Title: Prognostic impact of cardiac amyloidosis in patients with severe aortic stenosis who underwent percutaneous implantation of aortic valve prosthesis

Promoter: Hemodynamics Unit, Department of Cardiology in Santiago de Compostela Hospital

Main Researcher: Diego López Otero

Protocol Code: AMY-TAVI

Project Type: An analytical and observational study of prospective cohorts at a multicenter and international level.

INDEX

1. STUDY SUMMARY

Study title and code	Prognostic impact of cardiac amyloidosis in patients with severe aortic stenosis who underwent percutaneous implantation of an aortic prosthesis (AMY-TAVI)
Study type and design	Observational analytical of multicenter prospective cohorts.
Number and characteristics of patients	321 patients to whom a TAVI has been implanted by severe degenerative AoS.
Study duration	Recruitment period: All the consecutive patients to whom a non-invasive diagnostic study of amyloidosis (scintigraphy with tecnecio^{99} pyrophosphate and electrophoresis of proteins in the blood) and to whom a percutaneous aortic prosthesis for severe aortic stenosis has been implanted. Follow-up period: clinical and echocardiographic monitoring will be done before discharge, in consultation at three, six, twelve, and twenty-four months. Inclusion: 1 year Follow-up: 2 years Data analysis: 6 months Total: 3 years and 6 months
Events	Main event: cardiovascular events (cardiovascular death, stroke, heart attack), death from any cause, readmission for cardiac insufficiency, pacemaker implantation, functional assessment
Participating countries and centers	Various Spanish and international centers will be invited to participate.

2. GLOSSARY AND DEFINITIONS

AoS	Aortic stenosis
TAVI	Percutaneous implant of aortic prosthesis
GT99	Scintigraphy with talio^{99}
PE	Protein Electrophoresis
CMR	Cardiac Magnetic Resonance
AVR	Aortic valve replacement
ECG	Electrocardiogram
6MT	6-minute test
HR	Hazard ratio
TTE	Transthoracic echocardiography
DCL	Data Collection Logbook
CREC	Clinical research ethics committee
IHC	International harmonization conference

3. LIST OF RESEARCHERS AND TASKS TO BE COMPLETED

University Hospital Complex of Santiago de Compostela

- **Main researcher:**
  - Diego López Otero

- **Collaborating researchers:**
  - Ramiro Trillo Nouche, responsible for the Hemodynamic Department and the healthcare procedure for severe aortic stenosis and TAVI.
  - Javier López Pais, responsible for patient selection, coordination and data collection.
  - Virginia Pubul Núñez, responsible for the acquisition and interpretation of the GT99.
  - María Bastos Fernández, responsible for the acquisition and interpretation of the echocardiography.
  - Juan Carlos Sanmartín Pena, co-responsible for the healthcare procedure for severe aortic stenosis and TAVI.
  - Ana Belén Cid Álvarez, co-responsible for the healthcare procedure for severe aortic stenosis and TAVI.
  - Carmen Neiro Rey, co-responsible for the programming of tests, monitoring, and functional studies.
  - Montserrat Bello Seoane, co-responsible for the programming of tests, monitoring, and functional studies.
  - Mónica Gómez Fernández, co-responsible for the programming of tests, monitoring, and functional studies.

The intention is to increase the number of collaborating centers in order to carry out a study of an international multicentre by nature. (To begin with, other Sergas hospitals will not be taken into account).

4. INTRODUCTION AND CURRENT STATUS OF THE MATTER

The aortic stenosis.

The degenerative aortic stenosis (AS) is a progressive disease that is related to atherosclerosis, inflammation, hemodynamic factors, and active calcification. Its prevalence is high and depends on age. More than 3% of people over 75 have aortic stenosis(1) and reaches 8% in people over 84(2). When symptoms appear, the disease is rapidly progressive, and a surgical or percutaneous aortic valve replacement is necessary. It is, therefore, a disease with a major impact on society. Without a doubt, we are facing a serious health problem that will increase in the future, giving the aging population.

The percutaneous implantation of an aortic valve prosthesis.

Given the nature of the degenerative aortic stenosis, most of the patients affected to a severe degree and requiring treatment have an advanced age. The fragility of aging combined with the comorbidities presented in many cases makes up one-third of the patients who are not candidates for a surgical valve and another third present a high risk. The percutaneous implantation of an aortic prosthesis (TAVI) emerges as a less invasive alternative for the treatment of aortic stenosis. The randomized study PARTNER demonstrated in cohort B a reduction in mortality with this technique compared with standard medical treatment in inoperable patients(3). In cohort A of the PARTNER study, no differences were found in the survival of patients with high-risk surgical treatment randomized to conventional surgery or percutaneous implant(4). The studies PARTNER-II and SURTAVI have more recently shown that those patients, with intermediate surgical risk, the percutaneous aortic prosthesis implant has similar results to those of conventional surgery (5)(6). This has increased the number of percutaneous aortic prosthesis implants exponentially.

There are still many doubts about the long-term outcome after a TAVI implant. Avoiding the sternotomy and extracorporeal circulation by means of a percutaneous access has many advantages, but it also causes a series of technical and clinical problems: 1, cerebrovascular events, not only those due to the handling of large

vessels during the procedure but also those occurred in the monitoring where there is still no solid evidence regarding antithrombotic handling. 2, Atrioventricular conduction disorders requiring permanent pacemaker implantation in 20-40% of the patients. 3, Vascular complications of the accesses. 4, Residual valve by the malpositioning of the prosthesis (creating a high residual gradient or insufficiency). 5, Valvular degeneration and durability of the prosthesis.

Cardiac amyloidosis by transthyretin.

The amyloid myocardial infiltration by transthyretin monomers and oligomers (TTR) with misfolding(7) is a serious and under-diagnosed disease which predominantly affects men over 60 years old and presents itself with symptoms of heart failure, orthostatic hypotension, and syncope (8). The infiltration of the cardiac conduction system can generate branch blocks, atrioventricular blocks, and atrial fibrillation.

There are two types of amyloid that can infíltrate the myocardium: the amyloid light chain (AL or primary) within a monoclonal gammopathy, and the TTR amyloid. The latter in turn has two types of disease, familial, due to a mutation that causes transthyretin misfolding; and sporadic (formerly known as senile), which is due to incorrect folding of the common transthyretin. (8)

The prevalence of the TTR amyloidosis in patients with AoS to whom a TAVI is performed is estimated at 14-16%(9)(10) and 6% in those over 65 years old to whom a surgical valve replacement is performed(11). It is especially associated with the AoS phenotype of low-flow and low gradient (9). Cardiac amyloid deposits were found in nearly one-third of autopsies of patients with TAVI implants who died before 30 months(12), and the patients with AoS and amyloidosis have a prognosis similar to those with amyloidosis without AoS (13), It all states that the amyloidosis implies a worse prognosis (11). It is also suggested that amyloid infiltration may increase the requirements of permanent pacing (14).

