Hybrid techniques for myocardial regeneration: state of the art and future perspectives

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Abstract

Myocardium has a limited proliferative capacity, and adult hearts are considered incapable of regenerating after injury. A significant loss in the viable myocardium eventually diminishes the heart's ability to contract synchronously, leading to heart failure. Despite the development in interventional and pharmacological treatment for ischemic heart disease and heart failure, there is a significant number of highly symptomatic patients. For these individuals, treatments that stimulate myocardial regeneration can offer alleviation of dyspnea and angina and improvement in quality of life. Stem cells are known to promote neovascularization and endothelial repair. Various stem cell lines have been investigated over the years to establish those with the highest potential to differentiate into cardiomyocytes, including bone marrow-derived mononuclear cells, mesenchymal stromal cells, CD34+, CD133+, endothelial progenitor cells, and adipose-derived mesenchymal stromal cells. Stem cell studies were based on several delivery pathways: infusion into coronary vessels, direct injection into the injured region of the myocardium, and delivery within the novel bioengineered scaffolds. Acellular materials have also been investigated over the years. They demonstrate the therapeutic potential to promote angiogenesis and release of growth factors to improve the restoration of critical components of the extracellular matrix. This review summarizes hybrid cardiac regeneration treatments that combine novel bioengineering techniques with delivery approaches that cardiac surgeons can provide.

Key words: cardiac regeneration, cardiac surgery, stem cells, hybrid technique, scaffold, bioprinting.

Introduction

Myocardium has limited proliferative capacity and adult hearts are considered incapable of regenerating after injury [1, 2]. A large loss in the viable myocardium eventually diminishes the heart's ability to contract synchronously, leading to heart failure (HF). In Poland, HF affects about one million people, and an additional 250,000 cases are predicted over the next 25 years [3]. The prevalence of underlying causes for HF vary across the world; however, ischemic heart disease (IHD) is the most common one in Europe and North America [4]. Despite unquestionable progress in medical therapies and revascularization over the last 3 decades [5–7], the problem of ischemic heart failure is increasing, and is projected to further increase in the next decades [8].

Myocardial revascularization is known to effectively relieve angina and improve exercise capacity and quality of life [9]; however, some myocardial regions may be ineligible for direct revascularization due to the lack of a proper target vessel or lack of viability. Therefore, surgical techniques designed to restore myocardial contractility have their limits. Excluding infrequently performed procedures such as the Dor (pericardial-lined Dacron endoventricular circular plasty) and Batista (reduction ventriculoplasty) procedures, the only option beyond left ventricle assist device implantation is heart transplantation [10, 11].

Nevertheless, despite the undoubted development in conventional, both interventional and pharmacological, strategies, there is still a great number of patients who

experience significant IHD and HF symptoms. For these individuals, treatments that stimulate myocardial regeneration can offer alleviation of dyspnea and angina, and improvement in quality of life. Therefore, this subgroup of patients has been the most investigated one in regard to regenerative approaches over the last decades. However, based on the recent European Society of Cardiology guidelines, larger randomized controlled trials (RCTs) and registries are still required to define the role of additional treatment modalities to decrease non-responder rates and ascertain benefit beyond potential placebo effects [12].

In general, the scientific interest in regenerative medicine arose from fundamental studies, which showed that some vertebrates can regenerate myocardium throughout life. Orchestrated waves of inflammation, matrix deposition and remodeling, cardiomyocyte proliferation and tissue architecture restoration are commonly seen in heart regeneration models, including neonatal mice [13].

Although various approaches for regenerative treatment delivery have been studied, cardiosurgical access in the only one that allows direct delivery to the desirable part of the myocardium. In general, cardiac surgery allows the heart to be accessed through median sternotomy (Figure 1 A), ministernotomy (Figure 1 B) or left minithoracotomy (Figure 1 C). However, as studies on regenerative therapies show, minimally invasive approaches seem to be preferred. This review summarizes these hybrid techniques for regenerative treatments. An electronic search of MEDLINE, Embase, the Cochrane Library and ClinicalTrials.gov databases was performed on 15th July 2022 to identify relevant articles using the following key words: guided tissue regeneration, cardiac regeneration, cardiac surgery, surgical delivery, cardiac tissue engineering, bioprinting, stem cells, scaffold, biomaterial.

