CHAPTER II

LITERATURE REVIEW

Heart rate variability (HRV), that is, the amount of heart rate fluctuations around the mean hear rate, can be used as a mirror of the cardiorespiratory control system. It is a valuable tool to investigate the sympathetic and parasympathetic function of the autonomic nervous system. HRV measurements are easy to perform, are noninvasive, and have good reproducibility if used under standardized conditions (Kleiger et al,1991; Grossman et al, 1991).

The clinical relevance of HRV was first appreciated in 1965 when Hon and Lee note that fetal distress was preceded by alterations in interbeat intervals before any appreciable change occurred in heart rate itself. Twenty ago, Sayers, 1973 and Penaz et al,1968 focused attention on the existence of physiological rhythms imbedded I beat-to-beat heart rate signal. During the 1970, Ewing et al devised a number of simple bedside tests of short-term RR differences to detect autonomic neuropathy in diabetic patients. The association of higher risk of postinfarction mortality with reduced HRV was first show by Wolf et al in 1977. In 1981, Akselrod et al introduced power spectral analysis of RR interval fluctuation to quantitatively evaluate beat-to-beat cardiovascular control.

These frequency domain analyses contributed to the understanding of autonomic background of RR interval fluctuations in the heart rate record (Pomeranz et al 1985; Pagani et al 1986). The clinical importance of HRV has become appreciated in the late 1980, when it was confirmed that HRV was a strong and independent predictor of mortality after an acute myocardial infarction (Kleiger et al, 1987; Bigger et al, 1992).

1.Physiological Basis

Variation in heart rate is a physiological sign of the adaptive adjustments of the body to changes of internal and external environments. Such adjustments are vital for the body to face the challenge of every minute of life. Abnormality in HRV is often associated with malfunction of the body's adaptive adjustment. Measurement of HRV may provide a probe to physiological and/or pathophysiological adjustment of the body, especially to the mechanism of cardiovascular regulation.

The cardiovascular system transports blood to all organs of the body. It takes part in the maintenance of homeostasis by constantly altering the pump output of the heart, changing the diameter of resistance vessels or altering the amount of blood pooled in capacitance vessels. The pump output of the heart is altered by a beat-by-beat adjustment in it pump rate and force. Such a constant regulation of cardiovascular system provides the physiological basis for HRV and variation of other cardiac variables. The mechanism of cardiovascular regulation is complex. There are not only multiple control factors but also many complex feedback loops to regulate various parts of cardiovascular system and a complex interaction among different regulations.

1.1 Intrinsic Heart rate and its variation

Normally, heart rate is determined by the electrical discharge rate of the sinoatrial node and by the electrical conduction velocity in various parts of heart tissue.

The sinoatrial node is an anatomical region of the heart. In the fully developed heart, the sinoatrial node lies laterally in the epicardial groove of the sulcus terminalis. In the neonate and infant, the region of the sinoatrial node may extend more caudally toward the inferior vena cava (Michael, 1999). In the zone of the sinoatrial node, there are many cell nests. The major cell nest contains cell that are called "P" cell due to their pale appearance on electron micrographs. The p cell can spontaneously depolarize itself and initiate the electrical activation of the heart; hence it is called the pacemaker cell. The spontaneous firing rate of the sinoatrial node with no influence

from other factors is called the "intrinsic heart rate". The intrinsic heart rate may vary due to changes of discharge rate of the sinoatrial node or to the shift of the pacemaker cell within the sinoatrial node. The discharge rate of the sinoatrial node depends on the basic properties of pacemaker cells. There are four basic properties for all of cardiac tissue: automaticity, excitability, refractoriness, and conduction. Automaticity is the ability of a myocardial cell to initiate action potentials spontaneously. When the resting potential, channels in cell membrane open to the threshold potential, channels on the cell membrane open to let different kinds of ions go in or out of the cell and produce action potential. Due to changes in the ions channel, the pacemaker cell raises the transmembrane potential spontaneously. The rate of discharge is determined by threshold potentials, maximum resting potential and the rising rate of transmembrane potential. Variation in these three variables causes variability in the intrinsic heart rate. Pacemaker cells differ in their firing rates. Pacemaker cell in the head of the sinoatrial node have the highest firing rate, while pacemaker cells in tail of the sinoatrial node have lowest firing rate (Michael, 1996). Shifts by pacemaker cells within the sinoatrial node have been observed to occur cyclically throughout the day. Shifting pacemaker cells in sinoatrial node may also cause variation in the intrinsic heart rate.

Although the firing rate of the sinoatrial node is a major factor in determining normal intrinsic heart rate, the variation in velocity of electrical conduction within the sinoatrial node, from the sinoatrial node to the surrounding atrial tissues, within the atrium, from the atrium to the atrioventricular node, from atrioventricular node to the His- Purkinje system and to the ventricular myocardial cells, and within the ventricle- all of these variations may alter the intrinsic heart rate.

1.2 The Autonomic Nervous System and Heart Rate Variability

Besides variation in the intrinsic heart rate, changes in HRV result primarily from the regulation of heart rate by autonomic neural controls and other extrinsic factors.

The autonomic nervous system innervates the internal organs to modulate and control visceral functions of the body such as blood circulation, gastrointestinal motility, secretion, urinary output, and body temperature. The system is organized on the basis of the reflex area. Signals initiated within visceral receptors are relayed via afferent autonomic pathways to the central nervous system, integrated within it at various levels, and transmitted via efferent pathway to visceral effectors. The central part of this reflex is located in the spinal cord, brain stem, and hypothalamus. The hypothalamus is connected to the cortex of the brain via many loops. Based on their functions, the peripheral portions of the autonomic nervous system are classified into the afferent or efferent pathway (David and Lois, 1997). Anatomically, autonomic outflow is divided into 2 components: sympathetic and parasympathetic divisions. Figure 1 shows the anatomy of sympathetic control of the circulation. Sympathetic vasomotor nerve fibers leave the spinal cord through all the thoracic and the first one to two lumbar spinal nerves. They pass into the sympathetic chain and thence by two routes to the circulation: (1) through specific sympathetic nerves that innervate mainly the vasculature of the internal viscera and (2) through the spinal nerves that innervate mainly the vasculature of the peripheral areas (Gayton and Hall, 1996). Figure 2, shows the distribution of sympathetic nerve fiber to the blood vessels, demonstrating that all the vessels except of the capillaries, precapillary sphincters, and most of the metarterioles are innervated.

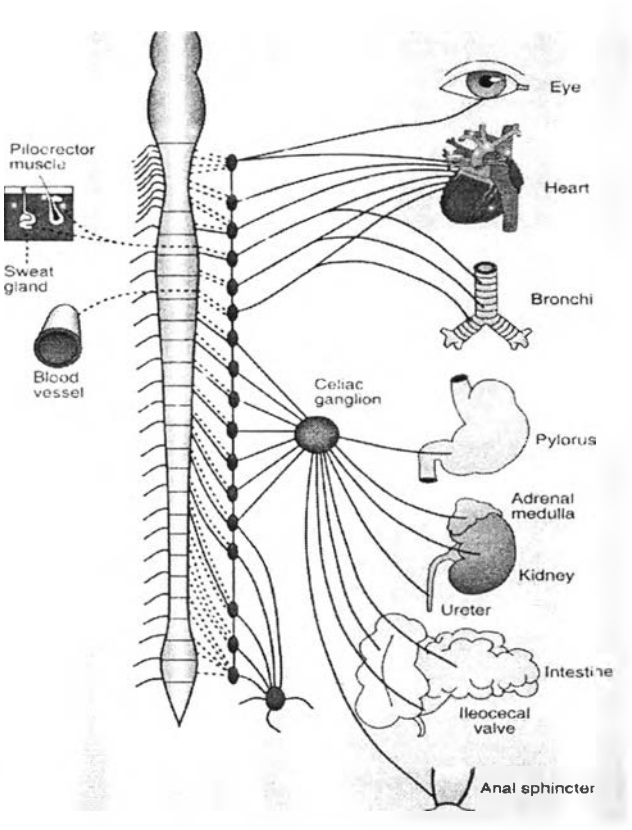

Figure 1. Anatomy of sympathetic nervous control of the circulation.