Recently the benefits of treatment with Tafamidis on patients with cardiac amyloidosis by TTR have been published. (15)

Non-invasive diagnosis of cardiac amyloidosis.

The echocardiogram is used to find interesting data of amyloidosis: increased thicknesses of the atrioventricular valves, interatrial septum valves or the free wall of

the right ventricle, the light pericardial effusions, high atrial dilation, a birefringence characteristic (*sparkling*) and a speed of S' waveform mitral in the *Doppler* tissue lower than 6cm/s(9), but in no case is it diagnosed. The mechanical ventricular assessed by *speckle tracking*(tracking marks) may be useful in monitoring patients with amyloidosis as there seems to be a link between the longitudinal *strain* of the ventricular and the cardiac involvement (16). The longitudinal *strain* is decreased in the basal and middle segments and stored in the apical segments(17). This typical pattern helps obtain the differential diagnosis with other causes of hypertrophy ventricular, with high sensitivity and specificity (90-95% and 80-85%, respectively). The value of the strain is also an independent prognostic factor (18).

The maps on T1 MRI may have a role in the diagnosis (19), but it is still a technique in the early stage of development.

Until recently, the diagnosis of amyloidosis required histological confirmation by biopsy and staining with Congo red of the sample. Recent studies have demonstrated the diagnostic capacity of the pyrophosphate tecnecio^{99} scintigraphy (20), with a sensitivity of more than 99% and a specificity of 86% for the TTR amyloidosis, being false positives due almost exclusively to the presence of AL amyloidosis. Therefore, the caption of radiotracer in grades 2 o 3 together with the absence of a monoclonal protein in the electrophoresis has a specificity and a positive predictive value to 100% TTR amyloidosis. (21)

5. BIBLIOGRAPHY

1. Muszil D, Avelino A, Matesz K, Manuscript A, Asymmetric LPK, Schank JR, et al. NIH Public Access. Changes. 2009;34(6):1443–53.

2. Lindroos M, Kupari M, Heikkilä J, Tilvis R. Prevalence of aortic valve abnormalities in the elderly: An echocardiographic study of a random population sample. J Am Coll Cardiol. 1993;21(5):1220–5.

3. Leon MB, Smith CR, Mack M, Miller DC, Moses JW, Svensson LG, et al. Transcatheter Aortic-Valve Implantation for Aortic Stenosis in Patients Who Cannot Undergo Surgery. N Engl J Med. 2010 Sep 22;363(17):1597–607.

4. Smith CR, Leon MB, Mack MJ, Miller DC, Moses JW, Svensson LG, et al. Transcatheter versus Surgical Aortic-Valve Replacement in High-Risk Patients. N

Engl J Med. 2011 Jun 5;364(23):2187–98.

5. Adams DH, Popma JJ, Reardon MJ, Yakubov SJ, Coselli JS, Deeb GM, et al. Transcatheter Aortic-Valve Replacement with a Self-Expanding Prosthesis. N Engl J Med. 2014;370(19):1790–8.

6. Leon MB, Smith CR, Mack MJ, Makkar RR, Svensson LG, Kodali SK, et al. Transcatheter or Surgical Aortic-Valve Replacement in Intermediate-Risk Patients. N Engl J Med. 2016 Apr 2;374(17):1609–20.

7. Rapezzi C, Quarta CC, Riva L, Longhi S, Gallelli I, Lorenzini M, et al. Transthyretin-related amyloidoses and the heart: a clinical overview. Nat Rev Cardiol. 2010 May 18;7:398.

8. Ruberg FL, Berk JL. Transthyretin (TRR) Cardiac Amyloidosis. Circulation. 2012;126(10):617–38.

9. Castano A, Narotsky DL, Hamid N, Khalique OK, Morgenstern R, DeLuca A, et al. Unveiling transthyretin cardiac amyloidosis and its predictors among elderly patients with severe aortic stenosis undergoing transcatheter aortic valve replacement. Eur Heart J. 2017;38(38):2879–87.

10. Scully PR, Treibel TA, Fontana M, Lloyd G, Mullen M, Pugliese F, et al. Prevalence of Cardiac Amyloidosis in Patients Referred for Transcatheter Aortic Valve Replacement. J Am Coll Cardiol. 2018;71(4):463–4.

11. Treibel TA, Fontana M, Gilbertson JA, Castelletti S, White SK, Scully PR, et al. Occult transthyretin cardiac amyloid in severe calcific aortic stenosis. Circ Cardiovasc Imaging. 2016;9(8):1–10.

12. Nietlispach F, Webb JG, Ye J, Cheung A, Lichtenstein S V., Carere RG, et al. Pathology of transcatheter valve therapy. JACC Cardiovasc Interv. 2012;5(5):582–90.

13. Sperry BW, Jones BM, Vranian MN, Hanna M, Jaber WA. Recognizing Transthyretin Cardiac Amyloidosis in Patients With Aortic Stenosis: Impact on Prognosis. JACC Cardiovasc Imaging. 2016;9(7):904–6.

14. Adam Castaño M, Sabahat Bokhari M, Mathew S. Maurer M. Reply: Could late enhancement and need for permanent pacemaker implantation in patients undergoing TAVR be explained by undiagnosed transthyretin cardiac amyloidosis? J Am Coll Cardiol. 2015;65(3):311.

15. Maurer MS, Schwartz JH, Gundapaneni B, Elliott PM, Merlini G, Waddington-Cruz M, et al. Tafamidis Treatment for Patients with Transthyretin Amyloid Cardiomyopathy. N Engl J Med. 2018;NEJMoa1805689.

16. Urbano-Moral JA, Gangadharamurthy D, Comenzo RL, Pandian NG, Patel AR. *Speckle tracking* three-dimensional echocardiography(tracking marks) in the cardiac amyloidosis of light chains: study of the parameters of the mechanical myocardial left and right ventricular. Rev Española Cardiol. 2015;68(08):657–64.

