"PRECISION AND PERSONALIZED MEDICINE": UN SOGNO CHE DIVENTA REALTÀ?

"PRECISION AND PERSONALIZED MEDICINE": A DREAM THAT COMES TRUE?

V. Favalli, A. Serio, L. Giuliani, M. Tagliani, E. Disabella, A. Pilotto, M. Digiovannantonio, M. Pasotti, M. Grasso, E. Arbustini*

Centre for Inherited Cardiovascular Diseases.
IRCCS Foundation University Hospital Policlinico San Matteo, Pavia.
*** IRCCS Fondazione Salvatore Maugeri, Montescano, Pavia.**

With the completion of human sequencing and availability of new biotechnology tools, molecular characterization of diseases at individual level is feasible and sustainable. In this context, "Precision medicine" aims at identifying precise etiology or target for each disease in order to develop disease-specific treatments, tailored on individual biomolecular profiles (Personalized medicine). The two terms, precision and personalized medicine, are often used interchangeably or combined [1,2].

The perception of the impact of precision and personalized medicine is easy for monogenic (both single disease gene and genetically heterogeneous diseases) and monofactorial diseases [3]. A typical example of monogenic diseases (one gene → one disease) is cystic fibrosis: new therapies modulating defective CFTR gene are entering the clinic [4]. In monogenic cardiovascular diseases (several genes are associated with one disease), precise diagnosis is largely dependent on genetic diagnosis (i.e. pure restrictive cardiomyopathy caused by defects in troponins-coding genes or desmin-coding gene). However, the burden of cardiovascular diseases is largely constituted of multifactorial, acquired diseases such as Heart Failure (HF), Ischemic Heart Disease (IHD), arrhythmias such as Atrial Fibrillation (AF), Valve Heart Diseases (VHD) or hypertension. For these large groups of disorders, current innovation is not related to their cure but results from novel strategies of risk assessment, incorporation in the clinical work-up of new diagnostics (endovascular diagnostics, non-invasive diagnostic of coronary atherosclerosis) and therapeutics (the development of percutaneous interventional treatments for acute coronary syndromes valve diseases, congenital heart diseases, etc.) up to resuscitation programs and artificial hearts. Personalized treatment guided by pharmacogenetics is one of the applications of precision-personalized medicine used in clinical practice (tab. I) [5].

Tabella I - The table lists genes and variants tested in clinical practice for dugs responsiveness, dose adjustments, other than side effects (statins), thrombogenic risk (clopidogrel), hemorrhagic risk (warfarin).

*Mim**	*Gene*	*Variant*
STATINS		
107741	APOE	rs7412 rs429358
142910	HMGCR	rs17244841 rs17238540 rs3846662
604843	SLCO1B1	rs149056
CLOPIDOGREL		
124020	CYP2C19	*2 (rs4244285) *17(rs3758581)
171050	ABCB1	C1236T (rs1128503) G2677T (rs2032582) C3435T (rs1045642)
600515	P2RY12	rs6798347 rs6787801 rs9859552 rs6801273 rs9848789 rs2046934
WARFARIN		
124020	CYP2C9	*2 (rs1799853) *3 (rs1057910)
604426	CYP4F2	rs2108622
608547	VKORC1	-1639 (rs9923231)

Multifactorial cardiovascular diseases

Current classifications of cardiovascular diseases are based on the principle of similarities of either pathologic bases or phenotypes: many cardiovascular diseases look alike and share similar pathologic background but their causes may differ. They often represent the end-phenotypes of different diseases: HF syndrome is the paradigmatic example. Almost all cardiac diseases may end in HF: systolic HF is the end-phenotype of diseases such as genetic cardiomyopathies in children or ischemic heart disease in elderly. Diastolic heart failure occurs in genetic cardiomyopathies such as troponinopathies or desminopathies as well as in hypertensive patients. Similar to HF, the diagnosis of IHD describes all diseases sharing defects of myocardial perfusion. The common pathologic bases are well recognized: coronary atherosclerosis and related complications are the primary cause. However, two identical coronary plaques in two different patients do not exist. The morphology of a same plaque evolves, influenced by ageing, risk factors, comorbilities and treatments. Serial in vivo imaging of coronary arteries can easily demonstrate the evolving patterns of coronary plaques [6,7]. Molecular imaging in the future will further add precise information on fine identification of cellular and extracellular pla-

que components [8]. Risk stratification including classical risk factors and their role in the development of atherosclerosis are major achievements of the basic and clinical research of the last fifty years [9]. However, the molecular bases of modifiable risk factors are only partly elucidated [10]; current treatments control the effect (cholesterol levels, blood pressure, diabetes, increased LDL) but often do not cure the primary causes. Precise innovation is emerging for phenotypically and genetically characterized diseases such as autosomal dominant familial hypercholesterolemia (FH, MIM #143.890) caused by mutations in LDLR gene (MIM *606.945): novel molecules such as microsomal triglyceride transfer protein inhibitors (lomitapide), oligonucleotide inhibitors of apoB-100 synthesis (mipomersen), and "proprotein convertase subtilisin/kexin type 9" (PCSK9) inhibitors (alirocumab and evolocumab) are entering FH management [11].