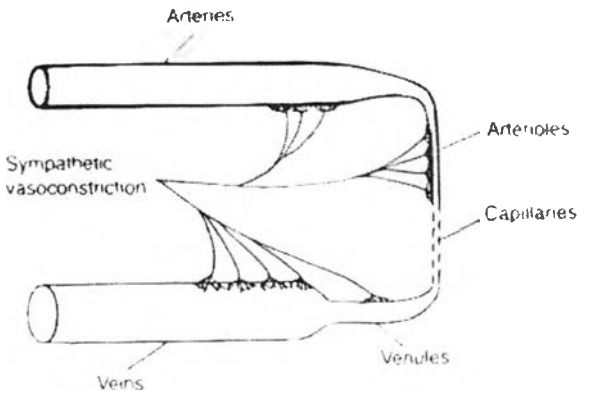

Figure 2. Sympathetic innervations of the systemic circulation.

The innervation of the small arteries and arterioles allows sympathetic stimulation to increase the resistance and thereby to decrease the blood flow through the tissues.

The innervation of large vessels, particularly of the veins, make it possible for sympathetic stimulation to decrease the volume of these vessels and thereby to alter the volume of the peripheral circulatory system. This can translocate blood into the heart and thereby play a major role in the regulation of cardivascular function (Gayton and Hall, 1996).

In general, mass activation of the sympathetic system prepares the body for intense physical activity in emergencies: the heart rate increase, blood glucose rises, and blood is diverted to the skeletal muscles and away from the visceral organs and skin. The theme of the body's sympathetic division has aptly been summarized in the phase "fight or flight". By contrast, the effects of parasympathetic activation are in many way opposite to the effects of sympathetic response: slowing the heart rate, dilating visceral blood vessels, and increasing the activity of the digestive tract.

Peripheral motor portion of the autonomic nervous system are made up of preganglionic and postganglionic neurons. Differing responses of visceral organ to sympathetic and parasympathetic activity are due to the fact that the postganglionic fibers of these two divisions release different neurotransmitters. Acetylcholine is the neurotransmitter of all preganglionic fibers, both sympathetic and parasympathetic. Acetylcholine is also the neurotransmitter by all parasympathetic postganglionic fibers at their synapses with effector cell. Transmission at the autonomic ganglio and the synapses of the postganglionic parasympathetic nerve fibers, is said to be cholinergic. The neurotransmitter released by most postganglionic sympathetic nerve fibers is norepinephrine(noradrenalin) (Gayton and Hall, 1996).

Although the autonomic nervous system has commonly been divided into sympathetic and parasympathetic system, the two divisions often work together in a cooperative integrated action rather than an antagonistic way and cannot be separated

from each other. Interactions between the sympathetic and parasympathetic systems are extremely complex. Two major types of peripheral interactions have been described. The first type is manifested as an accentuated antagonism between the two divisions. The second type is a reciprocal excitation, which means that the peripheral components of one division are activated as a consequence of activity in the other. The heart rate is well supplied with both sympathetic and parasympathetic nerves of autonomic nervous system. The distribution of sympathetic and parasympathetic nerve fibers within the myocardium is illustrated in Figure 3. The atria are supplied with large numbers of sympathetic and parasympathetic neurons, whereas the ventricles receive sympathetic fibers almost exclusively. These affect cardiac pumping in two ways, by changing the heart rate and by changing the strength of the cardiac contractions. In general, activation of sympathetic nerves increases the heart rate (chronotropic effect) and the force of cardiac contraction (inotropic effect). Activation of parasympathetic nerves decreases the heart rate and the force of cardiac contraction. The autonomic nervous system modulates the heart rate either by changing the firing rate of the sinoatrial node or by changing conduction velocity within various parts of the heart.

Acetylcholine released from parasympathetic nerve endings takes effect on its muscarimic receptors at the surface of cardiac cell. It increases the cell membrane's K^+ conductance, inhibits the hyperpolarization activated "pacemaker" current, and finally, hyperpolarizes the SA node. These previous well reduce reducing the rate of spontaneous firing and the velocity of conduction (Michael, 1999).

Epinephrine and norepinephrine released from sympathetic endings activated beta-adrenergic receptor. This action results in acceleration of diastolic depolarization. Sympathetic activation increases the rate of spontaneous firing and the velocity of conduction. Due to the rapid hydrolysis of acetylcholine, the effect of parasympathetic nerves or vagal nerves on heart rate is brief and fast. Norepinephrine is slowly

hydrolyzed, so the effect of sympathetic nerves on heart rate lasts longer (David and Lois, 1997).

Under resting conditions, vagal tone prevails and variation in heart period is largely dependent on vagal modulation. The vagal and sympathetic activity constantly interacts. Because the SA node is rich in acetylcholinesterase, the effect of any vagal impulse is brief because the acetylcholine is rapidly hydrolysed. Parasympathetic influences exceed sympathetic effects probably through two independent mechanism: (1) a cholinergically induced reduction of norepinephrine released in response to sympathetic activity and (2) a cholinergic attenuation of the response to a adrenergic stimulus (Malik, 1996).

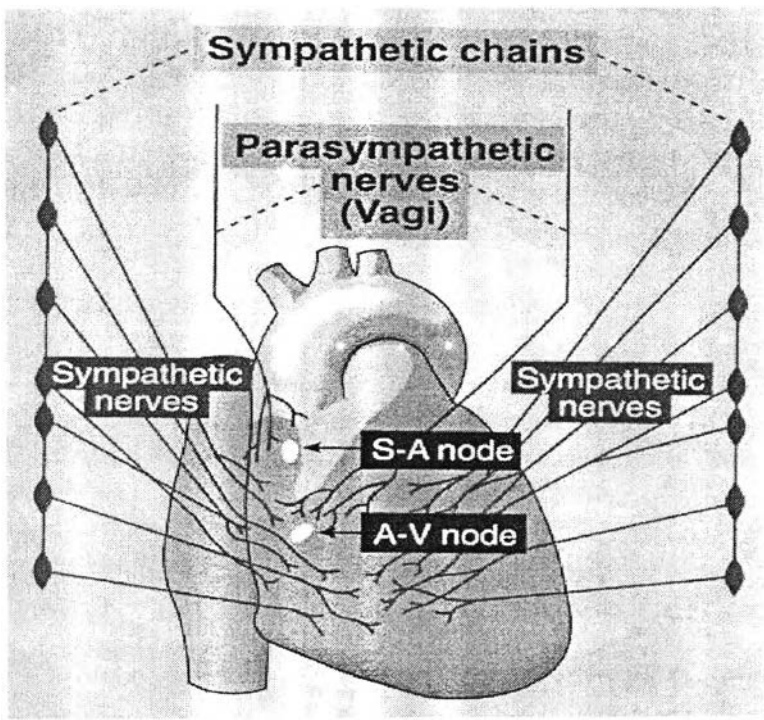

Figure 3. Distribution of sympathetic and parasympathetic nerve fiber within myocardium

1.3 Other Mechanisms of Heart Rate Variability

Many other factors modulate the heart rate and produce HRV. Factors in the sinus node of the heart it self can directly change the firing rate sinus node or the conduction of the heart. These effects are said to be localized. Local factors regulating the heart rate include changes in blood flow to the sinus node, in metabolic substances from the sinus node, in temperature at the sinus node, and in the blood ions (Michael, 1999).

