Long-term follow-up after autologous skeletal myoblast transplantation in ischaemic heart disease†

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Abstract

OBJECTIVES: Short-term follow-up after autologous skeletal myoblasts (ASM) transplantation (Tx) (Myoblast Autologous Grafting in Ischaemic Cardiomyopathy (MAGIC) Phase II Study) for the treatment of ischaemic cardiomyopathy revealed improved left ventricular (LV) remodelling. Our study reports the longest long-term worldwide follow-up of a single-centre cohort, focusing on the safety and efficacy of ASM-Tx.

METHODS: The multicentre MAGIC Phase II Study involved 120 patients and was conducted between 2004 and 2006. Out of the 120 patients involved in the entire study, the cohort treated at our institution contained 7 patients only. These 7 patients received ASM-Tx (injection volume: 400 million cells, n = 2 low dosage; 800 million cells, n = 2 high dosage) or placebo (n = 3) injections, in addition to coronary artery bypass grafting (CABG). After closure of the MAGIC registry, we conducted a long-term follow-up for our 7-patient cohort. The mean follow-up was 72.0 ± 5.3 months. The follow-up was complete for echo data, implanted cardioverter defibrillator (ICD) report, clinical investigation and New York Heart Association (NYHA) class.

RESULTS: At final follow-up, all the patients were alive, and 5 were in NYHA class 1 or 2. There were 6 hospitalizations for congestive heart failure during the follow-up (1 patient from each group). One patient (placebo group) was treated twice for ventricular fibrillation by the ICD. The LV ejection fraction remained stable in all the three groups (31.1 ± 3.9% preoperative vs 29.4 ± 4.4% at final follow-up). The LV volumes were reduced in the high-dosage group, remained unchanged in the low-dosage group and deteriorated in the placebo group.

CONCLUSIONS: Our long-term data confirm the findings of the MAGIC study. The LV function did not improve, but the long-term LV volumes in the high-dosage group were reduced. During the follow-up, there were also no additional arrhythmogenic incidences. Our data could imply that CABG in combination with ASM-Tx is safe and has beneficial therapeutic effects in the long-term. However, due to the small patient number, the clinical impact is limited.

Keywords: Cardiac cell therapy · Long-term follow-up · Chronic ischaemic heart disease

INTRODUCTION

At present, cardiovascular disease remains a major public health problem. In 2004, nearly half of all reported deaths in the German population were due to cardiovascular disease. Chronic ischaemic heart disease caused by myocardial remodelling presented the highest gender-specific mortality rate for women (11.1%) and men (9.3%) in 2004 in Germany [1, 2]. Chronic ischaemic heart disease [3] has a poor prognosis, and can only be positively affected by coronary revascularization if there is still viable myocardium [4]. Since treatment options are limited for patients with chronic ischaemic heart disease, it presents a need for innovative therapeutic concepts for these patients.

Myoblasts as a possible source of myocardial cell therapy have been investigated over the past decade. Menasché et al. [5] described intramyocardial cell delivery of myoblasts as a potential treatment option for chronic ischaemic heart disease. In 2004, the MAGIC Phase II Study, based on the promising results of the Phase 1 study [6], commenced. This study evaluated the safety and efficacy of myoblast Tx in a double-blinded, multicentre, prospective randomized placebo-controlled set-up [7]. In 2006, Philipp Menasché published the long-term follow-up data of the Phase 1 study experience of myoblast Tx in patients with chronic ischaemic heart disease. The Phase 1 cohort was the first to publish long-term data; however, the follow-up was conducted over a wide range from 18 to 58 months (median follow-up of 49.4 months) [8]. The MAGIC study protocol had a limited 12-month follow-up. Having identified the need for a long-term completed follow-up in this field of study, we continued to follow-up our patients who participated in the MAGIC Phase II Study up until July 2011.

Skeletal myoblasts as a possible source for regenerating cardiac therapy still continue to remain a topic of research in experimental [9, 10] and clinical set-ups [11].

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MATERIALS AND METHODS

Study design and group

The inclusion and exclusion criteria, intervention, randomization, blinding procedures and the results of the MAGIC Phase II Study have been described in detail by Menasché et al. [7].