Stem cell-based therapies

Stem cells are known to promote neovascularization and endothelial repair. Various cell populations such as bone marrow-derived mononuclear cells (BM-MSCs), mesenchymal stromal cells (MSCs), CD34+, CD133+, endothelial progenitor cells, and adipose-derived mesenchymal stromal cells (ADSCs) have been investigated so far [14–17]. MSCs along with other pluripotent stem cells have been proven to effectively stimulate angiogenesis and cardiac regeneration. BM-MSCs have been commonly used in stem cell-based therapies because of their wide availability and multipotency. ADSCs have been acknowledged for the feasibility and safety of repeated harvest and proliferation capacity that does not decline with aging.

Intracoronary stem cell delivery

The first evaluations of the efficacy of intracoronary stem cell delivery were not promising. Intracoronary infusion of BM-MSCs did not improve left ventricle (LV) function after ST-segment elevation myocardial infarction (STEMI) despite concomitant successful angioplasty of the infarct-related artery [18]. Similar conclusions were made after the results of intracoronary infusion of allogeneic cardiac stem cells (CDCs) were published (ALL-STAR trial) [19]. The technique was considered safe, but it did not confirm the hypothesized scar size reduction in patients with post-MI LV dysfunction. On the other hand, the CIRCULATE-AMI trial with MSCs showed significant improvement in myocardial contractility and LV remodeling inhibition but only at 3 years follow-up, contrary to nonsignificant changes found 12 months following the procedure [20, 21]. However, this observation may arise from the stem cell line used rather than the prolonged observation period, as consistent results were obtained in other studies with umbilical cord MSCs [22, 23].

Nevertheless, despite initial disappointments with intracoronary stem cell delivery, new trials have been designed to investigate the potential feasibility of intracoronary infusion during cardiac surgery. SCIPIO was a first-in-human, randomized placebo-controlled trial to assess the safety of autologous c-kit(+) CSC delivery in patients with IHD and HF undergoing coronary artery bypass grafting (CABG) (NCT00474461). CSCs were isolated from the right atrial appendage after being harvested and pro-
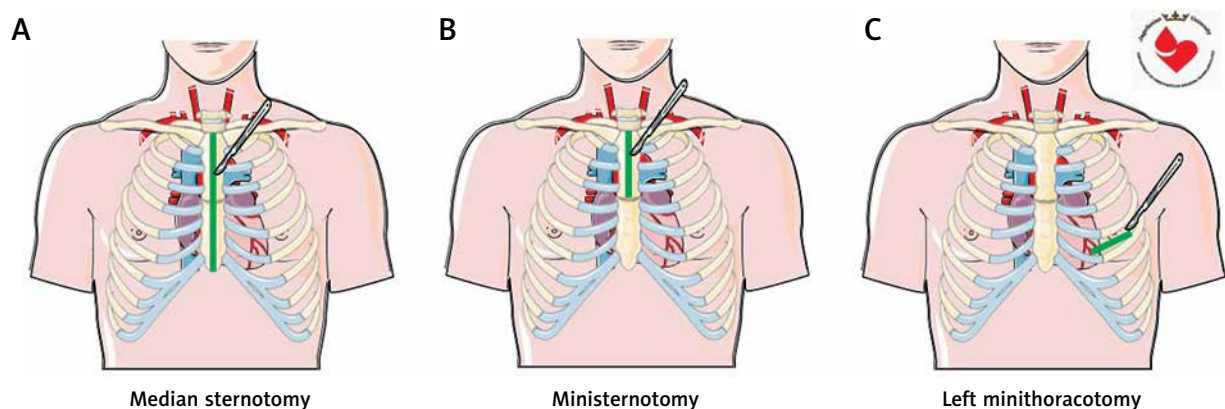

A Median sternotomy   **B** Ministernotomy   **C** Left minithoracotomy

Figure 1. Surgical approaches

cessed during surgery. Isolation of CSCs from cardiac tissue was proven to be feasible in the operating room and did not alter practices during CABG. Most importantly, cardiac magnetic resonance showed that patients treated with intracoronary CSC infusion experienced a striking improvement in both global and regional LV function, a reduction in infarct size, and an increase in viable tissue that was present at both 4- and 12-month follow-up [24].