The nervous system and circulating substances can also regulate HRV. Their effects are said to be systematic. The central nervous system of the body plays important role in HRV. Mental stress and many emotional as well as physical behaviors often cause remarkable changes in heart rate. These effects may take place through the complex connection between cortex of the brain and the high autonomic control centers such as the hypothalamus. Many circulation factors are cardiovascular active substances, such as kinins, endothelin, atrial natriuretic peptide, norepinephrine, epinephrin, angiotensin II, and thyriod hormones. Most of these substances have been found to cause changes in the heart rate either by a direct effect on their receptors at the heart rate or by direct effect of various regulation reflexes.

Cardiovascular regulation effects that result in HRV include carotid sinus and arch baroreflexes, atrial stretch receptor reflex, Bainbridge reflex, left ventricular receptor reflex, pulmonary chemoreceptor reflex, and somatosympathetic reflex. Baroreceptors are stretch receptor in the wall of the heart and blood vessels. The baroreceptors on the carotid sinus and aortic arch monitor arterial circulation. Changes in blood pressure will change the discharge rate of the baroreceptors. Though the nerve reflex, these changes result in variation in the heart rate and pumping force. Higher blood pressure results in a slowing heart rate, while lower blood pressure in an increasing heart rate. There are two types of stretch receptor in the atria namely type A and B. Type A discharges, primarily during atrial systole; while type B discharges in late diastole. Baroreceptors in the atria monitor the venous circulation. Changes in

central venous pressure result in reflex changes in the heart rate and pumping force. Rapid infusion of blood or saline produces a rise in heart rate. This effect is known as the Bainbridge reflex.

Baroreceptors in the left ventricle may play a role in the maintenance of the vagal tone that keeps the heart rate low at rest. When the left ventricle is distended, there is a fall in systemic arterial pressure and a rise in heart rate.

Chemoreflex also modulates heart rate. In experiment animals, injections of serotonin, veratridine, phenyldiguanide, and some other drugs into the coronary arteries cause apnea followed by rapid breathing, hypotension, and slower heart rate. This effect is known as coronary chemoreflex. A similar response has been observed when these drugs were injected in the pulmonary artery. Pulmonary chemoreflex can be blocked by vagotomy. Chemoreceptor discharge may also contribute to the production of Mayer waves. Mayer wave should not be confused with Traube-Hering waves, which are a variation in blood pressure synchronized with respiration. Mayer waves are slow regular fluctuation in arterial pressure that occur at the rate of about one per zoo to seconds during hypotension. Under these conditions, hypoxia stimulates the chemoreceptors.

Pain can have either a positive or negative influence on arterial pressure. Generally, superficial similar to that associated with the alerting response and perhaps over many of the same pathways. Deep pain from receptors in the viscera or joints, however, often causes a cardiovascular response similar to that which accompanies vasovagal syncope, i.e., decreased sympathetic tone, increased parasympathetic tone, and a serious decrease in blood pressure. This response may contribute to the state of shock that often accompanies crushing injuries and/or joint displacement.

In humans, the heart rate and other cardiovascular variables are under complex and organized regulation from interrelated and interactive mechanism that give rise to the fascinating rhythm of life and HRV. The net effect of a regulating factor is often not the direct effect of that factor. Although it is difficult to separate the effect of single

factor from others, it is possible to identify principle factors in a particular condition by analyzing the characteristics of HRV.

2. Measurement of HRV

2.1Time Domain Methods

The variations in heart rate may be evaluated by a number of methods. Perhaps the simplest to perform are the time domain measures. In these methods, either the heart rate at any point in time oı the intervals between successive normal complexes are determined. In a continuous electrocardiogram (ECG) record, each QRS complex is detected, and the so-called normal-to-normal (NN) intervals (that is, all intervals between adjacent QRS complexes resulting from sinus node depolarizations) or the instantaneous heart rate is determined. Simple time domain variables that can be calculated include the mean NN interval, the mean heart rate, the difference between the longest and shortest NN interval, the difference between night and day heart rates, and so forth. Other time domain measurements that can be used are variations in instantaneous heart rate secondary to respiration, tilt, valsalva maneuver, or phynylephrine infusion. These differeces can be described as either differences in heart rate or cycle length

Statistical Methods

From a series of instantaneous heart rate or cycle intervals, particularly those recorded over longer periods, traditionally 24 hours, more complex statistical time domain measures can be calculated. These may be divided into two classes: (1) those derived from direct measurements of the NN intervals or instantaneous heart rate and (2) those derived from the differences between NN intervals. These variables may be derived from analysis of the total ECG recording or may be calculated using smaller segments of the recording period. The latter method allows comparison of HRV to be made during varying activities, for example, rest, sleep, and so on.

The simplest variable to calculate is the standard deviation of the NN intervals (SDNN), that is, the square root of variance. Since variance is mathematically equal to total power of spectral analysis, SDNN reflects all the cyclic components responsible for variability in the period of recording. In many studies SDNN is calculated over a 24-hour period and thus encompasses short-term HF variations as well as the lowest-frequency components seen in a 24-hour period. As the period of monitoring decreases, SDNN estimates shorter and shorter cycle lengths. It also should be noted that the total variance of HRV increases with the length of analyzed recording. Thus, on arbitrarily selected ECG, SDNN is not a well-defined statistical quantity because of its dependence on the length of recording period. In practice, it is inappropriate to compare SDNN measures obtained from recordings of different durations. On the contrary, durations of the recordings used to determine SDNN values (and similarly other HRV measures) standardized. As discussed further in this document, short-term 5-minute recordings and nominal 24-hour long-term recordings appear to be appropriate options.

Other commonly used statistical variables calculated from segments of the total monitoring period include SDANN, the standard deviation of the average NN intervals calculated over short periods, usually 5 minutes, which is an estimated of the changes in heart rate due to cycles longer than 5 minutes, and the SDNN index, the mean of the 5-minutes standard deviations of NN intervals calculated over 24 hours, which measures the variability due to cycles shorter than 5 minutes.

The most commonly used measures derived from interval differences include RMSSD, the square root of the mean squared differences of successive NN intervals, NN50, the number of interval differences of successive NN intervals greater than 50 ms, and pNN50, the proportion derived by dividing NN 50 by the total number of NN intervals. All of these measurements of short-term variation estimate high frequency variation in heart rate and thus are highly correlated (Fig4) (Malik, 1996).

