Experimental animal models to induce cardiac arrhythmias

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ABSTRACT

Cardiac arrhythmias are of different types based on their mechanism and origin. The information gathered from animal studies has been instrumental in devising diagnostic and therapeutic strategies; so different animal models are needed for different types of arrhythmias. The origin and mechanism underlying clinical arrhythmias are of considerable significance, since knowledge of these processes may provide a basis for successful therapy. Various animal models that encompass different types of arrhythmias are reviewed. This review classifies various experimental models according to their origin, which are mainly supraventricular and ventricular. Also included are various transgenic animal models for arrhythmias.

KEY WORDS: Atrial fibrillation, atrial flutter, re-entrant arrhythmia, ventricular fibrillation

Introduction

Arrhythmias are disorders of heart rhythm. They are due to abnormalities in impulse generation, impulse conduction, or a combination of both. Abnormalities of impulse generation include abnormalities of automaticity and early or delayed after depolarization with triggered activity. Abnormalities of impulse propagation include conduction block and re-entry of the cardiac impulse. Combination of abnormalities of impulse formation and propagation can produce complex arrhythmias. In any arrhythmia, it is useful to know which cardiac tissue participates, the ionic mechanisms and structural abnormalities that promote it. Supraventricular and ventricular arrhythmias differ in origin, ECG changes and clinical manifestations, based on which one must be able to distinguish between supraventricular from ventricular arrhythmias. The mechanism underlying clinical cardiac arrhythmias are of considerable significance and it is unfortunate that these arrhythmias are not easily studied in clinical situations. Now a days, sophisticated electro-physiological techniques are available to study cardiac pathophysiology, both in vivo and in vitro. These techniques have enabled to study the underlying mechanisms of arrhythmias and conduction disturbances in both experimental models and in patients. Although our knowledge of the mechanisms of arrhythmias and conduction disturbances has greatly increased, much remains to be explored. Various animal models [Table 1] have been developed for supraventricular as well as ventricular tachycardia to understand the basic cause, origin, possible mechanisms, manifestations and for development of new therapeutic strategies. Supraventricular tachycardia in an animal model closely resembles the clinical features observed in the patients. But ventricular models are fraught with problems since they cannot be studied in human patients because of the unpredictable occurrence in situations, where electrophysiological changes may develop within minutes. Besides this, many other factors determine whether, and if so how often ventricular arrhythmias occur in the setting of acute ischaemia and/or a chronic myocardial infarction. In experimental models, usually only a single factor is taken into account. Though, an animal is not the same as a human patient, arrhythmogenic mechanisms derived from animal experiments have tremendously helped us to diagnose and adapt therapeutic strategies.

The therapeutic strategies to treat cardiac arrhythmias include pharmacologic approaches, ablation of specific foci involved in arrhythmogenesis, antiarrhythmic surgical approaches and implantable devices designed to respond to tachyarrhythmic events or to prevent symptomatic bradyarrhythmias. The antiarrhythmic drugs may be classified according to the modified Vaughan Williams system, which categorizes them on the basis of electropharmacologic and electrophysiological properties. Drugs having class-I action possess local anesthetic or membrane stabilizing activity. Their predominant action is to block the fast inward sodium channel. This produces a decrease in the maximum depolarization rate of the action potential (Phase 0) and slows intracardiac conduction. These agents can be further subclassified as class Ia, Ib or Ic on the basis of their effects on specific aspects of intracardiac conduction and refractoriness. Class-II drugs block β-receptor and thus reduce heart rate, decrease intracellular Ca++ overload and inhibit after depolarization-mediated automaticity. Class-III antiarrhythmic agents prolong action potential duration, presumably through blockade of K+ channels. Class IV antiarrhythmic agents inhibit the slow calcium influx during the plateau of the action potential through Ca++ channel blockade.
Animal models to induce cardiac arrhythmia

Supraventricular tachycardia

Wolf–Parkinson–White syndrome (WPW syndrome)