17. Phelan D, Collier P, Thavendiranathan P, Popović ZB, Hanna M, Plana JC, et al. Relative apical sparing of longitudinal strain using two-dimensional speckle-tracking echocardiography is both sensitive and specific for the diagnosis of cardiac amyloidosis. Heart. 2012 Oct 1;98(19):1442 LP-1448.

18. Tendler A, Helmke S, Teruya S, Alvarez J, Maurer MS. The myocardial contraction fraction is superior to ejection fraction in predicting survival in patients with AL cardiac amyloidosis. Amyloid. 2015 Jan 2;22(1):61–6.

19. Karamitsos TD, Piechnik SK, Banypersad SM, Fontana M, Ntusi NB, Ferreira VM, et al. Noncontrast T1 mapping for the diagnosis of cardiac amyloidosis. JACC Cardiovasc Imaging. 2013;6(4):488–97.

20. Bokhari S, Castaño A, Pozniakoff T, Deslisle S, Latif F, Maurer MS. (99m)Tc-Pyrophosphate scintigraphy for differentiating light-chain cardiac amyloidosis from the transthyretin-related familial and senile cardiac amyloidoses. Circ Cardiovasc Imaging. 2013 Mar 1;6(2):195–201.

21. Gillmore JD, Maurer MS, Falk RH, Merlini G, Damy T, Dispenzieri A, et al. Nonbiopsy diagnosis of cardiac transthyretin amyloidosis. Circulation. 2016;133(24):2404–12.

7. HYPOTHESIS

Amyloidosis is a disease which coexists in a high percentage of patients referred for TAVI by severe aortic stenosis; the pathophysiological mechanisms of this relationship have a negative impact on prognosis.

8. OBJECTIVES

8.1 PRIMARY OBJECTIVE

A multicentre prospective study of cohorts consisting of two cohorts from patients who have been implanted with percutaneous aortic prosthesis as treatment for the severe degenerative aortic stenosis and compare the overall mortality and that for

cardiovascular and admissions for heart failure in the 2-year follow-up of patients diagnosed with amyloidosis (by GT99 and EFP) versus those that do not have it.

8.1 SECONDARY OBJECTIVES

1. As part of the hypothesis, we will analyze separately the impact of the presence of amyloidosis on the requirements of permanent pacing (permanent pacemaker implantation, pacemaker dependence, changes in the conduction system)

2. Determine the impact on the functional class (measured by using the 6-minute test) of the coexistence of TTR amyloidosis in patients with a TAVI implant.

3. Validate echocardiographic patterns of cardiac amyloidosis diagnosed by ventricular strain in a population with EAS undergoing a TAVI implant, as well as study the evolution of these patterns mid-term.

9. DESIGN

A multicentre, analytical, observational study of two cohorts. One cohort will be constituted by the patients to whom a TAVI will be implanted, and that meet non-invasive diagnostic criteria of TTR amyloidosis (capting of radiotracer in grades 2 and 3 and absence of monoclonal gammopathy the PE of urine or serum). The other cohort will be constituted by patients whose TAVI implants do not meet the diagnostic criteria for TTR amyloidosis.

A clinical and echocardiographic follow-up will be done in all patients for two years.

9.1. POPULATION

Elderly patients, enlisted in the hospitalization area or from outpatients clinics that meet the following requirements will be included in the study: diagnosis of severe aortic stenosis by conventional echocardiographic criteria (maximum speed >4m/s, medium transvalvular gradient >40mmHg, area <1cm2).

9.2. INCLUSION CRITERIA

- Replacement of the aortic valve with a percutaneous prosthesis by severe aortic stenosis after evaluation by the Heart Team.

- Ability to understand and sign informed consent.

- Do not meet any of the exclusion criteria.

9.3. EXCLUSION CRITERIA

- Death as a result of a complication of the procedure during hospitalization.

- An associated mitral valve requiring intervention

- TAVI implant valve-in-valve.

- Does not sign informed consent.

9.4. INFORMED CONSENT

If the patient decides to participate, the patient information sheet will be given to the patient, and after reading the document and clarifying any doubts, if they agree to participate in the study, the patient will sign the informed consent.

10. STATISTICAL ANALYSIS PLAN AND SAMPLE SIZE

10.1 STATISTICAL ANALYSIS

A descriptive analysis of both cohorts defining their clinical and angiographic profiles and a comparative study of the two groups (cohorts) will be performed. The categorical variables are expressed in percentages and the continuous as medium value ± standard deviation. The relative risk and the "Hazard Ratio" will be used as measures of effect when variables are analyzed with a temporal component, always with a confidence interval of 95%. For the comparison of categorical variables, the Chi-square or the Fisher test will be used. To compare continuous variables, the "t" of student test or analysis of variance will be used. The variables of time to event, such as the analysis of event-free survival will be analyzed by using the Kaplan- Meier technique and will be supplemented by the log-rank test. The multivariate analysis of the adverse events predictors during the follow-up will be done by means of the Cox regression model, determining the Odds Ratio for each variable.

10.1 SAMPLE SIZE

Based on the information available, around 15% of the patients to whom a TAVI has been implanted show signs of amyloidosis. Mortality at two years after a TAVI is about 17% and in patients with amyloidosis, it is estimated to be 40%. Based on this data, for a significance level of 5% (error $\alpha = 0,05$) and a power of 80% (error $\beta = 0,20$), a sample size of 306 patients is estimated, assuming 5% of follow-up loses, it was decided to include 321 patients. The results in which the calculations are based are given at 2 years, if not significant; monitoring would continue up to 3 years.

11. PATIENTS FOLLOW-UPS

11.1 INITIAL PATIENT ASSESSMENT

An initial inclusion visit will assess whether the patient is a candidate who meets the inclusion criteria and none of the exclusion ones. After being informed, if they accept, the informed consent must be signed. The following data must be recorded:

- **Clinical parameters:** age, sex, height, weight, body mass index, body surface, cardiovascular risk factors, medication intake, background interests, history of heart disease, presence of ischemic heart disease, cerebrovascular accident, peripheral artery disease, chronic obstructive pulmonary disease, symptoms (angina, syncope, dyspnea according to grades of the NYHA, other symptoms), blood pressure, heart rate, evidence of heart failure, assessment of functional class by means of 6MT and fragility assessment.

- **Analytical parameters**: hematocrit, NT-proBNP, Troponin I, electrolytes, C-reactive protein, creatinine, glomerular filtrate.

-**Protein electrophoresis:** Rating presence of the monoclonal component

- **Scintigraphy with TC^{99}**: Assessment of values of tracer uptake.

- **Device parameters:** Type of prosthesis, dimensions, expandable/ self-expanding ball, post-dilatation.

