

Properties of the Frog Heart

Introduction Overview

The overall purpose of this lab was to take a closer look at various properties of the heart and show the physiological mechanisms that govern its behavior. To do this, 4 experiments designed to test different aspects of the heart were performed; electrical and mechanical activity of the heart, extrasystolic contractions, vagal stimulation, and the effects of epinephrine on the heart. By completing this lab, a deeper understanding of why the results that were collected should be acquired, why changing variables had the effect they did, as well as the physiological mechanisms that are responsible for these changes.

Introduction: Electrical and Mechanical Activity of the Heart

The first part of the experiment functioned mainly to establish a baseline reading of the mechanical and electrical traces of the heart as well as what systole and diastole looks like on the mechanical trace. It is expected for there to be a steady rhythm between the waveforms in the raw data, representing a stable heart cycle consisting of ventricular and atrial contractions observed through systolic and diastolic peaks (Sanders et al., 2011, 578).

Introduction: Extrasystolic Contractions

The second part of the experiment was designed in order to induce and observe the effects of premature ventricular contractions, also known as extrasystole during different parts of the heart cycle. It is expected for extrasystole to occur when the heart is stimulated during its relative refractory period but due to cardiac muscles relatively long refractory period compared to skeletal muscle, there will be a delay between the extrasystolic contraction and the next contraction caused by an action potential (Sherwood 2013, 317).

Introduction: Vagus Nerve Stimulation

In this part of the experiment, the vagus nerve was directly stimulated and its effects observed on heart rate and contraction size. Due to the vagus nerve controlling parasympathetic control of the heart, it is expected for weaker contractions to occur as well as bradycardia and cardiac arrest being induced (Sanders et al., 2011, 581).

Introduction: Effects of Epinephrine on the Heart

During the last portion of the experiment, epinephrine was applied to the heart in order to observe its effect on cardiac function. Due to epinephrine being the main neurotransmitter for the sympathetic nervous system for the heart, it is expected for the chemical to increase the heart rate as well as increase the force of contraction due to appropriate vasoconstriction and vasodilation of various blood vessels in order to achieve these two effects in cardiac function (Sanders et al., 2011, 581).

Materials and Methods

Electrical and Mechanical Activity of the Heart

In order to create a baseline measurement of cardiac activity, a force transducer in conjunction with electrical connections to the heart were used in order to measure the mechanical and electrical output of the heart belonging to a bullfrog and recorded with Biopac software. A more in-depth procedures and materials list can be found in the lab manual listed under the section “Electrical & Mechanical Activity of the Heart” (Bautista, 2009, 47).

Extrasystolic Contractions

With the goal of inducing extrasystolic contractions with different parameters, differences in the waveforms between extrasystole and baseline were observed. A more in-depth procedures and materials list can be found in the lab manual listed under the section “Extrasystolic Contractions” (Bautista, 2009, 49).

Vagal Stimulation

Using a hook electrode to stimulate the vagus nerve in conjunction with a force transducer and electrical connections to the heart of a bullfrog, different states of the heart were observed. A more in-depth procedures and materials list can be found in the lab manual listed under the section “Vagal Stimulation” (Bautista, 2009, 51).

The Effects of Epinephrine on the Heart

Several drops of epinephrine were directly dropped onto the ventricle of the bullfrog’s heart. A more in-depth procedures and materials list can be found in the lab manual listed under the section “The Effects of Epinephrine” (Bautista, 2009, 53).

Experimental Deviations

During the experiment, the electrical trace of all data contained noise due to the wire not being fully scraped off of insulation. Additionally, heart activity after the recovery from bradycardia was not recorded due to human error during the lab when the Biopac software was stopped before the data could be collected. Also, extrasystolic contraction data may have been skewed due to human error when manually eliciting stimulating during different parts of the cardiac cycle.