Whether the controlled infusion on a non-beating heart or the cardiac origin of stem cells was responsible for the success of this approach remains an issue to be solved by future studies. Although still awaiting results, similar design with intra-CABG c-kit(+) CSC infusion has been proposed by Sakakibara Heart Institute recently (NCT03351400). Moreover, the potential feasibility of another delivery route, via trans-bypass graft, is to be examined in a subgroup of patients in an ongoing randomized placebo-controlled trial with the use of Cardio-Cell (Regeneration of Ischemic Damages in Cardiovascular System Using Wharton's Jelly as an Unlimited Source of Mesenchymal Stem Cells for Regenerative Medicine; NCT03418233).

Direct myocardial stem cell implantation

The idea to directly implant cardiomyocytes into the injured area was initially tested in animal models. Contrary to adult cardiomyocytes, which did not survive under any conditions, fetal and neonatal cells formed new and mature myocardium in inbred rats [25]. The procedure can be done either under direct vision, usually using minimally invasive surgical access (Figure 1 C) or percutaneously (Figure 2).

Intramyocardial injections were first reported in clinical trials in 2007 with autologous BM-MSCs in patients with previous MI and severe LV dysfunction. Improved regional wall thickening and perfusion scores were observed at the 3-month follow-up [26]. The beneficial effect of BM-MSC implantation seems to remain present throughout longer observation periods, as other studies show [27–30].

The combination of ADSCs' direct implantation and creation of small channels using a medical laser through a left sided mini-thoracotomy was described by Konstanty-Kalandyk *et al.* [31, 32]. Excellent safety and feasibility results of a simultaneous harvest and implantation were provided. The Athena trial was also designed to investigate the efficacy of direct ADSC implantation. Initial results were promising for quality of life improvement at the 1-year follow-up, but the trial was prematurely terminated due to non-ADSC-related adverse events and subsequent prolonged enrollment time [33]. Efficacy results from large randomized placebo-controlled trials are still lacking; however, improvement in overall LV contractility and regional wall thickening combined with alleviation of symptoms has been reported throughout most of the studies. Currently, we are awaiting results from two large multicenter RCTs using direct intramyocardial injections of ADSCs [34, 35].

To conclude, most of the stem cell-based therapies were summarized in the systematic review from the Cochrane Database [36]. Based on the results from a total of 1010 participants from 21 randomized studies (with intramyocardial or intracoronary autologous adult stem/progenitor cell delivery) that reported long-term fol-
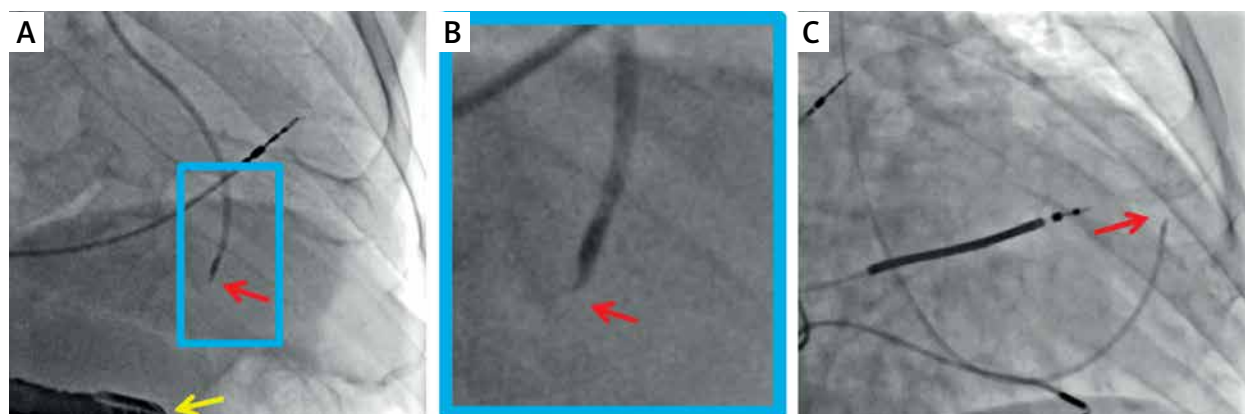