- **ECG parameters:** Rhythm, PR duration, pacemakers, implantable cardioverter defibrillator, QRS duration.

- The 6 minute-test: All patients in the study will have a test of 6 minutes before implantation and 6 months follow-up, according to the protocol after a TAVI, in order to carry out an objective functional assessment of the functional capacity. The distance covered will be determined, the variation of oxygen saturation by means of non-invasive pulse oximetry before and after the test and the appearance of symptoms with the effort according to the Borg scale.

11.2. CLINICAL AND IMAGE FOLLOW-UP

This will be done before discharge, in consultation at three, six, and 12 months and annually thereafter. All the visits have a window period to be carried out with additional tests within this protocol, one week before and one week after the due date.

First visit (Basal Visit).
- Assessment of patient history.
- Detailed physical assessment of the patient.
- Electrocardiogram.
- Analysis.
- Echocardiographic basal study, with ventricular *strain* measurements.
- Perform 6-minute test.

Follow-up visit at 3 months (more/less one week)
- Clinical assessment.
- Analysis.
- Events assessment.

Follow-up visit at 6 months (more/less one week)
- Clinical assessment.
- Analysis.
- Perform 6-minute test.
- Events assessment.

Follow-up visit at 12 months (more/less one week)
- Clinical assessment.
- Electrocardiogram.

- Analysis.
- Echocardiographic study with ventricular *strain* measurements.
- Events assessment.

Follow-up visit at 24 months (more/less one week)

- Clinical assessment.
- Analysis.
- Echocardiographic study.
- Events assessment.

11.3 WITHDRAWAL FROM THE STUDY

A) Withdrawal of a patient from the study because they withdraw the consent.

B) Withdrawal of the study due to transfer to another health area

12. WORK PLAN

	Basal	3 months	6 months	1 year	2 years
Clinical history	X				
Informed Consent	X				
Clinical assessment	X	X	X	X	X
ECG	X		X	X	X
Analysis[1]	X		X	X	X
Protein electrophoresis	X				
Scintigraphy with tecnecio^{99}	X				
Echocardiogram with *strain*	X		X	X	X
The 6-minute test	X		X		
Events assessment		X	X	X	X

1: includes: hemoglobin, hematocrit, lipid profile, NT-proBNP, TnT, electrolytes, C-reactive protein, creatinine, and glomerular filtrate.

13. DATA MANAGEMENT

13.1. DATA COLLECTION:

All the patients will receive standard treatment according to clinical practice guidelines and consensus in force. The criteria for inclusion/ exclusion will be assessed in all of them to confirm the feasibility of entering the study, which will be offered, explained, and then the appropriate informed consent collected. Subsequently, following the predefined protocol, a number of epidemiological variables, clinical, laboratory, electrocardiographic, echocardiographic, functionals, scintigraphy with Tc^{99}basals of the patient will be collected before discharge. All previously detailed. All the data will be collected in the Data Collection Logbook (DCL), which is designed to register all the data required in the protocol and collected by the researcher. The data on the DCL will be coded. The data entry will be done by the researcher or the person assigned by them, and then that data will be transferred to the database of the study. The researcher or the appointed person in their team commits to completing the DCL on each visit made by the participant. All data corrections in the DCL will be made by the researcher or the person assigned in their team in accordance with the instructions given. After the last visit from the participant and the closure of the DCL, the researcher must attest, by signing the DCL, the authenticity of the data collected in the DCL, the coherence between the data in the DCL and the data contained in the source document.

13.2DATA CONFIDENTIALITY:

All documents and information provided to the researchers by the promoter regarding the study are confidential. Both the researcher and their collaborators undertake to use the information provided only within the framework of this study to carry out this protocol. This commitment remains as long as the confidential information has not been made public by the promoter. The protocol delivered to the researcher can be used by them or their collaborators to obtain informed consent from the participants entering the study. It should not be disclosed to third parties without the written consent of the promoter. This information will be treated in accordance with the rules of professional secrecy. The screening list must be completed when the researcher verifies that the participant can enter the study after confirming their record and medical history.

14. PRACTICAL USE OF THE RESULTS

The progressive aging of our population carries an increasing prevalence of degenerative aortic valve stenosis in our country, constituting a public health problem

of crucial importance. There is scientific evidence that amyloidosis is an underdiagnosed disease in clinical practice, which coexists simultaneously in a large number of patients suffering from aortic stenosis, and that could overshadow the prognosis. To determine the prevalence and prognostic impact of amyloidosis referred for percutaneous implantation of an aortic prosthesis is important in order to understand the interrelationship of both diseases. This information will guide us to take the appropriate measures in need of a screening and the early treatment that will allow optimizing the resources available.

15. ETHICAL CONSIDERATIONS

The project will be made in compliance with the Declaration of Helsinki (Annex III) of the 1964 World Medical Association 1964 and ratifications of the following assemblies (Tokyo 75, Venice 83, Hong Kong 89, Somerset West 96, Scotland 00, Seoul 08 and Fortaleza 13) on ethical principles for medical research on human beings, the RD 1090/2015, of 24 December, clinical trials, specifically that stipulated in Article 38 on good clinical practice and the Agreement related to human rights and biomedicine), made in Oviedo the 4 of April 1997 and subsequent updates.

The participating researchers in this study undertake that all clinical data collected from the study subjects are separated from the personal identification data so that the anonymity of the patient is ensured; respecting the law on Personal Data Protection (Organic Law 15/1999, 13 of December), the RD 1720/2007 of 21 of December, approving the Regulations implementing the Organic Law 15/1999, the Law 41/2002, of 14 of November (regulating the autonomy of the patient, rights and obligations regarding clinical information and documentation), as well as Law 3/2001, of 28 of May, (regulating the informed consent and medical history of the patients), the Law 3/2005, of 7 of March, modification of Law 3/2001 and the 29/2009 Decree of 5 of February , by which electronic access to medical histories is regulated.

The clinical data of patients will be collected by the researcher in the Data Collection Logbook (DCL) specific to the study. Each DCL will be coded, protecting the identity of the patient. Only the research team and health authorities, who have a duty to maintain confidentiality, have access to all the data collected for the study. Third parties may only be provided with information that is not identified. Once the study is completed, the data will be destroyed.

