BULLETIN N° 190
ACADÉMIE EUROPEENNE INTERDISCIPLINAIRE DES SCIENCES

Lundi 1er décembre 2014 à 17h Maison de l'AX 5 rue Descartes 75005 Paris

1)Conférence:
Les ondes gravitationnelles : "*une nouvelle fenêtre sur l'Univers*"
par Patrice HELLO
Professeur à l'Université Paris Sud
Laboratoire de l'Accélérateur Linéaire à Orsay.
2) Proposition de création à titre expérimental d'un groupe de discussion AEIS sur la Toile

Prochaine séance :
Lundi 5 janvier 2015 à 17h Maison de l'AX 5 rue Descartes 75005 Paris

1)Conférence d'intérêt général :
"RECHERCHE ET DEVELOPPEMENT D'ORGANES BIOARTIFICIELS POUR LE TRAITEMENT DE MALADIES CARDIOVASCULAIRES"
par notre collègue Juan-Carlos CHACHQUES
Directeur du Programme de Bioassistance Cardiaque, Laboratoire de Recherches Biochirurgicales, Fondation Alain Carpentier.
Département de Chirurgie Cardiovasculaire, Hôpital Européen Georges Pompidou, Université Paris Descartes.
2) Premières réflexions sur les thématiques possibles d'un futur colloque (après 2016)

ACADEMIE EUROPEENNE INTERDISCIPLINAIRE DES SCIENCES

FONDATION DE LA MAISON DES SCIENCES DE L'HOMME

décembre 2014

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TABLE DES MATIERES

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ACADEMIE EUROPEENNE INTERDISCIPLINAIRE DES SCIENCES

Fondation de la Maison des Sciences de l'Homme, Paris.

Séance du Lundi 1 er décembre 2014
Maison de l'AX 17h

La séance est ouverte à 17h **sous la Présidence de Victor MASTRANGELO** et en la présence de nos Collègues Gilbert BELAUBRE, Alain CARDON, Françoise DUTHEIL, Claude ELBAZ, Michel GONDRAN, Irène HERPE-LITWIN, Gérard LEVY, Antoine LONG, Pierre MARCHAIS, Edith PERRIER, Alain STAHL, Jean-Pierre TREUIL.

Etaient excusés François BEGON, Bruno BLONDEL, Jean-Pierre BESSIS, Jean-Louis BOBIN, Michel CABANAC, Juan-Carlos CHACHQUES, Gilles COHEN-TANNOUDJI, Alain CORDIER, Daniel COURGEAU, Ernesto DI MAURO, Vincent FLEURY, Robert FRANCK, Jean -Pierre FRANCOISE, Jacques LEVY , Valérie LEFEVRE-SEGUIN, Claude MAURY, Pierre PESQUIES, Jean SCHMETS, Jean VERDETTI.

Etait également présente en tant que visiteuse Anne JAOUL, Professeur de classes préparatoires.

1. Conférence de Patrice HELLO "*Les ondes gravitationnelles* **:** *une nouvelle fenêtre sur l'Univers*"

a) Présentation du conférencier Patrice HELLO par notre Président Victor MASTRANGELO

Patrice HELLO est Professeur à l'Université Paris-Sud et il effectue ses recherches au Laboratoire de l'Accélérateur Linéaire à Orsay (LAL). Il est ancien élève de l'ENS de St-Cloud (Lyon maintenant), docteur (1990) et agrégé de sciences physiques et titulaire depuis 1996 d'une habilitation à diriger des recherches.Il est responsable de l'équipe Virgo au LAL depuis 3 ans. Il est membre de la collaboration internationale Virgo depuis de nombreuses années . Ses centres d'intérêt sont principalement la modélisation des optiques des grands interféromètres et l'analyse des données et le retour astrophysique de Virgo.

b) Résumé fourni par le Pr Patrice HELLO

Les ondes gravitationnelles sont prédites par la Relativité Générale et leur existence a été établie grâce à l'étude du pulsar binaire 1913+16 par J. Taylor et ses collaborateurs. Pour détecter directement ces ondes gravitationnelles, plusieurs interféromètres géants ont été construits à la fin des années 90 : Virgo (Italie) et LIGO (deux sites aux USA). Après une première phase (2005-2011) qui a vu les détecteurs Virgo et LIGO atteindre leurs sensibilités nominales, mais insuffisantes pour une première détection, les deux collaborations internationales se sont lancées dans une phase d'amélioration des détecteurs (Advanced Virgo et Advanced LIGO) dont le but est de gagner un ordre de grandeur en sensibilité.

Je commencerai par rappeler la nature des ondes gravitationnelles et leur effet sur la matière puis je passerai en revue les sources astrophysiques intéressantes pour LIGO et Virgo. Je décrirai ensuite Virgo en m'attachant aux principales sources de bruit et aux solutions techniques choisies pour y remédier. Je poursuivrai avec une sélection des principaux résultats scientifiques obtenus par le réseau LIGO-Virgo.

Je ferai finalement le point sur l'actualité et sur les perspectives des deux expériences (Advanced) Virgo et (Advanced) LIGO.

c) **Le compte rendu de la conférence du Pr HELLO rédigé par nos collègues Michel GONDRAN et Michel TREUIL ainsi que les diapositives présentées sont accessibles sur le site de l'AEIS http://www.science-inter.com dans la rubrique "Comptes-rendus conférences mensuelles".**

2. Proposition de création à titre expérimental d'un groupe de discussion AEIS sur la Toile

Notre Collègue Jean-Pierre TREUIL propose la création d'un groupe de discussion de l'AEIS sur la Toile qui s'appellerait aeis-forum@googlegroups.com permettant à 5 ou 6 personnes invitées de commenter les événements de l'AEIS. Les textes accessibles aux invités ont une adresse du type https://docs.google.com/document..... Des suggestions peuvent être apposées par les invités. Si ces suggestions sont approuvées par l'ensemble des propriétaires , le texte est modifié.

Par ailleurs, un résumé des échanges est automatiquement créé qui peut être accessible quotidiennement.

Les membres non- propriétaires ont des droits moindres que les administrateurs qui ont seuls le droit de modérer la discussion.

On créerait donc ainsi un outil puissant de rédaction collaborative des comptes-rendus des conférences notamment pour les binômes chargés de la rédaction.

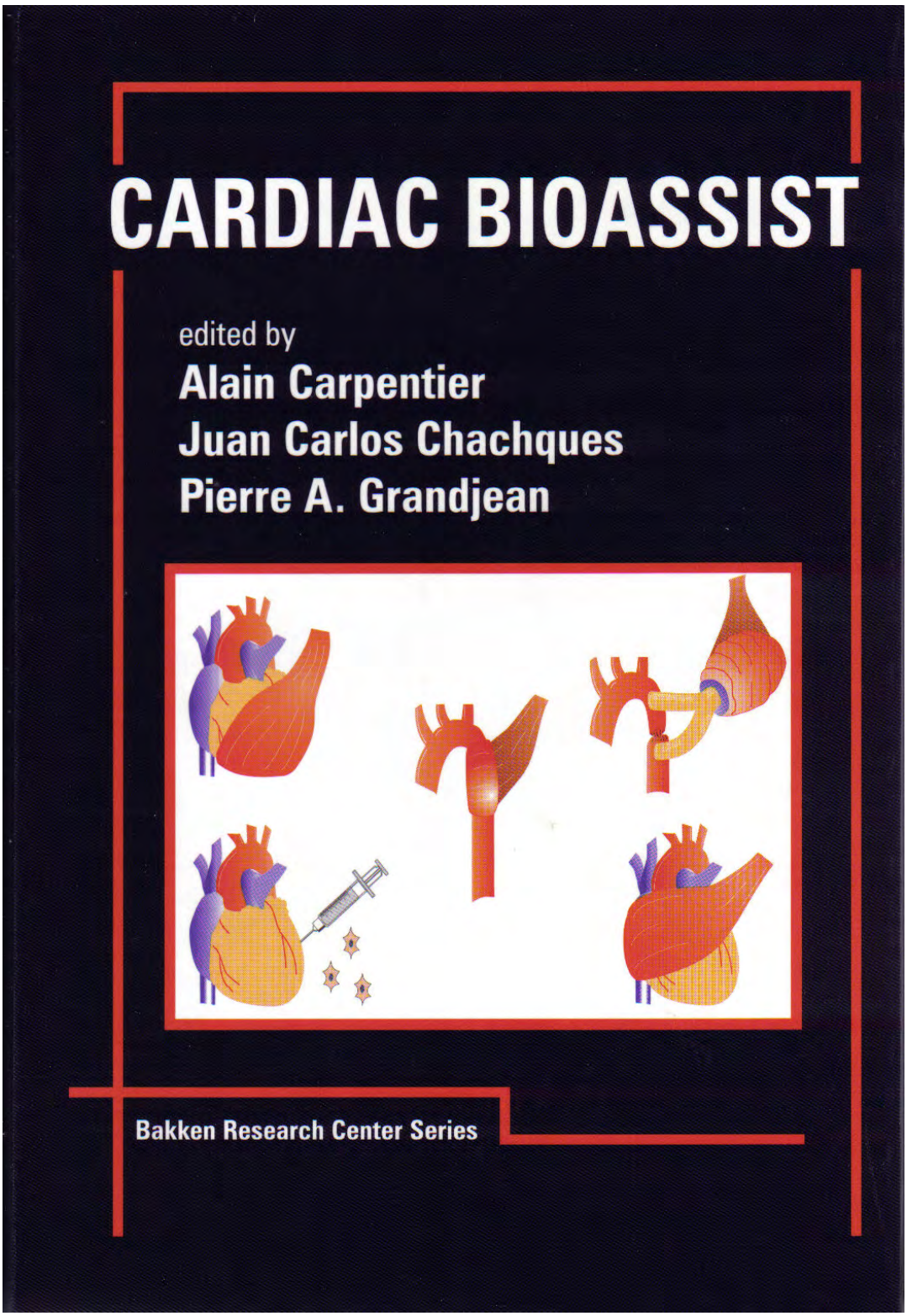
Notre Président Victor MASTRANGELO déclare qu'il s'agit certainement d'une idée intéressante dont doit être informé notre collègue Alain CORDIER responsable de la commission multimédia. Après une brève discussion, l'idée est retenue: les comptes-rendus des conférences pourront ainsi figurer sur le site de l'AEIS http://www.science-inter.com.

Après cette très riche séance, notre Président Victor MASTRANGELO déclare la clôture de notre séance .

Irène HERPE-LITWIN

Annonces

I. Notre Collègue Juan-Carlos CHACHQUES nous donne la page d'annonce de son ouvrage :



II. Notre collègue Christian HERVE nous fait part de la prochaine réunion de la SFFEM qui aura lieu le 17 janvier prochain :

"démarches palliatives et fin de vie chez l'enfant : 15 ans de réflexions et de pratiques à l'Hôpital Universitaire Necker Enfants Malades"

La réunion aura lieu:

de 9h 00 – 17 h 30
Amphithéâtre de l'institut IMAGINE
24 Boulevard de Montparnasse 75015 Paris

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Il nous invite également à visiter le site "Réseau Rodin" http://www.ethique.sorbonne-paris-cite.fr

Documents

En vue de sa conférence, ***"Recherche et développement d'organes bioartificiels pour le traitement de maladies cardiovasculaires"*** du 5 janvier 2015, notre collègue Juan Carlos CHACHQUES nous a confié quelques documents:

RECHERCHE ET DEVELOPPEMENT D'ORGANES BIOARTIFICIELS POUR LE TRAITEMENT DE MALADIES CARDIOVASCULAIRES

Juan-Carlos Chachques, MD, PhD

Directeur du Programme de Bioassistance Cardiaque, Laboratoire de Recherches Biochirurgicales, Fondation Alain Carpentier.
Département de Chirurgie Cardiovasculaire, Hôpital Européen Georges Pompidou, Université Paris Descartes.

La communauté scientifique spécialisée dans la recherche cardiovasculaire fondamentale, clinique et technologique, s'associe afin de développer des thérapies synergiques pour l'insuffisance cardiaque chronique, qui constitue actuellement une véritable épidémie.

La transplantation cardiaque est un traitement efficace pour les maladies cardiaques en phase terminale. Mais en raison de la pénurie de donneurs, la recherche se porte sur des solutions alternatives comme la transplantation de cellules souches associée à l'ingénierie tissulaire, permettant la création d'organes et tissus bio-artificiels. Le but ultime étant le développement de cœurs artificiels bio-mécaniques.

Des progrès récents dans la conception et la fabrication de matériaux biohybrides, ayant des caractéristiques bio-mimétiques, ont été accomplis dans le domaine des nanotechnologies. Des applications directes dans la médecine et l'ingénierie deviennent une réalité. Ainsi il est possible maintenant de créer des tissus et des organes ayant des propriétés structurelles et fonctionnelles similaires aux matrices extra-cellulaires naturelles, contenant des nano-structures tridimensionnelles, capables d'assurer la propagation de signaux cellulaires et la fonctionnalité tissulaire.

Ces nouvelles technologies émergentes permettent de créer des cœurs bio-mécaniques, constitués de prothèses implantables, hémocompatibles, connectées aux sources d'activation électrique externes ou aux systèmes d'activation pneumatiques. La régulation et la surveillance à distance des systèmes sont effectuées par des senseurs et dispositifs électroniques permettant un suivi postopératoire et ensuite une télésurveillance à partir du domicile des malades (home monitoring).

Research and development of bioartificial organs for cardiovascular diseases

by Juan-Carlos CHACHQUES,MD, PhD

Summary :

The basic and clinical cardiovascular research communities are developing and refining synergic therapies for epidemic chronic heart failure. Heart transplantation is an effective therapy for end-stage heart disease, but because of the shortage of donors, there is an increasing interest in more favourable alternatives like stem cell transplantation and tissue engineering for the creation of bioartificial organs and tissues. The ultimate translational goal is the development of biomechanical hearts. Recent progress on the design and fabrication of bio-hybrid materials having biometric characteristics has been accomplished in the field of nanotechnology for direct applications in medicine and engineering. The main purpose of these materials is to create tissues and organs and display structural and functional properties similar to natural extracellular matrices, containing truly three-dimensional nano-structures and controlling the delivery and effective concentration of bioactive signals and cells. New technology emerges to create fully functional, bio-mechanical replacement hearts, including hemocompatible implantable prostheses connected to external electric power supply or pneumatic activation systems, asociated with remote monitoring and electronic control systems for the hospital operative follow-up and for home-monitoring.

References :

J.-C. Chachques and co-authors, Creating the bioartificial myocardium for cardiac repair : challenges and clinical
targets, Expert Rev. Cardiovasc. Ther. 11(12), 1701--1711 (2013).
J.-C. Chachques and co-authors, Cardiac Bioassist : Results of the French Multicenter Cardiomyoplasty Study,
Asian Cardiovasc. Thorac Ann 2009; 17; 573--580.