Figure 2. Typical radiographic images of percutaneous trans-endocardial delivery of stem cells using a dedicated, steerable catheter with a curved needle (red arrow) that has an adjustable length and possesses side holes. Effective anchoring of the needle in the target site is confirmed with a minimal contrast injection. The delivery is performed to pre-specified myocardial segments (based on their viability and thickness) under transesophageal echo control (**A**, yellow arrow) or using an LV-gram-based myocardial map integrating information from echocardiography and/or magnetic resonance imaging (**C**). Examples show standardized cell suspension (cocktail with pro-angiogenic factors) delivery to a pre-specified site in the inferior wall (**A**) and anterior wall (**C**). **B** is a close-up of A. The delivery occurs independently of the cardiac cycle (i.e., throughout systole and diastole). Images courtesy of Prof. Piotr Musialek and Dr. Adam Mazurek

low-up (> 12 months), a mortality rate of 4.8% (28/587) was observed in participants who received cell therapy compared with 15.4% (65/423) in those who received no cells. A meta-analysis showed that cell therapy reduced the risk of long-term mortality (RR = 0.38, 95% CI: 0.25 to 0.58; participants = 1010; studies = 21; I^2 = 0%). However, further larger studies may be needed to draw robust conclusions as most of the included studies were very small, leading to a risk of small-study bias and spuriously inflated effect sizes. Nevertheless, the observed long-term survival benefit should be considered promising, especially as it was observed irrespectively of the underlying diagnosis (IHD, HF secondary to IHD, refractory angina) and co-interventions, and included only results up to 14th Dec 2015.

Matrix scaffolds and bioengineered myocardial patches

The creation of bioengineered biomaterials made it possible to design delivery systems that provided a scaffold for stem cells. This drew attention to the potential of replacing damaged tissue with fabricated and personalized novel tissue [37]. Recent technological development enabled the implementation of induced pluripotent stem cells (iPSCs) into a bioengineered 3-dimensional (3D) microtissue (referred to as 'cardiospheres', 'cardiac spheroids', or 'cardiac organoids'). The direct implantation of such bioengineered patches has been investigated in rodent and swine models with excellent results [38–40]. This technique offers a single, but probably highly important advantage over the previously discussed stem cell-based therapies, as it supplies the stem cells with a microenvironment that influences tissue growth and cell behavior [10].

Early trials with human participants were performed within the last decade and the first case report of a cellular patch applied to a human heart was described in 2015 by Menasche *et al.* [41]. The cell-loaded fibrin patch was developed using human embryonic stem cells (ESC), retrieved from Master Cell Bank, which were amplified and cardiac-committed. The procedure was performed in a 68-year old man through a median sternotomy with concomitant CABG to the non-infarcted area. With 6 more implantations in the ESCORT trial, all concomitant to conventional cardiac surgery, the researchers demonstrated feasibility of the procedure and overall improvement in HF symptoms and myocardial contractility [42].

Also another approach to combine stem cells with the matrix patch directly placed on the injured area of myocardium is currently being investigated (NCT04011059). The study is designed to assess neomyogenesis after the injection of Wharton's jelly-derived MSCs into the extracellular matrix placed on the epicardium during CABG in patients with previous MI.