The WPW syndrome, an electrocardiographic pre-excitation pattern, is associated in a fairly large percentage of cases with attacks of supraventricular tachycardia. At present, all the electrophysiological characteristics of accessory atrioventricular connections and their role in causing re-entrant tachycardia have been obtained from studies on human patients. Boineau and Moore described pre-excitation in dogs and studied propagation of activation across an accessory atrioventricular connection in types A and B pre-excitation. In type A, the effective refractory period of the accessory pathway exceeds that of the normal AV nodal His–Purkinje pathway. Therefore, a premature atrial impulse may get blocked at the accessory pathway and conducted anterogradely down the normal pathway. Ultimately, entering the accessory pathway in the retrograde direction and re-entering the atrium to establish a circus movement tachycardia referred to as orthodromic. In type B, a shorter refractory period in the anomalous pathway and then retrograde invasion of the normal pathway, with antegrade conduction down the anomalous pathway and then retrograde invasion of the normal AV nodal pathway to establish an antidromic tachycardia. In type A, the delta wave and QRS complex are predominantly upright in the precardial leads. The dominant R wave in lead V₁ may be misinterpreted as right bundle branch block. In type B, the delta wave and QRS complex are predominantly negative in leads V₁ and V₂ and positive in the other precardial leads, resembling left bundle branch block. In this study, they observed that atrial fibrillation induced in the dog caused ventricular fibrillation as well because the accessory pathway had a short refractory period and conducted many impulses, which otherwise would have been blocked in the AV node.

Human mutations in PRKAG2, the gene encoding the γ₃ subunit of AMP-activated protein kinase (AMPK), cause cardiomyopathy, characterized by ventricular hypertrophy, WPW syndrome and progressive conduction system disease. Michael et al. developed transgenic mice over-expressing the PRKAG2 cDNA with or without a missense N4881 human mutation. Transgenic mutant mice showed elevated AMP-activated protein kinase activity, accumulated large amount of cardiac glycogen, developed dramatic left ventricular hypertrophy and exhibited ventricular preexcitation and sinus node dysfunction.

Drugs, which can be screened through these models, are adenosine type drugs and class la drugs for acute therapy. For chronic therapy class I as well as class III drugs can be screened. Classes II and IV (phenylalkylamine and benzothiazepine like) drugs, which can cause AV nodal block, can also be screened for acute therapy.

Re-entrant arrhythmia of AV node

Paroxysmal supraventricular tachycardia (PSVT) due to AV nodal re-entry is the most common form of supraventricular arrhythmia. The underlying pathophysiology in AV nodal reentry is the presence of dual AV nodal pathways. The AV node in patients with dual-pathway physiology behaves as though there are two types of conduction pathways in the AV node, one capable of faster conduction, which usually has a longer refractory period, and the other more slowly conducting and having a shorter refractory period.

The pioneering clinical studies of the 1980s allowing successful surgical treatment or catheter ablation of the arrhythmia, all quoted the microelectrode studies on the isolated rabbit heart preparations that provided insight into arrhythmia mechanisms on a cellular basis. Janse et al., demonstrated circus movement within the AV node as a basis for supraventricular tachycardia. They employed multiple microelectrodes recording in the isolated rabbit heart for their study.

The animal model widely used for AV nodal re-entrant tachycardia is the isolated rabbit heart preparation. However, this model does not mimic the heart of patients suffering from AV nodal re-entrant tachycardia. Wit et al., demonstrated an in vitro model of paroxysmal supraventricular tachycardia. They used in vitro preparation of rabbit heart atrium, including the AV node and Bundle of His to evaluate the mechanism of paroxysmal supraventricular tachycardia. Microelectrode recordings from the atrium and AV node were observed. During sinus rhythm the atrial cycle was explored with atrial premature depolarization.

More et al., induced experimentally paroxysmal AV nodal tachycardia in the dog. Lin et al., experimentally created atrioventricular node re-entrant tachycardia in the dog by surgery. They blocked atrial impulses from the anterior input site to the AV node.

Wu et al., used optical mapping in isolated canine atrioventricular nodal re-entrant tachycardia. This study was performed to optically map Koch’s triangle and surrounding atrial tissue in an isolated canine AV nodal preparation. Multiple preferential AV nodal input pathways were observed in all preparation with continuous and discontinuous AV nodal function curves. AV nodal echo beats were induced in 54% (12/22) of preparations. The re-entrant circuit of the slow/fast echo beats (EB) (36%) started as a block in fast pathway and a delay in slow pathway (SP) conduction to the compact AV node, then excited from the AV node to the fast pathway and rapidly returned to the second pathway through the atrial tissue located at the base of Koch’s triangle. The re-entrant circuit of the fast/slow EB (9%, n=2) was in an opposite direction. In the slow/slow EB (9%, n=2), anterograde conduction was over the intermediate pathway (IP) and retrograde conduction was over the SP. Unidirectional conduction block occurred at the junction between the AV node and its input pathways. Conduction over the IP smoothed the transition from the FP to the SP resulting in a continuous AV nodal function curves. Complete or incomplete echoes were induced in isolated preparations by this method. Patterson et al., have demonstrated that longitudinal dissociation within the posterior AV nodal input can give rise to localized re-entry and AV nodal re-entrant tachycardia.