EUE

Les maladies cardio-vasculaires sont, avec le cancer, la première cause de mortalité dans les pays occidentaux. L'insuffisance cardiaque chronique touche approximativement 30 millions de personnes dans le monde, dont dix millions en Europe et sept millions aux États-Unis. Il constitue un problème majeur en matière de coût et de qualité de vie. Les patients souffrant d'insuffisance cardiaque nécessitent le développement de nouveaux traitements. On assiste à l'émergence d'une médecine dite « régénérative » exploitant les capacités de prolifération et de différenciation des cellules souches provenant de divers tissus comme la moelle osseuse, le tissu adipeux, le sang de cordon ombilical ou le liquide amniotique. L'ingénierie tissulaire associée aux cellules souches vise à réparer et à régénérer des tissus et des organes en utilisant des biomatériaux et/ou des molécules bioactives. Le défi principal est la création d'un myocarde bio-artificiel en appliquant des nanobiotechnologies, utilisant de matériaux composites. La fabrication d'organes bioartificiels sera bientôt rendue possible, ainsi la médecine régénérative est en voie de devenir une alternative à la transplantation d'organes.

Juan Carlos Chachques
Docteur en Médecine et Docteur ès Sciences (PhD). Chirurgien cardio-vasculaire à l'Hôpital Pompidou, Paris. Directeur de recherche, Laboratoire de recherches biochirurgicales, Fondation Alain Carpentier. Président-fondateur Cardiac Bioassist Association. Chevalier Légion d'Honneur de France. Professeur et Docteur Honoris-Causa des Universités

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ORGANES BIO-ARTIFICIELS EN CARDIOLOGIE

Juan Carlos Chachques
Abdel Shafy
Alain Carpentier

Nouveaux traitements de l'insuffisance cardiaque

De la pharmacologie aux cellules souches et aux organes bio-artificiels

Cardiac Bioassist: Results of the French Multicenter Cardiomyoplasty Study

Juan C Chachques, Olivier Jegaden, Thierry Mesana, Yves Glock, Pierre A Grandjean and Alain F Carpentier

Asian Cardiovasc Thorac Ann 2009;17:573-580
DOI: 10.1177/0218492309349371

ORIGINAL ARTICLE

Cardiac Bioassist: Results of the French Multicenter Cardiomyoplasty Study

Juan C Chachques, MD[1], Olivier Jegaden, MD[2], Thierry Mesana, MD[3], Yves Glock, MD[4], Pierre A Grandjean, MS[5], Alain F Carpentier, MD[1], for the French Cardiomyoplasty Investigators

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[4]*Department of Cardiovascular Surgery, Rangueil Hospital, Toulouse, France*
[5]*Bakken Research Center, Maastricht, The Netherlands*

ABSTRACT

The French multicenter experience (6 centers) of dynamic cardiomyoplasty was analyzed for long-term survival and functional outcome, the most important endpoints in congestive heart failure therapy. Cardiomyoplasty was performed in 212 patients with symptoms of chronic heart failure despite maximal pharmacological therapy. The etiology was ischemic (48%), idiopathic (45%) or other (7%). Cardiomyoplasty was performed using the latissimus dorsi muscle which was electrostimulated after surgery. During follow-up, 88% of patients improved clinically. Hospital death occurred in 29 (14%) patients and was related to the severity of preoperative heart failure symptoms. Late mortality occurred in 99 patients due to heart failure (44%), sudden death (37%), or noncardiac causes (18%). Combined dynamic cardiomyoplasty and implantation of a cardiac rhythm management system was safely achieved in 22 patients, and 26 underwent heart transplantation for recurrent heart failure. Long-term functional improvements were observed in most patients, and the best outcome was achieved in those with isolated right ventricular failure. Dynamic cardiomyoplasty can be considered as a destination therapy or a mid- to long-term biological bridge to heart transplantation.

(Asian Cardiovasc Thorac Ann 2009;17:573–80)

KEYWORDS: Cardiomyoplasty, Cardiomyopathies, Heart Failure, Myocardial Ischemia

INTRODUCTION

Heart failure with systolic dysfunction leads to progressive increases in the size of the cardiac chambers. The interaction of hemodynamic processes influences this dilatation, and the actin-myosin crossbridges stretch beyond their physiologic limits, causing a decrease instead of an increase in contractile strength as well as structural and hemodynamic changes. The increased end-diastolic pressure that accompanies dilatation may affect the coronary circulation during diastole, resulting in structural myocardial changes and extracardiac modifications such as metabolic and neuroendocrine changes at the systemic level. The growing prevalence of heart failure has prompted development of new therapies to support the failing heart. Important advances have been made in cardiac bioassist approaches both as a bridge to transplantation and recovery therapy.[1] Latissimus dorsi dynamic cardiomyoplasty (DCMP), introduced in the 1980s, has been followed by aortomyoplasty, atriomyoplasty, and skeletal muscle ventricles. With advances in cellular and molecular biology, the concept of ventricular assistance

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by biological means seems promising.[2] Worldwide, cardiomyoplasty has been performed in over 2,000 clinical cases, aortomyoplasty counterpulsation in 100 cases, cellular cardiomyoplasty in 3,000 cases, and cellular cardiomyoplasty associated with a bioartificial myocardium in 50 cases. DCMP aims to support cardiac function in patients with chronic heart failure by wrapping the (usually left) latissimus dorsi muscle around the failing ventricles and stimulating it electrically in synchrony with ventricular function.[3] The first successful clinical case was treated in 1985 in Broussais Hospital, Paris. This retrospective analysis gives an overview of the collective DCMP experience in France.

Patients and Methods

Since the first clinical case in January 1985, 212 cardiomyoplasty procedures have been performed in 6 centers in France. The last patient in this series was operated on in November 2006. Patient characteristics are summarized in Table 1. All patients had chronic heart failure refractory to pharmacological therapy. Their mean left ventricular ejection fraction was 22% ± 9%. The causes of right ventricular (RV) failure were arrhythmogenic cardiomyopathy in 7 patients, ischemic in 2, Uhl's disease in 1, and endomyocardial fibrosis in 1. Arrhythmogenic RV dysplasia/cardiomyopathy is characterized by progressive fibrofatty replacement of the myocardium, which predisposes to ventricular tachycardia and sudden death in young individuals and athletes. It primarily affects the RV, but with time, it may also involve the left ventricle (LV). DCMP was performed as an isolated procedure in 74% of patients, without the need for extracorporeal circulation; 26% had additional procedures including LV resection (7.3%), tricuspid repair or replacement (7%), coronary artery bypass grafting (6.6%), ventricular tumor resection (2.7%), and mitral valve repair or replacement (2.4%).

Table 1. **Characteristics of 212 patients undergoing cardiomyoplasty**

Variable	No. of Patients
Male	176
Female	36
Mean age (years) [range]	53 ± 11 [15–74]
Ischemic cardiomyopathy	48%
Idiopathic cardiomyopathy	45%
Other (valvular, tumor, RV dysplasia)	7%
Predominant LV dysfunction	68%
Predominant RV dysfunction	9%
Biventricular dysfunction	23%
NYHA functional class	
Mean preoperative class	3.0
Class II	9%
Class III	81%
Class IV	10%

LV = left ventricular, NYHA = New York Heart Association, RV = right ventricular.

The most common DCMP procedure was performed in 2 stages: elevation of the latissimus dorsi muscle, and cardiac wrapping. The patient was placed in the right decubitus position. The left latissimus dorsi muscle was dissected free with preservation of its axillary neurovascular pedicle (Figure 1A). Intramuscular pacing electrodes were implanted. The muscle and pacing leads were transferred to the left thoracic cavity after resection of a segment of the 2nd rib. The heart was exposed via a median sternotomy, and 1 or 2 epicardial leads were implanted for myocardial sensing. The latissimus dorsi muscle was wrapped around both ventricles and fixed to the pericardium with interrupted sutures (Figure 1B). Depending on the indication, different latissimus dorsi wrapping methods were used. The muscle was wrapped around dilated ventricles to reinforce the myocardium, or used to replace a portion of myocardium after aneurysm or tumor resection.[4] Anterior-to-posterior wrapping of the RV was carried out for specific diseases such as arrhythmogenic RV dysplasia, which was generally associated with tricuspid valve annuloplasty (Figure 2).[5]

To keep the latissimus dorsi muscle active, prevent atrophy, and support cardiac function, it must be activated in synchrony with ventricular contractions. Two specially designed intramuscular leads were used to induce contraction of the whole latissimus dorsi muscle using electrodes implanted near its motor nerve branches and synchronized with cardiac function. Muscle burst characteristics (amplitude, number of pulses, timing) could be modified by telemetry.[1,3] A progressive stimulation protocol developed at Broussais Hospital was used in all patients. This protocol was designed to allow the muscle to adapt to its new function. It consisted of a 2-week postoperative rest period (no muscle stimulation) to allow recovery and adhesion to the ventricle, followed by progressive activation by biweekly progressive increases in the number of pulses until the burst contained 6 pulses (31-ms intervals) and occurred at every other heart beat. The timing of the burst was adjusted by echocardiography.

Twenty-two (10%) patients concomitantly received cardiac rhythm management implants: 6 had a dual-chamber pacemaker for atrioventricular block and/or chronotropic incompetence, 10 had cardiac resynchronization devices for left bundle branch block (QRS > 130 ms) and heart failure symptoms, and 6 had defibrillators for ventricular tachycardia/arrhythmias.

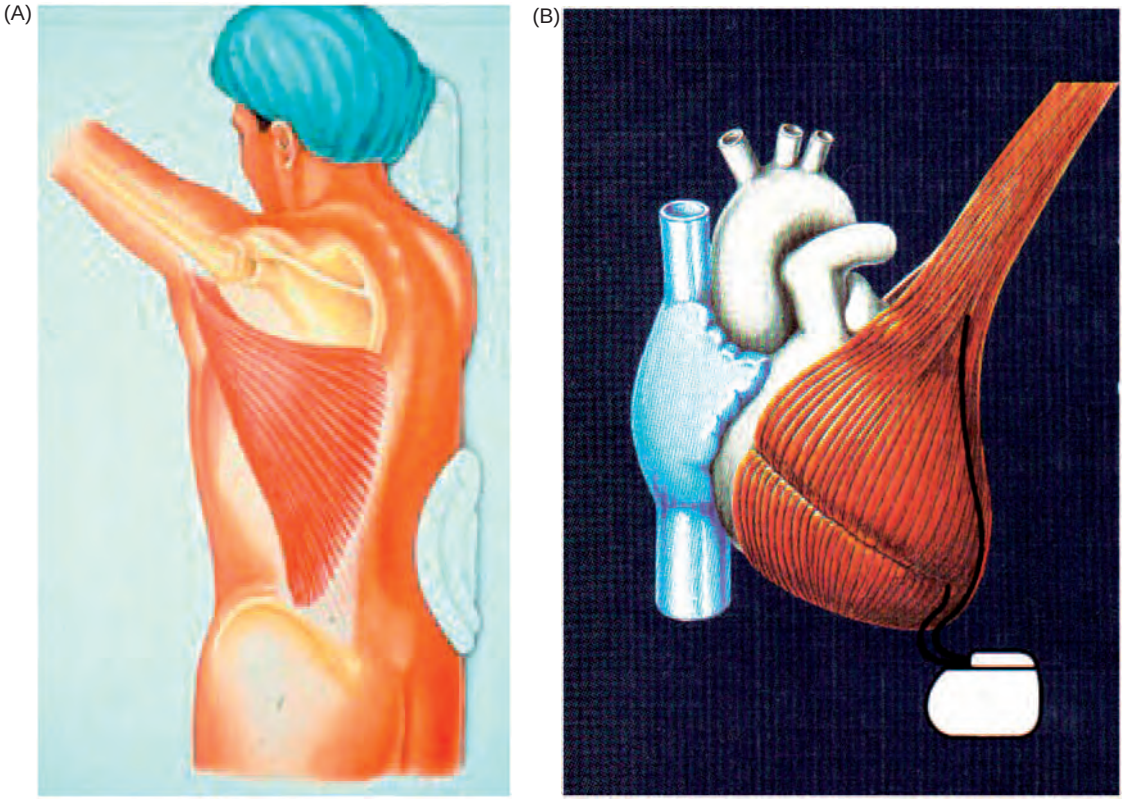

Figure 1. (**A**) Left latissimus dorsi muscle to be used for cardiac wrapping. (**B**) Latissimus dorsi dynamic cardiomyoplasty.

When indicated, heart transplants were performed in the standard fashion by anastomosing a donor heart to the remnant atria and great vessels. During surgery, the latissimus dorsi wrap was divided as far as possible inside the left pleural cavity, and its vascular pedicle was obturated. The proximal portion of the muscle as well as the intramuscular pacing electrodes were kept in place in the left pleural cavity. Adhesions between the muscle and the heart were not released so as to achieve en-bloc resection of the heart and muscle wrap. The latissimus dorsi was divided using electrocautery. The incidence of bleeding from the skeletal muscle stump was generally low. During removal of the recipient's heart, care was taken to avoid injuring the left phrenic nerve which was frequently close to the latissimus dorsi muscle.

RESULTS

Hospital death (within 30 days) occurred in 29 (14%) patients, and was related to the severity of preoperative heart failure symptoms: mortality was 31.8% for those in New York Heart Association (NYHA) functional class IV, 12.4% in class III, and 5.8% in class II. The most important cause of mortality was heart failure (83%). Survival data were to December 2008. From January 1985, 99 patients died (mean time to death, 4 years). Causes of death were heart failure (44%; mean time, 3.4 years), sudden death (37%; mean time, 4.1 years), and noncardiac causes (18%; mean time, 5.4 years). Noncardiac causes of death were stroke, cancer, and gastric bleeding. In addition, 26 patients underwent heart transplantation for recurring symptoms of heart failure. Data were analyzed for freedom from cardiac death or transplantation for the 3 types of predominant ventricular dysfunction (LV, RV, or biventricular). The best results were achieved for isolated RV dysfunction (5-year survival 78%; 10-year survival 69%); while LV and biventricular dysfunction had 48% and 39% survival at 5 years, and 30% at 10 years (Figure 3). Data were also analyzed for freedom from severe cardiac decompensation episodes: 61% of patients were free from severe decompensation at 5 years, 51% at 10 years, and 43% at 15 years. During follow-up, 88% of patients improved clinically as measured by NYHA class which improved by at least 1 class. The mean NYHA class of hospital survivors just before sudden death or death from a noncardiac cause was 1.66 (class III 11%, class II 44%, class I 45%; Figure 4). Mean NYHA class was 3.0 preoperatively and 1.7 postoperatively at a mean follow-up time of 7.2 years.