Table 1. Selected Time Domain Measures of HRV

Variable	Units	Description
		Statistical Measures
SDNN	ms	Standard deviation of all NN intervals
SDANN	ms	Standard deviation of the averages of NN interval in all 5 – minute segments of the entire recording
RMSSD	ms	The square root of the mean of the sum of the squares of differences between adjacent NN intervals
SDNN index	ms	Mean of the standard deviations all NN intervals for all 5-minute segments of the entire recording
pNN50	%	NN50 count divided by the total number of all NN interval

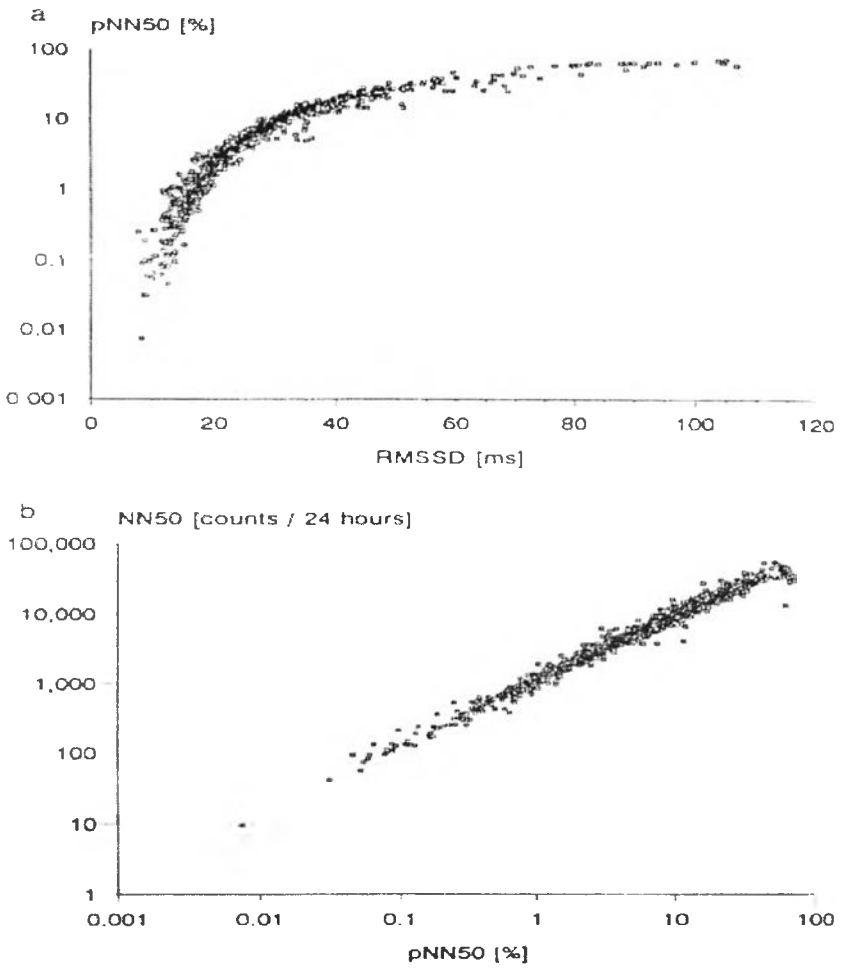

Figure 4. Relationship between the RMSSD and pNN50 (a) and pNN50 and NN50 (b) measures of HRV assessed from 857 nominal 24-hour Holter tapes recorded in survivors of acute myocardial infarction before hospital discharge. (Malik, 1996)

2.2 Frequency Domain Methods

Various spectral methods for the analysis of the tachogram have been applied since the late 1960s. Power spectral density (PSD) analysis provides the basic information of how power (variance) distributes as a function of frequency. Independent of the method used, proper mathematical algorithms can obtain only an estimate of true PSD of the signal.

Methods for the calculation of PSD may be generally classified as nonparametric and parametric. In most instances, both methods provide comparable results. The advantages of the nonparametric methods are (1) the simplicity of the algorithm used (fast Fourier transform [FFT] in most of the cases) and (2) the high processing speed, while the advantages of parametric methods are (1) smoother spectral components that can be distinguished independent of preselected frequency bands, (2) easy postprocessing of the spectrum with an automatic calculation of low – and high – frequency power components with an easy identification of the central frequency of each component, and (3) an accurate estimation of PSD even on a small number of samples on which the signal is supposed to maintain stationarity. The basic disadvantage of parametric methods is the need of verification of the suitability of the chosen model and of its complexity (that is, the order of the model).

Spectral components

Short – term recordings

Three main spectral components are distinguished in a spectrum calculated from short – term recording of 2 to 5 minutes (Pagani et al 1986; Malliani et al, 1991). VLF, LF and HF components. The distribution of the power and the central frequency of LF and HF are not fixed but may vary in relation to changes in automatic modulations of heart period (Fyrlan et al, 1990; Malliani et al, 1991). The physiological explanation of the VLF component is much less defined, and the existence of a specific physiological process attributable to these heart period changes

might even be questioned. The nonharmonic component, which does not have coherent properties and is affected by algorithms of baseline or trend removal, is commonly accepted as a major constituent of VLF. Thus, VLF assessed from short – term recordings (< 5 minutes) is a dubious measure and should be avoided when the PSD of short – term ECGs is interpreted.

The measurement of VLF, LF and HF power components is usually made in absolute values of power (milliseconds squared). LF and HF may also be measured in normalized units, which represent the relative value of each power component in proportion to the total power minus the VLF component (Pagani et al 1986; Malliani et al, 1991). The representation of LF and HF in normalized units emphasizes the controlled and balanced behavior of the two branches of the autonomic nervous system. Moreover, the normalization tends to minimize the effect of the changes in total power on the values of LF and HF components (Fig 5). Nevertheless, normalized units should always be quoted with absolute values of the LF and HF power in order to describe completely the distribution of power in spectral components.

Long – term recordings

Spectral analysis also may be used to analyze the sequence of NN intervals of the entire 24 – hour period. The result then includes a ULF component, in addition. the slope of 24 – hour spectrum also can be assessed on a log – log scale by linear fitting the spectral values. Table 2 lists selected frequency domain measures.

The problem of " stationarity" is frequently discussed with long-term recording. If mechanism responsible for heart period modulations of a certain frequency remains unchanged during the whole period of recording, the corresponding frequency component of HRV may be used as a measure of these modulations. If the modulations are not stable, the interpretation of the results of frequency analysis is less well defined. In particular, physiological mechanism of heart period modulations responsible for LF and HF power components cannot be considered stationary during

the 24 – hour period (Furlan et al, 1990). Thus, spectral analysis performed on the entire 24 – hour period as well as spectral results obtained from shorter segments (5 minutes) averaged over the entire 24 – hour period (the LF and HF results of these two computation are not different) provide averages of the modulation attributable to the LF and HF components (Fig 6). Such averages obscure the detailed informations about autonomic modulation of RR interval that is available in shorter recordings (Furlan et al, 1990). It should be remembered that the components of HRV provide measurement of the level of autonomic tone (Malik et al, 1993), and averages of modulations do not represent an averaged level of tone.