Adenosine is a potent source to terminate AV nodal re-entrant tachycardia. Hence, drugs such as adenosine can be screened by using these models. Class lc drugs can be screened for chronic use. AV nodal blockers [classes II and IV (such as verapamil and diltiazem)] may be screened for acute study.
Atrial flutter

Atrial flutter is a rapid regular atrial tachyarrhythmia that is less common than the PSVTs or atrial fibrillation. It is observed only very rarely in normal subjects but may occur at any age in the presence of underlying abnormalities such as those secondary to mitral valve disease, congenital heart disease, cardiomyopathies and less frequently coronary artery disease. Subgroups at particularly high risk for developing atrial flutter are children, adolescents and young adults, who have undergone corrective surgery for complex congenital heart disease, most commonly transposition of the great vessels, tetralogy of Fallot, or atrial septal defects. Lewis et al., concluded that atrial flutter was the result of circus movement in the atria. Successful animal preparations of atrial flutter have been developed over the years. Some important animal models to induce atrial flutter are as follows.

Canine right atrial crush injury model

Gregory et al., used this method for induction of atrial flutter. The atrial crush injury was made with a surgical clamp by lifting the anterior portion of the right atrial plaque after recording baseline sinus rhythm. The crush injury was placed on the right atrial free wall parallel to and approximately 1.5 cm above the atroventricular groove, extending from the base of the right atrial appendage 1.5-2.5 cm posterior towards the intercaval zone. The crush injury was typically 3-4 mm wide. After right atrial crush injury, attempts were made to induce sustained atrial flutter by programmed atrial stimulation introducing single (S,S), double (S,S,S), or triple (S,S,S,S) premature beats to atrial refractoriness. Sustained atrial flutter was defined as that lasting >10 min.

Atrial flutter induced by acetylcholine (ACh) and rapid pacing in the dog

Wu et al., induce atrial flutter in the isolated blood perfused canine heart. They produced episodes of rapid atrial flutter by continuous infusion of ACh and rapid atrial pacing. They isolated canine right atria and perfused it with 1–5 μM/L of ACh. Mapping of the endocardium was done by using 477 bipolar electrodes with simultaneously recording transmembrane potentials from the epicardium. The APD was measured during regular pacing with cycle lengths of 300 ms. Atrial arrhythmia was induced by a premature stimulus.

Atrial flutter by aconitine

Scherf et al., provoked atrial flutter in anesthetized dogs by application of a few crystals of aconitine or delphinine to the surface of the right atrium in the appendix area near the head of the sinus node. Nwangw et al., used aconitine as an arrhythmogenic agent to screen arrhythmic drugs in mice. Dadkar and Bhattacharya recommended aconitine antagonism in conscious mice as screening procedure. Winslow et al., established aconitine in anesthetized mice. Also, Winslow established the arrhythmogenic effects of aconitine in cats.

Right atrial enlargement model of atrial flutter

Restivo et al., developed the canine model in which right atrial enlargement was produced by banding of the pulmonary artery thereby producing tricuspid regurgitation which may have a clinical counterpart in patients with chronic obstructive pulmonary disease and tricuspid regurgitation. It is now established that atrial flutter is due to a re-entrant wave in the right atrium, and that a zone of slow conduction located inferiorly and posterior in the right atrium is the target for catheter ablation.

AV nodal blockade is a reliable mechanism to treat atrial flutter. Thus adenosine like drugs, Ca++ blockers and beta-adrenergic blockers can be screened on chronic basis through the models, which produce atrial flutter. Class Ia, Ib and class III drugs can be screened for acute therapy.