Between 2004 and 2006 a total of 120 patients were included in the MAGIC Phase II Study. Ten of these patients were included in the study at our institution, of whom 7 were randomized and treated, in addition to coronary bypass grafting, with a myoblast or placebo solution. The patient characteristics are summarized in Table 1. All the participating patients of the initial Phase 2 Study signed informed consent, which was approved by the local ethics committee. Our 7 patients also signed informed consent, and gave access to their complete medical history for the long-term follow-up.

To recapitulate the concept of the MAGIC Study: patients with chronic ischaemic heart disease, with a left ventricular ejection fraction (LVEF) from 35% down to 15% and an indication for a coronary artery bypass operation were included. Prior to randomization, all patients underwent transthoracic ultrasound (rest and dobutamine stress) to include only patients with a fixed akinesia affecting at least two accessible, contiguous LV segments [12]. After randomization, 10 g of skeletal muscle was taken from the upper thigh. A cell solution containing skeletal myoblasts was manufactured from the biopsy taken (vastus lateralis muscle). The cell solution, containing either 800 × 10^6 (high-dosage group) or 400 × 10^6 cells (low-dosage group) or placebo solution, was injected into the infarcted myocardium and the border zone of the infarction during the coronary bypass operation. All patients (placebo and treatment group) received an implanted cardioverter defibrillator (ICD) and treatment with amiodarone prior to discharge based on the experience of the Phase I Study.

Follow-up data

Between 2004 and July 2011, the patients were followed up on an annual basis. The follow-up included clinical investigation, ICD report and transthoracic echocardiogram when required. The echocardiographic examinations for LVEF and left ventricular end-diastolic volume (LVEDV) and left ventricular end-systolic volume (LVESV) were performed according to the standard protocol of the study. For the follow-up echocardiographic studies, the Vivid 7 (GE Healthcare) was used.

Statistics

All results are reported as an average and mean ± standard deviation (SD). The data include the original source data of the MAGIC Phase II Study (centrally evaluated echocardiogram studies, ICD reports) of our patients and locally generated and interpreted data after closure of the study register. The small patient numbers and the intrindividual variability of the results limit interpretation, making statistical analysis inappropriate. For this reason, descriptive statistical methods were used.

RESULTS

There were 3 patient withdrawals prior to randomization. Patient 4 was excluded due to heart failure prior to scheduled surgery. Patient 6 had an ejection fraction of >35% and was thus excluded. Patient 7 withdrew himself from the study for personal reasons.

We followed up all the 7 male patients who were randomized for the MAGIC Study, 2 of whom were allocated to the high-dosage group, 2 to the low-dosage and 3 to the placebo group. The mean follow-up was 72.0 ± 5.3 (range 80–64) months. The follow-up was complete (100%). Bypass grafting was performed following the preoperative angiogram. All patients had an initial posterior ischaemic area which was treated either with the cell or with the placebo solution. The pre- and perioperative details of all patients are given in Table 1.

Safety issues

There was no mortality during the follow-up. At the last follow-up (see Table 2), all the patients were in a good, stable medical condition at home, using specific medication including aspirin and/or clopidogrel or phenprocoumon, β-blocker, angiotensin converting enzyme-inhibitor or AT-1 receptor antagonist and statin. Additionally, 2 of the 7 patients were treated with digoxin, 4 of the 7 patients with diuretics, 2 of the 7 with an aldosterone antagonist, 1 of the 7 with central α-blockade and 1 of 7 patients with sotalol (either in combination or in isolation). Five patients had retired during the follow-up and 2 patients were still employed.

The patients of the high-dosage group remained in their New York Heart Association (NYHA) functional class during the follow-up. There was no readmission for heart failure or coronary reintervention in this group. Patient 1 experienced two episodes of ICD firing due to sensing lead failure. Patient 3 was treated once for slow ventricular tachycardia.

In the low-dosage group Patient 9 improved from NYHA class 3 to 2 and, Patient 2 deteriorated from NYHA class 1 to 2. The latter patient was also readmitted four times for heart failure within the follow-up period and received two ICD shocks in the first month (for supraventricular tachycardia and cluster ventricular tachycardia) and one in the third month (for high frequency atrial fibrillation). Patient 9 experienced ICD firing due to sensing lead failure and underwent percutaneous coronary intervention (PCI) because of progression of the coronary artery disease. He was, therefore, treated with PCI and stenting (Taxus® in first marginal artery).