All cardiac rhythm management implants were successful using standard endovenous implant procedures and testing. Device interactions were checked at implantation and at regular follow-up examinations, using the appropriate marker channel, electrocardiograms, and device memory readings. To minimize far-field sensing, sensing and pacing were programmed to bipolar mode

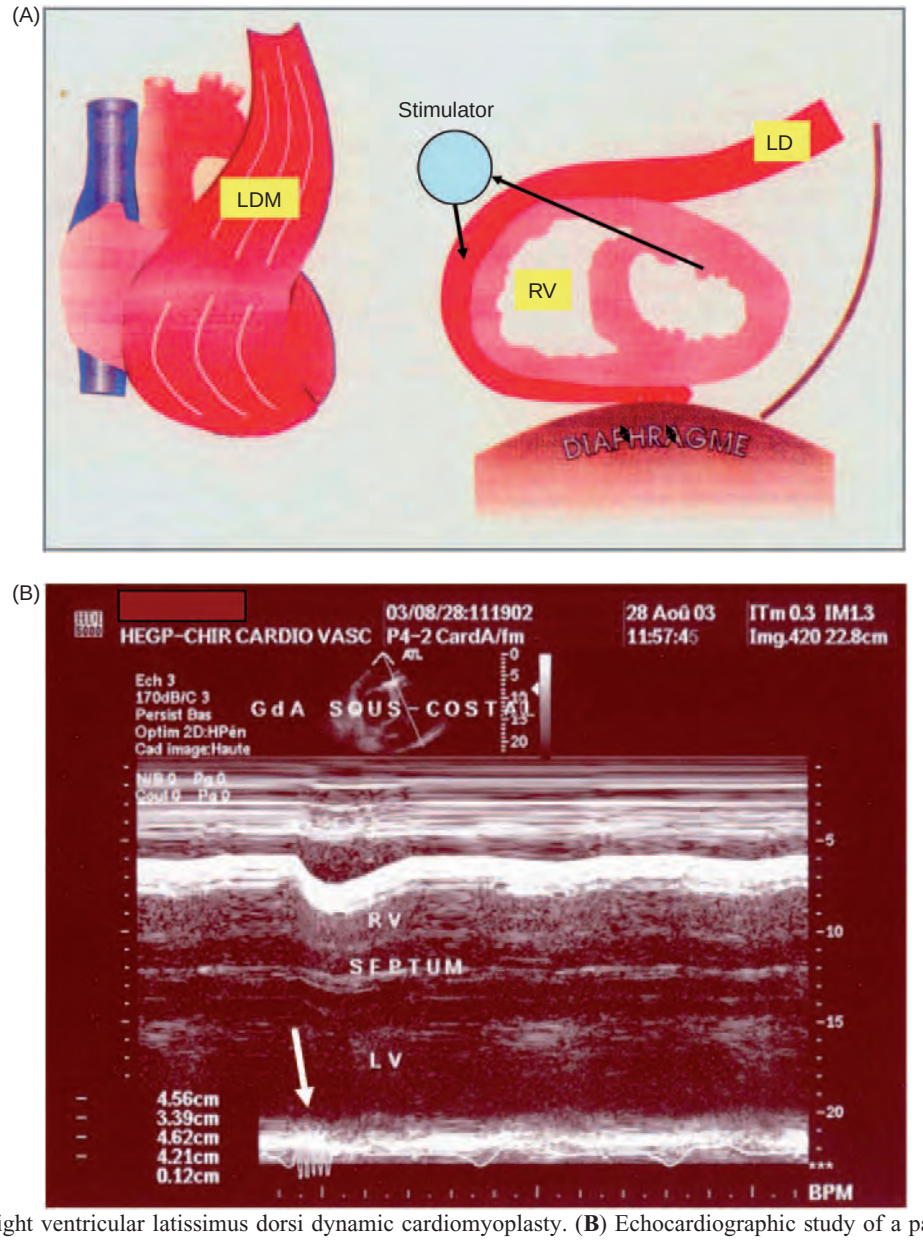

Figure 2. (**A**) Right ventricular latissimus dorsi dynamic cardiomyoplasty. (**B**) Echocardiographic study of a patient presenting with arrhythmogenic right ventricular dysplasia treated with right ventricular cardiomyoplasty and tricuspid valve annuloplasty (Carpentier ring). The arrow shows the electrostimulation pulse train inducing contraction of the right ventricular wall.

when possible. Muscle burst sensing could always be avoided by adjusting the sensitivity, number of pulses in the burst, pulse interval and/or burst delay. As an additional safety feature for patients with defibrillators, the cardiomyostimulator was programmed to turn its muscle stimulator off when in the ventricular tachycardia/fibrillation detection zone. In one patient, the shock from an implanted defibrillator disabled the muscle channel. It could be reprogrammed at the next follow-up visit.

Heart transplantation was indicated in 26 patients for persistent heart failure (no immediate improvement after DCMP) in 19% and for recurring heart failure symptoms, mostly due to progression of the underlying cardiac disease, in 81%. The mean age of these 26 patients was 51 ± 11 years, and the interval after DCMP was 2.3 ± 3 years (range, 0–16.7 years). The etiology in these cases was mostly ischemic (50%) and idiopathic (42%). LV ejection fraction at DCMP was $19\% \pm 6\%$, and the predominant ventricular failure was LV in 76%, RV in 5%, and biventricular in 19%. Survival after heart transplantation (mean, 5.5 years; longest, 13.5 years) is shown in Figure 5. For the urgent subgroup in which there was no clinical benefit after DCMP, the transplant was carried out within 4 months (mean, 1 month; range, 0.1–2.1 months). In the elective subgroup, transplantation was performed after a mean period of 3 years

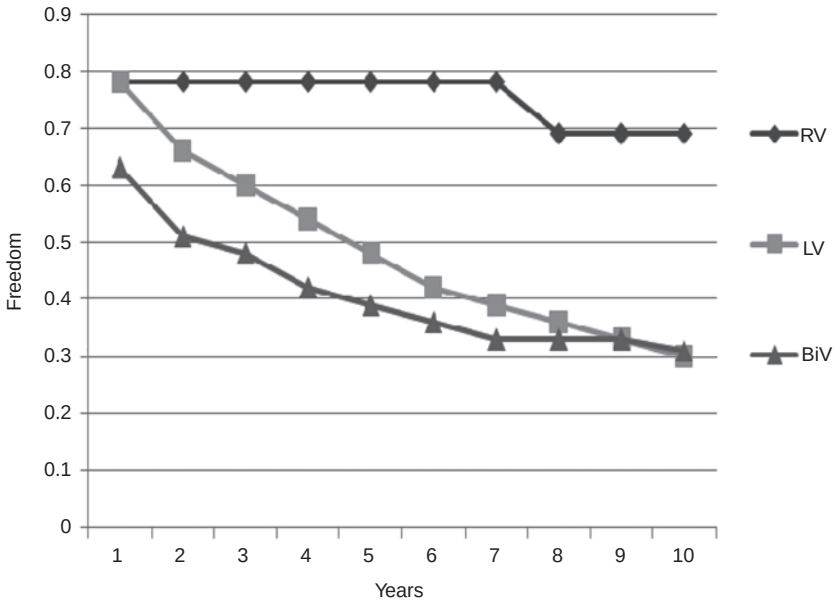

Figure 3. Freedom from cardiac death or heart transplantation after dynamic cardiomyoplasty.

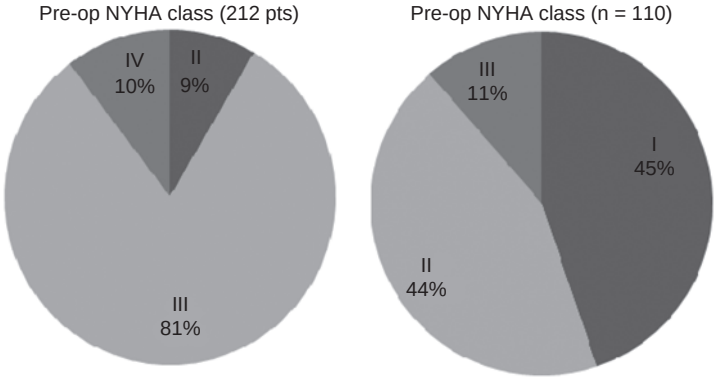

Figure 4. Pre- and postoperative functional class.

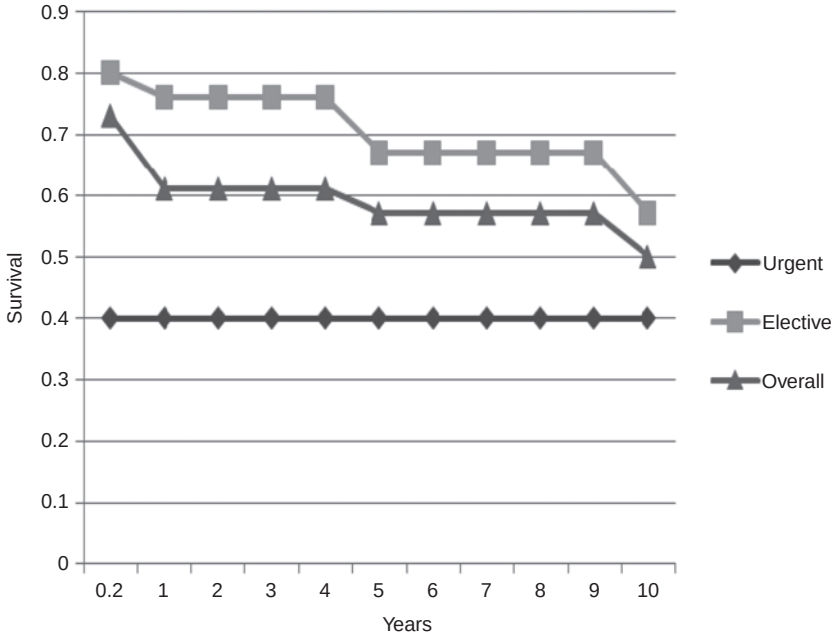

Figure 5. Survival after heart transplantation in cardiomyoplasty patients.

(range, 0.5–16.7 years). Hospital mortality was higher in the urgent subgroup compared to electively treated patients (60% vs. 17%). Early deaths were due to septicemia (2) and graft failure (5). Late deaths were caused by graft failure (4), rejection (1), and gastric bleeding (1). The muscle wrap was found to be well preserved in 9 patients, there was slight hypertrophy and mild fibrosis in 11, and severe ultrastructural impairment and atrophy in 9. Tight adhesions between the muscle wrap and the heart wall were frequently observed.

DISCUSSION

The 30-day hospital mortality includes data from the pilot and feasibility phase during which the surgical method was developed. As expected, hospital mortality was strongly linked to the preoperative NYHA class. With implementation of improvements, later multicenter studies (e.g. C-SMART in the USA) have focused on NYHA class III patients and used risk-stratification strategies, with 30-day hospital mortality of 4%.[6] As expected, our hospital mortality was higher when urgent transplantation was required due to postoperative heart failure or early inefficient DCMP support, but it was not significantly different to that after elective transplantation performed later due to progression of underlying cardiac disease resulting in DCMP inefficiency. This experience confirms that heart transplantation is feasible and safe after DCMP. For non-urgent transplants, the results were similar to those reported for primary transplant procedures.[7]

DCMP provided long-term cardiac benefits as demonstrated by a reduction of NYHA class by at least 1 class in 88% of patients discharged from hospital. Long-term event-free survival, defined as no cardiac death or transplantation, depended on cardiac indication; it was best for isolated RV failure. Muscle contraction can be maintained in the long term.[7–11] The effect of muscle stimulation, efficacy of contraction, and the need to be stimulated were not tested prospectively. However, some clinical observations could be undertaken, e.g., when the stimulator reached its end of service and the device was not replaced on time (battery depleted) because the patient lived overseas, or when muscle stimulation stopped due to lead fracture. Most of these events occurred >10 years after DCMP. In each of these cases, the patient returned to hospital with recurring symptoms of heart failure. Correction of the problem by exchanging the stimulator or repairing the lead allowed the patient to gradually recover to their previous clinical condition. These anecdotal observations confirm that muscle function can be preserved in the long term, and that long-term muscle stimulation is needed to maintain the clinical benefits.

In this study, all patients followed the original progressive stimulation protocol developed at Broussais Hospital. Implementation of recent improved understanding of muscle physiology and adapting stimulation protocols to the patient's physiological status, such as customizing the stimulation pattern to the muscle response and allowing for a mixed fiber composition, indicates that use of skeletal muscle pharmacological support would certainly improve results. A major impact might result from a protocol or procedure that would obviate the need for a muscle rest period after surgery as this would permit cardiac support during the early postoperative phase. Surgical improvements aimed at optimizing energy transfer from muscle to cardiac hemodynamics might also be important.

Nearly all of these cardiomyoplasty operations used cardiomyostimulators supplied by Medtronic, Inc. The commercial decision by Medtronic to withdraw from the field was damaging as it led to the widespread conclusion that the technique was inherently flawed. Ironically, it was the drive to meet regulatory requirements that discouraged departure from a fixed protocol, isolating clinical practice from progress in the underlying basic science. New devices are now available, such as the Microstim Myostimulator (Microstim, Germany) and the LD-PACE II (CCC, Uruguay); these may revive cardiomyoplasty and related research.[12,13]

Because 37% of late deaths during follow-up were sudden, a cardiomyostimulator that includes defibrillation therapy has the potential to improve clinical outcomes. This experience indicates that concomitant implantation of a cardiac rhythm management device can be undertaken safely, and confirms previous case reports of the benefit of a combination of cardiomyostimulators and implantable defibrillators. Appropriate long-term function of both devices can be maintained. The cumulative experience in this cohort amounted to 54 patient-years. Device interactions must be verified after implantation, and systems adjusted to avoid interaction of the bursts with cardiac rhythm detection. After defibrillation, checking cardiomyostimulator function by electrocardiography or telemetry is recommended. The addition of ventricular resynchronization could be an added benefit for heart failure patients, and should be considered in combination with a defibrillator function. Basic research in recent years on the vascular anatomy of the latissimus dorsi muscle and its adaptive response to electrostimulation has improved the viability and function of skeletal muscle grafts. DCMP performed with emerging pacing protocols could well reduce systolic wall stress and provide beat-to-beat assistance in the short term, together with reverse remodeling and extra myocardial revascularization in the long term.[14–16] Implementation of the latest findings on muscle

preservation, activation, energy transfer from muscle to heart, as well as combinations of defibrillators and resynchronization devices might improve survival and efficacy.[15] In the case of recurrent heart failure, heart transplantation is still feasible. Therefore DCMP could be considered as either a destination therapy or as a mid- to long-term biological bridge to heart transplantation.

The knowledge acquired with DCMP has been applied to regenerative cardiology. Stem cell therapy in ischemic and nonischemic cardiomyopathies is a rapidly growing field.[17] The benefits of stem cell delivery to injured myocardium have been demonstrated, and recent reports have described bioartificial matrices that may provide mechanical support and enhance myocardial regeneration. It is important to remark that current indications for cell-based regenerative therapy are small ischemic scars and not the large ischemic lesions responsible for end-stage ventricular failure. Electrostimulation combined with cellular cardiomyoplasty could transform passive cell therapy into ''dynamic cellular support''. The principles of electrophysiological conditioning of skeletal muscle fibers developed for DCMP are now applied in cellular cardiomyoplasty. The hypothesis is that electrostimulation of both ventricles following skeletal myoblast implantation induces contraction of the transplanted cells and greater expression of slow myosin which is better for chronic ventricular assistance. Electrostimulation seems to drive stem cells to differentiate into cardiac-type myogenic cells.[18] This type of differentiation should include the induction of gap junction formation, improving stem cell engraftment and reducing the risk of arrhythmogenic events. In-vitro electrostimulation of cell cultures induced both morphological and biochemical changes in mesenchymal stem cells, realizing a shift towards a striated muscle cell phenotype expressing cardiac-specific markers.[19] Cardiac tissue engineering is emerging as a new therapeutic tool, extending the amazing possibilities of cardiac bioassist procedures and promising the creation of a bioartificial myocardium. The stem cell niche, a specialized environment surrounding stem cells, provides crucial support for stem cell maintenance. Compromised niche function may lead to selection of stem cells that no longer depend on self-renewal factors. If the stem cell niche has aged, it might not be capable of supporting autogenous or transplanted stem cells. Cardiac tissue engineering is developing biodegradable scaffolds to support cell survival and early extracellular instruction. The MAGNUM trial showed that cellular cardiomyoplasty associated with a cell-seeded collagen matrix increased the thickness of the infarct scar with viable tissue, and helped to normalize cardiac wall stress in injured regions, thus limiting ventricular remodeling and improving diastolic function.[20] The creation of functional biocompatible contractile tissues remains challenging. Future developments include bioengineered platforms where stem cells are preconditioned to resist implantation into highly stressed myocardial tissue.