Monogenic cardiovascular diseases

For less common cardiovascular diseases such as genetic channelopathies [12], heritable cardiomyopathies [13], familial aortopathies [14], syndromes with heart involvement such as lysosome storage diseases (i.e. Pompe disease, Anderson-Fabry Disease) [15,16], or cardiac amyloidosis [17], the process of "precision diagnostic and therapeutic" is up and running. In each group of diseases, the level of advancement varies; precise diagnoses are feasible for an increasing number of genetic arrhythmias, inherited cardiomyopathies, genetic aneurysmal diseases and complex syndromes involving heart. For a few of these latter precise diagnoses now meet disease-specific treatments (i.e. Anderson Fabry disease, Pompe disease) [15,16] or novel strategies of treatment based on genetic diagnosis (i.e. Marfan Syndrome and drugs controlling TGFbeta) [18] or definite targets (i.e. amyloidogenic protein in cardiac amyloidosis) [19]. These few examples better describe the concept of precision medicine as finalized to treatments targeted to the needs of individual patients on the basis of disease-specific genotype or biomarkers, distinguishing each patient from all others with similar clinical presentations. The aims include improvement clinical outcomes and prevention of medical errors and side effects, in the ideal scenario of personalized medicine.

A dizzying acceleration in translation of precision medicine programs was given by key discoveries and completion of programs such as Human Genome project in 2013, and the commercial availability of high throughput and sensitivity technologies able to translate achievements of "omic" research in genetic testing, transcriptomic, proteomic and metabolomic assays for affected tissues or peripheral blood samples, and further development of research to detect novel biomarkers, both diagnostic or target or explore epigenetic mechanisms contributing to phenotypes. When comparing the achievements of oncology sciences with those of CV diseases, the concept of precision and personalized medicine becomes more and more clear. A paradigmatic example is the progression of knowledge in constitutive oncogenetics for breast and ovarian cancer: testing for BRCA1 and BRCA2 genes identifies women that are predisposed to develop cancer (MIM #604.370, #612.555). Mutations in these genes cause familial Breast-Ovarian Cancer (BROVCA). Unaffected women

testing positive for mutations in one of the two genes can undergo preventive surgery. More recently, the first PARP-inhibitor, Olaparib, targeting the nuclear enzyme poly (ADP-ribose) polymerase (PARP) discovered in 1963 [20], was approved and licensed in 2015 for the treatment of BRCA-mutated recurrent platinum-sensitive ovarian cancer [21]. For other types of cancer, somatic mutations in cancer cells now drive anti-cancer treatments [22].

Examples of multifactorial and monogenic cardiovascular diseases and precision diagnosis

IHD

About 50 years ago the first definition of factors of risk in Framingham study was, by itself, a revolution. The control of risk factors, the development of drugs such as ACEI, beta-blockers, aspirin, statins, and primary coronary intervention changed the scenarios of risk and events. Looking back at first Framingham, the advancements of knowledge and impact of related clinical achievements in multifactorial diseases such as IHD or HF are gigantic [9]. Individual risk assessment and prevention may further modify the future epidemiology. Family history is a proven non-modifiable risk factor. A few years ago, we attempted to answer the question increasingly raised by relatives of patients with AMI: "My parents died of myocardial infarction: is that my destiny?" [10]. Beyond technicalities and complex molecular genetic data, the manuscript highlighted a few key concepts that remain solid and clinically useful (tab. II).

For the future, the generation of individual "multigene score" of risk could contribute to precise and personalized risk assessment for IHD as well as for other multifactorial diseases. Genetic architecture is one of the keys: it gives the tool for addressing specific questions in different individuals and makes interpretable, at the molecular genetic level, consolidated evidence of family-related risk. As age and gender, genetic risk is un-modifiable; viceversa, other markers such as gene expression are dynamic and potentially modifiable once non-genetic factors influencing the gene expression itself are known. Gene expression signature can inform about the spectrum of coronary artery disease of responsiveness to antiplatelet drugs and contribute to predict and re-asses the risk of acute coronary syndromes [23,24].