Sample handling will follow what is established in Law 14/2007 of 3 of July for Biomedical Research, in the RD 1301/2006, of 10 of November, establishing the

regulations for quality and safety for the donation, procurement, evaluation, processing, preservation, storage and distribution of human cells and tissues and abiding the regulations for coordination and functioning for its use on humans and in the RD 1716/2011, of 18 of November, establishing the basic requirements for authorisation and functioning of biobanks for biomedical research and treatment of biological samples of human origin, and the functioning and National Registry of Biobanks for the biomedical research is regulated.

The treatment, communication, and transfer of data will be done according to what is stated in the General Data Protection Regulation (Regulation (UE) 2016/679 of the European Parliament and the Council, of 27 of April 2016). The center, where the information is obtained, is responsible for processing the data. The data collected will only be used for the purposes of the research study described in the protocol and maintained for the time needed to achieve the study objectives and in accordance with the applicable law.

ANNEX I: ECHOCARDIOGRAPHIC STUDY

SUGGESTIVE ECHOCARDIOGRAPHIC FINDINGS:

Cardiac involvement is generally characterized by the presence of an infiltrative/restrictive myocardiopathy with conventional echocardiographic findings that have been well described, but also with new echocardiographic findings identified using echocardiographic strain techniques. The main suggestive findings are:

- Increased wall thickness ≥ 12 mm with a myocardium of a shiny appearance, refractile, granular or mottled ("sparkling"), resulting from the deposits of amyloid fibrils which are more echogenic than a normal myocardium; wall thickening VI medium > 15 mm was an independent factor associated with a worse prognosis.
- Normal or small left ventricular cavity.
- Ejection fraction of VI (FEVI) preserved or borderline > 50% (at least in the early stages of the disease; depressed in advanced stages)
- Tissue Doppler: waves S´ and E´ reduced to basal level of the septal or lateral myocardium, reflecting a depressed longitudinal and an altered relaxation of VI.
- Abnormal mitral filling pattern: from diastolic dysfunction of VI mild or moderate (transmitral pattern type I or II); and in advanced stages, a typical restrictive transmitral pattern, with a ratio E/A >2, E/E´ increased, and small wave A due to the atrial dysfunction.
- The high filling pressure of VI may gradually lead to enlargement of the left atrium (diameter > 23 mm/m2; area > 20 cm2 or maximum volume > 28 ml/m2), which is useful for the differential diagnosis and it is also an independent factor associated with worse prognosis in AL.
- Enlarged right atrium and dilated inferior vena cava, reflecting the filling pressure of the right ventricle (RV).
- Increased thickness of the interatrial septum (this variable is much easier to measure in cardiac magnetic resonance (MRI) than in echocardiography).
- Wall thickness free of increased RV (> 7 mm), with systolic and diastolic dysfunction of RV, associated with worse survival.
- Thickening of the left and right valves, generally responsible for light insufficiencies.

- Reduced aortic ejection time (< 273 ms), described as a prognostic factor.
- Mild pericardial effusion in 50% of the cases. The presence of effusion is an independent factor associated with worse survival;
- Atrial thrombus can be found despite maintaining a sinus rhythm;

ACQUISITION PROTOCOL

.

Acquire 3 beats in sinus rhythm and 5 beats in Atrial Fibrillation.
Ensure good ECG signal.
For Doppler curves, use adjusted scales to the corresponding valve gradients, scan speed 75 mm/s.
Optimize sector width, gain, and compression to ensure maximum image quality.
Do not perform any measurement in the study to be sent to the Core Lab (only the collection of images and imaging loops)

Parasternal long axis (PLAX):
2D
Colour in mitral valve and aortic valve
2D Zoom in aortic valve
If aortic insufficiency: zoom color in the aortic valve.
Mode M in aortic valve
Modo M in VI

Parasternal short axis (PSAX).
2D
Zoom 2D in aortic valve
Colour in the aortic valve.
Zoom 2D in pulmonary valve
Colour in the pulmonary valve
Pulsed Doppler in the pulmonary valve
Colour in the pulmonary valve
Colour in tricuspid valve
Continuous Doppler in tricuspid valve
Mode M in aortic valve and in VI (if PLAX has not been performed)

Parasternal in Right Cavities.

2D

Colour Doppler in the tricuspid valve.

Continuous Doppler in the tricuspid valve.

Apical 4C:

2D

2D with extended left Ventricle

Colour Doppler in Mitral Valve

Pulsed Doppler in Mitral Valve

Pulsed Tissue Doppler in septal and lateral Mitral annulus.

Continuous Doppler in Mitral Valve, if significant insufficiency

Zoom color with PISA scale in Mitral Valve, if significant insufficiency.

Colour Doppler in Tricuspid Valve

Continuous Doppler in Tricuspid Valve

Mode M in tricuspid annulus.

Pulsed Tissue Doppler in the tricuspid annulus.

3D: Acquisition of complete volume of the left ventricle (minimize the presence of artifacts and optimize profit adjustment)

3D: Acquisition of complete volume centered in the right ventricle.

Apical 5C

2D

Colour Doppler in aortic valve

Pulsed Doppler in TSVI

Continuous Doppler in aortic valve

APICAL 2C

2D

Colour Doppler in Mitral Valve.

Zoom color with PISA scale in the mitral valve, if significant insufficiency.

APICAL 3C

2D

Zoom in TSVI

Colour Doppler in aortic valve.

Pulsed Doppler in TSVI
Continuous Doppler in aortic valve
Colour Doppler in Mitral valve.
If significant mitral insufficiency: continuous in Mitral and zoom color with PISA scale in Mitral.

Subcostal plan

2D 4C.
Zoom VD free wall (for measuring the thickness of the right ventricle)
2D in VCI
Mode M in VCI, from basal to deep inspiration.

ACQUISITION PROTOCOL FOR ECHOCARDIOGRAPHY IMAGES FOR THE STUDY OF THE STRAIN.

Aspects of images ACQUISITION:

Configure the study to gain 3 consecutive beats in apnea. If the patient is in atrial fibrillation, acquire 5 beats, avoid extrasystole.
Use a temporary resolution /frame rate of > 50 (ideally 60-80 frames/s).

Plans for measuring of the STRAIN

Acquire imaging of left ventricle expanded in the plans apical 4C, Apical 2C, and Apical 3C, focussed on the left ventricle, ensuring a correct visualisation of the endocardial borders.
Acquire imaging of the left atrium in the plans apical 4C, apical 2C, and apical 3C, focused on the atrium, ensuring correct visualization of all the atrium borders, avoiding an auricle and pulmonary veins.
Acquire imaging centered in the right ventricle from the plan apical 4C, ensuring adequate visualization of the endocardial border of the free wall.