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References

1. Carpentier A, Chachques JC, Grandjean P, editors. Cardiac Bioassist. Futura Publishing, New York, 1997:1–632.
2. Chachques JC, Shafy A, Duarte F, Cattadori B, Goussef N, Shen L, et al. From dynamic to cellular cardiomyoplasty. J Card Surg 2002;17:194–200.
3. Chachques JC, Acar C, Tapia M, Radermecker MA, Guibourt P, Fiemeyer A, et al. Quality of life and long-term survival following cardiomyoplasty. Asian Cardiovasc Thorac Ann 1993;1:55–62.
4. Chachques JC, Argyriadis PG, Latremouille C, D'Attellis N, Fornes P, Carpentier AF, et al. Cardiomyoplasty: ventricular reconstruction after tumor resection. J Thorac Cardiovasc Surg 2002;123:889–94.
5. Chachques JC, Argyriadis PG, Fontaine G, Hebert JL, Frank R, D'Attellis N, et al. Right ventricular cardiomyoplasty: 10-year follow-up. Ann Thorac Surg 2003;75:1464–8.
6. Furnary AP, Jessup FM, Moreira LP. Multicenter trial of dynamic cardiomyoplasty for chronic heart failure. The American Cardiomyoplasty Group. J Am Coll Cardiol 1996; 28:1175–80.
7. Chachques JC, Jegaden OJ, Bors V, Mesana T, Latremouille C, Grandjean PA, et al. Heart transplantation following cardiomyoplasty: a biological bridge. Eur J Cardiothorac Surg 2008;33: 685–90.
8. Schreuder JJ, van der Veen FH, van der Velde ET, Delahaye F, Alfieri O, Jegaden O, et al. Left ventricular pressure-volume relationships before and after cardiomyoplasty in patients with heart failure. Circulation 1997;96:2978–86.
9. Jondeau G, Dorent R, Bors V, Dib JC, Dubourg O, Gandjbakhch I, et al. Dynamic cardiomyoplasty: effect of discontinuing latissimus dorsi muscle stimulation on left ventricular systolic and diastolic performance and exercise capacity. J Am Coll Cardiol 1995;26:129–34.
10. Jegaden O, Delahaye F, Finet G, Van der Veen F, Montagna P, Mikaeloff PH, et al. Late hemodynamic results after cardiomyoplasty in congestive heart failure. Ann Thorac Surg 1994; 57:1151–7.
11. Mesana TG, Ferzoco SJ, Reul RM, Sayeed-Shah U, Karamichalis JM, Cohn LH, et al. Innovative techniques in skeletal muscle cardiac assistance: first experimental study on minimally invasive aortomyoplasty and cardiomyoplasty. ASAIO J 1997;43:M791–6.
12. Salmons S. Cardiac assistance from skeletal muscle: a reappraisal. Eur J Cardiothorac Surg 2009;35:204–13.
13. Chekanov VS, Chachques JC, Brum F, Arzuaga J, Arzuaga P, Krum DP, et al. LD-PACE II: a new cardiomyostimulator for cardiac bioassist. ASAIO J 2001;47:50–5.

14. Rigatelli G, Carraro U, Riccardi R, Rigatelli G. Demand dynamic biogirdling: ten-year results. J Thorac Cardiovasc Surg 2009;137:e58–9.

15. Benicio A, Moreira LF, Bacal F, Stolf NA, Oliveira SA. Reevaluation of long-term outcomes of dynamic cardiomyoplasty. Ann Thorac Surg 2003;76:821–7.

16. Lorusso R, Marchini A, Bianchetti F, Curnis A, Visioli O, Zogno M. Cardiomyoplasty and implantable cardioverter defibrillator: efficacy and safety of concomitant device implantation: sudden death and cardiomyoplasty. J Card Surg 1998;13:150–5.

17. Chachques JC, Salanson-Lajos C, Lajos P, Shafy A, Alshamry A, Carpentier A. Cellular cardiomyoplasty for myocardial regeneration. Asian Cardiovasc Thorac Ann 2005;13:287–96.

18. Shafy A, Lavergne T, Latremouille C, Cortes-Morichetti M, Carpentier A, Chachques JC. Association of electrostimulation with cell transplantation in ischemic heart disease. J Thorac Cardiovasc Surg 2009;138: (in press).

19. Genovese JA, Spadaccio C, Chachques E, Schussler O, Carpentier A, Chachques JC, et al. Cardiac pre-differentiation of human mesenchymal stem cells by electrostimulation. Front Biosci 2009;14:2996–3002.

20. Chachques JC, Trainini JC, Lago N, Cortes-Morichetti M, Schussler O, Carpentier A. Myocardial Assistance by Grafting a New Bioartificial Upgraded Myocardium (MAGNUM trial): clinical feasibility study. Ann Thorac Surg 2008;85:901–8.

EXPERT REVIEWS

Creating the bioartificial myocardium for cardiac repair: challenges and clinical targets

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The association of stem cells with tissue-engineered scaffolds constitutes an attractive approach for the repair of myocardial tissue with positive effects to avoid ventricular chamber dilatation, which changes from a natural elliptical to spherical shape in heart failure patients. Biohybrid scaffolds using nanomaterials combined with stem cells emerge as new therapeutic tool for the creation of 'bioartificial myocardium' and 'cardiac wrap bioprostheses' for myocardial regeneration and ventricular support. Biohybrids are created introducing stem cells and self-assembling peptide nanofibers inside a porous elastomeric membrane, forming cell niches. Our studies lead to the creation of semi-degradable 'ventricular support bioprostheses' for adaptative LV and/or RV wrapping, designed with the concept of 'helical myocardial bands'. The goal is to restore LV elliptical shape, and contribute to systolic contraction and diastolic filling (suction mechanism). Cardiac wrapping with ventricular bioprostheses may reduce the risk of heart failure progression and the indication for heart transplantation.

KEYWORDS: bioartificial myocardium • biohybrid scaffolds • bionanomaterials • cardiac bioregeneration • cardiomyoplasty • cell transplantation • heart failure • myocardial infarction • myocardial regeneration • tissue engineering

Myocardial ischemia is leading cause of heart failure (HF) in all over the world. Following myocardial infarction the irreparable loss or dysfunction of cardiomyocytes occurs due to sudden deprivation of oxygen supply to the heart. Heart has very limited regeneration capacity as most of the myocytes seems to be terminally differentiated, only small fraction of myocytes retain the capacity to replicate. Until now, drug therapy, surgical procedures (e.g., CABGs, ventricular remodeling and restoration, dynamic cardiomyoplasty), heart transplantation and mechanical circulatory assist devices are used as treatment when hearts are irreparably damaged. It is evident that it is necessary to develop more effective, less invasive, therapeutic strategies for HF. Stem cells-based therapies give new hope in the field of regenerative medicine, as stem cells have ability to differentiate into same as well as different tissue types and to regenerate themselves without losing their differentiation potential. This property of differentiation is being explored for the regeneration of several damaged tissues [1,2].

Post-ischemic ventricular dilatation & remodeling

Cardiac remodeling refers to the changes in size, shape, structure and physiology of the heart after injury to the myocardium. Dilated cardiomyopathy from many causes results in a change in ventricular geometry, whereby the elliptical chamber becomes more spherical. This change in architecture alters myocardial fiber direction and diminishes function. The early changes of increased spherical configuration lead to impairment of ventricular function and may lead to mitral valve regurgitation.

The extracellular matrix (ECM) is mainly composed of collagen which gives structural strength to the left ventricle (LV). After myocardial infarction, not only the changes affect the contractile element of the myocardium (cardiomyocytes) but also the ECM. Cardiomyocyte death and scar formation, which are

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Box 1. Current limitations of cell-based cardiac treatments.

- Cell bioretention and engraftment within infarct scar tissue is low.
- Mortality of implanted cells in ischemic myocardium is high.
- In ischemic heart disease, the extracellular matrix is pathologically modified.
- Genetic abnormalities (karyotype instability) have been described for *in vitro* expanded mesenchymal stem cell.

ECM: Extracellular matrix; MSC: Mesenchymal stem cell.

caused by coronary artery occlusion, modulate cardiac remodeling. Ischemic heart disease may progress inducing geometric alteration of the ventricular cavity, heart dilatation is a negative symptom in the evolution of HF patients, related with morbidity and mortality.

Strategies to repair or regenerate injured cardiac tissue offer new hope for the treatment of congestive HF; stem cell therapy associated with myocardial tissue engineering should play an important role in modern regenerative cardiology [3,4].

Myocardial tissue engineering

The poor survival of grafted cells following cellular cardiomyoplasty has been a great concern of researchers as transplanted cells cannot survive for long period due to the ischemic environment, the pathologically modified ECM, the proapoptotic factors and the inflammatory response. It seems suitable to provide a safe environment (niche) for cell proliferation and differentiation [5–7]. The use of anti-oxidant, anti-inflammatory and anti-apoptotic proteins may give better survival of transplanted cells.

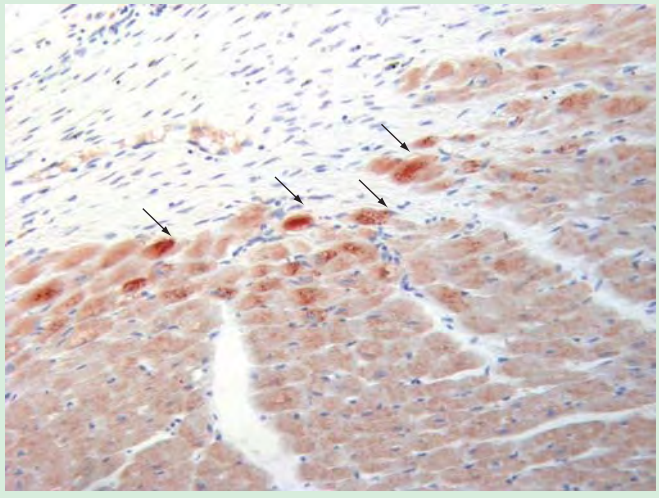

Figure 1. Myocardial infarction treated with collagen scaffold seeded with adipose mesenchymal stem cells. This figure shows growing myocardial tissue adjacent to the infarct scar including BrdU-labeled cells (arrows). Immunohistological study using anti-BrdU antibody (Sigma monoclonal B2531) on left ventricular wall x20.

Poor cell retention without graft viability in the target tissue was observed during clinical application of cellular cardiomyoplasty. A significant number of cells (more than 90%) die within the early days post-transplantation or widely spread to other tissues [1,2]. A possible solution would be to encapsulate the appropriate cells in an injectable gel, but this is not seen as a viable option for the low mechanical properties of gels, which makes it difficult to handle for fixation onto the pathologic myocardium (Box 1).

Alternative tissue engineering strategies [7–10] combine cells with three-dimensional scaffolds or patches to lodge them and improve their survival, to induce the formation of new blood vessels and ECM and at the same time to mechanically support the native tissue. Isolated injection of stem cells in patients presenting post-infarction myocardial scars showed poor results in terms of improving ventricular function and clinical functional class. Several experimental studies showed the benefits of engineered materials charged with stem cells for ischemic models. In addition, in some of these studies the cell transplantation was associated with epicardial grafting of scaffolds onto the ventricular surface [11–14]. Subsequently, our group coordinated the MAGNUM Clinical Trial (Myocardial Assistance by Grafting a New bioartificial Upgraded Myocardium). This trial was the first clinical application of myocardial tissue engineering [15]. In this biosurgical approach performed during OP-CABGs, a collagen type I matrix was seeded with autologous mononuclear bone marrow cells and grafted onto the LV wall. Long-term results showed that a combined cell transplantation and matrix scaffolds approach offer further benefits with respect to cell therapy alone. The cell-seeded collagen matrix increases the thickness of the infarct scar with viable tissue and helps to normalize cardiac wall stress in injured regions, thus limiting ventricular remodeling and improving diastolic function.

The main limitation of collagen scaffolds for heart repair is the low mechanical characteristics of the material to engineer the myocardial tissue and the complete bioresorption and degradation of the material at mid-term. For these reasons, recent studies combined biodegradable cell-seeded scaffolds with synthetic mesh wrap devices [16] showing promising results in terms of myocardial tissue repair (Figure 1).

Natural and synthetic scaffolds are in development, the main goal of these tissue-engineered materials is to allow cell attachment and migration, deliver and retain cells and biochemical factors and enable diffusion of vital cell nutrients. These materials should offer a suitable 'cellular niche', microenvironment needed for cell survival and function (Box 2) [17–19].

Biologic scaffold materials composed of ECM can be derived by processes that involve decellularization of tissues or organs. Preservation of the complex composition and 3D ultrastructure of the ECM is highly desirable but methods of decellularization result in some disruption of the architecture and potential loss of surface structure and composition. Because the native ECM guides organ development, repair and physiologic regeneration, it provides a promising alternative to synthetic scaffolds. In

summary, perfusion decellularization is a novel technology that generates native ECM scaffolds with 3D anatomical architecture and vasculature. These bioscaffolds can then be recellularized to create potentially functional organs, through progressive flow from large vessels branching into medium-sized arterioles [20–23].

Biohybrid scaffolds

One of the main mechanisms by which cellular therapy could bring functional benefits would be that the implanted cells should provide a supporting 'band-aid' scaffolding effect, which can limit the spread of the infarcted area, preventing excessive remodeling and dilatation of the ventricle.

On this direction, we created the European Consortium RECATABI (Regeneration of Cardiac Tissue Assisted by Bioactive Implants) [201], where the main goal was to develop a bioengineered platform and obtaining newly designed biodegradable and semi-degradable scaffolds (constructs) to support implanted stem cell survival and mobilization into the ischemic tissue, promoting slow progressive tissue remodeling and tissue replacement with minimal ventricle dilatation. RECATABI is an interdisciplinary group of experts in areas such as material sciences, tissue engineering, stem cell technologies and clinical cardiovascular research, integrating and synergizing their capacities in order to obtain a simple one-time patch technology application.

This was accomplished by fabricating nanoscale-engineered biomaterials and scaffolds to match the exact biomechanical and biophysical requirements of the implanted tissue. The goal of this bioactive construct is to induce rapid vascularization to ensure tissue remodeling and regeneration into a newly functional myocardium.

Bioactive implants were developed combining self-assembling peptide [24], new elastomeric membranes and stem cells [17,18,25,26]. These biohybrid scaffolds are basically created introducing adipose mesenchymal stem cells (MSCs) and self-assembling peptide nanofibers inside a porous elastomeric membrane. In this way, the membrane acts as carrier but most importantly, protects mechanically the cells into the soft nanofiber scaffold or hydrogel, creating cell niches.

The biohybrid tissue-engineered scaffolds developed by the RECATABI Consortium were the following:

- Non-degradable polymer: porous membrane of poly ethyl acrylate (PEA), associated with self-assembly nanofibers peptide hydrogel (Figure 2).
- Partially degradable polymer: caprolactone methacryloxyl ethyl ester (CLMA) and hyaluronic acid membranes, associated with self-assembly nanofibers peptide hydrogel (Figure 3).