Atrial fibrillation

The prevalence, presentation, clinical significance and long-term implications of atrial fibrillation depend heavily upon the clinical circumstance in which it occurs. Among the cross-sectional studies of prevalence, there is a large gradient across age categories, ranging from less then 0.5% through the decades from 40 to 70 years and reaching rates in excess of 10% is some beyond age 70. The haemodynamic consequences of atrial fibrillation are due to two factors: (i) the loss of atrial systole may impair ventricular function in the noncompliant ventricle (e.g. aortic stenosis, left ventricle hypertrophy or the dilated ventricle with systolic dysfunction) and (ii) a rapid ventricular rate encroaches upon diastolic filling of the left ventricle and the coronary arteries. The risk of embolism and stroke is a long-term concern of special importance. The left superior vena cava can be the arrhythmogenic source of AF. The left superior vena cava (LSVC) is the embryological precursor of the ligament of Marshall, which has been implicated in the initiation and maintenance of atrial fibrillation. Rarely the LSVC may persist and has been associated with some organized arrhythmias. Li, Fern reported five patients in whom the LSVC was a source of ectopy, initiating atrial fibrillation.

Atrial fibrillation by atrial ischaemia in dogs

Hani et al., induced AF by atrial ischaemia after occluding the right intermediate atrial artery (a branch of the right coronary artery) that perfuses the right atrial free wall. Atrial–arterial occlusion increased the duration of AF induced by burst pacing from 57-32 to 803-214 sec after 0.5 h of occlusion and to 887-209 sec after 3 h of occlusion. Prolonged AF was induced in none of the 16 dogs under control nonischemic conditions, 7 of 16 dogs (44%, P<0.01) at 0.5–3 h after occlusion, and 5 of 13 dogs (38%, P<0.01) 3–5 h after occlusion.

Pituitary adenylate cyclase activating polypeptide-27 (PACAP-27) induced biphasic chronotropic effect and atrial fibrillation

PACAP-27 causes negative chronotropic effect through postganglionic nerve activation and it produces the positive chronotropic effect mediated by PACAP receptors with an activation of nonadrenergic nonvasoactive intestinal peptideergic nerves at least in part in the dog heart. Neuroly released acetylcholine induced by PACAP-27 participates in the induction of atrial fibrillation.

Atrial fibrillation in dogs by atrial burst pacing

Danshi et al., induced atrial fibrillation by atrial burst pacing (10 Hz, 1 to 5 sec). Atrial fibrillation >20 min requiring electrical cardioversion for termination was considered persistent. To estimate mean atrial fibrillation duration, atrial fibrillation was induced 10 times if the duration was less than 10 minutes and 5 times if it was 10–20 min. If persistent atrial fibrillation was induced twice, no further atrial fibrillation inductions were performed.

Canine model of chronic atrial fibrillation

Thomas et al., has induced chronic atrial fibrillation in dogs
by creating moderate mitral regurgitation and rapidly pacing the right atrium at 640 bpm for > 8 weeks. Chronic atrial fibrillation was established with the combination of rapid atrial pacing and creation of moderate regurgitation. Catheters were introduced into the left and right heart of female mongrel dogs via femoral venous and arterial sheath. Baseline hemodynamic measurements were recorded. A 7F steerable catheter with a stiff 2 mm wire hook at its terminus was placed in the left ventricle and manipulated until mitral chordae tendineae were ensnared and then avulsed. An active fixation atrial J permanent pacemaker lead was placed in the right atrial appendage by a jugular venous approach. The pacemakers were programmed at the rate of 640 bpm or 400 bpm and an output of 2–3 times atrial diastolic threshold. After 6 weeks and weekly thereafter, the pacemakers were reprogrammed to low rates and to threshold outputs. After 24 h without pacing, a six lead surface ECG was obtained to verify the presence of atrial fibrillation.  

**Vagal atrial fibrillation**

Parasympathetic stimulation has been used for decades for the induction and maintenance of atrial fibrillation in experimental protocols. Parasympathetic stimulation dramatically shortens the atrial effective refractory period thereby decreasing the wavelength of atrial excitation wave fronts. The shorter the wavelength, the higher is the probability that multiple re-entrant circuits can exist simultaneously in the atrial myocardium; the presence of these multiple circuits, in turn, increases the stability of atrial fibrillation.

For cervical vagal nerve stimulation, the cervical vagosympathetic trunks were cut, and stainless steel wires were introduced in the cranial end of the vagosympathetic trunk. Stimulation of both the vagi was performed with separate isolated constant current sources (SS-202J, Nihon Kohden) driven by a programmable stimulator (SEN-7203, Nihon Kohden). During bilateral vagal stimulation, AF was induced by atrial extrastimulation using a digital programmable stimulator (SEC-2102, Nihon Kohden). AF lasting more than 10 min was repeatedly induced in dog.  