Atrial and ventricular arrhythmias

Atrial Fibrillation (AF) affects 1.5% population in Europe and worldwide and shows familial in up to 30% of cases [25]. Heterogeneous, multifactorial etiology of Atrial Fibrillation (AF) includes heritability (fig. 1) [26]. Several disease genes have been identified to date [26] but less than 1% of patients with AF receive genetic advice or are genetically tested. Screening programs in familial AF (ATFB in OMIM catalogue) may be less informative than those run for familial cardiomyopathies: in fact AF paroxysms can be clinically silent and unrecognized; unless implanting an ECG recording system, clinically silent episodes of AF can be missed. Nonetheless, familial ATFB is progressing through a precise molecular classification. As far as the causes are identified, an ordered number of categories or groups of ATFB fills the existing gaps

Table II - AMI and genetics. Modified from: Med Clin North Am 2012; 96:67-86. "My parents died of myocardial infarction: is that my destiny?" (ref. [10]).

The table summarizes major concepts on heritability of risk factors and acute coronary syndromes.

1. Although most risk factors for coronary ATheroSclerosis (ATS) (the rule) are also risk factors for Myocardial Infarction (MI) (the exception), MI-specific risk is additional to or independent from ATS risk. In addition to classical risk factors, Genome-Wide Association Studies identified chromosomal loci with candidate genes that do not code for known risk factors, and thereby open new frontiers of research for MI-specific risk markers. Endo-phenotypes, such as plasma levels or activity of gene products, could be more closely associated with genetic variations than is the eventual end phenotype (MI), which is a cumulative multifactorial event.
2. Because a positive family history for ATS risk factors does not fully coincide with a positive family history for MI, the risk stratification for MI should include family history data on number, age, and gender of affected family members, modifiable and non-modifiable known risk factors, and unpredictable triggers.
3. Heritable factors playing a role in the coagulation, inflammatory, and adrenergic pathways, as well as associated epistatic and gene-environment interactions may be MI-specific contributors, but their individual role in MI cannot currently be translated, due to multilayered influences (by both genes and environment) on each factor.
4. Genetic counseling may contribute to implement family-tailored preventive strategies, taking into consideration the patient's clinical history, family history, and lifestyle. While waiting for genetic tests that add predictive contribution to the calculation of the genetic risk, cardiologists can incorporate family history and clinical data to provide the best individualized monitoring and preventive programs.
5. Members from families with more than one affected relatives who have experienced MI:
   - are exposed to higher risk than individuals with negative family history;
   - should be reassured that their destiny is not predefined solely by the genetic ground;
   - should undergo clinical monitoring of their health state.

between phenotype (are all AF alike?) and causes. Sick Sinus Syndrome (SSS) may also cluster in families, either as autosomal dominant or recessive disease [27,28]. Rare autosomal recessive Atrial Dilated atrial CardioMyopathies (ADCM), are now recognized as entities with possible precise genetic causes [29]. A precise diagnosis of the cause of AF or of SSS or ADCM may influence treatments, such as ablation, antiarrhythmic drugs and anticoagulation. By itself, the diagnosis of AF describes a rhythm disorder and not the underlying disease. We treat the arrhythmias but we do not cure the disease. Ablation can be successful in large series of cases, but not in all cases: when the AF has a precise underlying "atrial" disease that is unknown and not cured, the arrhythmias will recur: we cannot predict when, but this is what we are learning from anatomical electro-mapping atrial studies that are demonstrating in vivo that atrial fibrosis is a common substrate for AF, SSS, AS [27-30]. A first personalized advantage from precision diagnosis is the contribution to clinical decisions, both ablation and drugs.

In the complex group ventricular arrhythmias, examples of precision diagnosis and specific treatments include LQT Syndrome, Brugada Syndrome, or