Biomaterials for cardiac tissue engineering need to feature biocompatibility and mechanical properties to be elastic enough to match the myocardium contraction–distraction activity to allow deep structural and functional biointegration. Thus, 'viscoelasticity' is a key physical factor in the selection of materials for myocardial tissue engineering, stiffening of ventricular chambers can be related with systolic and diastolic dysfunctions, progressing to HF. Therefore, we evaluated viscoelastic properties of biohybrid scaffolds, results showed mechanical properties [101].

To progress from bench to bedside, small animal studies were followed by the application of biohybrids on large animal ischemic models. Specific readjustments were taken care of in order to scale up the implant, since the increase on the membrane size might affect self-assembling peptide distribution into the elastomeric membrane as well as the amount of seeded stem cells and their survival capacity.

Technically, the bioactive implants were successfully attached by surgery to the infarcted areas. The application of bioactive implants on infarcted sheep hearts showed enhanced parameters of systolic and diastolic functions in a 6-month trial. LV chamber dilatation was reversed and infarct size was reduced in treated animals (Figure 4). In addition, MRI showed in the biohybrid scaffold group a significant reduction of the infarct volume related with the LV myocardial mass (Figure 5).

Box 2. Challenges in myocardial tissue engineering.

- It is difficult to repair a large myocardial scar
- Extracellular matrix degradation and myocardial fibrosis contribute to progressive ventricular dilatation in heart failure patients
- Materials selected for myocardial tissue engineering associated with stem cells should provide adequate niches for cell survival, multiplication and differentiation
- The therapeutic limitation of heart dilatation and the recovery of the native elliptical shape of ventricular chambers are key prognostic factors for patient's survival
- Complete biodegradation of grafted scaffolds may compromise long-term limitation of post-ischemic ventricular remodeling

ECM: Extracellular matrix; HF: Heart failure.

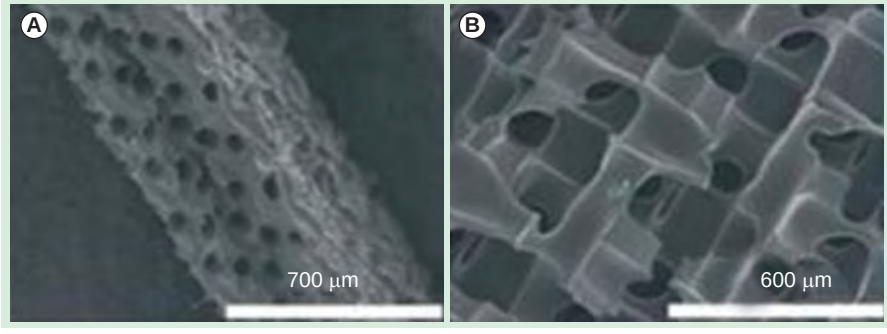

Figure 2. Microscopic images of the poly ethyl acrylate scaffolds with cylindrical orthogonal pores. (A) SEM image, cross-section. **(B)** SEM image, surface.

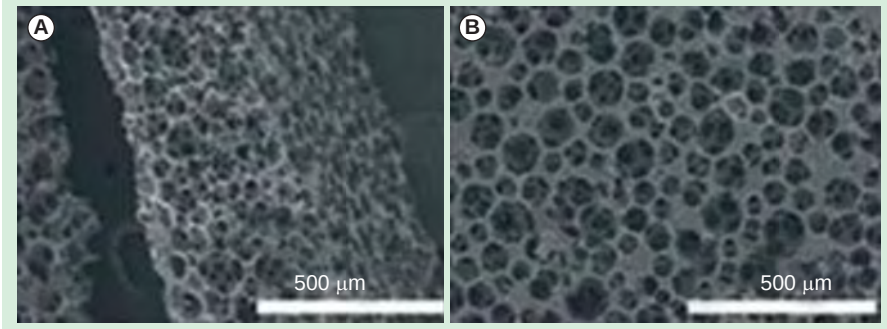

Figure 3. Microscopic images of the caprolactone methacryloxyl ethyl ester scaffolds with spherical pores. (A) SEM image, cross-section. **(B)** SEM image, surface.

Electrophysiology of cardiac scaffolds

Electrical impedance has shown to be an effective indicator of myocardial tissue characteristics. Significant modifications have been demonstrated during myocardial ischemia and in infarct scars. Electrical resistivity (also known as specific electrical resistance or volume resistivity) is a measure of how strongly a material opposes the flow of electric current. A low resistivity indicates a material that readily allows the movement of electrical charge. The SI unit of electrical resistivity is the ohm (Ω).

Electrophysiological studies were performed implanting two electrodes into the opposite borders of elastomeric scaffolds and of a collagen type I matrix, electrodes were connected to a Pacing System Analyzer Model 5311 (Medtronic Inc., MN, USA). Bipolar charge balanced electrostimulation was delivered just for testing, electrical impedance was assessed.

Results showed that all the evaluated materials present electrical conduction properties (i.e., resistance) similar to those encountered with cardiac tissues, thus these scaffolds have the potential to be used for myocardial substitution (Table 1).

Stem cell differentiation in 3D systems

Undifferentiated stem cells reside within differentiated tissues and can become specialized cell types when they received signals. Stem cells have the ability to differentiate into various types of lineages which makes them attractive for the creation of bioartificial myocardium when seeded in specific scaffolds [3,4].

Stem cell differentiation can be achieved by extrinsic physical stimuli (electrostimulation, magnetic fields), cyclic compressive strain as well as by chemical (cytokines) and biological/genetic stimuli (cell co-cultures, genetic manipulations) [27–31]. Experimental studies demonstrated that electrostimulation of cell-seeded collagen matrix changed stem cell morphology and biochemical characteristics, increasing the expression of cardiac markers [32,33]. This preconditioned biological matrix scaffold could be useful for myocardial support and regeneration [102].

Thus, electrostimulation seems to be a safe method to induce physical and biochemical changes in stem cells moving toward cardiac cells without using any demethylating agents or viral vector, representing a safe alternative to drugs like 5-azacytidine (5-azaC; a nucleoside-based DNA methyltransferase inhibitor that induces demethylation and gene reactivation) and chemical cytokines (Box 3) [34,35].

On another approach, it was shown that the functionalization of collagen scaffold with RGD molecule allows terminal differentiation of neonatal rat cardiomyocytes. Thus, an efficient and stable contractile tissue could be elaborated without the need of tumoral extracts such as Matrigel. In a regenerative matrix model combined with RGD peptides, a spontaneous differentiation toward a contractile cell phenotype such as myofibroblasts [36] was observed.

Table 1. Electrophysiological studies of biohybrid scaffolds compared with normal myocardium.

	Collagen matrix	PEA scaffold	CLMA scaffold	Normal myocardium
Impedance (Ω)	230	340	440	300–700
Current (mA)	4.35	2.94	2.1	2

Pacing pulse; 1.0 V, 0.5 ms
CLMA: Caprolactone methacryloxyl ethyl ester; PEA: Poly ethyl acrylate.

Translational research

Cardiac support bioprostheses for myocardial regeneration & ventricular remodeling

HF patients develop oversized, dilated hearts due to increased filling pressures. Over time, the increased workload of the heart can lead to a change called remodeling, which is the chamber enlargement and wall thinning of the ventricles. The failing cardiac muscle need to be supported to decrease the ventricular wall stress. For example, biological approaches like latissimus dorsi (LD) cardiomyoplasty have been used for 'ventricular support therapy' [103]. Limitation of cardiac enlargement and reduction in mechanical ventricular wall stress was observed experimentally and clinically in cases treated with LD dynamic cardiomyoplasty, a biological support/restraint therapy in which the heart is wrapped by the electrostimulated LD muscle [37,38]. This autologous source of circulatory assistance in which an electrically stimulated grafted skeletal muscle works in concert with myocardium requires a rather long and complex surgical procedure. For this reason, less invasive alternative approaches have been proposed like ventricular restraint therapy using polyester or nitinol devices and more recently biological approaches for myocardial support and regeneration such as stem cell transplantation associated with tissue-engineered scaffolds.

Synthetic mesh wrap devices have been clinically implanted around the heart; these devices are intended to prevent and reverse the progression of HF by the limitation of heart dilatation. Two devices were investigated: a polyester net-like sack designed for placement around the heart fabricated into a multifilament mesh knit (CorCap device, Acorn); and a nitinol

mesh for ventricular wrapping (HeartNet device, Paracor). Permanent implantation experience of both synthetic devices showed lack of improvement of systolic function, without evidence of myocardial healing and adverse effects like restriction in diastolic function, probably due to foreign body reaction fibrosis [39,40].

Recently, the RECATABI European Study demonstrated the feasibility and safety of bioactive implants manufactures with PEA and CLMA for the treatment of myocardial infarction in large animal models. Bioactive implants improved systolic and diastolic functions, reducing adverse cardiac dilatation. Better results were obtained combining semi-degradable CLMA material with a non-degradable PEA trellis mesh net. No inflammatory reaction was observed after long-term implantation of both materials; in addition, grafted marked stem cells have been found in histological studies of small and big experimental treated animals. These cells were localized at 6 months follow-up in bioactive implants, in infarcted myocardium and in vessels' wall. Stem cells actively migrate from the biohybrid scaffold to the host cardiac tissue (Figure 6).

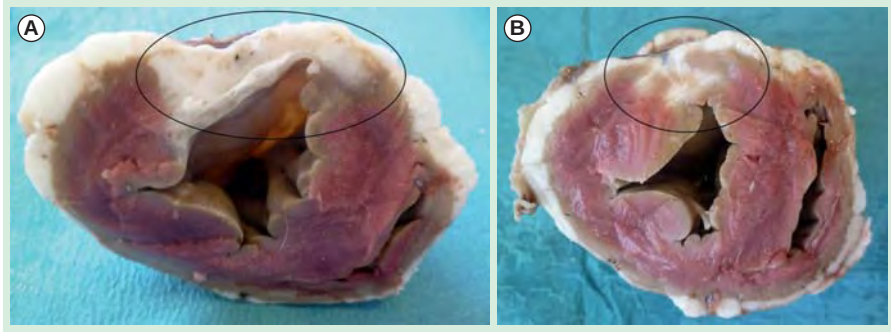

Figure 4. Pathological studies after myocardial infarction. (A) Heart macroscopic view showing a large left ventricular myocardial scar (encircled) following experimental infarction without treatment (sheep control group). Follow-up at 6 months. (**B)** Small infarct scar including islets of regenerated myocardium (encircled) in a heart treated with bioactive implant (caprolactone methacryloxyl ethyl ester scaffold patch with self-assembly peptides and adipose mesenchymal stem cells). Follow-up at 6 months.

Bioprostheses for left and /or right ventricular wrapping

Our proposal is to create bioprostheses using biohybrid scaffolds for adapted ventricular wrapping. Devices are designed for LV and/or right ventricular (RV) support and regeneration, including different sizes for partial or complete ventricular wrappings. The implant characteristics (mechanical, physical, chemical, biological) are adapted for the LV or the RV geometry, physiology and pathology.

For example, for biventricular HF patients, bioprostheses are made using the same semi-degradable material to wrap both ventricles; for RV failure cases, bioprostheses are made using high rate of biodegradable material for the normal LV; and for LV failure cases, bioprostheses are made of high rate of biodegradable material to wrap the normal RV (Figure 7).

Helical loop to restore ventricular shape & function

The anatomical model proposed by Torrent-Guasp *et al.* [41] considers the heart one muscle band plied in a double helical loop, this explains how the ventricles contract and get an efficient pumping in every heartbeat, achieving an ejection fraction of the 60% when sarcomeres individually contract 15% only [42].

Our studies lead to the creation of semi-degradable 'ventricular support bioprostheses' designed with the concept of 'helical myocardial bands' [101]. Bioprosthetic helical loops follow the anatomical heart configuration (native muscular ventricular bands) beginning at the insertion of the pulmonary artery in the RV and ending at the aortic valve annulus. In cases of severe heart dilatation, double basal and apical ventricular helical loops could be used to restore conical shape (Figure 8). The role of helical myocardial band is to limit LV dilatation, preserving elliptical shape, and contribute to systolic contraction and diastolic filling (suction mechanism generated by a piston-like downstream motion) [41,42].

Artificial prosthetic helical bands are a complement of ventricular support bioprostheses. These myocardial bands might be repopulated with MSCs or cardiac stem cells. Afterward, cardiomyogenic cell differentiation could be induced by physical approaches like electrostimulation and/or cyclic compressive strain (from the heart contractions).

Expert commentary

Tissue engineering and nanobiomaterials (containing nanoparticles smaller than 100 nm) emerge as new therapeutic tool becoming a promising way for the creation of 'Bioartificial Myocardium'. Biohybrid scaffolds have been implanted onto the heart for cardiac support and myocardial regeneration, these implants should provide a suitable environment for cell homing, growth and differentiation, as well as mechanical support to the heart. Semi-degradable nanofibers should allow the implanted cells to progressively interconnect, organize and contract. Partial degradation of the elastomers forming the implants should

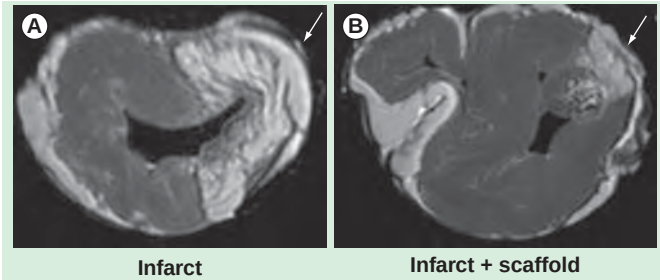

Figure 5. MRI of infarcted sheep, 6 months follow-up. MRI was performed after 10 min gadolinium injection. **(A)** Without treatment: an infarct volume of 15% related to left ventricular myocardial mass. **(B)** Treated with biohybrid scaffold: an infarct volume of 5% related to left ventricular myocardial mass. Arrows: white myocardial infarction area stained by gadolinium.

Box 3. Methods to induce cardiomyogenic cell differentiation.

- Chemical: cytokines, 5-azacytidine.
- Biological: stem cell co-cultures with cardiomyocytes.
- Genetic engineering.
- Physical: electric stimulation, magnetic fields, mechanical stimulation (cyclic compressive strain).

reduce the risk of development of chronic fibrosis involved in the restriction of diastolic function. Electrostimulation seems to be a safe method to induce physical and biochemical changes in isolated stem cells and in scaffolds repopulated with MSCs, moving toward cardiac cells.

New cells have been evaluated for regenerative cardiology like MSCs originating from adipose tissue. Theses cells can be found in a high concentration per ml tissue and can be easily harvested by liposuction [16,43]. Induced pluripotent stem cells (iPSCs, potentially autologous) and embryonic stem cells (ESCs) are promising cell sources for myocardial tissue engineering [44,45]. Also non-embryonic parthenogenetic stem cells (PSCs) can be directed toward the cardiac lineage and applied to tissue-engineered heart repair [46]. Recognition of the existence of cardiac stem cells and of the ability of mature myocytes to re-enter the cell cycle and proliferate has motivated the development of new approaches to regenerative cardiology [47,48]. In this direction, clinical trials are in progress like the SCIPIO trial using autologous cardiac stem cells [49] and the CADUCEUS trial using cardiosphere-derived cells [50].