Table 2 Selected Frequency Domain Measures of HRV

Variable	Units	Description	Frequency Range
		Analysis of Short – term Recordings (5 min)	
5-min total power	ms^2	The variance of NN interval over the temporal segment	~≤0.4 Hz
VLF	ms^2	Power in VLF range	≤0.04 Hz
LF	ms^2	Power in LF range	0.04 – 0.15 Hz
LF norm	nu	LF power in normalized units LF/(total power - VLF) x 100	
HF	ms^2	Power in HF range	0.15-0.4 Hz
HF norm	ms^2	HF power in normalized units HF/(total – VLF) x 100	
LF/HF		Ratio LF[ms^2]/HF[ms^2]	
		Analysis of Entire 24 Hours	
Total power	ms^2	Variance of all NN interval	~≤0.4 Hz
ULF	ms^2	Power in the ULF range	≤0.003 Hz
VLF	ms^2	Power in the LF range	0.003 – 0.04 Hz
LF	ms^2	Power in the LF range	0.04-0.15 Hz
HF	ms^2	Power in the HF range	0.15-0.4 Hz
α		Slop of the linear interpolation of the spectrum in a log-log scale	~≤0.04 Hz

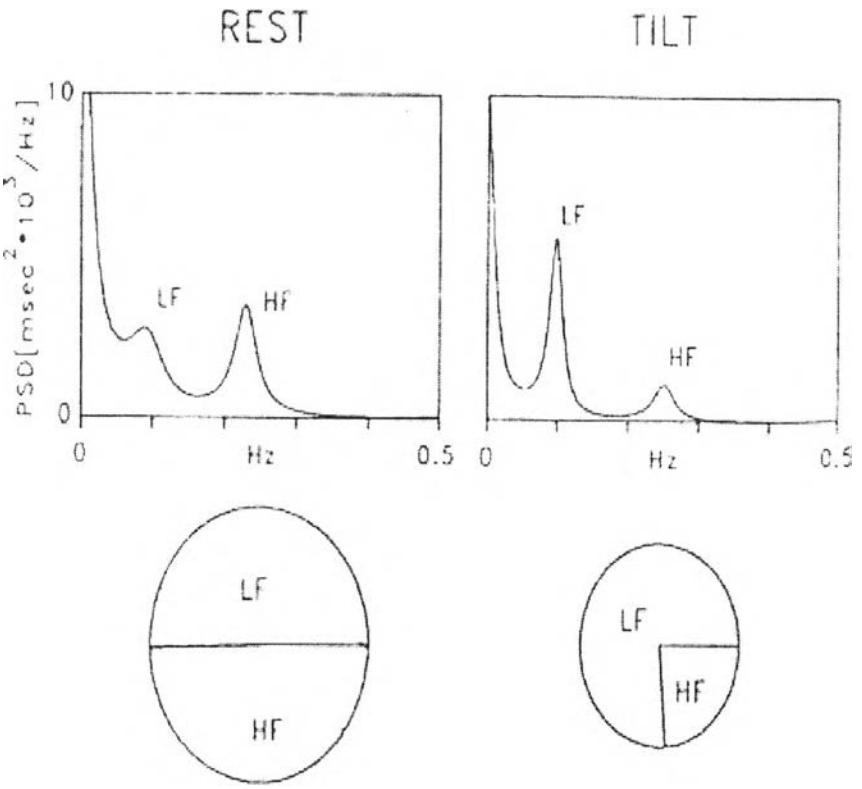

Fig 5. Spectral analysis (autoregressive model, order 12) or RR interval variability in a healthy at rest and during 90° head – up tilt. At rest, two major components of similar power are detectable at low and high frequencies. During tilt, the LF component becomes dominant, but as total variance is reduced, the absolute power of LF appears unchanged compared with rest. Normalization procedure leads to predominant LF and smaller HF component, which express the alteration of spectral components due to tilt. Pie charts show the relative distribution together with the absolute power of the two components represented by the area. During rest, the total variance of the spectrum was 1201 ms2, and its VLF, LF, and HF components were 586 ms2, 310 ms2, and 302 ms2, respectively. Expressed in normalized units (nu), the LF and HF were 48.95 and 47.78 nu, respectively. The LF/HF ratio was 1.02. During tilt, the total variance was 671 ms2, and its VLF, LF and HF components were 265 ms2, 308 ms2, and 95 ms2, respectively. The LF and HF were 75.96, and 23.48 nu, respectively. The LF/HF ratio was 3.34. Thus, note that for instance, the absolute power of the LF component was slightly decreased during tilt while the normalized units of LF were substantially increased.

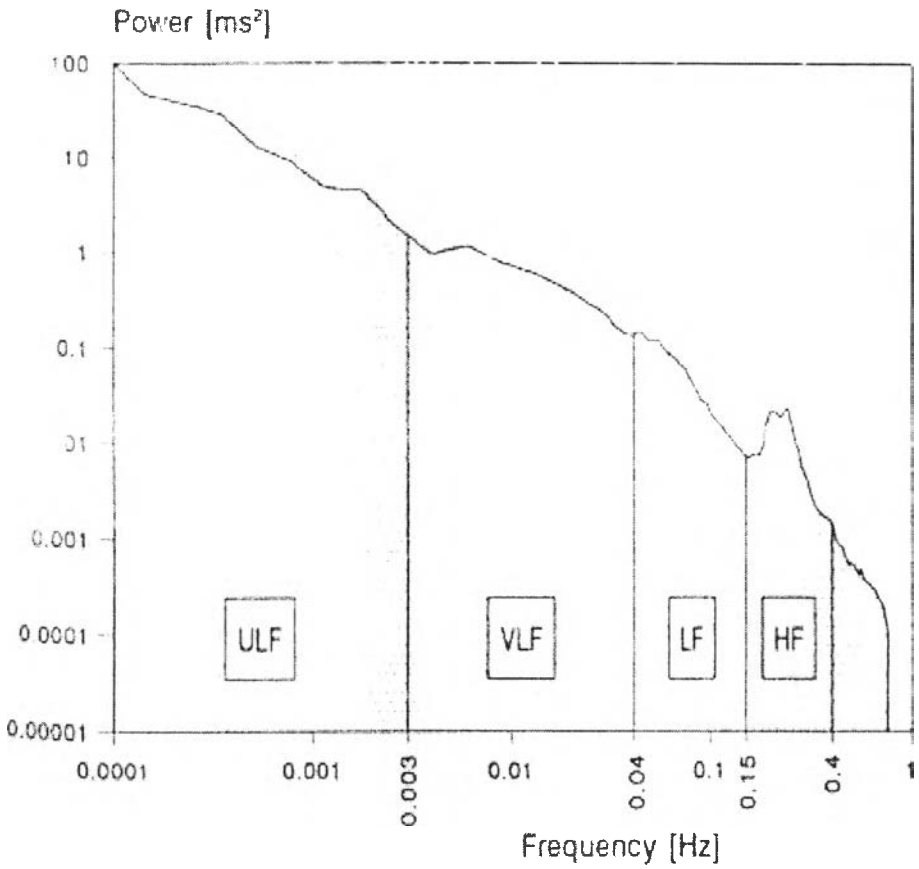

Fig 6. Example of an estimate of power spectral density obtained from the entire 24 – hour interval of a long-term Holter recording . Only the LF and HF components correspond to peaks of the spectrum, while the VLF and ULF can be approximated by a line in this plot with logarithmic scales on both axes. The slope of such a line is the α measure of HRV.

3. Technical Requirements and Recommendations

Because of the important differences in the interpretation of the results, the spectral analyses of short-term and long-term ECGs should always be strictly distinguished, as reported in Table 2.