Nevertheless, the issues of scalable manufacturing, to personalize the size of the patch, and patient-compatible 3D bioprinting, to avoid the need of immunosuppression, have not been resolved by clinical trials so far. However, the successful use of bioinks to create microfibrous scaffolds that can be engineered into human cardiac organoids was described by Zhang *et al.* in 2016 [43]. Shortly after, Noor *et al.* presented promising results for iPSCs, generated from the patient's own omental stromal cells, combined with personalized bioinks created from fatty tissue extracted from the patient [44]. In this study, the bioengineered patch was designed to match the exact region of myocardial injury and to fit the blood vessel geometry to optimize the blood supply to the patch. This was established based on the computed tomography scans of volunteer patients, and prints were made as proof of concept. Although *in vivo* animal implantation experiments are still to be performed, this technique may make it possible to supply the patient with a fully personalized and biocompatible myocardial patch that can be implanted through minimally invasive cardiosurgical access in the future.

Other approaches to aid the blood supply to bioengineered patches included built-in vasculature and implantation with direct surgical anastomosis to the vessels [38, 45]. However, such techniques have not yet reached clinical trials.

Acellular materials

Acellular materials have also demonstrated preclinical therapeutic potential to promote angiogenesis and release of growth factors to improve restoration of key components of the extracellular matrix [46]. As cardiac surgeons already have experience with synthetic materials for structural purposes, such as GORE-TEX and other textile grafts, a thin mesothelial patch can easily be sewn on the epicardium. A bioinductive extracellular matrix (CorMatrix Cardiovascular Inc., United State) has already been tested in a porcine model and significant myocardial recovery was observed after epicardial repair [47].

Future perspectives

Other sources for stem cells are currently being investigated. In the study by Wang *et al.*, a portion of the thymus, that is usually removed and discarded during neonatal cardiac surgery, was evaluated for the possibility of MSCs' isolation and cardiac regenerative potential. The primary rationale of the study was that newborns undergoing cardiac surgery are at risk of developing end-stage HF over time. Collected MSCs were implanted in rat models with promising results with no need for additional scaffolds [48].

Moreover, the potential of both bioengineered and decellularized 3D neoscaffolds to create a full bioartificial heart is still being investigated. Initial reports show

promising results, but future clinical strategies and applicability seem to be years ahead [49–52].

Nevertheless, a possibility of a few much simpler regenerative approaches has arisen, since the endogenous potential of cardiomyocytes has been further understood in fish, amphibians and neonatal mammals. Several translational and transcriptomic markers have been identified as responsible for the capability to repair cardiac damage. Therefore, direct delivery of selected microRNAs, to endow pro-proliferative activity, and cyclins, to reactivate the cell cycle, hold great potential for cardiac regeneration [53].

Conclusions

Although all of those techniques present several challenges, at least some have the potential to develop into a clinically available approach in the coming years. If successful, regenerative technologies have the capacity to re-shape therapeutic goals and to promote cardiac regeneration as an additional part of cardiac surgery. The number of potential treatment options is increasing, but so far robust evidence to support their efficacy and wide use varies from non-existent to only promising.

Conflict of interest

The authors declare no conflict of interest.