Among paracrine factors, that could be responsible for the beneficial effect of the stem cell therapy, the most significant are believed to be VEGF, IGF-1 and basic fibroblast growth factor that interestingly are upregulated by hypoxia. In fact, it has been demonstrated that hypoxic preconditioning of stem cells prior to implantation, promotes their therapeutic potential as determined by their proangiogenic properties [51,52].

The use of MSCs as potential 'universal donor cells' should be carefully evaluated [6]. Some genetic abnormalities have been described for *in vitro* expanded MSC. Different methods should be applied to overcome the problem of karyotype instability and a universal protocol for identification of the same is warranted, to exclude cells carrying chromosomal abnormalities for clinical application [53].

Autologous human serum has been proposed for cell cultivation and expansion; this serum can be obtained by plasmapheresis. This approach avoids the antigen–antibody reaction and inflammatory reactions caused by heterologous proteins, and cell destruction as a consequence of the repeated lavages. In addition, cells cultured can be performed without the risk of contamination by prion disease, viruses and zoonosis inherent to the use of fetal bovine serum. To avoid the preparation of large volumes of autologous serum, it can be only used in the last-passage of cell cultures [54,55].

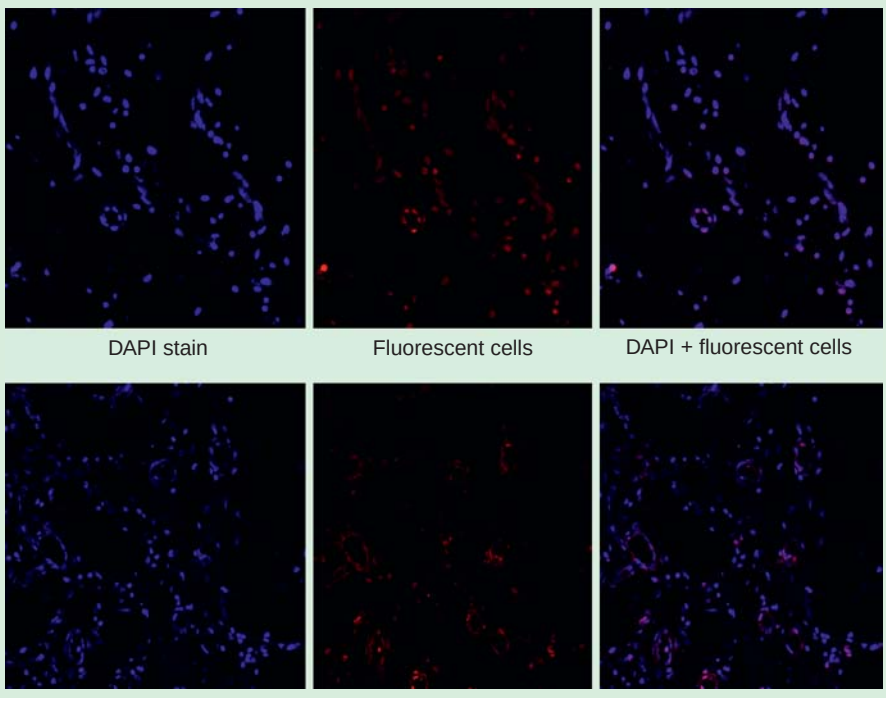

Figure 6. Left ventricular myocardial infarction in sheep treated with biohybrid scaffolds. Histochemical studies at 6 months. Left pictures show DAPI marked cells into infarcted areas. Central pictures show previous labeled fluorescent adipose stem cells in the same infarct areas. Right pictures show DAPI cells superimposed with fluorescent adipose stem cells. These cells are integrated into vessels walls (angiogenesis) within the infarct zone.
DAPI: 4′,6-diamidino-2-phenylindole.

Five-year view

Biohybrid nanomaterials are in development to be used as scaffolds for pathologic myocardium. Some show considerable interest, for example, semi-degradable materials like CLMA and self-assembling peptides (that produce nanostructures), all reinforced with a PEA trellis mesh netting. Bioactive patch carrying stem cells embedded in a self-assembling nanofiber network demonstrated good cell survival, regular cell distribution and to properly deliver cells into the ischemic cardiac tissue of small and large animal models.

Bioactive molecules and nanobiotechnologies may contribute for the creation of new bioprostheses for myocardial regeneration and LV and/or RV support. The properties of biohybrids could be improved associating electrostimulation, this might be a way to transform a

passive scaffold effect into a 'dynamic ventricular support'.

Ventricular bioprostheses derived from biohybrid scaffolds may reduce the indication for heart transplantation and the risk of death in chronic HF. Long-term effectiveness studies of bioactive implants evaluating the efficacy to prevent HF progression are needed to guide future clinical translation. Ventricular support bioprostheses could be indicated as a single procedure or after ventricular restoration surgery or the Revivent myocardial anchoring system.

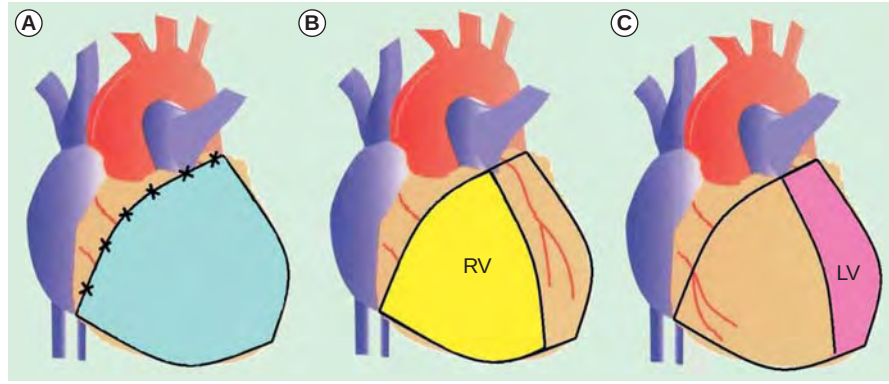

Figure 7. Ventricular support bioprostheses for cardiac wrapping. (A) For biventricular heart failure patients, where bioprosthesis is made of the same biohybrid material for both ventricles. **(B)** For RV failure patients, where bioprosthesis is made of elastomeric membrane for the right side and of biodegradable material for the normal left heart. **(C)** For LV failure patients, where bioprosthesis is made of elastomeric membrane for the left side and of biodegradable material for the normal RV.
LV: Left ventricle; RV: Right ventricle.

Nanomaterials can be useful for both *in vivo* and *in vitro* biomedical research and applications [56,57]. The size of nanomaterials (containing nanoparticles smaller than 100 nm) is similar to that of most biological molecules and structures of human body. Nanobiotechnology tools are progressing in areas of diagnosis, imaging and therapy, the integration of nanomaterials with biology has led to the development of diagnostic devices, contrast agents for cell imaging, analytical tools, nanoelectronic biosensors,

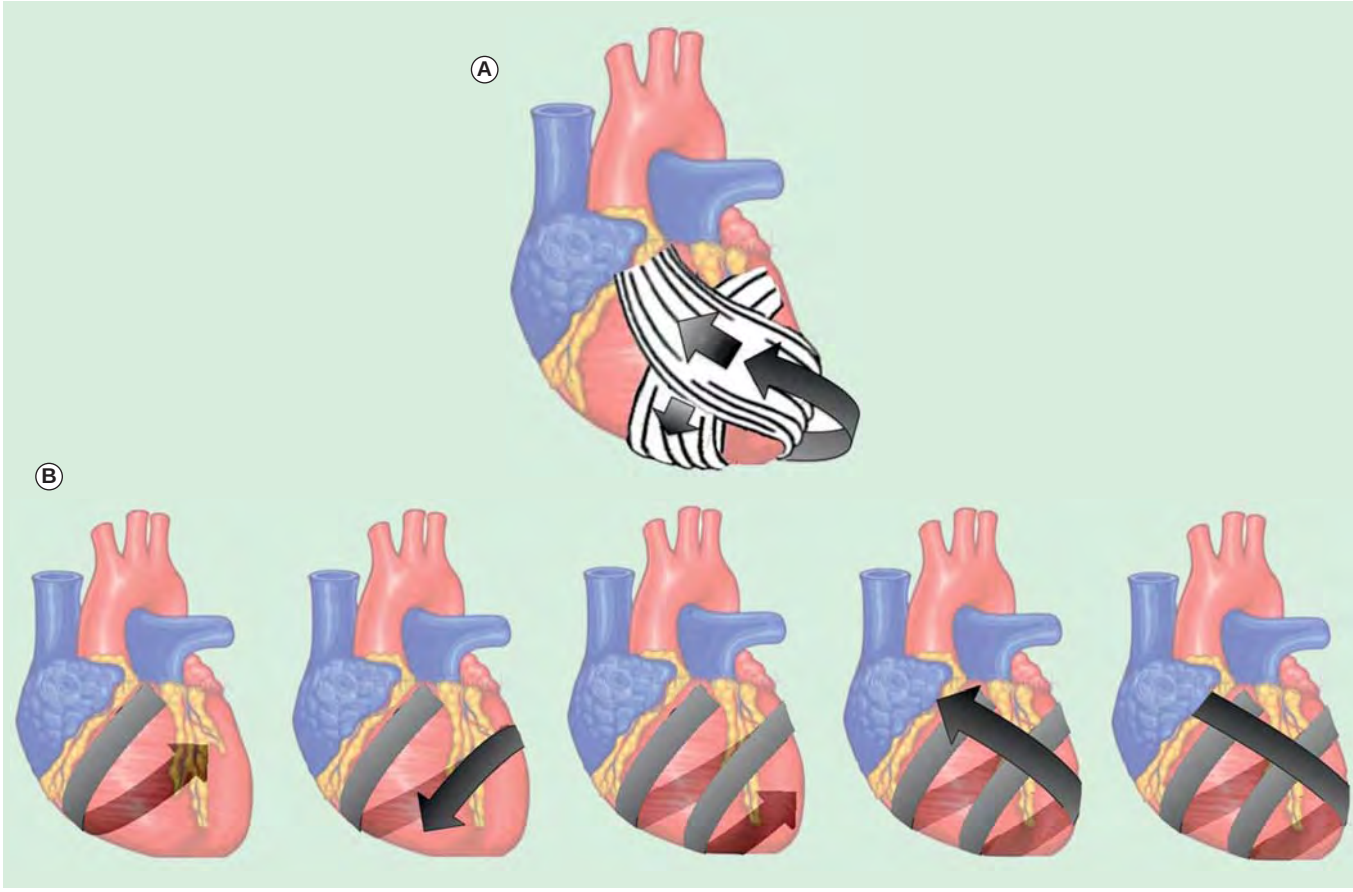

Figure 8. Bioprostheses are reinforced by helical loops made of a band of semi-degradable polymers to recover the conical shape of dilated spherical ventricles. (A) Single helical loop: For moderate heart dilatation, a single apical loop is used to wrap ventricles, starting at the level of the left atrial appendage and ending at the aortic root. **(B)** Double helical loop: For severe heart dilatation, double basal and apical ventricular helical loops are used to restore conical shape and to improve systolic and diastolic functions.

physical therapy applications, drug delivery vehicles and development of biohybrid materials for cardiac support and myocardial replacement [58–60]. The main challenges in the development of nanotechnologies seem to be related to fabrication strategies, biocompatibility, toxicity as well as environmental, ethical and societal impacts.

Bioartificial organs are in development in view of the limitations of the donor's pool for organ transplantation. Decellularized bioscaffolds from donor organs or tissues can be recellularized again using cells from recipients to create potentially functional organs. Pursuing the studies on nanostructured biomaterial strategies for the treatment of ischemic heart disease, should be important to progress from bench to bedside.

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Key issues

- Cardiac remodeling following myocardial infarction is one of the most common causes of heart failure (HF), cardiomyocyte death and scar formation modulate cardiac remodeling. Ischemic heart disease may progress inducing geometric alteration of the ventricular cavity, the original left ventricular (LV) elliptical chamber geometry becomes spherical. Heart dilatation is a negative symptom in the evolution of HF patients, related with morbidity and mortality. The underlying concept of rebuilding the ventricle by ventricular restoration is suggested to be reconstruction of form, rather than focusing on only the underlying disease.
- Tissue engineering and nanobiomaterials (containing nanoparticles smaller than 100 nm) emerge as new therapeutic tool becoming a promising way for the creation of 'bioartificial myocardium'. Biohybrid scaffolds are basically created introducing stem cells and self-assembling peptide nanofibers inside a porous elastomeric membrane. In this way, the membrane acts as carrier but most importantly, protects mechanically the cells into the soft nanofiber scaffold or hydrogel, creating cell niches.
- Biohybrid scaffolds carrying stem cells embedded in a self-assembling nanofiber network demonstrated good cell survival, regular cell distribution and to properly deliver cells into the ischemic cardiac tissue of large animal models. The implanted stem cells started active migration into the host tissue suggesting that it could contribute to induce regeneration, angiogenesis and cardiac tissue neo-formation, improving systolic and diastolic functions, limiting therefore adverse remodeling of ventricular chambers.
- Biohybrid scaffolds and nanobiotechnologies may contribute for the creation of cardiac wrap bioprostheses for myocardial regeneration and ventricular support that may reduce the risk of HF progression and cardiac death. Ventricular support bioprostheses could be indicated as a single procedure or following ventricular restoration surgery or the Revivent myocardial anchoring system.
- Ventricular support bioprostheses are designed with the concept of 'helical myocardial bands', following the anatomical heart configuration, where muscular ventricular bands provide conical configuration to LV chamber, beginning at the insertion of the pulmonary artery in the right ventricle and ending at the aortic valve annulus. For severe heart dilatation, double basal and apical ventricular helical loops are used. The role of myocardial band is to limit ventricular dilatation, preserving elliptical shape and contribute to systolic contraction and diastolic filling (suction mechanism). These myocardial bands may be repopulated with mesenchymal stem cells or cardiac contractile cells, afterward the construct might be electrically coupled to the host myocardium.
- Stem cell differentiation can be achieved by extrinsic physical stimuli (electrostimulation, magnetic fields), cyclic compressive strain as well as by chemical (cytokines) and biological/genetic stimuli (cell co-cultures, genetic manipulations). Chronic electrostimulated MSC grafted in 3D scaffolds seems to be a way for the creation of bioartificial myocardium.
- There is an increased demand for human organs to replace acute and chronically damaged tissues. This demand cannot be met by the currently available pool of donor organs. Efforts to provide an alternative source have led to the development of organ engineering, a discipline that combines progressive organ/matrix decellularization followed by cell perfusion for recellularization. These bioscaffolds can then be used to create potentially functional organs.

References

Papers of special note have been highlighted as:
• of interest
•• of considerable interest

1 Chachques JC. Cellular cardiac regenerative therapy in which patients? *Expert Rev. Cardiovasc.Ther.* 7, 911–919 (2009).

2 Rajnoch C, Chachques JC, Berrebi A, Bruneval P, Benoit MO, Carpentier A. Cellular therapy reverses myocardial dysfunction. *J. Thorac. Cardiovasc. Surg.* 121, 871–878 (2001).

3 Schussler O, Chachques JC, Mesana TG, Suuronen EJ, Lecarpentier Y, Ruel M. 3-dimensional structures to enhance cell therapy and engineer contractile tissue. *Asian Cardiovasc. Thorac. Ann.* 18, 188–198 (2010).