At final follow-up, the NYHA class of the 3 patients of the placebo group remained unchanged. Patient 10, however, did improve in the early postoperative phase, but deteriorated again 40 months postoperatively from NYHA 2 to 3. Following echocardiography, an anterior infarction might have been the underlying cause of the deterioration, but a final diagnosis could not be established due to poor patient compliance. None of the patients in this group underwent coronary intervention, but Patient 8 and 10 were each readmitted once for heart failure. The ICD of Patient 5 was explanted after the system got infected with Staphylococcus aureus and to date has not been replaced. Patient 10 was treated for ventricular fibrillation at 5 and 24 months.

During the follow-up, there was no clinical evidence for any patient of either relevant long-term side-effects after myoblast wash out from the heart to peripheral organs or cancer.

Functional issues

The functional analysis is based on the results of the echocardiographic investigations throughout this study. The investigation at
Table 1: Patient characteristics

<table>
<thead>
<tr>
<th>Patient profile</th>
<th>Randomization</th>
<th>Myoblast target</th>
<th>Age</th>
<th>Body mass index</th>
<th>NYHA</th>
<th>Previous Infarction</th>
<th>Ejection fraction [%]</th>
<th>Rhythm preoperative</th>
<th>Arrhythmia preoperative</th>
<th>Coronary artery disease</th>
<th>Pathological wall motion preoperation</th>
<th>Bypass grafting</th>
<th>Diabetes mellitus</th>
<th>Hypertension</th>
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<th>COPD</th>
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<tbody>
<tr>
<td>1 2 3 4 5 6 7 8 9 10</td>
<td>High dose Posterior</td>
<td>Low dose Posterior</td>
<td>High dose Posterior</td>
<td>Excluded na</td>
<td>Placebo Posterior</td>
<td>Excluded na</td>
<td>Withdrawn na</td>
<td>Placebo Posterior</td>
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<td>Placebo Posterior</td>
<td>Low-dose Posterior</td>
<td>Placebo Posterior</td>
<td>Placebo Posterior</td>
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<td>54 64 53 67 55 64 56 68 55 63</td>
<td>25 5 25 24 27 2 2 2 3 3</td>
<td>Yes Yes Yes Yes Yes Yes Yes Yes Yes</td>
<td>33 34 25 24 31 35 20 34 32 26</td>
<td>SR Multif. VES Multif. VES SR VES Multif. VES Multif. VES</td>
<td>Multif. VES Multif. VES Multif. VES Multif. VES</td>
<td>Anterior + posterolateral Anterior + inferior Posterospetal Posterior</td>
<td>OM CX LAD, diag., OM LAD, CX LAD, OM LAD, diag., OM, RIVP LAD, OM LAD, diag., OM, RIVP</td>
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NYHA: New York Heart Association; COPD: chronic obstructive pulmonary disease; PAVD: peripheral artery vessel disease; SR: sinus rhythm; AF: atrial fibrillation; VES: ventricular extra systole; multif.: multiform; ant.: anterior; OM: obtuse marginal branch; CX: circumflex artery; LAD: left anterior descending; diag.: diagonal branch; RIVP: right marginal artery; na: not applicable.
6-month follow-up for Patient 10 and the investigation for Patient 8 at 12 months and 69 months could not be adequately interpreted due to poor echo quality.

The geometry of the left ventricle changed during the follow-up. There was an initial improvement in the three groups for both LVESV and LVEDV at 6 and 12 months follow-up. This positive tendency continued in the long-term follow-up for the high-dosage group only. The low-dosage group, however, did not show this positive tendency in the long term when compared with the pre-operative levels. The diameters in the placebo group deteriorated over time and showed an increase of the LV volumes, particularly after 12 months. Details of the echocardiographic findings described above are given in Table 3 and Figs 1 and 2.

The contractility of the left ventricle and the ventricular wall dynamics showed different tendencies. The LVEF improved in the high-dosage group within the first year from 29 to 35.5%. During the long-term follow-up it decreased to 32%. The low-dosage group showed its peak at 6 months when the LVEF reached 35%. At 12-month follow-up, it decreased to 29%, but thereafter remained stable. In the placebo group, the LVEF showed an overall decrease during the follow-up: at 6 months the LVEF decreased from 31 to 25%, at 12 months it increased again to 32% and at the final follow-up decreased again to 28%.