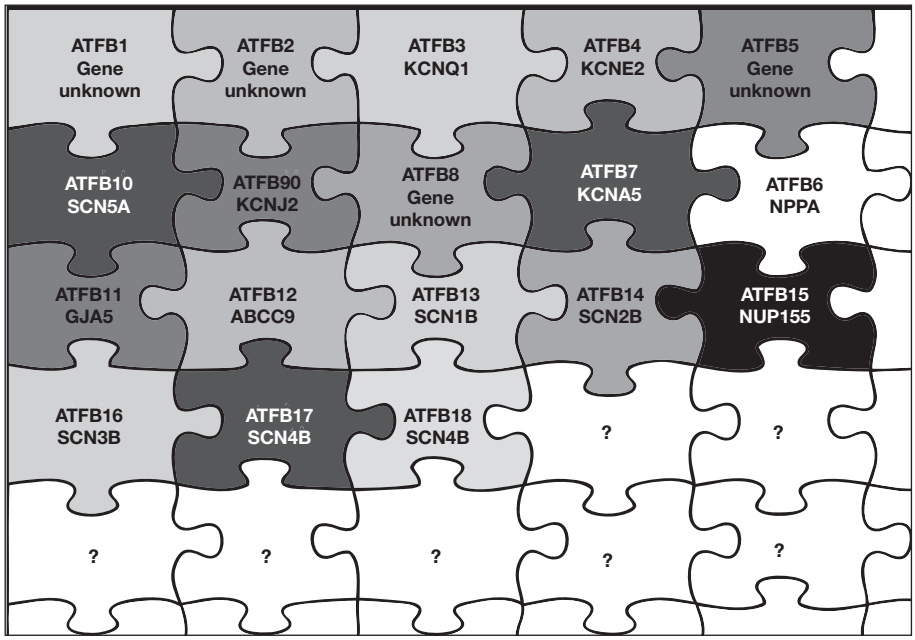

Fig. 1. The puzzle shows genes associated with atrial fibrillation (ATFB in OMIM Catalogue); different shades of grey indicate that although the phenotype is the same, the genetic bases are heterogeneous. For some of the disease genes more diseases (both autosomal dominant and recessive) are allelic at the same locus. The paradigmatic example is SCN5A gene: allelic disorders include Brugada syndrome 1 (autosomal dominant, MIM #601144), DCM (autosomal dominant, MIM #601154), non-progressive heart block (autosomal dominant, MIM #113900), progressive heart block IA (autosomal dominant, MIM #113900), LQT syndrome-3 (autosomal dominant, MIM #603830), Sick Sinus Syndrome 1 (autosomal recessive, MIM #608567), Familial Ventricular Fibrillation 1 (autosomal dominant, MIM #603829), susceptibility to Sudden Infant Death (SID) syndrome (autosomal recessive, #272120). Additional genes are expected to be identified (? cells).

Short QT Syndrome [12]. Several disease genes are now systematically analyzed in patients diagnosed with LQT syndrome. Genotype is one of the factors included in the assessment of risk stratification, along with gender and baseline QTc duration. Corrected QT interval (QTc) in males diagnosed with LQT3 (SCN5A gene), or females diagnosed with LQT2 (KCNH2) carry the highest arrhythmogenic risk: guidelines advise aggressive treatments [31]. Additional risk is associated with the protein domain of the mutated residues. However, the genetic diagnosis may not coincide with clinical diagnosis as shown in families in which healthy parents are carriers of the same mutation of the proband. A second mutation inherited from the non-carrier parents may contribute to explain the absence of clinical manifestations in mutated members of same families. Modifier genes (i.e. variants in NOS1AP modulate risk of events in LQTS) [32] or common SNPs in LQT genes influencing the length of the QT interval are now matter of investigation in order to demonstrate a possible dose effect of independent mutations/variants/SNPs. Therefore, precise genetic diagnosis is now a major clinical contributor in the overall managements of patients and families with LQTS. Same considerations apply to other heritable

channelopathies [33,34]. Beyond achievements for monitoring, preventing and treating ventricular arrhythmias, genetic predisposition can also contribute to the stratification of arrhythmogenic risk in multifactorial diseases.

CardioMyoPathies (CMP)

CMPs, defined as primary diseases of the heart muscle, are a further example of how precision medicine is entering our clinical practice. The genetic origin of the majority of CMPs, hypertrophic, dilated restrictive, arrhythmogenic (right, left and biventricular) is now consolidated [35]. Major diagnostic complications are the extreme genetic heterogeneity (more than 100 disease genes identified to date), the high probability that new fast high throughput tools for genetic testing identify more mutations in a same patient, the lack of functional tests able to provide (in the same short interval now needed to get genetic data), information supporting the role of the mutation/s in each given patient/families. In this context, genotyping is now part of diagnostic work-up: the challenge is interpretation of results [13,36]. The correct interpretation of the role of mutations identified in each single patient and family largely relies on segregation studies in families [36]. Precision in this context starts with deep phenotyping in clinical family screening that provides the basis for segregation analysis. Genetic test in probands and relatives can go in parallel, but they are two independent steps of the same genetic work-up. The impact of precise diagnoses varies for the different cardiomyopathies: risk stratification, such as in dilated cardiolaminopathes [37], disease-specific treatment such as lysosomal storage diseases involving the heart [15,16], prevention of treatment errors in mitochondrial diseases [38], timely indications to heart transplantation in dilated cardiodystrophinopathies [39], identification of high arrhythmogenic risk in troponinopathies with mild left ventricular hypertrophy [40], management of gastrointestinal disturbances in patients with restrictive desminopathies [35,41,42], are all example of how a precise diagnosis may condition the personalized managements of patients. Obviously, the ideal scenario should see the development of disease-specific drugs.