**Atrial fibrillation in the isolated Langendorff-perfused rabbit heart**

Atrial arrhythmias frequently occur under conditions associated with atrial dilation. In patients with acute myocardial infarction, the onset of atrial arrhythmias is thought to be related to the elevated left ventricular end diastolic pressure, resulting in stretch of the atrial wall. In the Langendorff perfused rabbit heart, the interatrial septum was perforated, and after occlusion of the caval and pulmonary veins, bilateral pressure was increased by raising the level of an outflow cannula in the pulmonary artery. Right and left arterial effective refractory periods, monophasic action potentials, and inducibility of atrial flutter by single premature stimuli were measured as a function of atrial pressure. Increasing the atrial pressure from 0.5±0.7 to 16.2±2.2 cm H$_2$O resulted in a progressive shortening of the right atrial effective refractory period (AERP) from 82.2±9.8 to 48.0±5.1 ms. In the left atrium, an increase in pressure up to 7.4±0.3 cm H$_2$O had no effect on the AERP. At higher pressures, however, the left AERP also shortened, from 67.5±7.5 to 49.3±2.0 ms. The duration of monophasic action potentials (MAP) also decreased by an increase in atrial pressure, showing a high correlation with the shortening in AERP (r=0.94, P<0.01). All these changes were completely reversible within 3 min after release of the atrial stretch. Dilatation of the atria was a major determinant for the vulnerability to AF.

**Atrial fibrillation by fibrillation pacemaker**

Goats were chronically instrumented with multiple electrodes sutured to the epicardium of both atria. Two to three weeks after implantation, the animals were connected to a fibrillation pacemaker which artificially maintained atrial fibrillation. Control episodes of AF were short lasting (6±3 sec) but artificial maintenance of AF resulted in a progressive increase in the duration of AF to become sustained (>24 h) after 7.1±4.8 days.

**Atrial fibrillation by rapid atrial pacing and acetylcholine**

Burashnikov et al., induced atrial fibrillation in isolated sheep hearts by burst rapid pacing from the epicardial surface of either the right atria after the addition of acetylcholine (1 µM/L) to the perfusate. Transmembrane action potentials, pseudo-ECG (pseudo-ECGs are constructed from optical recordings by integrating the transmembrane fluorescence signal over the left and right halves of the mapped region and taking the difference) and tension development were recorded. Allan et al., recorded the optical recordings of atrial movements to demonstrate wave propagation and lines of block, which changed on a beat to beat basis.

**Atrial fibrillation by aconitine**

The plant alkaloid aconitine persistently activates sodium channels. Nakayama et al., compared the effects of various beta adrenergic blocking agents with known antiarrhythmics on aconitine arrhythmia. They produced supraventricular arrhythmias by topical application of aconitine in a small cup placed on the right atrium of dogs. Yamamoto et al., used urethane-anesthetized rats under artificial respiration with tubocurarine pretreatment. After thoracotomy and incision of the pericardium, a piece of filter paper soaked with aconitine solution was applied to the right atrium. Test drugs were applied by continuous i.v. infusion. ECG lead II and intra atrial ECG were monitored.

Therapy of atrial fibrillation is almost same as atrial flutter. Thus choice of drugs, which can be screened through these models of atrial fibrillation, is same as atrial flutter.

**Ventricular fibrillation**

**Porcine model of VF**

Iyad et al., used this model to show ventricular anti-fibrillation activity of cariporide (sodium–hydrogen exchanger isoform-1 inhibitor). A 5F pacing electrode was advanced through the right cephalic vein into the right ventricle for induction of VF. VF was induced by an alternating current (1.5 mA) delivered to the right ventricular endocardium, and mechanical ventilation was discontinued. The effects on ischemic contracture were investigated by the use of transesophageal echocardiography (TEE). The effects on APD and ventricular ectopic activity were investigated by the use of a monophasic action potential (MAP) recording/pacing catheter.
### Animal models to induce cardiac arrhythmias

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**Ventricular fibrillation induction by 60-Hz alternating current in isolated swine right ventricle.**

Voroshilovsky et al., studied seven isolated perfused swine right ventricles *in vitro*. The action potential duration restitution curve was determined. Alternating current captured the right ventricles at 100± 65 µA, which is significantly lower than the direct current pacing threshold (0.77 ± 0.45 mA P<0.05). Alternating current induced ventricular tachycardia or ventricular fibrillation at 477 ± 266 µA, when the stimulated response to alternating current had (1) short activation cycle lengths (128±14 ms), (2) short diastolic intervals (16±9 ms) and (3) short diastolic intervals associated with a steep action potential duration restitution curve. Optical mapping studies showed that during rapid ventricular stimulation by alternating current, a wave front might encounter the refractory tail of an earlier wave front, resulting in the formation of a wave break and ventricular fibrillation.