The analyzed ECG signal should satisfy several requirements in order to obtain a reliable spectral estimation. Any departure from the following requirements may lead to unreproducible results that are difficult to interpret.

To attribute individual spectral components to well defined physiological mechanism, such mechanisms modulating the heart rate should not change during the recording. Transient physiological phenomena may perhaps be analyzed by specific methods that currently constitute a challenging research topic but are not yet ready to be used in applied research. To check the stability of the signal in terms of certain spectral components, traditional statistical tests may be used.

The sampling rate must be properly chosen. A low sampling rate may produce a jitter in the estimation of the R-wave fiducial point, which alters the spectrum considerably. The optimal range is 250 to 500 Hz or perhaps even higher (Malik, 1996), while a lower sampling rate (in any case > 100 Hz) may behave satisfactorily only if an algorithm of interpolation (parabolic) is used to refine the R-wave fiducial point (Malik, 1996).

Baseline and trend removal (if used) may affect the lower component in the spectrum. It is advisable to check the frequency response of the filter or the behavior of the regression algorithm and to verify that the spectral components of interest are not significantly affected.

The choice of QRS fiducial point may be critical. It is necessary to use a well-tested algorithm (derivative plus threshold, template, correlation method) to locate a stable and noise-independent reference point. A fiducial point localized far within the QRS complex may also be influence by varying ventricular conduction disturbances.

Ectopic beats, arrhythmic events, missing data, and noise effects may alter the estimation of the PSD of HRV. Proper interpolation (or linear regression or similar algorithms) on preceding/successive beats on the HRV signal or on its autocorrelation function may decrease this error. Preferentially, short-term recordings that are free of ectopy, missing data, and noise should be used. In some circumstances, however, acceptance of only ectopic-free, short-term recordings may introduce significant selection bias. In such cases, proper interoperation should be used and the possibility of the results being influenced by ectopy should be considered. The relative number and relative duration of RR intervals that were omitted and interpolated should also be quoted.

4. Algorithmic Standards and Recommendations

The series of data subjected to spectral analysis can be obtained in different ways. A useful pictorial representation of the data is the discrete event series (DES), that is, the plot of R_iR_{i-1} interval versus time (indicated at R_i occurrence), which is an irregularly time-sampled signal. Nevertheless, spectral analysis of the sequence of instantaneous heart rate has also been used in many studies.

The spectrum of the HRV signal is generally calculated either from the RR interval tachogram (RR durations versus number of progressive beats; see Fig 5a,b) or by interpolating the DES, thus obtaining a continuous signal as a function of time, or by calculating the spectrum of the counts-unitary pulses as a function of time corresponding to each recognized QRS complex. Such a choice may have implication on the morphology, the measurement units of the spectra, and the measurement of the relevant spectral parameters. To standardize the methods used, the use of RR interval tachogram with the parametric method, or the use of the regularly sampled interpolation of DES with the nonparametric method may be suggested; nevertheless, regularly sampled interpolation of DES is also suitable for parametric methods. The

sampling frequency of interpolation of DES must be sufficiently high that the Nyquist frequency of the spectrum is not within the frequency range of interest.

Standards for nonparametric methods (based on the FFT algorithm) should include the values in Table 2, the formula of DES interpolation, the frequency of sampling the DES interpolation, the number of sample used for the spectrum calculation, and the spectral window used (Hann, Hamming, and triangular windows). The method of calculating the power in respect of the window also should be quoted. In addition to requirements described in other parts of this document, each study using the nonparametric spectral analysis of HRV should quote all these parameters.

Standards for parametric methods shall include the values reported in Table 2, the type of the model used, the number of samples, the central frequency for each spectral component (LH and HF), and the value of the model order (numbers of parametrics). Furthermore, statistical figures must be calculated in order to test the reliability of the model. The prediction error whiteness test (PEWT) provides information about goodness of the fitting model, while the optimal order test (OOT) checks the suitability of the order of the model used. There are different possibilities of performing OOT that include final prediction error and Akaike information criteria. The following operative criterion for choosing the order P of and autoregressive model might be proposed: the order shall be the range of 8 to 20, fulfilling the PEWT test and complying with the OOT test (Malik, 1996).