REFERENCES FOR ANNEX II

1. Lang RM, Badano LP, Mor-Avi V, el al. Recommendations for cardiac chamber quantification by echocardiography in adults: an update from the American

Society of Echocardiography and the European Association of Cardiovascular Imaging. *Eur Heart J Cardiovasc Imaging*. 2015; 16:233–270. doi: 10.1093/ehjci/jev014.

2. Pagourelias ED, et al. Echo Parameters for Differential Diagnosis in Cardiac Amyloidosis. Circ Cardiovasc Imaging. 2017; 10:e005588. DOI: 10.1161/CIRCIMAGING.116.005588.)

3. González-Lopez E, López-Sainz A, García-Pavia P. Diagnosis and treatment in cardiac amyloidosis by transthyretin. Progress and hope. Rev Esp Cardiol. 2017;70(11):991–1004

4. White JA, Fine NM. Recent Advances in Cardiovascular Imaging Relevant to the Management of Patients with Suspected Cardiac Amyloidosis. Curr Cardiol Rep (2016) 18: 77 DOI 10.1007/s11886-016-0752-7

5. Phelan D, et al. Relative apical sparing of longitudinal strain using two-dimensional speckle- tracking echocardiography are both sensitive and specific for the diagnosis of cardiac amyloidosis .Heart 2012; 98:1442e1448. doi:10.1136/heartjnl-2012-302353

6. Bhogal S, et al. Cardiac Amyloidosis: An Updated Review With Emphasis on Diagnosis and Future Directions .Curr Probl Cardiol 2018; 43:10–34.

7. Tendler A, Helmke S, Teruya S, Alvarez J, Maurer MS. The myocardial contraction fraction is superior to ejection fraction in predicting survival in patients with AL cardiac amyloidosis. Amyloid 2015;22:61-6

8. Rubin, Jonah et al. Myocardial Contraction Fraction by M-Mode Echocardiography Is Superior to Ejection Fraction in Predicting Mortality in Transthyretin Amyloidosis. Journal of Cardiac Failure , Volume 24 , Issue 8 , 504 - 511

ANNEX II: SCINTIGRAPHIC STUDY

BASIC INFORMATION

Amyloidosis is a heterogeneous group of diseases in which there are abnormal extracellular insoluble fibril deposits, which are given the anatomopathological name of amyloid deposits. The amyloidosis can be classified into systemic or localized ways in an organ. Some types of this disease can infiltrate the heart, although only some types of these twenty known amyloidotic proteins produce cardiac affectation. These correspond to the varieties: *senile, the secondary type AA, the primary type AL*, and some inherited ways like the one related to *transthyretin (ATTR*), among others *(AAPoA-I and Afib).* Of these, the most frequently related to the heart are the ATTR and AC-AL.

The ^{99m}Tc-DPD (^{99m}Tc acid 3,3-diphosphono-1,2-propanedicarboxylic) is a bisphosphonate conventionally used in nuclear medicine for assessing bone pathology, but it has also demonstrated utility as an important technique to distinguish between the cardiac deposits in AC-TTR versus those in AC-AL, as they pose evolution, treatment and different prognoses as in the case of amyloidosis due to genetic defect with great transcendence for the family.

The cardiac scintigraphy with ^{99m}Tc-DPD is a tool that has proven useful for differentiating between the amyloidosis related with the transthyretin and the deposit by light chains of monoclonal immunoglobulin.

PATIENT PREPARATION

Previous preparation is not necessary.

Avoid caffeine and toxins like tobacco 48 hours before the study.

RADIOPHARMACEUTICALS, DOSAGE, AND ADMINISTRATION

Compound: The radiopharmaceutical used is the *^{99m}Tc-DPD*. The *^{99m}Tc-MPD* can also be used.

Dosage:

- Adults: 20 mCi (740 Mbq).
- For children, the dosage is calculated according to the weight and the clinical guidelines in the European Society of Nuclear Medicine for the ^{99m}Tc.

Administration: intravenous.

Dosimetry: The effective dosage (E) resulting from the administration of a quantity of 700 MBq of acid 3,3-diphosphono-1,2-propanedicarboxylic labeled with technetium (99mTc) for an adult of 70 kg in weight is of 4 mSv. For an administered activity of 700 MBq, the dosage of radiation absorbed by the target organ (bone) is 44,1 mGy, and 33,6 mGy by the critical organ (bladder).

Dosage absorbed to organs (mGy/MBq): Bone 0,063; Heart 0,0012; Bladder 0,050; Kidneys 0,007; Bone marrow 0,0096, Ovaries 0,0035, testicles 0,0024 and Kidneys 0,007.

INSTRUMENTATION

Collimator: LEHR

Window: 20% centered in 140 KeV

Mode: selective cardiac images.

Matrix: 256 x 256, 1.000 Kc.

Zoom: 1,3.

IMAGES ACQUISITION

Patient supine with the head of the gamma camera focused on the area of interest (heart region).
Planar Study and selective SPECT of the cardiac area.
Projection:
Full body: front and rear
Selective cardiac areas: Anterior, 45º anterior left oblique, and lateral left.
Images acquisition:
Beginning of exploration: at 3 hours post-injection of the tracer.
Study duration: - Accumulate 1000 kcts./ previous and subsequent projection.
- Accumulate 250 kcts./oblique projection.

INTERPRETATION

During the examination, on the one hand, the existence and intensity of the deposit in the myocardium will be assessed, and on the other hand, its distribution. Furthermore, the relation in the distribution of the radiotracer among the bone structures and soft parts as an indication of its proper clearance and image quality.

The intensity of the deposit is graduated according to a visual rating scale of 0 to 3, in which the absence of capture is valued with a score of 0; the capture lower than bone (referred to as the adjacent rib), with: 1; similar to bone: 2 and higher capture than bone: 3.

The distribution of the deposit in the myocardium is assessed as focal deposit, diffuse deposit, deposit in a segment of the ventricular wall, diffuse ventricular deposit or diffuse biventricular deposit. The captured relation between the bone and the

background when this is very similar is graded 0, and when the bone is more or much more highlighted than the background, it is graded 1.

REFERENCES TO ANNEX II

Sanjay M. Banypersad, MRCP; James C. Moon, MD, MRCP; Carol Whelan, MD, MRCP; Philip N. Hawkins, PhD, FmedSci; Ashutosh D. Wechalekar, DM, MRCP, FRCPath. Updates in Cardiac Amyloidosis: A Review. December 20, 2016.