Results

Electrical & Mechanical Activity of the Heart

The mechanical and electrical activity of a bullfrog’s heart was measured in order to observe its normal function without the effects of any change in variables. Heart rate (BPM), atrial contractile force (g), ventricular contractile force (g), and time between heartbeats (sec) were measured.

Table 1. Heart Rate (BPM), atrial contractile force (g), ventricular contractile force (g), and latency between heartbeats (sec) were recorded in a bullfrog’s heart and averaged between 3 data

points. Heart rate was analyzed using the BPM tool in Biopac software; P-P tool for atrial and ventricular contractions (g); and Delta T for time between heartbeats (sec). Ventricular contractile force was greater than atrial contractile force with a latency in between beats.

Heart Rate (BPM)	32
Atrial Contractile Force (g)	0.245
Ventricular Contractile Force (g)	2.85
Time Between Heartbeats (sec)	0.668

Extrasystolic Contractions of the Heart

Extrasystolic contractions were induced in a bullfrog's heart and measured in order to observe differences in the force before and after extrasystolic contraction (g), as well as the compensatory pause that follows one (sec). A decrease in contractile force during both before/after extrasystole as well as the compensatory pause could be observed in 2x threshold during late diastole when compared to late diastole at threshold. Early diastole elicited contractile forces and compensatory pause values in-between late diastole at threshold and late diastole at 2x threshold.

Table 2. Extrasystolic contractions were induced during different parts of the cardiac cycle in a bullfrog's heart; late diastole at threshold, late diastole with two times threshold voltage, and early diastole at threshold. Ventricular force before and after extrasystole was measured and averaged between 3 different data points, and analyzed using the P-P tool in Biopac software. The compensatory pause that follows extrasystolic contractions was analyzed between an extrasystolic contraction and normal ventricular contraction that was induced in each part of the cardiac cycle tested during the experiment using the Delta T tool in Biopac software. Increases in ventricular force after an extrasystolic contraction occurred in all parts of the cardiac cycle. A compensatory pause was also observed.

	Voltage (V)	Force Before Extrasystole (g)	Force After Extrasystole (g)	Compensatory Pause (sec)
Late Diastole	2.6	2.864	2.89	1.445
2x Threshold Late Diastole	5.2	1.9	2.07	0.81
Early Diastole	2.2	2.33	2.41	1.33

Vagal Stimulation of the Heart

The effects of prolonged stimulation to the vagus nerve of a bullfrog's heart were observed and cardiac function measured in voltage (V), atrial contractile force (g), and ventricular contractile force (g) were measured during 3 cardiac states known as bradycardia, cardiac arrest, and vagal escape. During bradycardia, a decrease in heart rate but increase in atrial and ventricular force was observed compared to baseline. During cardiac arrest, no heart rate or atrial or ventricular force could be recorded due to the heart not contracting during this state. When the heart underwent vagal escape, the heart rate as well as atrial and ventricular contractile force returned to near baseline levels.

Table 3. Prolonged stimulation to a bullfrog's heart via the vagus nerve until bradycardia, cardiac arrest, and vagal escape were induced in order to observe differences in heart rate (bpm), voltage (V), atrial contractile force (g), and ventricular contractile force (g) in comparison to baseline

values without any stimulation. Heart rate was analyzed using the BPM tool in Biopac software and atrial/ventricular contractile force using the P-P tool averaged among 3 different data points. Voltage was gathered during the experiment. During bradycardia, heart rate decreased about 11 bpm from baseline while vagal escape returned to near baseline levels with a bpm of 2 slower than baseline. Atrial and contractile force increased during both bradycardia and vagal escape from baseline. During cardiac arrest, neither heart rate nor contractile force could be measured due to cardiac output decreasing to 0 during arrest.