References

1. Zhang Y, Mignone J, Robb Maclellan W. Cardiac regeneration and stem cells. Physiol Rev 2015; 95: 1189-204.
2. Isomi M, Sadahiro T, Ieda M. Progress and challenge of cardiac regeneration to treat heart failure. J Cardiol 2019; 73: 97-101.
3. Dobrowolska M, Miękus P, Świątczak M, et al. Two-year prognosis of patients hospitalized with decompensated heart failure in a district general hospital. Kardiol Pol 2021; 79: 302-10.
4. Virani SS, Alonso A, Aparicio HJ, et al. Heart Disease and Stroke Statistics –2021 update: a report from the American Heart Association. Circulation 2021; 143: e254-743.
5. Konstanty-Kalandyk J, Kędziora A, Legutko J, et al. Hybrid coronary revascularization in multivessel coronary artery disease: who can benefit most? A pilot study. Kardiol Pol 2021; 79: 449-51.
6. Ganyukov V, Kochergin N, Shilov A, et al. Randomized clinical trial of surgical vs. percutaneous vs. hybrid revascularization in multivessel coronary artery disease: residual myocardial ischemia and clinical outcomes at one year – Hybrid coronary REvascularization Versus Stenting or Surgery (HREVS). J Interv Cardiol 2020; 2020: 5458064.
7. Ganyukov VI, Kochergin NA, Shilov AA, et al. Randomized clinical trial of surgical versus percutaneous versus hybrid multivessel coronary revascularization: 3 years' follow-up. JACC Cardiovasc Interv 2021; 14: 1163-5.
8. Pearson J, Sipido KR, Musialek P, et al. The Cardiovascular Research community calls for action to address the growing burden of cardiovascular disease. Cardiovasc Res 2019; 115: e96-8.
9. Neumann FJ, Sousa-Uva M, Ahlsson A, et al. 2018 ESC/EACTS Guidelines on myocardial revascularization The Task Force on myocardial revascularization of the European Society of Cardiology (ESC) and European Association for Cardio-Thoracic Surgery (EACTS) Developed with the special contribution of the European Association for Percutaneous Cardiovascular Interventions (EAPCI). Eur Heart J 2019; 40: 87-165.
10. Roche CD, Brereton RJL, Ashton AW, et al. Current challenges in three-dimensional bioprinting heart tissues for cardiac surgery. Eur J Cardiothorac Surg 2020; 58: 500-10.
11. McDonagh TA, Metra M, Adamo M, et al. 2021 ESC Guidelines for the diagnosis and treatment of acute and chronic heart failure: developed by the Task Force for the diagnosis and treatment of acute and chronic heart failure of the European Society of Cardiology (ESC). With the special contribution of the Heart Failure Association (HFA) of the ESC. Eur J Heart Fail 2022; 24: 4-131.
12. Knuuti J, Wijns W, Saraste A, et al. 2019 ESC Guidelines for the diagnosis and management of chronic coronary syndromes. Eur Heart J 2020; 41: 407-77.
13. Uygur A, Lee RT. Mechanisms of cardiac regeneration. Dev Cell 2016; 36: 362-74.
14. Litwinowicz R, Kapelak B, Sadowski J, et al. The use of stem cells in ischemic heart disease treatment. Kardiochir Torakochir Pol 2018; 15: 196-9.
15. Song J, He K, Hou J. Autologous bone marrow stem cell transplantation for patients undergoing coronary artery bypass grafting: a meta-analysis of 22 randomized controlled trials. J Cardiothorac Surg 2022; 17: 167.
16. Ang KL, Chin D, Leyva F, et al. Randomized, controlled trial of intramuscular or intracoronary injection of autologous bone marrow cells into scarred myocardium during CABG versus CABG alone. Nat Clin Pract Cardiovasc Med 2008; 5: 663-70.
17. Konstanty-Kalandyk J, Bartus K, Piątek J, et al. Midterm outcomes of transmyocardial laser revascularization with intramyocardial injection of adipose derived stromal cells for severe refractory angina. Front Cardiovasc Med 2018; 14: 176-82.
18. Nicolau JC, Furtado RHM, Silva SA, et al. Stem-cell therapy in ST-segment elevation myocardial infarction with reduced ejection fraction: a multicenter, double-blind randomized trial. Clin Cardiol 2018; 41: 392-9.
19. Makkar RR, Kereiakes DJ, Aguirre F, et al. Intracoronary ALLogeneic heart STem cells to Achieve myocardial Regeneration (ALLSTAR): a randomized, placebo-controlled, double-blinded trial. Eur Heart J 2020; 41: 3451-8.
20. Kwiecien E, Drabik L, Mazurek A, et al. P4604: Insights into left ventricular remodelling and clinical outcomes after Wharton's jelly multipotent stem cells transcoronary administration in a pilot cohort of CIRCULATE-AMI Trial (NCT03404063). Eur Heart J 2019; Suppl_1: ehz745.0987.
21. Kwiecien E, Drabik L, Mazurek A, et al. Evolution of left ventricular function after Wharton's jelly mesenchymal stem cells transcoronary administration: 5-year follow up in a pilot cohort of CIRCULATE-AMI randomized trial. Eur Heart J 2020; Suppl_2: ehaa946.1719.
22. Bartolucci J, Verdugo FJ, González PL, et al. Safety and efficacy of the intravenous infusion of umbilical cord mesenchymal stem cells in patients with heart failure: a phase 1/2 randomized controlled trial (RIMECARD trial [Randomized clinical trial of intravenous infusion umbilical cord mesenchymal stem cells on cardiopathy]). Circ Res 2017; 121: 1192-204.
23. Gao LR, Chen Y, Zhang NK, et al. Intracoronary infusion of Wharton's jelly-derived mesenchymal stem cells in acute myocardial

