic spine prior to the construction of the SMV [5,7,9,]. In two sheep a new concept of muscle prefabrication was put into practice. In these two animals the LDM was detached from the thoracic wall, while its insertion to the humeral bone and the supplying neurovascular pedicle were preserved carefully. The LDM was divided longitudinally from its distal end up to the entry of the neurovascular bundle in order to create two muscle flaps of equal size. At the end of the procedure the LDM was reattached into its anatomical position. In four sheep the LDM was kept unaffected. In all sheep the clinically approved stimulation protocol for cardiomyoplasty was applied for preconditioning of the LDM. In the goat the LDM was not preconditioned, electrodes and stimulation unit were implanted at the time of SMV construction. All unaffected LDM were divided into two branches during SMV construction.

Prior to the main surgical procedure the pericardium and the entire thoracic aorta had been excised from fresh sheep and goat cadavers. These ”homografts” were cryopreserved according to approved techniques [4,8].

The SMV construction was performed as a standardized procedure using a left side lateral flank incision. The serratus muscle was detached from the thoracic wall. A segment of the third rib was resected subperiostally and the fifth and sixth rib were removed and preserved as pedicled vascularized grafts. Enlarging the circumference of the homograft with homologous pericardium created a neoventricle. This biological conduit was anastomosed in parallel to the descending aorta. The branches of the LDM were wrapped around the neoventricle in counter-rotating direction, applying near physiologic resting tension to the muscle and the free ends of the muscular branches were fixed to the sixth rib. Two ECG-sensing electrodes were fixed directly to the heart and the electrode leads were connected with the implanted stimulation unit. Finally the thorax was reconstructed by means of the rib grafts and the mobilized serratus muscle.

At the end of the procedure functional electrical stimulation (FES) was started. Rectangular pulses with 0.6 msec duration at a frequency of 28 Hz were used to perform bipolar burst stimulation. The current could be adjusted from 0 to 4 mAmp and actually was set to achieve maximum tetanic contraction of the LDM. R-wave triggered burst stimulation at a rate of 1:3 with the native heart rate and limited to maximum 30 contractions per minute was applied during cardiac diastole to simulate aortic counterpulsation.

3. Results

The newly developed fully implantable stimulation unit was tested in six sheep during muscle conditioning and SMV construction and afterwards. The follow-up periods reached from four up to six month, the accumulative period amounted to 30 months. The implants operated reliable and stable, technical problems were not observed.

“Prefabrication” and preconditioning of the LDM produced a poor result. In visual inspection these muscles did not produce significant muscle contraction under FES. Macroscopical and histochemical examination revealed severe signs of muscle degeneration. Preconditioning of the LDM alone produced fatigue resistant muscles consisting of slowly contracting type I muscle fibers only.

The standardized construction of the SMV was performed successfully and without complications in all animals. Activation of the biological SMV in counterpulsation mode augmented the diastole to nearby systolic values. Postoperatively the survival period in sheep ranged from one to three days only. Two sheep died from acute hemorrhage. One sheep suffered from severe neurological deficiencies and three sheep could not be weaned from mechanical ventilation. Accordingly these four sheep were sacrificed. Post mortem examination revealed a patent SMV in all sheep. The goat survived the surgical procedure without complications. Muscle conditioning was started after the operation and completed three months later. At that time a diminishing muscle function terminated the experiment. A regenerate of the third rib was found to compromise the LDM and the neurovascular pedicle on its way into the thorax. The proximal and distal part of the biological conduit was found calcified whereas the actual neoventricle did not exhibit signs of calcification. The conduit was patent with some thrombus formation adhering to the neo-ventricular wall.

4. Discussion

Principles from plastic surgery, vascular delay and tissue prefabrication were combined with the concept of muscle preconditioning in order to create a muscle which is capable to perform chronic work immediately after construction of the SMV [1,2,10]. This concept proved to be a failure. Splitting and mobilization of the LDM prior to preconditioning in order to achieve vascular delay and preconditioning as a concept of muscle prefabrication should be strictly avoided.

Preconditioning of the LDM alone led to a well known result, producing fatigue resistant muscles [6]. The contraction rate was limited to maximum 30 contractions per minute in order to avoid a complete transformation of the LDM into slowly contracting type I muscle fibers and to maintain some of the contraction velocity of the unconditioned LDM. Actually the conditioned LDM consisted of type I muscle fibres only.

A standardized surgical procedure to construct the SMV was developed during the first series of acute experiments and optimized during the presented series in sheep and goat. Several modifications were applied concerning time schedule during surgery and additional use of technical aids like cell-saver and surgical details. Especially the aortic root of the homograft was omitted and no longer used as an inflow valve into the conduit. The presented results show, that the procedure is suitable for long term animal experiments. Stimulation of the LDM in counterpulsation mode yielded about 75% of the hemodynamic changes compared with the results from the series of acute experiments [3]. Probably the well known reduction in force and contraction velocity of the conditioned LDM due to the conditioning program is responsible for the reduced hemodynamic efficacy. However, the construction produced distinct hemodynamic changes required for aortic counterpulsation, although the experiments did not provide enough information to evaluate the overall influence to the circulation. First useful and positive experiences were gained concerning the patency of the fully biological SMV. Thrombosis formation did not represent a major problem, even without sufficient anticoagulation. The follow up periods indeed were too short to answer the important question of reendothelialisation of the homograft.

Regarding the intractable weaning problems in sheep the use of this species was abandoned. The first experiment with goat was promising, enabling to observe the biological SMV over ninety days in circulation. Thus the experiments will be continued with goats.

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