Phenocopies mimicking the major CMP morpho-functional phenotypes should be now matter of a precise distinction and separation to move up from description to causes and treatments. The maintenance of confusing nosology is one of the major limits in progression of R&D of disease-specific treatments. This is the reason why precise nosology, the MOGE(S) system, had been proposed [35,42]. Unexpectedly, recent studies suggest that MOGE(S) system can contribute to risk stratification in CMPs with more than one etiology contributors [43].

Heritable aneurysmal diseases

More than 30 different heritable diseases include aortic aneurysm and dissection in their phenotypical spectrum [14,44]. Although in our models of care the clinical work-up for aortic aneurysm is fragmented (cardiologists, cardiac surgeons, vascular surgeons), cardiologists are directly involved both in the study of cardiac valves and in the evaluation of aortic root aneurysm, as well as measurements of the aortic diameters at the level of the arch, thoracic descending aorta, and abdominal aorta. Several heritable aneurysmal diseases are in

the clinical context of syndromes that are usually diagnosed independently of the aortic disease (ie. Marfan Syndrome, Loeys-Dietz Syndromes, Ehlers-Danlos type IV, Shprintzen-Goldberg craniosynostosis Syndrome). However, some of the known familial diseases uniquely or predominantly involve the aorta and occur in patients and families in which additional phenotypical traits can be either absent (i.e. MYH11, or ACTA2) or identified only when specifically investigated and known as associated with aortic disease (i.e. iris flocculi) [45].

The different genetic diseases carry a different risk of dissection, which is assessed both on the basis of anatomic data (aortic diameter) and type of genetic disease [46]. Syndromes caused by defects in genes playing in the TGF-beta pathway (i.e. FBN1) have been proposed as candidate to treatments with agents blocking TGF-beta [47]. Ten randomized, controlled trials are exploring the effects of angiotensin receptor antagonists (ARB) in Marfan Syndrome, both in combination with β-blockers [48] and monotherapy [49]. A jointed international initiative (Marfan Treatment Trialists' Collaboration) will generate a prospective, collaborative meta-analysis based on data from all trials that are randomizing patients treated with (a) ARBs versus placebo (or open-label control) and (b) ARBs versus β-blockers [18]. Regardless of the results, the major message is that precise diagnoses and novel treatments by themselves increase the quality of care, level of attention and progression of research.

Conclusions

The implementation of precision and personalized cardiology relies on deep phenotyping programs of diseases sharing causes, or resulting from defects of genes playing in same pathways, or expressing disease-specific markers possible target of new treatments. Omic data are expected to play a role in precise diagnosis and novel assessment of the risk for cardiovascular diseases because genetic factors are either cause or contributors to most of them. As such, precision and personalized medicine is now matter of families rather than of single patients. This evidence calls for novel strategies of health management, introducing patient/family-centered models of care. The challenges for future developments are the clinical interpretation of existing and novel omic data and clinically-based governance of technologies.

Acknowledgements: research on precision medicine for cardiomyopathies, atrial fibrillation, pharmacogenetics, and heritable aneurysmal diseases have been and are supported by grants: EU 241924 INHERITANCE; CARITRO 2009-0569; RC IRCCS San Matteo "Chronic Thromboembolic Pulmonary Hypertension"; Ricerca Finalizzata GR-2009-1608713.

REFERENCES

1) *Blaus A, Madabushi R, Pacanowski M, Rose M, Schuck RN, Stockbridge N, Temple R, Unger EF*. Personalized Cardiovascular Medicine Today: A Food and Drug Administration/Center for Drug Evaluation and Research Perspective. Circulation 2015; 132:1425-32