infarction: double-blind, randomized controlled trial. BMC Med 2015; 13: 162.

24. Chugh AR, Beache GM, Loughran JH, et al. Administration of cardiac stem cells in patients with ischemic cardiomyopathy: the SCIPIO trial: surgical aspects and interim analysis of myocardial function and viability by magnetic resonance. Circulation 2012; 126: 54-64.

25. Reinecke H, Zhang M, Bartosek T, et al. Survival, integration, and differentiation of cardiomyocyte grafts: a study in normal and injured rat hearts. Circulation 1999; 100: 193-202.

26. Beeres SLMA, Bax JJ, Dibbets-Schneider P, et al. Intramyocardial injection of autologous bone marrow mononuclear cells in patients with chronic myocardial infarction and severe left ventricular dysfunction. Am J Cardiol 2007; 100: 1094-8.

27. Rodrigo SF, Van Ramshorst J, Hoogslag GE, et al. Intramyocardial injection of autologous bone marrow-derived ex vivo expanded mesenchymal stem cells in acute myocardial infarction patients is feasible and safe up to 5 years of follow-up. J Cardiovasc Transl Res 2013; 6: 816-25.

28. Konstanty-Kalandyk J, Piątek J, Kędziora A, et al. Long-term follow-up after Holmium:YAG laser revascularization combined with autologous bone marrow derived stem cells implantation. Przegl Lek 2017; 74: 91-5.

29. Naseri MH, Madani H, Tafti SHA, et al.; COMPARE CPM-RMI Trial. Intramyocardial transplantation of autologous bone marrow-derived CD133+ cells and MNCs during CABG in patients with recent MI: a phase II/III, multicenter, placebo-controlled, randomized, double-blind clinical trial. Cell J 2018; 20: 267-77.

30. Mathiasen AB, Qayyum AA, Jørgensen E, et al. Bone marrow-derived mesenchymal stromal cell treatment in patients with ischaemic heart failure: final 4-year follow-up of the MSC-HF trial. Eur J Heart Fail 2020; 22: 884-92.

31. Konstanty-Kalandyk J, Piątek J, Chrapusta A, et al. Use of adipose-derived stromal cells in the treatment of chronic ischaemic heart disease: safety and feasibility study. Kardiol Pol 2018; 76: 911-3.

32. Konstanty-Kalandyk J, Sadowski J, Kędziora A, et al. Functional recovery after intramyocardial injection of adipose-derived stromal cells assessed by cardiac magnetic resonance imaging. Stem Cells Int 2021; 2021: 5556800.

33. Henry TD, Pepine CJ, Lambert CR, et al.; the Athena trials. Autologous adipose-derived regenerative cells for refractory chronic myocardial ischemia with left ventricular dysfunction. Catheter Cardiovasc Interv 2017; 89: 169-77.

34. Kastrup J, Schou M, Gustafsson I, et al. Rationale and design of the first double-blind, placebo-controlled trial with allogeneic adipose tissue-derived stromal cell therapy in patients with ischemic heart failure: a phase II Danish Multicentre Study. Stem Cells Int 2017; 2017: 8506370.

