CHAPTER 3

RESEARCH METHODOLOGY

3.1. Research questions and Objectives

3.1.1 Research Questions:

- **Primary question**

In children (age 3 years or below) with congestive heart failure caused by congenital and acquired heart diseases, whether Enalapril in addition to conventional therapy, can increase 30% patients with their cardiac contractility improved as compared to conventional therapy alone?

- **Secondary questions:**

(1). Are there any clinical changes between two groups?

(2). What are the side effects in patients treated with enalapril?

3.1.2 Objectives

- **General objectives**

(1). To assess the hemodynamic and clinical effects of enalapril added as an additional drug in the treatment of children with congestive heart failure in conjunction with conventional therapy.

(2). To evaluate the possible side effects of enalapril in pediatric patients.

- **Specific Objectives:**

(1). To evaluate the changes of left ventricular systolic function by measuring the Left Ventricular End-Systolic Wall stress/Rate corrected fiber shortening relation (LVESWS/VCFc RELATION) between two groups.

(2). To observe the clinical and symptomatic changes in terms of weight, heart rate, respiratory rate, heart size (cardiac-thoracic ratio) and liver size.

(3). To monitor the possible side effects Enalapril which may cause dropping blood pressure, electrolyte disorders, severe cough, syncope or even renal failure.

3.1.3 Hypothesis :

Enalapril added as an additional drug in conjunction with conventional therapy has significant beneficial hemodynamic and clinical effects on children with congestive heart failure caused by congenital and acquired heart diseases.

3.2 Conceptual framework

Congestive heart failure in childhood is a clinical syndrome that reflects the inability of the myocardium to meet the metabolic requirements of the body, including those needs incurred by the growth process. In pediatric clinical setting the following issues must be considered before designing a trial to deal with some complicated situation.

First, The nature of status in childhood may arise as a consequence of: excessive work load imposed on cardiac muscle, usually by structural defects; intrinsic alterations in myocardial performance, or a combinations in mechanical and myocardial elements. This is one of major difference to compare with the status in adults(most CHF are due to hypertension, myocardial infarction with the consequence of myocaridial dysfunction).

Secondary, in pediatric population, there has a strong age related drug response and disposition, especially between the groups of young age children and old children, the dosage and serum half life time will be different respectively. Third, unlike the adults, measureing the cardiac function is a very tough task. In different age groups, it may need different approach to evaluate the heart performance. So, for carrying a clinical study related heart function, these facts must be considered seriously. That is also the reason why this study only choose pediatric population age below 3 years old, and only enrolled CHF caused by congenital and acquit heart diseases, this will make subjects more homogeneous. Since majority of patients with CHF may have a chance to be further surgically corrected, the one of main purpose for treatment of CHF, is try to improve and stabilize cardiac function as soon as we can, choose two weeks as a

check point to measure the heart function is clinically meaningful, however the further or long-term follow-up study should be done afterward.

Left ventricular function frequently influences the choice and timing of cardiac surgery, as well as long-term survival, in children with congenital heart disease. Abnormality of ventricular shape, hypertrophy and loading conditions exist and may be altered by medical and surgical interventions, thus the commonly used load-dependent indexes of systolic function, such as ejection fraction, may be difficult to interpret. It is difficult and also crucial to choose a sensitive and appropriate parameter to represent hemodynamic status in child which some times has the combination of congenital structural defects, myocardial dysfunction and medical intervention, left ventricular end-systolic wall stress and its relation with rate-corrected velocity of fibber shortening index, so far, it is the most suitable and acceptable for using in pediatric patients although it is not a perfect one.

3.2.1 Operational definition:

1. Left Ventricular End-Systolic Wall stress/Rate corrected fibber shortening relation(LVESWS/VCFc RELATION). Where: Left ventricular end-systolic wall stress represents the force developed by the myocardium after the onset of contraction; rate-corrected velocity of fibber shortening represent the index myocardium performance during ejection, and was normalised by the heart rate. This index is a sensitive measure of contractile state that is independent of preload and incorporates afterload [18-20].