2) *Kirchhof P, Sipido KR, Cowie MR, Eschenhagen T, Fox KA, Katus H, Schroeder S, Schunkert H, Priori S; ESC CRT R&D and European Affairs Work Shop on Personalized Medicine*. The continuum of personalized cardiovascular medicine: a position paper of the European Society of Cardiology. Eur Heart J 2014; 35:3250-7
3) *Delude CM*. Deep phenotyping: The details of disease. Nature 2015; 527:S14-5
4) *Amaral MD*. Novel personalized therapies for cystic fibrosis: treating the basic defect in all patients. J Intern Med 2015; 277:155-66
5) *Stankov KM, Stanimirov BG, Mikov MM*. Pharmacogenomics determinants of response to cardiovascular drugs. Med Pregl 2015; 68:259-65
6) *Liu LC, Voors AA, Valente MA, van der Meer P*. A novel approach to drug development in heart failure: towards personalized medicine. Can J Cardiol 2014; 30:288-95
7) *Souteyrand G, Arbustini E, Motreff P, Gatto L, Di Vito L, Marco V, Amabile N, Chisari A, Kodama T, Romagnoli E, Tavazzi L, Crea F, Narula J, Prati F*. Serial optical coherence tomography imaging of ACS-causing culprit plaques. Euro Intervention 2015; 11:319-24
8) *Wang T, McElroy A, Halaney D, Vela D, Fung E, Hossain S, Phipps J, Wang B, Yin B, Feldman MD, Milner TE*. Dual-modality fiber-based OCT-TPL imaging system for simultaneous microstructural and molecular analysis of atherosclerotic plaques. Biomed Opt Express 2015; 6:1665-78
9) *Tsao CW, Vasan RS*. The Framingham Heart Study: past, present and future. Int J Epidemiol 2015; 44:1763-6
10) *Narula N, Rapezzi C, Tavazzi L, Arbustini E*. "My parents died of myocardial infarction: is that my destiny?". Med Clin North Am 2012; 96:67-86
11) *Patel RS, Scopelliti EM, Savelloni J*. Therapeutic Management of Familial Hypercholesterolemia: Current and Emerging Drug Therapies. Pharmacotherapy 2015; 35:1189-203
12) *Campuzano O, Sarquella-Brugada G, Brugada R, Brugada J*. Genetics of channelopathies associated with sudden cardiac death. Glob Cardiol Sci Pract 2015; 2015:39
13) *Towbin JA*. Inherited cardiomyopathies. Circ J 2014; 78:2347-56
14) *Paterick TE, Humphries JA, Ammar KA, Jan MF, Loberg R, Bush M, Khandheria BK, Tajik AJ*. Aortopathies: etiologies, genetics, differential diagnosis, prognosis and management. Am J Med 2013; 126:670-8
15) *Kim JA, Kakhlon O, Li L, Myerowitz R, Raben N*. Pompe disease: Shared and unshared features of lysosomal storage disorders. Rare Dis 2015; 3:e1068978
16) *Seydelmann N, Wanner C, Störk S, Ertl G, Weidemann F*. Fabry disease and the heart. Best Pract Res Clin Endocrinol Metab 2015; 29:195-204
17) *Bejar D, Colombo PC, Latif F, Yuzefpolskaya M*. Infiltrative Cardiomyopathies. Clin Med Insights Cardiol 2015; 9 (Suppl 2):29-38
18) *Pitcher A, Emberson J, Lacro RV, Sleeper LA, Stylianou M, Mahony L, Pearson GD, Groenink M, Mulder BJ, Zwinderman AH, De Backer J, De Paepe AM, Arbustini E, Erdem G, Jin XY, Flather MD, Mullen MJ, Child AH, Forteza A, Evangelista A, Chiu HH, Wu MH, Sandor G, Bhatt AB, Creager MA, Devereux RB, Loeys B, Forfar JC, Neubauer S, Watkins H, Boileau C, Jondeau G, Dietz HC, Baigent C*. Design and rationale of a prospective, collaborative meta-analysis of all randomized controlled trials of angiotensin receptor antagonists in Marfan syndrome, based on individual patient data: A report from the Marfan Treatment Trialists' Collaboration. Am Heart J 2015; 169:605-12
19) *Patel KS, Hawkins PN*. Cardiac amyloidosis: where are we today? J Intern Med 2015; 278:126-44
20) *Chambon P, Weill JD, Mandel P*. Nicotinamide mononucleotide activation of new DNA-dependent polyadenylic acid synthesizing nuclear enzyme. Biochem Biophys Res Commun 1963; 11:39-43