Suzane Garcia Ferreira a,b,∗, Alexandre Marins Rocha a, Osvaldo José Moreira do Nascimento a, Claudio Tinoco Mesquita. Role of 99mTc-DPD scintigraphy on discrimination of familial cardiac amyloidosis. November 2015.

Brett W. Sperry, MD; Michael N. Vranian, MD; Rory Hachamovitch, MD; Hariom Joshi, MD; Asad Ikram, MBBS; Dermot Phelan, MD; Mazen Hanna, MD. Subtype-Specific Interactions and Prognosis in Cardiac Amyloidosis. December, 2016.

Enrica Perugini, MD,* Pier Luigi Guidalotti, MD,† Fabrizio Salvi, MD,‡ Robin M. T. Cooke, MA,*Cinzia Pettinato, MD,† Letizia Riva, MD. Noninvasive Etiologic Diagnosis of Cardiac Amyloidosis Using 99mTc-3,3-Diphosphono-1,2-Propanodicarboxylic Acid Scintigraphy. Journal of American College of Cardiology 2005.

Bjo¨ rn Pilebroa, Ole B. Suhrb, Ulf Na¨slunda, Per Westermarkc, Per Lindqvistd, and Torbjo¨ rn Sundstro¨me. 99mTc-DPD uptake reflects amyloid fibril composition in hereditary transthyretin amyloidosis. UPSALA JOURNAL OF MEDICAL SCIENCES, 2016.

Wael A. AlJaroudi, MD, FACC, FAHA, FESC, FASE, FASNC,a Milind Y. Desai, MD,b, W. H. Wilson Tang, MD,b Dermot Phelan, MB, BCH, PhD,b Manuel D. Cerqueira, MD,b and Wael A. Jaber, MDb. Role of imaging in the diagnosis and management of patients with cardiac amyloidosis: State of the art review and focus on emerging nuclear techniques. 2013 American Society of Nuclear Cardiology.

F. Javier de Haro-del Moral, Alicia Sánchez-Lajusticia. Role of cardiac scintigraphy with 99Tc-DPD in the discrimination of the cardiac amyloidosis subtype. Rev Esp Cardiol. 2012; 65 (5); 440-446.

J. Mallol. Radiopharmaceutical Manual. Iberoamerican Academy of Pharmacy; M.5.797- 2008.

ANNEX III: DECLARATION OF HELSINKI

Initially submitted in 1964
Original: English

DECLARATION OF HELSINKI IN THE WORLD MEDICAL ASSOCIATION
Ethical principles for medical research in human beings

Adopted by the 18th General Assembly in the World Medical Association (WMA) Helsinki,Finland, June 1964 and amended by the
29th General Assembly of the WMA, Tokyo, Japan, October 1975
35th General Assembly of the WMA, Venice, Italy, October 1983
41st General Assembly of the WMA, Hong Kong, September 1989
48th General Assembly of the WMA, Somerset West, South Africa, October 1996
and the 52nd General Assembly of the WMA, Edinburgh, Scotland, October 2000 and
Explanatory note to paragraph 29, added by the General Assembly of the WMA, Washington 2002
Explanatory note to paragraph 30, added by the General Assembly of the WMA, Tokyo 2004

A. INTRODUCTION
1. The World Medical Association has passed the Declaration of Helsinki as a statement of ethical principles to provide guidance to doctors and other people who carry out medical research in humans. Medical research in humans includes research on human material or identifiable information.
2. The duty of the doctor is to promote and safeguard the health of people. The knowledge and the awareness of the doctor are dedicated to the fulfillment of the duty.
3. The Declaration of Geneva in the World Medical Association binds the doctor with the words "the health of my patient will be my first consideration", and the International Code of Medical Ethics declares: "The doctor must act only in the interest of the participant when providing medical care which might have the effect of weakening the physical and mental condition of the participant".
4. Medical progress is based on research, which ultimately must rest in part on experimentation on humans.
5. Medical research on human beings, concern for the welfare of human beings must take precedence over the interests of science and society.
6. The main purpose of medical research involving humans is to improve preventive, diagnoses and therapeutic procedures, and also the understanding of the etiology and pathogenesis of the diseases. Even the best proven preventive, diagnoses and therapeutic methods must continuously be challenged through research to be efficient, effective, accessible and good quality.

7. In the present practice of medicine and medical research, most of the preventive, diagnoses and therapeutic procedures involve risks and costs.
8. Medical research is subject to ethical standards that promote respect to all human beings and protect their health and individual rights. Some populations subject to the research are vulnerable and need special protection. The particular needs must be recognised of those that have economic or medical disadvantages. Attention must also be paid to those who cannot give or refuse consent for themselves, those who can give consent under pressure, those who will not personally benefit from the research and those that have the research combined with medical care.
9. Researchers must know the ethical, lawful and legal requirements for the research on human beings in their own countries as well as the applicable international requirements. There is no allowance for an ethical or legal requirement to reduce or eliminate any measure of protection for human beings set in this Declaration.

B. BASIC PRINCIPLES FOR ALL MEDICAL RESEARCH
10. In a medical investigation it is the duty of the doctor to protect the life, health, privacy and dignity of the human being.
11. The medical research in human beings must conform to the generally accepted scientific principles and must be based on thorough knowledge of scientific literature, other relevant sources of information, as well as laboratory experiments correctly carried out, and on animals, when appropriate.
12. When researching, adequate attention must be paid to the factors that may harm the environment. The welfare of the animals used in the experiments must also be looked after.
13. The project and method of all experimental procedure in human beings must be clearly formulated in an experimental protocol. This must be submitted for consideration, comment, advice, and when appropriate, approval, to an ethical assessment committee specially appointed, that must be independent of the researcher, promoter or any other kind of undue influence. It is understood that this independent committee should be in accordance with the applicable laws and regulations in the country where the experimental research takes place. The committee has the right to control ongoing trials. The researcher has the obligation to provide monitoring information to the committee, especially any serious adverse events. The researcher must also submit to the committee the information on funding, sponsorships, institutional affiliations, other possible conflicts of interest and incentives for the people in the study.
14. The Project and method of all experimental procedures in human beings must be clearly formulated in an experimental protocol. This must be sent, for consideration, comment, advice, and when appropriate, approval, to a specially assigned committee of ethical assessment that must be independent of the researcher, promoter or any other kind of undue influence. It is understood that this independent committee should be in accordance with the applicable laws and regulations in the country where the experimental research takes place. The committee has the right to control ongoing trails. The researcher has an obligation to provide monitoring information to the committee, especially any serious adverse events. The researcher should also submit

to the committee the information on funding, sponsorships, institutional affiliations, other possible conflicts of interest and incentives for the people in the study.