4 Spadaccio C, Rainer A, Chachques JC, Covino E, Herreros J, Genovese JA. Stem cells cardiac differentiation in 3D systems. *Front. Biosci. (Schol. Ed.)* 3901–3918 (2011).

5 Chachques JC. Development of bioartificial myocardium using stem cells and nanobiotechnology templates. *Cardiol. Res. Pract.* 2011,806795 (2010).

6 Zhang Y, Liang X, Lian Q, Tse HF. Perspective and challenges of mesenchymal stem cells for cardiovascular regeneration. *Expert Rev. Cardiovasc.Ther.* 11, 505–517 (2013).

7 Christalla P, Hudson JE, Zimmermann WH. The cardiogenic niche as a fundamental building block of engineered myocardium. *Cells Tissues Organs.* 195, 82–93 (2012).

•• **Important information about the fundamental role of the cardiogenic niche for engineered myocardium.**

8 Kensah G, Roa Lara A, Dahlmann J *et al.* Murine and human pluripotent stem cell-derived cardiac bodies form contractile myocardial tissue *in vitro*. *Eur. Heart J.* 34, 1134–1346 (2013).

9 Tulloch NL, Muskheli V, Murry CE *et al.* Growth of engineered human myocardium with mechanical loading and vascular coculture. *Circ. Res.* 109, 47–59 (2011).

10 Dahlmann J, Krause A, Möller L *et al.* Fully defined in situ cross-linkable alginate and hyaluronic acid hydrogels for myocardial tissue engineering. *Biomaterials* 34, 940–951 (2013).

11 Cortes-Morichetti M, Frati G, Schussler O, Van Huyen JP, Carpentier A, Chachques JC. Association between a cell-seeded collagen matrix and cellular cardiomyoplasty for myocardial support and regeneration. *Tissue Eng.* 13, 2681–2687 (2007).

12 Lilyanna S, Martinez EC, Kofidis T *et al.* Cord lining-mesenchymal stem cells graft supplemented with an omental flap induces myocardial revascularization and ameliorates cardiac dysfunction in a rat model of chronic ischemic heart failure. *Tissue Eng. Part A* (Epub ahead of print) (2013).

13 Turner WS, Wang X, Johnson S *et al.* Cardiac tissue development for delivery of embryonic stem cell-derived endothelial and cardiac cells in natural matrices. *J. Biomed. Mater. Res. B Appl. Biomater.* 100, 2060–2072 (2012).

14 Habib M, Shapira-Schweitzer K, Gepstein L *et al.* A combined cell therapy and in-situ tissue-engineering approach for myocardial repair. *Biomaterials* 32, 7514–7523 (2011).

15 Chachques JC, Trainini JC, Lago N, Cortes-Morichetti M, Schussler O, Carpentier A. Myocardial assistance by grafting a new bioartificial upgraded myocardium (MAGNUM trial): clinical feasibility study. *Ann. Thorac. Surg.* 85, 901–908 (2008).

•• **First world clinical application of myocardial tissue engineering for ischemic heart disease.**

16 Shafy A, Fink T, Zachar V, Lila N, Carpentier A, Chachques JC. Development of cardiac support bioprostheses for ventricular restoration and myocardial regeneration. *Eur. J. Cardiothorac. Surg.* 43, 1211–1219 (2013).

•• **Research and development of cardiac support bioprostheses for myocardial repair and limitation of ventricular dilatation.**

17 Castells-Sala C, Semino CE. Biomaterials for stem cell culture and seeding for the generation and delivery of cardiac myocytes. *Curr. Opin. Organ. Transplant.* 17, 681–687 (2012).

18 Vallés-Lluch A, Arnal-Pastor M, Martínez-Ramos C *et al.* Combining self-assembling peptide gels with three-dimensional elastomer scaffolds. *Acta Biomater.* 9, 9451–9460 (2013).

• **Investigation of new approaches for the creation of biohybrid scaffolds.**

19 Sicari BM, Johnson SA, Badylak S *et al.* The effect of source animal age upon the *in vivo* remodeling characteristics of an extracellular matrix scaffold. *Biomaterials* 33, 5524–5533 (2012).

20 Ott HC, Matthiesen TS, Goh SK *et al.* Perfusion-decellularized matrix: using nature's platform to engineer a bioartificial heart. *Nat. Med.* 14, 213–221 (2008).

21 Taylor DA. From stem cells and cadaveric matrix to engineered organs. *Curr. Opin. Biotechnol.* 20, 598–605 (2009).

22 Song JJ, Ott HC. Organ engineering based on decellularized matrix scaffolds. *Trends Mol. Med.* 17, 424–432 (2011).

23 Crapo PM, Gilbert TW, Badylak SF. An overview of tissue and whole organ decellularization processes. *Biomaterials* 32, 3233–3243 (2011).

24 Mari-Buye N, Semino CE. Differentiation of mouse embryonic stem cells in self-assembling peptide scaffolds. *Methods Mol. Biol.* 690, 217–237 (2011).

• **Original biological evaluation of self-assembling peptide nanofibers analyzing embryonic cell differentiation.**

25 Izal I, Aranda P, Sanz-Ramos P *et al.* Culture of human bone marrow-derived mesenchymal stem cells on of poly(L: -lactic acid) scaffolds: potential application for the tissue engineering of cartilage. *Knee Surg. Sports Traumatol. Arthrosc.* 21, 1737–1750 (2013).

26 Soler-Botija C, Bagó JR, Bayes-Genis A. A bird's-eye view of cell therapy and tissue engineering for cardiac regeneration. *Ann. N. Y. Acad. Sci.* 1254, 57–65 (2012).

• **Describes clinical translational feasibility of newly developed tissue-engineered biomaterials.**

27 Yan J, Dong L, Zhang B, Qi N. Effects of extremely low-frequency magnetic field on growth and differentiation of human mesenchymal stem cells. *Electromagn. Biol. Med.* 29, 165–176 (2010).

28 Flaibani M, Boldrin L, Cimetta E, Piccoli M, De Coppi P, Elvassore N. Muscle differentiation and myotubes alignment is influenced by micropatterned surfaces and exogenous electrical stimulation. *Tissue Eng. Part A* 15, 2447–257 (2009).

29 Michalopoulos E, Knight RL, Korossis S, Kearney JN, Fisher J, Ingham E. Development of methods for studying the differentiation of human mesenchymal stem cells under cyclic compressive strain. *Tissue Eng. Part C Methods* 18, 252–262 (2012).

30 Schraufstatter IU, Discipio RG, Khaldoyanidi S. Mesenchymal stem cells and their microenvironment. *Front. Biosci.* 17, 2271–2288 (2011).

31 Ling SK, Wang R, Dai ZQ *et al.* Pretreatment of rat bone marrow mesenchymal stem cells with a combination of hypergravity and 5-azacytidine enhances therapeutic efficacy for myocardial

infarction. *Biotechnol. Prog.* 27, 473–482 (2011).

32 Haneef K, Lila N, Benadda S, Legrand F, Carpentier A, Chachques JC. Development of bioartificial myocardium by electrostimulation of 3D collagen scaffolds seeded with stem cells. *Heart Int.* 7, e14 (2012).

•• **Original article showing that stem cells grafted in three-dimensional (3D) biological scaffolds can differentiate in cardiac cells using electrostimulation, becoming a convenient source for the creation of bioartificial myocardium.**

33 Shafy A, Lavergne T, Latrémouille C, Cortes-Morichetti M, Carpentier A, Chachques JC. Association of electrostimulation with cell transplantation in ischemic heart disease. *J. Thorac. Cardiovasc. Surg.* 138, 994–1001 (2009).

34 Genovese JA, Spadaccio C, Chachques E, Schussler O, Carpentier A, Chachques JC. Cardiac pre-differentiation of human mesenchymal stem cells by electrostimulation. *Front. Biosci.* 14, 2996–3002 (2009).

35 Llucià-Valldeperas A, Sanchez B, Soler-Botija C, Gálvez-Montón C, Prat-Vidal C, Bayes-Genis A. Electrical stimulation of cardiac adipose tissue-derived progenitor cells modulates cell phenotype and genetic machinery. *J. Tissue Eng. Regen. Med.* (Epub ahead of print) (2013) (doi: 10.1002/term.1710).

36 Schussler O, Coirault C, Louis-Tisserand M *et al.* Use of Arginine-Glycine-Aspartic acid adhesion peptides coupled with a new collagen scaffold to engineer a myocardium-like tissue graft. *Nat. Clin. Pract. Cardiovasc. Med.* 6, 240–249 (2009).

37 Chachques JC, Jegaden O, Mesana T, Glock Y, Grandjean PA, Carpentier AF; French Cardiomyoplasty Investigators. Cardiac bioassist: results of the French multicenter cardiomyoplasty study. *Asian Cardiovasc. Thorac. Ann.* 17, 573–580 (2009).

38 Chachques JC. Cardiomyoplasty: is it still a viable option in patients with end-stage heart failure? *Eur. J. Cardiothorac. Surg.* 35, 201–203 (2009).

39 Wenk JF, Ge L, Zhang Z, Mojsejenko D, Guccione JM, Ratcliffe MB. Biventricular finite element modeling of the Acorn CorCap Cardiac Support Device on a failing heart. *Ann. Thorac. Surg.* 95, 2022–2027 (2013).

40 Klodell CT Jr, Aranda JM Jr, McGiffin DC *et al.* Worldwide surgical experience with the Paracor HeartNet cardiac restraint device. *J. Thorac. Cardiovasc. Surg.* 135, 188–195 (2008).

41 Torrent-Guasp F, Kocica MJ, Corno AF, Komeda M, Cosin-Aguillar J, Wen H. Towards new understanding of the heart structure and function. *Eur. J. Cardiothorac. Surg.* 27,191–201 (2005).

42 Buckberg G, Hoffman JI, Nanda NC, Coghlan C, Saleh S, Athanasuleas C. Ventricular torsion and untwisting: further insights into mechanics and timing interdependence: a viewpoint. *Echocardiography* 28, 782–804 (2011).

43 Houtgraaf JH, den Dekker WK, Fernandez-Aviles F, Zijlsta F, Serruys PW, Duckers HJ. First experience in humans using adipose tissue-derived regenerative cells in the treatment of patients with ST-segment elevation myocardial infarction. *J. Am. Coll. Cardiol.* 59, 539–540 (2012).

44 Díez Villanueva P, Sanz-Ruiz R, NúñezGarcía A, Fernández Santos ME, Sánchez PL, Fernández-Avilés F. Functional multipotency of stem cells: what do we need from them in the heart? *Stem Cells Int.* 2012,817364 (2012).

• **Review that defines new concepts of stem cell potency and differentiation needed for cardiac repair.**

45 Zhang D, Shadrin IY, Lam J, Xian HQ, Snodgrass HR, Bursac N. Tissue-engineered cardiac patch for advanced functional maturation of human ESC-derived cardiomyocytes. *Biomaterials* 34, 5813–5820 (2013).

46 Didié M, Christalla P, Zimmermann WH *et al.* Parthenogenetic stem cells for tissue-engineered heart repair. *J. Clin. Invest.* 123, 1285–1298 (2013).

47 Smith RR, Marbán E, Marbán L. Enhancing retention and efficacy of cardiosphere-derived cells administered after myocardial infarction using a hyaluronan-gelatin hydrogel. *Biomatter* (Epub ahead of print) (2013) (doi: pii: e24490).

•• **Describes the development of a next generation cardiac engineered therapy, involving the combination of cardiosphere-derived cells and hydrogels.**

48 Chimenti I, Gaetani R, Barile L, Frati G, Messina E, Giacomello A. Isolation and expansion of adult cardiac stem/progenitor cells in the form of cardiospheres from human cardiac biopsies and murine hearts. *Methods Mol. Biol.* 879, 327–338 (2012).

49 Chugh AR, Beache GM, Loughran JH, Mewton N, Anversa P, Bolli R. 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* 126 (11 Suppl. 1), S54–S64 (2012).

50 Makkar RR, Smith RR, Marbán E *et al.* Intracoronary cardiosphere-derived cells for heart regeneration after myocardial infarction (CADUCEUS): a prospective, randomised phase 1 trial. *Lancet* 379, 895–904 (2012).

51 Bath C, Yang S, Muttuvelu D, Fink T, Emmersen J, Vorum H, Hjortdal J, Zachar V. Hypoxia is a key regulator of limbal epithelial stem cell growth and differentiation. *Stem Cell Res.* 10, 349–360 (2013).

52 Hu X, Yu SP, Fraser JL *et al.* Transplantation of hypoxia-preconditioned mesenchymal stem cells improves infarcted heart function via enhanced survival of implanted cells and angiogenesis. *J. Thorac. Cardiovasc. Surg.* 135, 799–808 (2008).

53 Ferreira RJ, Irioda AC, Cunha RC, Guarita-Souza LC, Chachques JC, de Carvalho KA. Controversies about the chromosomal stability of cultivated mesenchymal stem cells: their clinical use is it safe? *Curr. Stem Cell Res. Ther.* 7, 356–363 (2012).

54 Chachques JC, Herreros J, Trainini J, Juffe A, Prosper F, Genovese J. Autologous human serum for cell culture avoids the implantation of cardioverter-defibrillators in cellular cardiomyoplasty. *Int. J. Cardiol.* 95 (Suppl. 1), S29–S33 (2004).

55 Tekkatte C, Gunasingh GP, Cherian KM, Sankaranarayanan K. "Humanized" stem cell culture techniques: the animal serum controversy. *Stem Cells Int.* 2011, 504723 (2011).

56 Pennisi CP, Zachar V, Fink T, Gurevich L, Fojan P. Patterned polymeric surfaces to study the influence of nanotopography on the growth and differentiation of mesenchymal stem cells. *Methods Mol. Biol.* 1058, 77–88 (2013).

57 Sawada T, Mihara H, Serizawa T. Peptides as new smart bionanomaterials: molecular-recognition and self-assembly capabilities. *Chem. Rec.* 13, 172–186 (2013).

58 Laaksonen P, Szilvay GR, Linder MB. Genetic engineering in biomimetic composites. *Trends Biotechnol.* 30, 191–197 (2012).

59 An J, Chua CK, Yu T, Li H, Tan LP. Advanced nanobiomaterial strategies for the

development of organized tissue engineering constructs. *Nanomedicine (Lond.)* 8, 591–602 (2013).

60 Chachques JC. Bio-hybrid tissue engineering for cellular cardiomyoplasty: future directions. *Methods Mol. Biol.* 1036, 151–62 (2013).

Patents

101 Chachques JC, Bayes-Genis A, Monleon Pradas M, Semino CE, Zur Nieden N, Jenny PH. Bioactive implants for myocardial regeneration and ventricular chamber restoration. WO134957 (2011); EP-2422823-A1; US20130116789.

102 Chachques JC, Leonhardt H: Method of providing a dynamic cellular cardiac support. US7341062 (2008).

103 Chachques JC, Grandjean PA, Smits K. Cardiomyoplasty: method and apparatus including a sliding insulation lead for cardiac assistance. US4735205 (1988).