21) *Kaufman B, Shapira-Frommer R, Schmutzler RK, Audeh MW, Friedlander M, Balmaña J, Mitchell G, Fried G, Stemmer SM, Hubert A, Rosengarten O, Steiner M, Loman N, Bowen K, Fielding A, Domchek SM*. Olaparib monotherapy in patients with advanced cancer and a germ-line BRCA1/2 mutation. J Clin Oncol 2015; 33:244-250
22) *Saeidi K*. Myeloproliferative neoplasms: Current molecular biology and genetics. Crit Rev Oncol Hematol 2015 Nov 28. pii: S1040-8428(15)30074-3
23) *Friede KA, Ginsburg GS, Voora D*. Gene Expression Signatures and the Spectrum of Coronary Artery Disease. J Cardiovasc Transl Res 2015; 8:339-52
24) *Williams MS, Weiss EJ, Sabatine MS, Simon DI, Bahou WF, Becker LC, Parise LV, Dauerman HL, French PA, Smyth SS, Becker RC; 2010 Platelet Colloquium Participants*. Genetic regulation of platelet receptor expression and function: application in clinical practice and drug development. Arterioscler Thromb Vasc Biol 2010; 30:2372-84
25) *Disertori M, Mazzola S, Quintarelli S, Arbustini E*. New frontiers for an ancient disease: genetics of atrial fibrillation. G Ital Cardiol 2013; 14:46-54
26) *Tucker NR, Ellinor PT*. Emerging directions in the genetics of atrial fibrillation. Circ Res 2014; 114:1469-82
27) *Kodama T, Serio A, Disertori M, Bronzetti G, Diegoli M, Narula N, Grasso M, Mazzola S, Arbustini E*. Autosomal recessive paediatric sick sinus syndrome associated with novel compound mutations in SCN5A. Int J Cardiol 2013; 167:3078-80
28) *De Regibus V, Rordorf R, Giorgianni C, Canclini C, Vicentini A, Taravelli E, Petracci B, Savastano S, De Servi S, Arbustini E*. Autosomal recessive atrial disease presenting with Sick Sinus Syndrome (SSS), right atrial fibrosis and biatrial dilatation: clinical impact of genetic diagnosis. Int J Cardiol 2016; doi:10.1016/j.ijcard.2016.01.160
29) *Disertori M, Quintarelli S, Grasso M, Pilotto A, Narula N, Favalli V, Canclini C, Diegoli M, Mazzola S, Marini M, Del Greco M, Bonmassari R, Masè M, Ravelli F, Specchia C, Arbustini E*. Autosomal recessive atrial dilated cardiomyopathy with standstill evolution associated with mutation of Natriuretic Peptide Precursor A. Circ Cardiovasc Genet 2013; 6:27-36
30) *Kottkamp H*. Human atrial fibrillation substrate: towards a specific fibrotic atrial cardiomyopathy. Eur Heart J 2013; 34:2731-8
31) *Ackerman MJ, Priori SG, Willems S, Berul C, Brugada R, Calkins H, Camm AJ, Ellinor PT, Gollob M, Hamilton R, Hershberger RE, Judge DP, Le Marec H,McKenna WJ, Schulze-Bahr E, Semsarian C, Towbin JA, Watkins H, Wilde A, Wolpert C, Zipes DP; Heart Rhythm Society (HRS); European Heart Rhythm Association (EHRA)*. HRS/EHRA expert consensus statement on the state of genetic testing for the channelopathies and cardiomyopathies: this document was developed as a partnership between the Heart Rhythm Society (HRS) and the European Heart Rhythm Association (EHRA). Europace 2011; 13:1077-109
32) *Kolder IC, Tanck MW, Postema PG, Barc J, Sinner MF, Zumhagen S, Husemann A, Stallmeyer B, Koopmann TT, Hofman N, Pfeufer A, Lichtner P, Meitinger T, Beckmann BM, Myerburg RJ, Bishopric NH, Roden DM, Kääb S, Wilde AA, Schott JJ, Schulze-Bahr E, Bezzina CR*. Analysis for Genetic Modifiers of Disease Severity in Patients With Long-QT Syndrome Type 2. Circ Cardiovasc Genet 2015; 8:447-56
33) *Adler A*. Brugada syndrome: diagnosis, risk stratification, and management. Curr Opin Cardiol 2016; 31:37-45
34) *Spoonamore KG, Ware SM*. Genetic testing and genetic counseling in patients with sudden death risk due to heritable arrhythmias. Heart Rhythm 2015 Nov 12. pii: S1547-5271(15)01420-4
35) *Arbustini E, Narula N, Tavazzi L, Serio A, Grasso M, Favalli V, Bellazzi R, Tajik JA, Bonow RO, Fuster V, Narula J*. The MOGE(S) classification of cardiomyo-