3. Heart rate: Defined as heart beat per minute measured by ECG. Measured RR interval in seconds and divided by 60.

4. Respiratory rate: Defined as respiration time per minute measured by chest auscultation.

5. Syncope: Defined as temporary loss of consciousness and postural tone.

6. Abnormal renal function: Defined as serum creatinine two times higher than high normal range for that age.

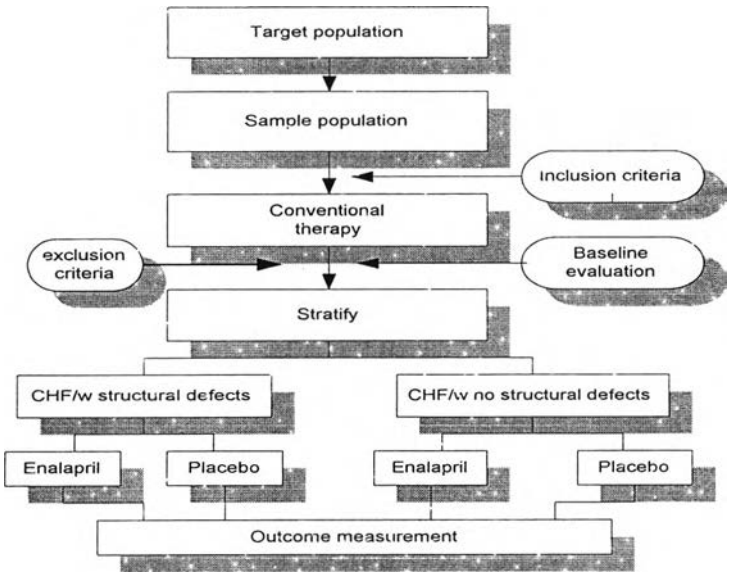
7. hypotension: Defined as two times higher than low normal range for that age under resting condition.

3.3 Research Design

This study was fully conducted as an randomized, double-blind, controlled clinical trial.

3.3.1 Research design model

Fig 3.1 Flow chat of design model



3.4 The sample

3.4.1 Target population

Patients with congestive heart failure age 3 years or below.

3.4.2 Sample population

Patients with congestive heart failure age 3 years or below. who will be hospitalized for CHF treatment at Children's Hospital of Shanghai Medical University.

3.4.3 Eligibility Criteria

- **Inclusion criteria:**

Table 3.1 Inclusion criteria

♦ Congestive heart failure patients, who meet the Diagnosis Criteria Issued by Chinese Medical Association[22], which is corresponding to the diagnosis criteria described by the most text books of Pediatrics . (See appendix A).
♦ All patients must meet at least one of following categories of the cause of CHF: a. Existing left-to-right shunt b. Existing valve regurgitation c. Left ventricular outflow obstruction d. Ventricular dysfunction without structural defects: (dilated cardiomyopathy post operative ventricular dysfunction, endocardial fibroelastosis…).
♦ Age 3 years or below.
♦ Hospitalized patients for the treatment of CHF.
♦ Agree to participate study with written informed consent from patient's parents.

- **Exclusion Criteria**

Table 3.2 Exclusion criteria

♦Patients with abnormal renal function. (Creatinine>0.15mmol/L, or urea>10mmol/L)
♦Patients with hypotension at the baseline measuring.

♦Patients with severe infection and cause circulation system collapse, which means unstable blood pressure, peripheral circulation dysfunction.

♦Patients with severe arrhythmia need additional anti-arrhythmia medication. (irregular beat>15 times/min)

♦Patients with pericardial disease or restrict cardiomyopathy

♦CHF due to non-cardiac encountered causes.

♦For ethical consideration:

a. When baseline checking, patient's contractility is within normal range (95%CI)

b. Patients need emergency cardiac surgery.