35. Paitazoglou C, Bergmann MW, Vrtovec B, et al. Rationale and design of the European multicentre study on Stem Cell therapy in IschEmic Non-treatable Cardiac diseasE (SCIENCE). Eur J Heart Fail 2019; 21: 1032-41.

36. Fisher SA, Doree C, Mathur A, et al. Stem cell therapy for chronic ischaemic heart disease and congestive heart failure. Cochrane Database Syst Rev 2016; 12: CD007888.

37. Hashimoto H, Olson EN, Bassel-Duby R. Therapeutic approaches for cardiac regeneration and repair. Nat Rev Cardiol 2018; 15: 585-600.

38. Maiullari F, Costantini M, Milan M, et al. A multi-cellular 3D bioprinting approach for vascularized heart tissue engineering

based on HUVECs and iPSC-derived cardiomyocytes. Sci Rep 2018; 8: 13532.

39. Gao L, Gregorich ZR, Zhu W, et al. Large cardiac muscle patches engineered from human induced-pluripotent stem cell-derived cardiac cells improve recovery from myocardial infarction in swine. Circulation 2018; 137: 1712-30.

40. Mattapally S, Zhu W, Fast VG, et al. Spheroids of cardiomyocytes derived from human-induced pluripotent stem cells improve recovery from myocardial injury in mice. Am J Physiol Heart Circ Physiol 2018; 315: H327-39.

41. Menasché P, Vanneaux V, Hagège A, et al. Human embryonic stem cell-derived cardiac progenitors for severe heart failure treatment: first clinical case report. Eur Heart J 2015; 36: 2011-7.

42. Menasché P, Vanneaux V, Hagège A, et al. Transplantation of human embryonic stem cell-derived cardiovascular progenitors for severe ischemic left ventricular dysfunction. J Am Coll Cardiol 2018; 71: 429-38.

43. Zhang YS, Aleman J, Arneri A, et al. Bioprinting 3D microfibrous scaffolds for engineering endothelialized myocardium and heart-on-a-chip. Biomaterials 2016; 110: 45-59.

44. Noor N, Shapira A, Edri R, et al. 3D printing of personalized thick and perfusable cardiac patches and hearts. Adv Sci 2019; 6: 1900344.

45. Zhang B, Montgomery M, Chamberlain MD, et al. Biodegradable scaffold with built-in vasculature for organ-on-a-chip engineering and direct surgical anastomosis. Nat Mater 2016; 15: 669-78.

46. Vasanthan V, Fatehi Hassanabad A, Pattar S, et al. Promoting cardiac regeneration and repair using acellular biomaterials. Front Bioeng Biotechnol 2020; 8: 291.

47. Mewhort HEM, Turnbull JD, Satriano A, et al. Epicardial infarct repair with bioinductive extracellular matrix promotes vasculogenesis and myocardial recovery. J Heart Lung Transplant 2016; 35: 661-70.

48. Wang S, Huang S, Gong L, et al. Human neonatal thymus mesenchymal stem cells promote neovascularization and cardiac regeneration. Stem Cells Int 2018; 2018: 8503468.

49. Weymann A, Loganathan S, Takahashi H, et al. Development and evaluation of a perfusion decellularization porcine heart model: generation of 3-dimensional myocardial neoscaffolds. Circ J 2011; 75: 852-60.

50. Weymann A, Patil NP, Sabashnikov A, et al. Bioartificial heart: a human-sized porcine model – the way ahead. PLoS One 2014; 9: e111591.

51. Tong C, Li C, Xie B, et al. Generation of bioartificial hearts using decellularized scaffolds and mixed cells. Biomed Eng Online 2019; 18: 71.

52. Ozlu B, Ergin M, Budak S, et al. A bioartificial rat heart tissue: perfusion decellularization and characterization. Int J Artif Organs 2019; 42: 757-64.

53. Ali H, Braga L, Giacca M. Cardiac regeneration and remodelling of the cardiomyocyte cytoarchitecture. FEBS J 2020; 287: 417-38.