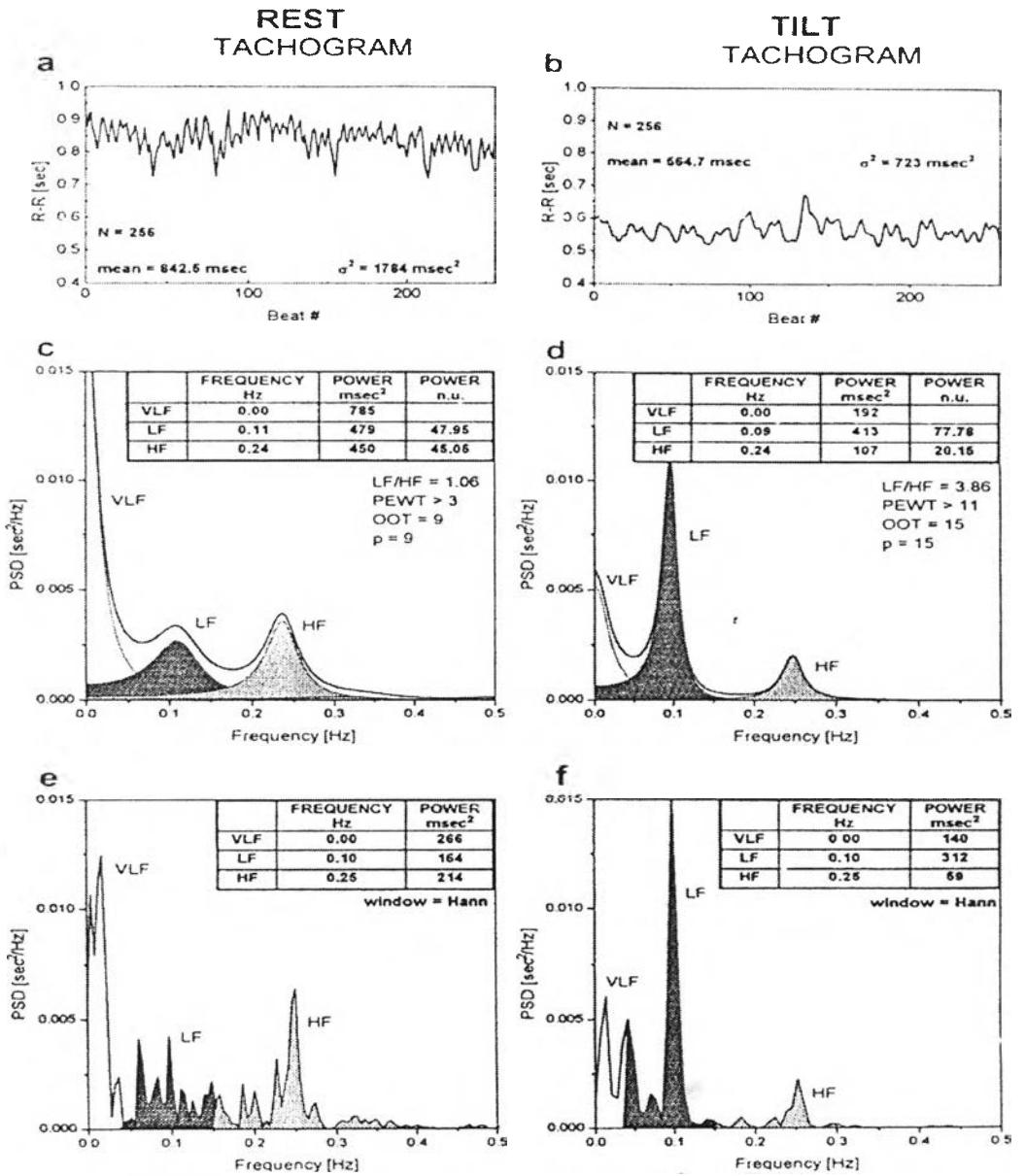

Fig 7. Interval tachogram of 256 consecutive RR values in a normal subject at supine rest (a) and after head-up tilt (b). the HRV spectra are shown, calculated by parametric autoregressive modeling (c and d) and by a fast Fourier transform-based nonparametric algorithm (e and f). mean values (m), variances (s2), and the number (N) of samples are indicated. For c and d, VLF, LF, and HF central frequency, power in absolute value and power in normalized units (n.u.) are also indicated together with order p of the chosen model and minimal values of the prediction error whiteness test (PEWT) and optimal order test (OOT) that satisfy the tests. In e and f, the peak frequency and the power of VLF, LF, and HF were calculated by integrating the power spectral density (PSD) in the defined frequency bands. The window type is also specified. In c through f, the LF component is indicated by dark shaded areas and the HF component by light shaded areas.

5. Recording Requirements

5.1 ECG Signal

The fiducial point recognized on the ECG tracing that identifies a QRS complex may be based on the maximum or baricentrum of the complex, on the determination of the maximum of an interpolating curve, or found by matching with a template or other event markers. To localize the fiducial point, voluntary standards for diagnostic ECG equipment are satisfactory in term of signal-to-noise ratio, common mode rejection, bandwidth, and so forth. An upper-band frequency cutoff substantially lower than that established for diagnostic equipment (~200 Hz) may create a jitter in the recognition of the QRS complex fiducial point, introducing an error of measured RR intervals. Similarly, limited sampling rate induces an error in the HRV spectrum that increases with frequency, thus affecting more high frequency. An interpolation of the under sampled ECG signal may decrease this error. With proper interpolation, even a 100 Hz sampling rate can be sufficient.

When solid state storage recorders are used, data compression techniques must be carefully considered in term of both the effective sampling rate and the quality of reconstruction methods that may yield amplitude and phase distortion (Kennedy et al, 1992).

5.2 Duration and circumstances of ECG Recording

In studies researching HRV, the duration of recording is dictated by the nature of each investigation. Standardization is needed particularly in studies investigating the physiological and clinical potential of HRV. Frequency domain methods should be preferred to the time domain methods when short-term recordings are investigated. The recording should last for at least 10 times the wavelength of the lower frequency bound of the investigated comment, and in order to ensure the stability of the signal, should not be substantially extended. Thus, recording of approximately 1 minute is needed to assess the HF components of HRV, while approximately 2 minutes are

needed to address the LF component. To standardize different studies investigating short-term HRV, 5-minute recordings of a stationary system are preferred unless the nature of the study dictates another design.

Averaging of spectral components obtained from sequential periods of time is able to minimize the error imposed by the analysis of very short segments. Nevertheless, if the nature and degree of physiological heart period modulations changes from one short segment of the recording to another, the physiological interpretation, of such averaged spectral components suffers from the same intrinsic problems as that of the spectral analysis of long-term recordings and warrants further elucidation. A display of stacked series of sequential power spectra (for example, over 20 minutes) may help confirm steady state for a given physiological state.

Although the time domain methods, especially the SDNN and RMSSD methods, can be used to investigate recordings of short durations, the frequency methods are usually able to provide results that are more easily interpretable in term of physiological regulations. In general, the time domain methods are ideal for the analysis of long-term recordings (the lower stability of heart rate modulations during long-term recordings makes the results of frequency methods less easily interpretable). The experience shows that a substantial part of the long-term HRV value is contributed by the day-night differences. Thus, the long-term recording analyzed by the time domain methods should contain at least 18 hours of analyzable ECG data that include the whole night.

Little is known about the effects of the environment (type and nature of physical activity and emotional circumstances) during long-term ECG recordings. For some experimental designs, environmental variables should be controlled and in each study, the character of the environment also should ensure that the recording environment of individual subjects is similar. In physiological studies comparing HRV in different well-defined groups, the differences between underlying heart rate also should be properly acknowledged.

5.3 Editing of the RR Interval Sequence

The errors imposed by the imprecision of the NN interval sequence are known to affect substantially the results of statistical time domain and all frequency domain methods. It is known that casual editing of the RR interval data is sufficient for the approximate assessment of total HRV by the geometric methods, but it is not known how precise the editing should be to ensure correct results from other methods. Thus, when the statistical time domain and/or frequency domain methods are used, the manual editing of RR data should be performed to a very high standard, ensuring correct identification and classification of every QRS complex. Automatic "filters" that exclude some interval from the original RR sequence (for example, those differing by more than 20% from the previous interval) should not replace manual editing because they are know to behave unsatisfactorily and to have undesirable effects leading potentially (Malik, 1996).

6. Components of HRV

The RR interval variations present during resting conditions represent a fine-tuning of the beat-to-beat control mechanism (Akselrod et al, 1985; Saul et al, 1990). Vagal afferent stimulation leads to reflex excitation of vagal efferent activity and inhibition of sympathetic efferent activity. The opposite reflex effects are mediated by the stimulation of sympathetic afferent activity. Efferent vagal activity also appears to be under "tonic" restraint by cardiac afferent sympathetic activity (Cerati et al, 1991). Efferent sympathetic and vagal activities directed to the sinus node are characterized by discharge largely synchronous with each cardiac cycle that can be modulated by central (vasomotor and respiratory centers) and peripheral (oscillation in arterial pressure and respiratory movements) oscillators (Malliani et al, 1991). These oscillators generate rhythmic fluctuations in efferent neural discharge that manifest as short and long term oscillation in the heart period. Analysis of these rhythms may

permit inferences on the state and function of (a) the central oscillators, (b) the sympathetic and vagal efferent activity, (c) humoral factors, and (d) the sinus node.

An understanding of the modulatory effects of neural mechanisms on the sinus node has been enhanced by spectral analysis of HRV. The efferent vagal activity is a major contributor to the HF component, as seen in clinical and experimental observations of autonomic maneuvers such as electrical vagal stimulation, muscarinic receptor blockade, and vagotomy (Pomeranz et al, 1985; Malliani et al, 1991). More controversial is the interpretation of the LF component, which is considered by some (Montano et al, 1994). As a marker of sympathetic modulation (especially when expressed in normalized units) and by others (Appel et al, 1989). As a parameter that includes both sympathetic and vagal influences. This discrepancy is due to the fact that in some conditions associated with sympathetic excitation, a decrease in the absolute power of the LF component is observed. It is important to recall that during sympathetic activation the resulting tachycardia is usually accompanied by a marked reduction in total power, whereas the reverse occurs during vagal activation. When the spectral components are expressed in absolute units (milliseconds squared), the changes in total power influence LF and HF in the same direction and prevent the appreciation of the fractional distribution of the energy. This explains why in supine subjects under controlled respiration, atropine reduces both LF and HF (Pomeranz et al, 1985) and why during exercise LF is markedly reduced (Malliani et al, 1991).
This concept is exemplified in Fig 3, showing the spectral analysis of HRV in a normal subject during control supine conditions and 90 head-up tilt. Because of the reduction in total power, LF appears as unchanged if considered in absolute units. However, after normalization an increase in LF becomes evident. Similar results apply to the LF/HF ratio (Malliani et al, 1994).