**Ventricular arrhythmia by various chemicals**

Vogel and Vogel described a method in which they administered continuous aconitine infusion into the saphenous vein to produce ventricular arrhythmias. Vaille et al., used CaCl2 continuous i.v. infusion screening method in rats for evaluation of antiarrhythmic calcium antagonists. Lawson induced ventricular fibrillation in mice by using chloroform as an antiarrhythmic agent. Tripathi and Thomas produced ventricular tachycardia in rats and guinea pigs by exposing the animals to benzene vapours for 2 min followed by an intravenous adrenaline injection. Digoxin a cardiac glycoside, if given in overdose produces ventricular extrasystoles, ventricular fibrillation, and finally death. Nagasawa et al., used digitalis induced arrhythmia model in dogs for screening of a new antiarrhythmic drug which was Na+/Ca++ exchange inhibitor. Sono et al. induced ventricular fibrillation by isoprenaline in isolated rat hearts. Tomokazu et al., used ouabain or strophanthin K, which is a cardiac glycoside as an arrhythmogenic substance. Ouabain induced ventricular tachycardia and multifocal ventricular arrhythmias in dogs. Ra et al., demonstrated a modified method for the production of cardiac arrhythmias by ouabain in anesthetized cats. Takei described experimental arrhythmia in guinea pigs induced by grayanotoxin-I, a biologically active diterpenoid from the plant family of Ericaceae.

**Ischaemia-induced ventricular arrhythmia**

Cardiac ischaemia leading to myocardial infarction is a most common cause of morbidity and mortality. Chemical or surgical interventions allow the recovery of the ischaemic myocardium by restoration of blood flow or reperfusion. This reperfusion, however, is known to be associated with...
ventricular arrhythmias and myocardial dysfunction that can lead to severe cardiac impairment and cell death. Reactive oxygen species (ROS) such as superoxide anion, hydrogen peroxide, hydroxyl radical, and singlet oxygen have been implicated as important factors in the pathogenesis of cellular injury in the postischemic heart.

Coronary artery occlusion and reperfusion in the isolated perfused rat heart, was widely used as a model for assessment of antifibrillatory action of antiarrhythmic agents. Later, coronary occlusion/reperfusion arrhythmias have been shown in anesthetized animals. Clark et al., demonstrated coronary artery ligation in anesthetized rats as a method for the production of experimental dysrhythmias and for the determination of infract size. Harris et al., studied influences of hypothermia, cold and ischemia stress on the severity of coronary artery ligation-induced arrhythmias in rats. Bernier et al., used reperfusion-induced arrhythmias and studied oxygen derived free radicals, which causes myocardial infarction and ventricular arrhythmia, in isolated perfused rat heart. Lepran et al., used the coronary artery ligation technique in rats after 7–10 days of surgery. They placed a loose silk loop around the left coronary artery and passed the threshold through a cylinder-shaped polyethylene tube outside the thorax. The loose ligature was tightened and arrhythmia record by ECG. Lubbe et al., used coronary artery occlusion and reperfusion in isolated perfused rat heart for assessment of antifibrillatory action of antiarrhythmic agents. Maclod et al., induced arrhythmia by ischaemia and reperfusion in conscious and anesthetized rats, and they studied the effect on epicardial intracellular action potentials.

In the ischaemia–reperfusion model different parameters can be evaluated, such as mortality, haemodynamic parameter, ventricular extrasystoles, ventricular tachycardia, ventricular fibrillation and infract size. The number of ventricular premature beats, ventricular tachycardia and ventricular fibrillation are counted in the occlusion and reperfusion periods and evaluated according to the guidelines of Lambeth convention. Capasso et al., reported heterogeneity of ventricular remodelling after acute myocardial infarction in rats which was produced by the ischaemia–reperfusion technique. Ruben et al., studied the distribution of extracellular potassium and its relation to electrophysiological changes during acute myocardial ischaemia in the isolated perfused porcine heart. Bradykinin perfusion reduced the incidence of ventricular fibrillation and reduced the release of cytosolic enzyme and preserved glycogen stores. Ventricular arrhythmias occurring secondary to impeded myocardial perfusion is the cause of death in more then one-half of the subjects associated with myocardial infarction and is also an important cause of sudden cardiac death. Two distinct phases of ventricular arrhythmias occur during the first 30 minutes after induction of regional ischaemia by acute occlusion of a coronary artery in canine and porcine heart.