3.4.4 Sample size estimation

- **Sample size estimation**

The sample size was estimated by using the formula for two independent samples with proportion variable. According to our clinical survey, average cardiac contractility improvement rate after one month treatment by conventional therapy is about 50% up and downward, after reviewed and discussed with pediatric cardiologists, a difference of 30% absolute cases increased is regarded as the effective size which will make a clinical significance.

$$2N = 2 [Z\alpha\sqrt{2P(1-P)} + Z\beta\sqrt{Pc(1-Pc)+Pi(1-Pi)}]^2 /(Pc-Pi)^2$$

Assuming: $\alpha = 0.05$ $Z_\alpha = 1.96$ (two-tailed)

$\beta = 0.1$ $Z_\beta = 1.28$

$Pc = 0.5$ $Pi = 0.8$

$2N = 2 [1.96 \sqrt{2(0.65)(0.35)} + 1.28 \sqrt{0.5(0.5) + 0.8(0.2)}]^2 / (0.8-0.5)^2$

$2N = 102$

Estimated drop rate = 5%, total estimated number should considered = 114

Overall, 114 cases supposed should be recruited in the study with 57 patients in each group.

3.5 Experimental maneuver

3.5.1 Sampling of population

Consecutive sampling was used on CHF patients diagnosed at Children's Hospital of Shanghai Medical University. Every patient was checked by pediatric cardiologist and those meet the eligible criteria were then enrolled.

3.5.2 Randomization method

Patients meet the eligible criteria were assigned to 2 strata, according to the causes of CHF(CHF with structural defects or CHF caused by impaired cardiomyol function). expecting the study will be finished within a relatively short period of time, to avoid the imbalance in the number of subjects assigned to each group, block randomization was adopted. The block size of 4 was selected by a senior physician.

This person was response to arrange all the possible combinations of the chosen block size according to the random number table, selected one of the arrangement randomly each time and assigned the patient accordingly. Each patient's random number was put into one envelope labeled in orders. The clinical investigator then open the envelope once an eligible patient was selected and prescribed the random number of the patients. Pharmacist distributed the drugs according to the prescribed number. The same procedure was repeated as many time as needed until all patients.

3.5.3 Blindness

In order to avoid potential problems of bias during data collection and assessment double blind design was adopted. We tried very hard to let both patient's parents and investigators including technicians, nurses be blinded to identify of patients' assignment until the end of trial. Nevertheless, a true double blind trial always is tough to implement.

The method to assign patients to either treatment group was kept by a senior physician. The drug company prepared the active drugs and their corresponding identical placebos, but the coding of the drugs and placebos was performed by one assistant and then all coded bottles was kept by the pharmacist. Each patient had an unique drug or placebo code which remained with the patient for the duration of trial. Once the eligible patient is selected, the assigned the patient to the group according to the prepared arrangement and tell the investigator the corresponding labelling number of drugs. This method would guarantee that in case of emergency due to severe side effects, unblinding of one patient will not result in unbinding others.

3.5.4 Intervention

All the eligible patients were on conventional anti-failure therapy at least 48 hours before the baseline assessment. The suggested maximal total digitalizing dose is 0.04mg/kg, maximal dose for diuretics: Lasix is 4mg/kg/day; Dihydrochlorothiazide is 5mg/kg/dasy and all the patients were digitalized within first 24 hours. Preliminary baseline assessment was then carried out after 48 hours. Followed by a echocardiography left ventricular systolic function assessment, patients were assigned randomly by blocked randomization procedure to either Enalapril+conventional therapy or Placebo+conventional therapy group. All medications, clinical symptoms and laboratory tests for each individual were recorded on a special prepared sheet.

During the trial, Digoxin and diuretic regimens were allowed to be remained unchanged. The diuretic dosage, however was adjusted if necessary based on congestive symptoms and /or signs. this change was reported at the final results. The double-blinding will be maintained through a double-dummy design, but even though when the dosage of diuretics is re-justified, the blindness may be challenged.