Within our cohort a total of 10 akinetic segments were treated. There were five segments (2 patients) treated with a high-dose myoblast solution, and five segments (2 patients) with the low-dose cell solution. Additionally, seven segments (3 patients) were injected with the placebo solution (as demonstrated in Table 3). At 6-month follow-up, 1 of the 10 grafted segments (Patient 1—high-dosage group) showed a new contractility. At the final follow-up, 3 of the 10 grafted segments had improved (1 of the high-dosage group and 2 of the low-dosage group). In the placebo group, there was an improvement at 6-month follow-up in 1 of 7 segments, and at the final follow-up in 2 of 7 segments. Due to poor investigational quality, 2 patients could not be re-evaluated at follow-up.

<table>
<thead>
<tr>
<th>Table 2: Safety issues</th>
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<tbody>
<tr>
<td><strong>Patient</strong></td>
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<tr>
<td>1 (hd)</td>
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<td>2 (ld)</td>
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<td>3 (hd)</td>
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<tr>
<td>5 (pla.)</td>
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<tr>
<td>8 (pla.)</td>
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<td>9 (ld)</td>
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hd: high dosage; ld: low dosage; pla.: placebo control; NYHA: New York Heart Association.

<table>
<thead>
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<th>Table 3: Functional issues</th>
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<tbody>
<tr>
<td><strong>Patient</strong></td>
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<tr>
<td>1 (hd)</td>
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<td>2 (ld)</td>
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<tr>
<td>3 (hd)</td>
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<td>8 (pla.)</td>
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<tr>
<td>9 (ld)</td>
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<td>10 (pla.)</td>
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hd: high dosage; ld: low dosage; pla.: placebo control; LVEF: left ventricular ejection fraction; LVESV: left ventricular end-systolic volume; LVEDV: left ventricular end-diastolic volume; Pre: preoperative; M6: 6-month follow-up; M12: 12-month follow-up; FU: final-month follow-up; XX: no values available.

![Figure 1: Left ventricular end-diastolic and volume (LVEDV) over time.](https://academic.oup.com/icvts/article-abstract/18/1/61/674476/616167)

![Figure 2: Left ventricular end-systolic volume (LVESV) over time.](https://academic.oup.com/icvts/article-abstract/18/1/61/674476/616167)
DISCUSSION

The MAGIC Phase II Study was the first multicentre double-blinded, prospective randomized, placebo-controlled study evaluating the safety and efficacy of skeletal myoblast Tx for ischaemic heart disease. The patients of this long-term follow-up were part of the MAGIC Phase II Study at our centre. This study did not demonstrate a significant improvement in regional or global LV function, but showed beneficial effects of myoblast engraftment on LV dimensions. There was no statistical significance demonstrated for an increased risk of arrhythmias after myoblast Tx [7].

This long-term study revealed the same findings as those of the MAGIC Phase II Study. The improved remodelling in the high-dosage group with reduced LV volumes remained unchanged and there was also no increased incidence of ventricular arrhythmia treated or detected by the implanted ICD. No side-effects after myoblast wash out were detected clinically.

Long-term survival after isolated coronary artery bypass operation for patients with ischaemic heart disease and an impaired LV function is stated to be ～70% [13] or have an annual mortality rate of 3–4% [14]. In our small study cohort, 1 patient of the low-dosage group was readmitted three times for congestive heart failure within 6 months of the initial treatment. Late readmissions (i.e. after 2 years) were predominantly seen in the placebo group. Epidemiological studies quote a risk of rehospitalization of ～33% within 2 years [14]. Over our completed 5-year follow-up period, all the patients who participated in our study remained clinically stable. This clinically stable condition of all our patients could be attributable to the operative revascularization in combination with adequate medical treatment.