15. The research protocol should always make reference to the ethical considerations that belong to the case and must indicate that the enunciated principles in this Declaration have been complied with.
16. The medical investigation in human beings must be carried out only by qualified scientific people and under the supervision of a clinically competent doctor. The responsibility of human beings must always rest with a person with medical training and never on the participants of the research, even though they have given consent.
17. Every medical research project in human beings must be preceded by a careful comparison of the expected risks with the foreseeable benefits for the individual or others. This does not preclude the participation of healthy volunteers in medical research. The design of all studies must be available to the public.
18. Doctors must abstain from engaging in research projects on human beings unless they are confident that the risks involved have been adequately assessed and that it is possible to face them in a satisfactory manner. The experiment taking place must be suspended if the risks involved are more important than the expected benefits and if there is conclusive proof of positive or beneficial results.
19. The medical research on human beings must only be carried out if the importance of the objective is greater than the inherent risk and the costs to the individual. This is especially important when humans are healthy volunteers.
20. The medical research is only justified if there is a reasonable likelihood that the population on which the research is done, may benefit from the results.
21. To take part in a research project, the individuals must be voluntary and informed participants.
22. Always respect the right of participants in the research to protect their integrity. Every caution must be taken to protect the privacy of individuals, the confidentiality of the information on the participant and to minimise the consequences of the research on their physical and mental integrity and personality.
23. In any research on human beings, each potential individual must receive adequate information about the objectives, methods, financial sources, possible conflicts of interest, institutional affiliations of the researcher, expected benefits, foreseeable risks and discomforts from the experiment. The person must be informed of the right to participate or not in the research and to withdraw their consent at any time without reprisal. After ensuring that the individual has understood the information, the doctor must then obtain, preferably in writing, the informed and voluntary consent of the person. If the consent cannot be obtained in writing, the process to obtain it must be formally documented and witnessed.
24. When obtaining informed consent for the research project, the doctor must be particularly cautious when the individual is linked with them due to dependency or providing consent under pressure. In such a case, the informed consent must be obtained by a well informed doctor who does not participate in the research and is independent from this relationship.

25. When a person is deemed legally incompetent, physically or mentally unfit to obtain an informed consent or a minor, the researcher must obtain the informed consent from the legal representative and in accordance with the applicable law. These groups must not be included in the research unless it is necessary to promote the health of the population represented and this research cannot be carried out on legally competent people.
26. If a person is deemed legally incompetent, as is the case of a minor, they are able to give their assent to participate or not in the research, the researcher must obtain it apart from the consent from the legal representative.
27. Research on individuals from whom consent cannot be obtained even by representative or in advance, must only be done if the physical/mental condition that prevents obtaining the informed consent is a necessary characteristic of the population being researched. The specific reasons why research participants are used which cannot provide their informed consent must be stated in the experimental protocol for consideration and approval by the assessment committee. The protocol must state that the consent of the individual or legal representative to remain in the research must be obtained as soon as possible.
28. Both authors and publishers have ethical obligations. When publishing the results of their research, the doctor is obliged to maintain the accuracy of the data and results. Both negative and positive results must be published or otherwise available to the public. The publication must cite the source of funding, institutional affiliations and any possible conflicts of interest. Reports on research that do not adhere to the principles described in the Declaration must not be accepted for publication.

C. APPLICABLE PRINCIPLES WHEN MEDICAL RESEARCH IS COMBINED WITH MEDICAL CARE

29. The doctor may combine medical research with medical care only to the extent that such research is justified by its preventive, diagnosis and therapeutic potential. When medical research is combined with medical care, the additional standards apply to protect the participants participating in the research.
30. The possible benefits, risks, costs and effectiveness of new procedures must be assessed by its comparison with the best preventive, diagnoses and therapeutic methods available. This does not exclude a placebo, or any treatment, in studies where preventive, diagnoses and therapeutic procedures tested are not available.(*See explanatory note*).*
31. At the end of the research, all the participants that participate in the study must be assured of the best preventive, diagnoses and therapeutic methods available, identified by the study (*See explanatory note*)*.
32. The doctor should fully inform the participants about the aspects of care related to the study. The refusal from the participant to participate in a research must never interfere with the doctor-participant relationship.
33. When the preventive, diagnoses and therapeutic methods do not exist or result ineffective in the attention of a patient, the doctor, with the informed consent of the participant, may use preventive, diagnoses and therapeutic procedures which are new or not tested, if, in their opinion, this offers hope of saving life, restoring health or

alleviating the suffering. Whenever possible, those measures must be researched to evaluate its safety and effectiveness. In all cases, that new information must be registered and, when appropriate, published. All the relevant guidelines of this Declaration must be followed.

***Explanatory note to paragraph 29 in the declaration of Helsinki in the WMA**
The WMA reaffirms that there must be great care when using trials with placebo, and in general, this methodology can only be applied if not counting on a tested and existing therapy. However, the trials with placebo are ethically acceptable in some cases, even if there is a tested therapy and the following conditions are met:
-when for methodological, scientific and compelling reasons, its use is necessary to determine the effectiveness and safety of a preventive, diagnosis and therapeutic method; or
- when a preventive, diagnosis or therapeutic method is tested for a disease of less importance which does not imply an additional risk, serious adverse effects or irreversible harm for the participants who receive the placebo.
All the other provisions of the Declaration of Helsinki, especially the need for appropriate ethical and scientific reviews must be followed.

***Explanatory note to paragraph 30 in the declaration of Helsinki in the WMA**
Hereby, the WMA reaffirms its position that it is necessary, during the process of the study plan, to identify the access after the testing of the participants in the study to preventive, diagnoses and therapeutic procedures that have resulted beneficial in the study or the access to another appropriate attention. The provisions for the access after testing or other attention must be described in the protocol of the study so that the ethical review committee can consider such arrangements during its review.

9.10.2004
The declaration de Helsinki (Document 17.C) is an official document of the World Medical Association, representing doctors in the world. Adapted in 1964 in Helsinki (Finland), was revised in 1975 in Tokyo (Japan), in 1983 in Venice (Italy), in 1989 in Hong Kong, in 1996 in Somerset West (South Africa), in 2000 in Edinburgh (Scotland), by the General Assembly of the WMA, Washington 2002 (adding an explanatory note for paragraph 29) and by the General Assembly of the WMA, Tokyo 2004 (adding an explanatory note for paragraph 30).