- **Initiation of therapy with Enalapril:**

After baseline evaluation, all enrolled patients were remained in the hospital for at least 2 weeks. During first 3 days, the Enalapril therapy was initiated and the dose triturated to 0.25mg/kg/day, if tolerated. Lack of tolerance was considered to be a

significant decrease of blood pressure, if patient's blood pressure decrease more than 15% from the initial level, patient will be allowed remain in lower dosage.

Dosage for child:

0.1mg/kg/day (oral) for first day;

0.15mg/kg/day (oral) for second day;

0.25mg/kg/day (oral) for third day then same dosage (adjusted by the clinical presentation) will be given each day.

During first 3 days dosage escalation period, pulse, respiratory rate and blood pressure will be measured and recorded 3 times everyday. To keep the principle investigator blinded, during this initiation period and 2 weeks maintenance therapy, patients were under the control of "third party " physician.

Time for taking the drug: once a day in the morning (8AM).

3.6 Measurement

3.6.1 Outcome variable

(1). Primary outcome:

Ideally, contractility should be expressed in a way that is independent of preload and afterload. By relating the left ventricular end-systolic wall stress with rate-corrected velocity of circumferential fiber shortening, it provided an index that is less dependent on preload and may some time incorporates afterload, heart rate and dimension. The index showed a constant inverse linear regression during childhood

and with a narrow 95% confidence interval, and showed no change with age or body surface area[23].

Left Ventricular End-Systolic Wall Stress(g/cm^2)

This index was calculated by the method of Grossman[21]

$Stress_{es}= (1.33)(P_{es})(D_{es})/(4)(h_{es})[1+(h_{es}/D_{es})]$

where: $Stress_{es}$=left ventricular wall stress(g/cm^2) at end-systolic; P_{es}=the left ventricular pressure(mm Hg) at end-systole; D_{es} and h_{es}=left ventricular internal dimension and posterior wall thickness (cm) at end-systole; 1.35 is a conversion factor (mm Hg to g/cm^2); and 4 is a geometric factor that results form conversion of radios to internal dimension.

Rate-corrected velocity of circumferential fiber shortening(VCFc)

$VCFc = LVEDD\text{-}LVESD/[LVEDD*(LVET/\sqrt{R\text{-}R})]$

Where: LVEDD = left ventricular end diastolic dimension; LVESD = left ventricular end-systolic dimension; LVET = left ventricular ejection time; $\sqrt{R\text{-}R}$ = square root of the RR interval.

The echocardiography data of each patient then was plotted in The normal range of LVESES/VCFc relation presented as a linear regression line with 95% confidence interval.

All hemodynamic studies were conducted at Echocardiography Lab of Children's Hospital of Shanghai Medical University by one senior technician. This person has 20 years experience in echocardiography. His performance was double checked by a third person who has same back ground in echocardiography. The instrument used in study is HP Sonos 2500 Echo Machine. The measurement was conducted at baseline, and 4^{th} day, two weeks after trial started.

(2). Clinical data:

All the variables were assessed at baseline and on day 4 and day14 after trial started.

Table 3.3 clinical variables

• Heart rate, respiratory rate, Liver size, cardiothoracic ratio(chest X-ray) and body weight were measured at the baseline and on 4th day, and two weeks after trial started.
• Percentage of patients, those with congenital heart disease, became eligible for further cardiac surgery based upon their cardiac function after 2 weeks treatment. To evaluate the eligible cases for cardiac surgery, a 3 members of evaluation group was formed to conduct this evaluation.

(3). Laboratory safety data:

These tests include: serum sodium; serum potassium; serum urea; serum creatinine;

All the tests will be assessed at baseline and 4th day, two weeks after trial started. These tests were conducted at clinical lab center of Children's hospital of Shanghai Medical University.

(4). Side effects observation:

syncope: temporary loss of consciousness and postural tone.

Abnormal serum creatinine, serum urea.