Spectral analysis of 24 –hour recordings (Malliani et al, 1991; Furlan et al, 1990) shows that in normal subjects, LF and HF expressed in normalized units exhibit a circadian pattern and reciprocal fluctuations, with higher values LF in the daytime

and of HF at night. These patterns become undetectable when a single spectrum of the entire 24- hour period is used or when spectra of subsequent shorter segments are averaged. In long- term recordings, the HF and LF component for only approximately 5% of total power. Although the ULF and VLF component for the remaining 95% of total power, their physiological correlates are still unknown. LF and HF can increase under different conditions. An increased LF (expressed in normalized units) is observed during 90 tilt, standing, mental stress, and moderate exercise in healthy subjects, and during moderate hypotention, physical activity, and occlusion of a coronary artery or common carotid arteries in conscious dogs (Rimoldi et al, 1990) conversely, an increase in HF is induced by controlled respiration, cold stimulation of the face, and rotational stimuli (Malliani et al, 1991).

7. Exercise Training and HRV

Exercise training may decrease cardiovascular mortality and sudden cardiac death (Connor et al, 1989). Regular exercise training is also thought capable of modifying the autonomic balance (Furlan et al, 1993; Arai et al, 1989). A recent experimental study designed to assess the effects of exercise training on markers of vagal activity has simultaneously provided information on changes in cardiac electrical stability (Hull et al, 1994). Conscious dogs documented to be at high risk by the previous occurrence of ventricular fibrillation during acute myocardial ischemia were randomly assigned to 6 weeks of either daily exercise training or cage rest followed by exercise training (Hull et al, 1994). After training, HRV (SDNN) increased by 74%, and all animals survived a new ischemic test. Exercise training can also accelerate recovery of the physiological sympathovagal interaction, as shown in post MI patients (La Rovere et al, 1992).

8. Mental Stress and HRV

Analysis of HRV in normal subject during mental stress results in: 1) a reduction in total HRV power; and 2) a shift in sympathetic-parasympathetic balance toward greater sympathetic dominance and parasympathetic withdrawal (Pagani et al, 1991). This withdrawal of parasympathetic influent with the decrease in baroreflex sensitivity during mental stress demonstrated by other investigators (Robbe et al,1987). While multiple studies have suggested that analysis of HRV may be useful as a physiologic indicator mental load, such measurements must be interpreted with caution. Although measurements during sedentary activities may be useful, it must be remembered that concomitant alterations in the pattern of respiration and/or physical activity may also significantly influence HRV measurement (Pagani et al, 1991).

9. Acute Hemodynamic Effects of Dynamic Muscular Training

A dynamic muscular training program involving endurance sports (running cycling etc.) performed predominantly at an intensity of 60%-70% of maximum oxygen uptake (VO_2max) result in a steady rise in oxygen utilization by the peripheral musculature. The increased oxygen requirement of the regional musculature induces an acute vasodilatation of the supplying arterioles and preexisting capillaries. Thereby, arterial vascular resistance falls in the region of dynamically exercised muscle groups, which results in an increase in regional arterial blood flow and higher venous blood backflow from the active muscle groups to the heart. The reduction of local arterial vascular resistance and the increase in venous blood flow leads to a reduction in afterload and to a rise in preload of the heart (Blomguist and Saltin, 1983). Under these conditions, the diastolic filling and systolic emptying of the heart are augmente(Colan et al, 1985), resulting in higher stroke volume. The continuously augmented oxygen demand of the exercising muscle groups with increasing exercise intensity leads to a reduction in vagal tone and an activation of the sympathetic nervous system (Saltin,

1985). The cardiac effects include a rise in heart rate and an increase in myocardial contractility. In the arterial vascular system, selective vasoconstriction occurs in nonactive muscle groups and in the splanchnic bed (unpaired organs of the gastrointestinal tract). This result in a redistribution of blood with a further increment in arterial blood flow to the exercising muscle groups.

Chronic Hemodynamic Effects of Dynamic Muscular Training

Dynamic muscular training for more than 3h per week induces functional adaptations within exercise peripheral muscles. This concerns an improvement intramuscular and intermuscular coordination (e.g., a uniform pedal stroke in cyclists) and consequently an oxygen requirement reduction at constant exercise intensity. This adaptation results in low blood levels of epinephrine and norepinephrine(Lehmann et al, 1984) with a reduction in sympathetic activity of heart at rest and at comparable levels of exertion (Keul et al, 1982). The degree of this functional adaptation of the heart is limited. Thus, an enhancement in duration of dynamic muscular exercise (to more than 5h per week) and the involvement of more than 1/6 th of the total skeletal muscle mass leads to structural cardiac adaptations. Eccentric myocardial hypertrophy with an increase in all cardiac chambers dimensions can be demonstrated. Longitudinal studies have shown that a 20% increase in cardiac chamber diameters and a 70%-80% rise in cardiac muscle mass represent the upper limits of structural myocardial adaptations which cannot be exceeded under physiological conditions (Dickhuth et al,1987).

10. Acute Hemodynamic Effects of Static Muscular Training

Static muscular training is mainly performed by power athletes (weightlifters, bodybuilders etc.), but also to some extent in other sport disciplines such as sprinting, jumping and multiple competition (e.g., decathlon). In contrast to a dynamic exercise, which involves a high frequency of muscle contraction and a low level of power, a low

rate of contraction and a high level of power characterize static exercise. The development of power in the involved muscle fibers causes a marked augmentation in intramuscular tone. In turn this leads to mechanical compression of the intramuscular arterial vessels and thus to an acute rise in local arterial resistance.

Furthermore, submaximal to maximal contraction or eccentric muscular contraction resulting form power training demands high levels of acute energy supply, so that anaerobic glycolysis is stimulated. Therefore, an increase lactate acidosis and an activation of the sympathetic activation occur. This leads to a generalized arterial vasocontriction and a further rise in local vascular resistance with in exercise muscle groups is impaired and the venous blood backflow is reduced. The effect on the heart includes an acute increase in afterload, a reduction in preload combined with a cathecholamine-induced increase in heart rate and myocardial contractility (Keul et al, 1981). This lead to impair diastolic filling and consequently to reduced systolic emptying of the heart resulting in a decreased stroke volume (Rost et al, 1983)

Chronic Hemodynamic Effects of Static Muscular Training

To date, only a few studies have investigated the chronic functional adaptation of the cardiovascular system to regular static muscular training. While endurance trained athletes have been shown to have a lower density of beta-receptors than sedentary controls, no similar effect has been found in power-trained athletes (Jost et al, 1989). Echocardiography studies of statically trained athletes have demonstrated moderate structural cardiac adaptations with a tendency towards a concentric myocardial hypertrophy (Longhurst et al, 1981). In contrast endurance trained athletes, who show a disproportional increase in cardiac muscle mass in relation to skeletal muscle mass, a linear correlation between skeletal mass and cardiac muscle mass can be observed in power trained athletes (Longhurst et al, 1980)