In the first phase, la (up to approximately 8–10 min of coronary occlusion), there is a rapid change in electrical membrane properties associated with metabolic acidification (anaerobic glycolysis), and cellular loss and extracellular accumulation of $[K^+]$. The impacts of these changes are a rapid depolarization of the ischemic myocytes, and a loss of amplitude and duration of the transmembrane potential. In second phase, lb, which occurs after 10–15 min of occlusion is related to the electrical uncoupling of the myocytes resulting in smaller size of circus movement. William et al., reported that the lb phase of ventricular arrhythmias in ischemic in situ porcine heart is related to changes in cell-to-cell electrical coupling. Borrett et al., described a myocardial ischaemia model for arrhythmia in rabbits. Vara et al., induced ventricular fibrillation by coronary occlusion during hypothermia in dogs. Baboo open chest model of myocardial ischaemia was described by Premaratne et al., in which a 2 h ischaemia period is followed by 22 h of reperfusion. Nashlund et al., presented a closed chest model in pigs. In this model, occlusion was induced in closed chest, pentobarbital anaesthetized, and mechanically ventilated pigs by injection of a 2 mm ball into a preselected coronary artery. Reperfusion was achieved by retraction of the ball via an attached filament.

**Ventricular arrhythmia during exercise by ischaemia**

Ventricular fibrillation due to myocardial ischaemia during exercise is the model which resemble most closely the situation in coronary patients. In this model a major surgery is done in dogs, in which transducers are fixed in body followed by two-stage LAD ligation. After 28 days, animals were prepared for test, in which animals are allowed to walk on a motor driven treadmill. The animals run on the treadmill simultaneously the workload was increased in every 3 min for 18 min.

**Stretch-induced arrhythmias in isolated canine ventricle**

It is commonly accepted that serious ventricular arrhythmias are caused by abnormalities in impulse formation and conduction. These electrophysiological mechanism fail to explain why lethal arrhythmia most commonly arise in patients with severe heart failure and dilated ventricles. David et al., presented a hypothesis that alternation in loading conditions and muscle length influenced the electrophysiology of ventricular myocardium and these alterations might play a role in arrhythmogenesis in globally dilated or dyskinetic ventricles. To test the hypothesis that stretch can initiate arrhythmias in normal myocardium, the response to graded mechanical stretch was studied in seven isolated blood perfused canine ventricles. After eight conditioning contraction produced by His bundle pacing (2 Hz), global stretch of the ventricle was produced by a servo-controlled pump that abruptly increased ventricle volume by a precise amount during early diastole and then returned ventricular volume to the initial holding volume. The probability of a stretch-induced arrhythmia was determined from multiple alternating sequences in which a stretch of known amplitude or no stretch was delivered.

**Model for sudden cardiac death**

Male mongrel dogs weighing 14-22 kg are used for this model. Programmed electrical stimulation is performed between days 3 and 5 after the induction of anterior myocardial infarction by occlusion/perfusion on the left anterior descending coronary artery. A direct anodal 15 μA current from a 9V nickel–cadmium battery was passed through a 250 Ohm resistor and applied to the electrode in the lumen of the left circumflex coronary artery. After 24 h of constant anodal current or development of ventricular fibrillation, the animals were killed, the hearts were excised and the thrombus mass removed and weighed.
Canine model of two-stage ligation

Harris showed that mortality in dogs after coronary occlusion with a two-stage ligation procedure was lower than with one-stage ligation. Left descending coronary artery is partially occluded for 30 min, after which total ligation is performed. This model resembles late arrhythmias occurring in post-infarction patients. Akira et al., used this model to check antiarrhythmic activity of a new compound. Dubray et al. presented methods for producing experimental complete atrioventricular block in dogs.

Drugs, which can be screened for ventricular tachycardia, are class Ia, Ib and class III drugs for both acute and chronic therapy. Ventricular fibrillation is a result of disorganized re-entry and classes Ia and Ib drugs are useful for acute therapy and can be screened for acute uses. For chronic use classes I–III can be screened.