Website

201 RECATABI PROJECT (Regeneration of Cardiac Tissue Assisted by Bioactive Implants), financially supported by the 7th Framework Programme (FP7) of the European Commission. www.recatabi.com/

REPÈRES

RÉGÉNÉRATION. Parmi les nombreuses voies de recherche explorées pour pallier la défaillance d'un organe, et compte tenu du fait que le nombre de greffons (que ce soit la cornée, le rein, le foie, le poumon, le cœur ou la moelle osseuse) reste plus faible que le nombre de patients en attente de greffe, l'utilisation de cellules souches embryonnaires suscite, à moyen ou long terme, de réels espoirs. Ainsi, des chercheurs français ont il y a peu réussi, chez le rat, à faire se développer en deux mois des cellules cardiaques humaines, issues de cellules souches embryonnaires, dans la zone endommagée. Dans cet environnement propice, les cellules souches se sont différenciées en cardiomyocytes et les tissus cardiaques ont commencé à se régénérer. **AMPHIBIENS.** Des animaux comme la salamandre ont la capacité de régénérer spontanément certaines parties de leur corps, ce qui n'est pas le cas des oiseaux et des mammifères. Les chercheurs s'efforcent de découvrir les commandes génétiques de cette régénération spontanée. Ils ont déjà montré en laboratoire que des poulets par exemple avaient les gènes nécessaires. **CERVEAU.** De nombreux travaux ont également lieu pour tenter de régénérer des neurones ou leurs prolongements principaux, les axones, s'ils sont lésés. Ces recherches pourraient trouver des applications dans les maladies dégénératives du cerveau comme la maladie d'Alzheimer ainsi que d'autres troubles neurologiques. **FOIE.** Certaines équipes de recherche travaillent sur la réalisation de matrices artificielles sur ou dans lesquelles pourraient proliférer diverses sortes de cellules. Autre idée, dans le cas du foie par exemple, pouvoir se servir de la matrice tridimensionnelle décellularisée de l'organe pour la repeupler avec des hépatocytes, comme cela a été fait chez l'animal. Mais on est encore loin de pouvoir le réaliser chez l'homme. **J.-L. N.**

Face au manque de greffons, le bond en avant des organes artificiels

Ils devraient permettre à terme d'offrir une alternative à la transplantation.

MARTINE LOCHOUARN

GREFFES Face à la défaillance d'un organe qui met en péril la vie du malade, la greffe s'impose souvent. Le recours aux organes artificiels ou bioartificiels apparaît toutefois comme une alternative pour pallier la pénurie chronique de greffons, lorsqu'un cancer interdit le traitement immunosuppresseur nécessaire après une greffe ou quand celle-ci n'est pas faisable. Issus des techniques de suppléance vitale, des biotechnologies et de l'ingénierie cellulaire, ces organes de remplacement sont encore souvent à l'état de projets portés par des chirurgiens, des spécialistes d'organes, des réanimateurs ou des scientifiques.

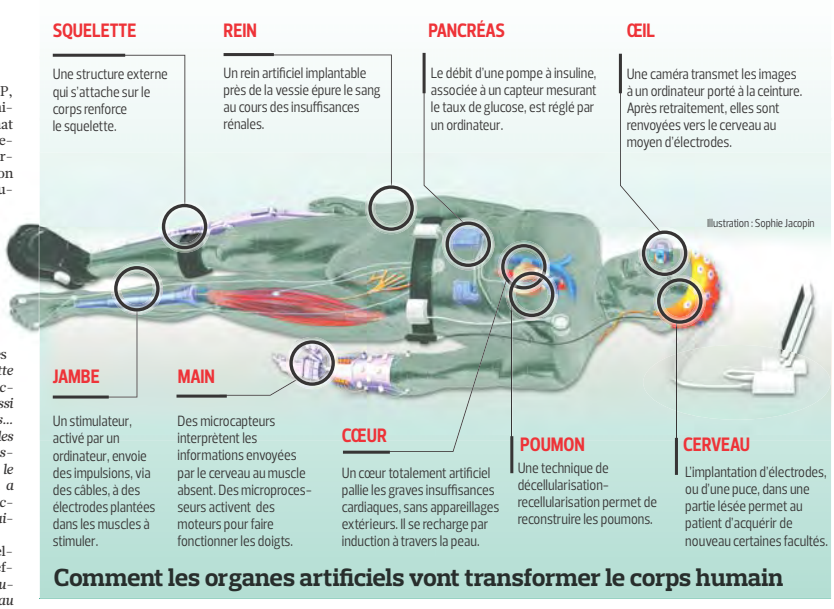
Le cœur artificiel suscite depuis longtemps l'intérêt car la transplantation, seul traitement curatif de l'insuffisance cardiaque avancée, se heurte à un manque criant de greffons. L'un des projets phares est celui du cœur implantable du Pr Alain Carpentier (HEGP, Paris), « père » des premières valves cardiaques en péricarde bovin, non immunogène. « *Le cœur Carmat qu'il développe avec le groupe EADS sera le premier cœur implantable bioartificiel, puisqu'il utilise ces valves et que ses cavités sont aussi tapissées de péricarde bovin* », souligne le Pr Juan Carlos Chachques, lui aussi chirurgien cardio-vasculaire (HEGP, Paris). Muni d'un système embarqué miniaturisé très sophistiqué, le Carmat comprend la bioprothèse implantée pesant 900 g et des éléments externes portables de monitoring et d'alimentation électrique. Première implantation humaine prévue fin 2011.

« Le cœur Carmat, développé avec le groupe EADS, sera le premier cœur implantable bioartificiel »
Pr JUAN CARLOS CHACHQUES (HEGP, PARIS)

Comment stimuler la régénération

Des tentatives très novatrices ont été menées par l'équipe hispano-américaine de Doris Taylor, d'abord sur le rat et tout récemment sur cœur humain. Le cœur est d'abord perfusé par des détergents qui éliminent les cellules mais laissent intacte la matrice. « *Cette matrice de collagène garde son architecture et son système de valves, donc aussi des fibres d'élastine, des polysaccharides... Puis elle est ensemencée avec des cellules cardiaques et endothéliales (1) qui reconstituent peu à peu l'organe. Chez le rat, le cœur obtenu, maintenu sous perfusion, a montré après quelques jours des contractions et une fonction de pompe rudimentaire* », explique le Pr Chachques.

À partir d'une culture des propres cellules du patient, on obtiendrait un greffon parfaitement compatible. « *Il faudrait d'abord évaluer le risque lié au détergent, réserver cette technique à des cœurs inutilisables pour la greffe, et la tester en premier pour régénérer une zone restreinte du cœur* », estime le Pr Chachques. Ce qu'il tente autrement sur le myocarde de malades lésé par l'infarctus en le recouvrant de « patchs » bioactifs à base de cellules souches immobilisées dans du collagène ou des nanofibres, pour stimuler sa régénération.



Comment les organes artificiels vont transformer le corps humain

La même technique de décellularisation-recellularisation a permis l'an dernier à deux équipes américaines (Petersen et Ott), en conservant l'architecture pulmonaire et vasculaire, de reconstruire des poumons chez le rat. Greffés, ces poumons ont assuré durant quelques heures les échanges gazeux avec le sang. Mais nombre de questions sur le type de cellules à utiliser, la fiabilité de la régénération et les mécanismes en jeu, devront être résolues avant un éventuel et lointain passage à l'homme.

Il n'existe pas aujourd'hui d'alternative à la greffe pulmonaire : impossible d'obtenir un système d'échanges gazeux aussi efficace que le poumon qui déploie, dans l'espace réduit de la cage thoracique, une surface d'échanges de 70 m^2. Seules techniques disponibles, celles de suppléance externe utilisées en réanimation. « *Les systèmes de circulation extracorporelle permettent maintenant de faire aussi de l'assistance pulmonaire en cas de défaillance respiratoire grave. Mais cela reste une technique de pointe, peu miniaturisable, aux indications très sélectionnées, en attente de récupération ou de transplantation* », explique le Pr Alain Combes (CHU Pitié-Salpêtrière, Paris). « *Malgré d'importants progrès, cette assistance demeure donc aujourd'hui temporaire* », précise le réanimateur.

Première mondiale, la greffe de bronche réalisée par le Pr Emmanuel Martinod (hôpital Avicenne, Paris) se heurtait à une contradiction : un tissu bronchique très immunogène et l'impossibilité d'un traitement immunosuppresseur chez un malade cancéreux. « *D'où notre choix d'utiliser un segment d'aorte dont la seule couche immunogène, l'endothélium, est détruite par la cryopréservation, rendant l'immunosuppression inutile*, explique le chirurgien. *Après mise en place, le segment, rigidifié par un stent, se recouvre peu à peu d'épithélium bronchique par colonisation à partir des extrémités de la greffe.* » La procédure va être renouvelée sur une série de malades très sélectionnée. ■

(1) L'endothélium est la couche interne qui tapisse l'intérieur des vaisseaux et du cœur et se trouve au contact du sang.

Rein et vessie : une multitude de techniques

LA GREFFE, meilleure solution dans l'insuffisance rénale terminale, ne couvre pas les besoins. Nombre de malades dépendent d'hémodialyses trois ou quatre fois par semaine pour épurer leur sang des déchets qui passent à travers une membrane dans un liquide de dialysat.

En dix ans, la technologie a beaucoup progressé. « *L'appareil sait désormais capter et analyser de nombreux paramètres du patient pour adapter en continu la dialyse, d'où une meilleure tolérance physique. Les dialysats ultrapurs ont réduit l'inflammation, donc les complications. Obtenues par nanotechnologie, de nouvelles membranes inertes au sang, aux pores calibrés, permettent d'adapter la filtration aux besoins*, explique le Pr Bernard Canaud (CHRU Montpellier). *À l'avenir, des vaisseaux bioartificiels provenant de cellules souches du patient, déjà testés chez quelques dialysés, pourraient permettre d'améliorer l'accès veineux.* »

Malgré des performances supérieures, la dialyse ne peut rivaliser avec le rein qui fonctionne en continu

Malgré ses performances supérieures, la dialyse intermittente ne peut rivaliser avec le rein qui fonctionne en continu. « *Il faut se rapprocher de la situation physiologique, avec des dialyses plus longues, plus fréquentes, donc développer des machines transportables, utilisables à domicile et capables de régénérer le liquide de dialysat*, insiste le néphrologue pour qui un rein artificiel implantable pourrait bien voir le jour dans les dix ans qui viennent. *La taille du dialyseur, déjà divisée par dix en dix ans, va atteindre celle d'un paquet de cigarettes. Si on résout le problème de l'accès au sang et de la coagulation, son implantation en continu près de la vessie devient envisageable.* »

Plus futuristes, des prototypes anglo-américains de reins artificiels portables régénérant le sang en continu ont été testés quelques heures sur des patients. Mais leur rendement reste faible et leur sécurité incertaine. « *Un usage courant semble très éloigné* », estime le Pr Canaud. Autre voie, le rein bioartificiel, incorporant des cellules rénales au dialyseur. Recréer un rein, très complexe, à partir de cellules souches semble encore plus lointain...

En urologie, la vessie est parfois enlevée, comme lors d'un cancer. « *Sa reconstruction à partir d'un segment d'intestin est devenue la règle et restaure avec de très bons résultats le réservoir vésical*, explique le Pr François Haab (hôpital Tenon, Paris). *Le développement de vessies artificielles a donc été délaissé, mais de récents essais cliniques ont reconstruit, en partie ou en totalité, une vessie à l'aide d'une matrice biologique acellulaire ensemencée avec des cellules vésicales du patient.* »

Autre remplacement, celui du sphincter vésical, le muscle circulaire qui obture la vessie. « *Dans l'incontinence urinaire sévère, la pose d'un implant sphincterien gonflable comprimant le canal urétral, permet au patient d'uriner selon ses besoins en appuyant sur une petite pompe cachée sous la peau*, indique l'urologue. *Quatre équipes, dont la nôtre, cherchent maintenant à restaurer la force du sphincter naturel défaillant en y injectant des cellules musculaires du malade. Les résultats, encore hétérogènes, nous font envisager plutôt l'usage de cellules moins différenciées.* » ■

M. L.

Vers des pancréas et des foies de substitution

« Dans les essais menés à l'hôpital, ce pancréas artificiel divise par cinq le risque d'hypoglycémie »
Pr ÉRIC RENARD, CHU MONTPELLIER

CERTAINS organes assurent aussi une fonction de synthèse. Par exemple, le pancréas dont les îlots de Langerhans sécrètent l'insuline, qui régule avec une infinie précision le taux de glucose dans le sang. « *Des greffes d'îlots permettent à certains diabétiques, transplantés rénaux ou dont les hypoglycémies sévères menacent la vie, de stabiliser leur diabète et souvent d'interrompre les injections. Mais c'est au prix d'une immunosuppression, et la production d'insuline des greffons diminue souvent au fil des ans* », explique le Pr Éric Renard (CHU Montpellier).

Parmi les alternatives, le pancréas artificiel. « *Il associe une pompe à insuline et un capteur qui mesure le taux de glucose en continu, avec un algorithme, un programme complexe géré par ordinateur qui règle en permanence le débit de la pompe en fonction du capteur.* »

Reste à mimer au mieux la physiologie. « *Le capteur de glucose placé sous la peau a un court délai de latence sur le taux de glucose sanguin, ce qui rend délicat l'équilibre de la glycémie après les repas, et l'insuline, délivrée aussi sous la peau, demande un petit délai pour diffuser jusqu'au sang* », explique l'hépatologue. Toute la difficulté, c'est d'élaborer un algorithme qui prenne bien en compte ces contraintes. « *Dans les essais menés à l'hôpital, ce pancréas artificiel divise par cinq le risque d'hypoglycémie. Nous allons lancer un essai à domicile, de nuit, où ce risque est le plus élevé, avec une plate-forme de télémonitoring permettant de surveiller et d'intervenir à distance sur le système.* » Ce projet européen devrait aboutir dans les trois ans.

L'objectif : encapsuler des îlots humains

Autre piste, le pancréas bioartificiel. La greffe d'îlots de porc encapsulés a été abandonnée à cause du risque d'obstruction et la nécessité d'un traitement immunosuppresseur limite celle d'îlots humains. « *L'objectif serait d'encapsuler des îlots humains pour réduire les réactions immunitaires, donc l'immunosuppression nécessaire. Des recherches très préliminaires évaluent pour cela des membranes proches de celles du rein artificiel, perméables au glucose et à l'insuline mais pas aux anticorps.* » Quant à la différenciation de cellules souches en îlots de Langerhans, elle est beaucoup plus problématique...

Organe complexe de transformation, de stockage, de synthèse et de filtration, le foie pose encore plus de problèmes. « *Il n'existe pas aujourd'hui de suppléance hépatique efficace, capable de remplacer la greffe de foie*, explique le Pr Didier Samuel (hôpital Paul-Brousse, Villejuif). *Des systèmes artificiels de dialyse à l'albumine permettent de détoxifier le plasma de patients en insuffisance hépatique sévère, et parfois de passer un cap dangereux. Mais ils ne remplacent pas les autres fonctions du foie, que seules des cellules humaines peuvent assurer.* »

Des dispositifs de foie bioartificiel extracorporel à base de cellules hépatiques cultivées sont en essais cliniques très préliminaires aux États-Unis et en Chine. En France, Cécile Legallais (CNRS, UTC Compiègne) a développé un bioréacteur permettant de cultiver des hépatocytes encapsulés dans des fibres creuses d'alginate, qui restent fonctionnels plus de quinze jours. « *Un projet commun avec l'équipe de Villejuif devrait nous permettre de l'évaluer assez rapidement en clinique* », espère-t-elle. ■

M. L.