pathy for clinicians. J Am Coll Cardiol 2014; 64:304-18
36) *Favalli V, Serio A, Grasso M, Arbustini E*. Genetic causes of dilated cardiomyopathies. Heart 2016, in press
37) *Authors/Task Force Members, Priori SG, Blomström-Lundqvist C, Mazzanti A, Blom N, Borggrefe M, Camm J, Elliott PM, Fitzsimons D, Hatala R, Hindricks G, Kirchhof P, Kjeldsen K, Kuck KH, Hernandez-Madrid A, Nikolaou N, Norekvål TM, Spaulding C, Van Veldhuisen DJ*. 2015 ESC Guidelines for the management of patients with ventricular arrhythmias and the prevention of sudden cardiac death: The Task Force for the Management of Patients with Ventricular Arrhythmias and the Prevention of Sudden Cardiac Death of the European Society of Cardiology (ESC). Endorsed by: Association for European Paediatric and Congenital Cardiology (AEPC). Europace 2015; 17:1601-87
38) *Varga ZV, Ferdinandy P, Liaudet L, Pacher P*. Drug-induced mitochondrial dysfunction and cardiotoxicity. Am J Physiol Heart Circ Physiol 2015; 309:H1453-67
39) *Diegoli M, Grasso M, Favalli V, Serio A, Gambarin FI, Klersy C, Pasotti M, Agozzino E, Scelsi L, Ferlini A, Febo O, Piccolo G, Tavazzi L, Narula J, Arbustini E*. Diagnostic work-up and risk stratification in X-linked dilated cardiomyopathies caused by dystrophin defects. J Am Coll Cardiol 2011; 58:925-34
40) *Mogensen J, Arbustini E*. Restrictive cardiomyopathy. Curr Opin Cardiol 2009; 24:214-20
41) *Walter MC, Reilich P, Huebner A, Fischer D, Schröder R, Vorgerd M, Kress W, Born C, Schoser BG, Krause KH, Klutzny U, Bulst S, Frey JR, Lochmüller H*. Scapuloperoneal syndrome type Kaeser and a wide phenotypic spectrum of adult-onset, dominant myopathies are associated with the desmin mutation R350P. Brain 2007; 130:1485-96
42) *Arbustini E, Narula N, Dec GW, Reddy KS, Greenberg B, Kushwaha S, Marwick T, Pinney S, Bellazzi R, Favalli V, Kramer C, Roberts R, Zoghbi WA, Bonow R, Tavazzi L, Fuster V, Narula J*. The MOGE(S) classification for a phenotype-genotype nomenclature of cardiomyopathy: endorsed by the World Heart Federation. J Am Coll Cardiol 2013; 62:2046-72
43) *Hazebroek MR, Moors S, Dennert R, et al*. Prognostic Relevance of Gene-Environment Interactions in Patients With Dilated Cardiomyopathy: Applying the MOGE(S) Classification. J Am Coll Cardiol 2015; 66:1313-23
44) *Arslan-Kirchner M, Arbustini E, Boileau C, Charron P, Child AH, Collod-Beroud G, De Backer J, De Paepe A, Dierking A, Faivre L, Hoffjan S, Jondeau G, Keyser B, Loeys B, Mayer K, Robinson PN, Schmidtke J*. Clinical utility gene card for: Hereditary thoracic aortic aneurysm and dissection including next-generation sequencing-based approaches. Eur J Hum Genet 2016 Jan; 24(1)
45) *Disabella E, Grasso M, Gambarin FI, Narula N, Dore R, Favalli V, Serio A, Antoniazzi E, Mosconi M, Pasotti M, Odero A, Arbustini E*. Risk of dissection in thoracic aneurysms associated with mutations of smooth muscle alpha-actin 2 (ACTA2). Heart 2011; 97:321-6
46) *Erbel R, Aboyans V, Boileau C, Bossone E, Bartolomeo RD, Eggebrecht H, Evangelista A, Falk V, Frank H, Gaemperli O, Grabenwöger M, Haverich A, Iung B,Manolis AJ, Meijboom F, Nienaber CA, Roffi M, Rousseau H, Sechtem U, Sirnes PA, Allmen RS, Vrints CJ; ESC Committee for Practice Guidelines*. 2014 ESC Guidelines on the diagnosis and treatment of aortic diseases: Document covering acute and chronic aortic diseases of the thoracic and abdominal aorta of the adult. The Task Force for the Diagnosis and Treatment of Aortic Diseases of the European Society of Cardiology (ESC). Eur Heart J 2014; 35:2873-926
47) *Brooke BS, Habashi JP, Judge DP*. Angiotensin II blockade and aortic-root dilation in Marfan's syndrome. N Engl J Med 2008; 358:2787-95
48) *Gambarin FI, Favalli V, Serio A et al*. Rationale and design of a trial evaluating the effects of losartan vs. nebivolol vs. the association of both on the progression

of aortic root dilation in Marfan syndrome with FBN1 gene mutations. J Cardiovasc Med (Hagerstown) 2009; 10:354-362

49) *Pees C, Laccone F, Hagl M*. Usefulness of losartan on the size of the ascending aorta in an unselected cohort of children, adolescents, and young adults with Marfan syndrome. Am J Cardiol 2013; 112:1477-83