Transgenic mice for arrhythmia

Genetically engineered animal models hold promise for understanding the pathophysiology of mutations that cause human disease. In the last decade various artificial mutation in mice genotype yielded a number of transgenic mice, which are useful in screening for anti-arrhythmics. Berul et al., used cardiac electrophysiology method to study mice harbouring an α-myosin heavy chain Arg 43 Gln missense mutation (α-MHC 403/405), which resulted in histological and haemodynamic abnormalities characteristic of familial hypertrophy and sudden death of uncertain etiology during exercise. Wu et al., studied a mouse model of cardiac hypertrophy attributable to transgenic over-expression of a constitutively active form of CaMκ IV that also has increased endogenous CaMκ II activity. ECG telemetered transgenic mice had significantly more arrhythmias then wild type littermate controls at baseline. The KCNE 1 gene encodes a channel regulator Iκ which, in association with the K₇,QT1K⁺ channel protein, determines the slow component of the delayed rectifier current. Charpentier et al., investigated the cellular electrophysiological characteristics of adult KCNE1 knockout mouse hearts by means of the standard microelectrode technique. They concluded that invalidation of the mouse KCNE1 gene by homologous recombination leads to a mild cardiac phenotype at the cellular level. Berul et al., presented a mouse model of dilated cardiomyopathy resulting from a homozygous mutation in the myosin-binding protein C (MyBP-C). They also presented a model of familial hypertrophic cardiomyopathy due to heterozygous mutation in the same gene (MyBP-C) that were used to characterize the electrophysiological phenotype and correlate ‘vulnerability to arrhythmia’ with quantitative histopathological changes. As described earlier mutations in the γ subunit (PRKAG2) of AMP activated protein kinase produce an unusual human cardiomyopathy characterized by ventricular hypertrophy and electrophysiological abnormalities such as: Wolf–Parkinson–White syndrome and programmed degenerative conduction system disease. Mutations of the K⁺ channel genes HERG and KvLQT 1 cause the autosomal dominant long QT syndrome, presumably by interfering with the cardiac currents Iₚ and Iₚ-, as an antiarrhythmic agent. They concluded that KvLQT invalidates transgenic mice discriminating in vivo drugs that blocks Iₚ from drugs that block the transient outward current, the sodium current or the calcium current. Transgenic mice over-expressing the inflammatory cytokine tumour necrosis factor TNF-α (TNF-α mice) in the heart develop a progressive heart failure syndrome characterized by biventricular dilation, decreased ejection fraction, atrial and ventricular arrhythmias on ambulatory telemetry monitoring, and decreased survival compared with nontransgenic litter mates. These transgenic animals are more prone to re-entrant arrhythmia.

Antiarrhythmic drugs and screening models

Antiarrhythmic drugs generally affect arrhythmia by modulating conduction velocity, or effective refractory period or both. Conduction velocity, depends on the passive electrical properties of cardiac tissue, also is a characteristic of the Na⁺ channels and Ca²⁺ channels. Antiarrhythmic drugs that prolongs the action potential duration, and thereby the refractory period are effective against re-entry arrhythmias in two ways: by prolonging the wavelength [the product of (a) refractory period and (b) conduction velocity]. The initiation of a re-entrant arrhythmia by a premature impulse may be prevented or an existing arrhythmia may terminate because the wavelength becomes too large with respect to the re-entrant circuit, so that by closing the excitable gap, the head of the re-entrant wavelength will hit the wall of refractoriness and propagation stops. It is well known that there is significant difference in effective refractory period of different species. Depending on action potential duration and effective refractory period selection of animal could be one important aspect. Porcine ventricle myocardium appears to have a diastolic interval similar to that in human ventricle. In contrast to all other species, it may be appreciated that in the rat there is no shortening of refractory periods at the shorter cycle length and rat ventricle is not the first choice if one aims at filling up the diastolic interval by means of a class I or class III antiarrhythmic agent. The rabbit is often used for electrophysiological research, probably because it
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constitutes a reasonable compromise in terms of cardiac dimensions, basic electrophysiological characteristics and cost. [122,123] There are clear species differences that determine arrhythmogenesis. These differences should also be considered while choosing an animal model for arrhythmia corresponding to antiarrhythmic agent.

Conclusion

Although no animal model can accurately resemble with human disease condition and species differences also exist, close similarities with humans suffering from or threatened by arrhythmias can be developed by selecting appropriate model and species. Rather than a single model or experimental technique, combinations of investigations, like isolated heart (Langendorff arrangement or working heart), whole hearts in anesthetized or conscious animals, excised cardiac preparations, testing the function of molecules involved in electrical excitation, single cardiac cell preparation, can be performed.

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