Following the experience of the Phase I Study, the major safety concern using skeletal myoblasts as a possible source for myocardial cell therapy was the arrhythmogenic potential [6]. The electrophysiological reason for this increased tendency for arrhythmias is the absence of electromechanical coupling (i.e. the absence of gap junctions) of the transplanted myoblasts [15]. Furthermore, the MADIT II Study revealed an increased risk of ventricular arrhythmia in patients with a severely reduced ejection fraction in combination with an ischaemic history [16]. In all the 7 patients of our cohort, we detected ventricular arrhythmia prior to the study intervention (see Table 1). The Magic Phase II Study did not reveal a statistically significant difference for an increased risk of arrhythmia among the three groups after intervention [7]. Possic et al. [11] also documented an increase of ventricular arrhythmias in the myoblast groups, but just by number. However, their results were not statistically significant. The 2 patients of our cohort who had adequate ICD firing in the early phase at 1, 3 and 5 months follow-up were in the low dosage and in the placebo-controlled group. Both patients had already revealed multiform ventricular extrasystoles and salvos in the preoperative Holter ElectroCardioGram and, as every MAGIC patient received amiodarone treatment after myoblast Tx for a minimum of 6 months. The late ICD firing in Patient 10 could probably be attributed to the fact that the patient did not take his prescribed medication on a regular basis, as well as taking unsupervised sildenafil [17].

Other safety concerns of cardiac cell therapy include arrhythmogenesity [6], as well as oncogenic transformation [18], multiorgan seeding [19], aberrant cell differentiation [20] and accelerated atherosclerosis, which could lead to a higher incidence of coronary events [21].

Cell therapies in general may lead to an improved LV remoulding measured mainly with reduced LV volumes and slightly improved ejection fraction [11]. The mechanisms by which cell therapy could lead to an improved remodelling are not clearly understood. Various theories have been introduced and rejected. Seidel et al. summarized three possible mechanisms by which transplanted myoblasts, with respect to their cellular activity, might improve cardiac function: (i) the implanted cells act as a strengthening scaffold within the ventricular wall and lead to a limitation of the infarct size and an improved remodelling; (ii) due to their contractile properties, transplanted myoblasts contribute directly to the cardiac systolic function; (iii) the transplanted myoblasts constitute a potent source of growth and/or angiogenic factors as well as a paracrine effect stimulated through the transplanted cells. At present, a cytokine-paracrine hypothesis is favoured, with the assumption that >30 factors involved in different pathways and cascades are responsible for the improved remodelling [19]. However, the observed changes are most likely multifactorial [22]. After an initial improvement of the myocardial volumes at 6-month follow-up, the 4 patients of the treatment groups showed a slight decline towards the preoperative levels at 12-month follow-up. At final follow-up, the high-dosage group showed smaller volumes than the low-dosage group. The placebo group also showed an initial improvement at 6-month follow-up, but continued to increase the LV volumes again from 6 months onwards. Similar findings were documented for the LVEF for the follow-up period. Left ventricles have also a chance for improvement after isolated coronary artery bypass [23]. Therefore, it must be taken into consideration that the documented LV changes in our small cohort could be incidental.

The evaluation of the graft segments for wall motion with echocardiography is questionable in this study set-up. Despite the fact that we followed the standardized protocol, echo investigational quality was often poor in certain patients and received data were thus incomplete. It is a known fact that echocardiographic imaging can be of poor quality with a high intra- and interobserver variability [24]. Magnetic resonance imaging is known to provide reliable information with acceptable variability, but unfortunately could not be considered an option due to the ICD implant [25].

LIMITATIONS

Patient numbers in the groups are low and comparisons are thus of a descriptive nature rather than statistical. The results of this long-term follow-up study are thus limited to the small patient numbers, and should be taken into consideration when applying it to bigger patient populations. Interobserver variability needs to be taken into account since follow-up echocardiographies were not interpreted in the MAGIC Phase II Study core laboratory.

CONCLUSION

Cardiac cell therapies, such as skeletal myoblasts, are known to limit infarction size and improve cardiac remodelling. Our long-term data confirm the findings of the MAGIC study. The LV function did not improve, but the long-term LV volumes in the high-dosage group were reduced. During the follow-up, there were also no additional arrhythmogenic incidences in the high-dosage group or the low-dosage group. Our data suggest that coronary artery bypass grafting (CABG) in combination with autologous skeletal myoblasts...
transplantation could be safe and might have beneficial therapeutic effects in the long-term. However, due to the small patient number, the clinical impact is limited. The mechanisms by which cell-based therapy improves cardiac function are not yet clearly understood and there is an obvious need to conduct further long-term studies with bigger patient groups to prove the feasibility and safety of these modern concepts of cell delivery for the treatment of ischaemic cardiomyopathy. Therefore, skeletal myoblasts remain a possible source of treatment for chronic ischaemic heart disease.

Conflict of interest: We certify that there is no conflict of interest with any financial organization regarding the material discussed in the manuscript.

REFERENCES