	Baseline	Bradycardia	Cardiac Arrest	Vagal Escape
Heart Rate (bpm)	32	20.984	N/A	30.76
Voltage (V)	2	12	15	15
Atrial Contractile Force (g)	0.245	0.327	N/A	0.247
Ventricular Contractile Force (g)	2.85	4.055	N/A	4.19

Effects of Epinephrine on the Heart

The effects of applying epinephrine to a bullfrog's heart was observed by measuring heart rate (BPM) and ventricular contractile force before and after application of epinephrine. Heart rate and ventricular force was averaged between 3 different data points and analyzed using the BPM tool in Biopac software for the former and the P-P tool for the latter. After epinephrine was applied, an increase in heart rate of around 17 bpm was observed, as well as a .415g increase in contractile force.

Table 4. Effects of epinephrine on a bullfrog's heart rate (bpm) and ventricular contractile force (g) before and after application. Heart rate was analyzed using the BPM tool in Biopac, and the P-P tool for ventricular contractile force averaged among 3 different data points. Both heart rate and ventricular contractile force increased after application of epinephrine.

	Before Epinephrine	After Epinephrine
Heart Rate (BPM)	32.7	49.401
Ventricular Contractile Force (g)	2.16	2.575

Discussion

Electrical and Mechanical Activity of the Heart

During the first part of the experiment, a baseline mechanical and electrical trace reading of a bullfrog's heart was established in order to observe normal function without the modification of any variables. As seen in Table 1, the average heart rate was 32 bpm with an atrial contractile force of .245g and ventricular contractile force of 2.85g with a latency of .688 seconds. Although

the heart rate would be considered extremely slow in humans, the physiology of a frog's heart as well as the conditions in which it was operating are much different, such as the frog being incapacitated during the experiment. However, the atrial and ventricular contractile force are what we expect to see in a normal functioning heart. This is due to the atria being responsible for only filling the ventricles with blood while the ventricles are the main movers of blood throughout the whole body (Hall, 2011, 107). Even though the atria mainly serve only to fill the ventricles with blood, most of the blood in the ventricles has already passively flowed into them from the atria, with the atria contracting only to top them off (Hall, 2011, 108). During a period in the cardiac cycle known as isovolumetric contraction during systole, ventricular pressure rises sharply and continues rising in order to open the semilunar valves and allow for blood to be ejected into circulation (Hall, 2011, 108). This build-up in pressure exceeds that of atrial pressure due to the need for blood to be able to flow from the heart all throughout the body to the veins and back to the heart in one circulation. This explains why during this experiment, ventricular contractile pressure was greater than atrial pressure.

Extrasystolic Contractions of the Heart

Extrasystolic contractions, also known as premature ventricular contractions (PVC's) occur when sudden depolarizations occurs in the ventricle, rather than the SA node, which is the usual site for ventricular contractions, inducing an early contraction (Cha, et. al, 2012, 229). During the experiment, extrasystolic contractions were induced during 3 different parts of the cardiac cycle, late diastole at threshold voltage, late diastole at double threshold voltage, and early diastole. Ventricular force (g) before and after the extrasystolic contraction was measured, as well as the compensatory pause (sec) that followed before the next normal ventricular contraction induced

by an action potential. As seen in table 2, the compensatory pause between late diastole at threshold (1.445 seconds) was longer than early diastole (1.33 seconds). However, late diastole at 2x threshold (0.81 seconds) was observed to be shorter. The expected result would be for both late diastole experiments to be shorter than that during early diastole. Although in all 3 scenarios, extrasystolic contractions were able to be induced, suggesting that stimulation was during the relative refractory period (RRP), it is expected that since a PVC is induced earlier in the relative refractory period, such as in early diastole, the compensatory pause before the next action potential will be longer, or in the inverse scenario, where a PVC is induced later in the relative refractory period, there will be a shorter delay such as during late diastole since the PVC was induced closer towards the end of the RRP interval. The data collected during the experiment may not have lined up with what was expected to be seen due to human error during the application of the stimulus to different parts of the cardiac cycle.

Aside from a difference in compensatory pause, ventricular contractile force before and after systole also produced changes. During all 3 trials, ventricular contractile force increased in the heartbeat following the extrasystolic contraction. Although not measured or observed directly during the experiment, this increase in ventricular force will result in an increase in the end diastolic volume, due to the principles of the Frank-Starling law of the heart, which states that the amount of blood pumped by the heart is in proportion to the blood flow into the heart from the veins, also known as venous return (Hall, 2011, 111). Starling's law is possible due to the increased stretch in cardiac muscle up to a certain length when filling increases, which in turn increases the contractile force of the heart, resulting in higher ejection volumes (Shiels, 2008, 2005). In Dr. Shiels and Whites research on the Frank-Starling law in other types of animals,

they found that the frog heart is able to develop more force and perform cardiac functions due to longer optimal stretch volumes than mammals (Shiels, 2008, 2009). This increased stretch volume, according to the Frank-Starling law, would allow for higher venous return, cardiac muscle stretching, and increased cardiac output (Shiels, 2008, 2009).

Vagal Stimulation of the Heart

Many functions of the heart are controlled by both sympathetic and sympathetic effects controlled by nerves that innervate the heart (Hall, 2011, 112). In this experiment, the effects of parasympathetic effects of the heart was observed by stimulating the vagus nerve in order to observe three cardiac states; bradycardia, cardiac arrest, and vagal escape. As seen in table 3, during bradycardia, a decrease in heart rate (~21bpm) compared to baseline (32bpm) was observed. This is due to continuous vagal stimulation, which exerts inhibitory effects on the heart (Vasalle, 1985, 36B). During stimulation of the vagus nerve, an increase in acetylcholine increases the influx of potassium, making it more difficult for the dominant pacemaker, the SV node, to reach threshold as often (Vasalle, 1985, 36B). Due to less thresholds being able to be reached by the SV, less pacemaker action potentials will be fired and as a result, a decrease in heart rate will occur. Should vagal stimulation occur even further past this point, complete inhibition of the primary pacemaker will eventually occur due to no depolarizations being able to be reached, resulting in cardiac arrest (Vasalle, 1985, 36B). Despite cardiac arrest being induced, a physiological mechanism known as vagal escape was observed during the lab. After approximately 60 seconds of cardiac arrest at the same voltage that induced cardiac arrest, a gradual return of heart rate to near baseline levels occurred. Vagal escape is possible due nerve innervation not being uniform throughout the primary and other pacemakers (Vasalle, 1985,

36B). In Dr. Vasalle's research on inhibitory mechanisms of a dog heart, maximum sympathetic stimulation increased the heart rate of the primary pacemaker to 180 beats/min but the secondary pacemakers only to 60 beats/min (Vasalle, 1985, 36B). Although his research was done on the sympathetic effects on the heart, similar principles can be applied to the parasympathetic nervous system. Because the innervation is not uniform throughout the heart, secondary pacemaker activity can develop in another part of the heart, allowing for a gradual return of the heart rate, a mechanism known as vagal escape (Wallace, et al., 1964, 93). During both bradycardia and vagal escape, an increase in ventricular contractile force was observed from baseline. This may be due to hypertension occurring in response to the slower heart rates during these two cardiac states, allowing more time for ventricular filling, resulting in greater stretch of the cardiac muscles, and an increased force of contraction, per the Frank-Starling law of the heart (Ghasemi, et al., 2010, 116).

Effects of Epinephrine on the Heart

Sympathetic control of the heart was observed during the experiment by applying epinephrine, the primary neurotransmitter for sympathetic control onto the heart. As expected, both heart rate and ventricular contractile force increased from baseline. According to Dr. Hennacy's research on the effects of epinephrine on frog ventricles, epinephrine has the possibility of increasing cardiac output by increasing end-diastolic volume (EDV), decreasing end-systolic volume (ESV), and increasing the heart rate (Hennacy, 1960, 831). These cardiac effects are possible due to factors such as Ca^{+} concentration increasing within the heart, allowing for not only an increased number of pacemaker potentials to be reached but also increases in the cardiac output (Blank, et al., 2002, 1884).