Abnormal electrilyte: serum patossium, sodium

Hypotension: low blood pressure with clinical symptoms

3.6.2 Demographic data and baseline Data

Age: Defined as month in the identification card and recorded by physician at the time of entry ward. Sex: Recorded from identification card. Dosage of digoxin; dosage of diuretics; Additional medication if applicable; cause of CHF.

3.7 Consideration of some confounding factors and bias:

3.7.1 Selection bias

This trial is based on the hospitalized patients. The clinical setting which is a teaching hospital, all the staffs involved in this study had a clear consensus to recruit samples based on explicit inclusion and exclusion criteria. The eligible subjects were sampled and allocation randomly into two treatment group in order to keep baseline comparable. This had minimized the selection bias successfully.

3.7.2 Measurement bias

For most variables measured in this study were objective, Lab test and Echocardiography assessment. There has been a quite stable standard for those lab tests, and quality control was conducted by local medical laboratory control organization. Echocardiography skill would have some impact on the outcome. We used internal and external variation assessment to reduce this problem and had a successful result.

3.8 Data collection :

The trained resident was responsible for collecting and recording the demographic, clinical and side effects evaluation data. A special senior physician was response to check the record whether is correct or not. The echocardiography and serum test results was collected carefully by a research assistant. For the computer entry of the data, each variable was given a name that will identify its field in the data set. All the possible values was given a code in number so that it can be entered in the computer.

3.9 Data analysis

3.9.1 Hypothesis testing

Intention-to-treat analysis was applied in analyzing the outcome variables. For the drop out patients, the reasons for drop out will be reported and they are included by using the last observation on treatment for all time points subsequent to drop out. Patients. All statistical tests are two-tailed with significance level taken at 0.05. All data analysis will be performed by using SPSS/PC$^+$ program.

Table 3.4 primary and secondary question analysis

Questions	Outcome	Statistical methods
Primary Question		
Comparison of proportion of patients with cardiac contractility(LVESWS-VCFc) within normal range between two groups		
	Proportions	Z- test
*Comparison of LVESWS between two groups		
	Mean +/- SD	Student's t- test
*Comparison of VCFc between two groups		
	Mean +/- SD	Student's t- test
Secondary Questions		
Comparison of clinical effects between two groups		
Heart Rate	Mean +/- SD	Student's t- test
Respiratory Rate	Mean +/- SD	Student's t- test
Liver size	Mean +/- SD	Student's t- test
Cardiothoracic ratio	Mean +/- SD	Student's t- test
Weight	Mean +/- SD	Student's t- test
Comparison of percentage of patients become eligible for cardiac surgery:		
	Proportions	Chi-square test
Comparison of clinical effects between two groups		
Side effects:		
Hypotension	Proportions	Chi-square test*

Hypotension	Proportions	Chi-square test*
Syncope	Proportions	Chi-square test*
Abnormal renal function	Proportions	Chi-square test*
Cough	Proportions	Chi-square test*

* if necessary

Demographic data and baseline:

Age	Mean +/- SD	Student's t- test
Sex	Proportions	Z-test
Dosage of digoxin	Mean +/- SD	Student's t- test
Dosage of diruretics	Mean +/- SD	Student's t- test
Etiology	Proportions	Z-test
Serum sodium	Mean +/- SD	Student's t- test
Serum potassium	Mean +/- SD	Student's t- test
Serum urea	Mean +/- SD	Student's t- test
Serum creatinine	Mean +/- SD	Student's t- test

Estimation of the magnitude of difference:

95% confidence interval will be calculated for the mean difference of hemodynamic and clinical effects between two groups.

The measurement took place at baseline, day 4 and day 14 after trial started. Selecting day 4 as a check point was on the purpose of monitoring immediate response when most of case had reached their stable drug serum level.

3.10 Ethical consideration:

Angiotensin converting enzyme inhibitors are established for treatment in adults with congestive heart failure with firmly positive results. They are known to provide significant hemodynamic, symptomatic, and clinical improvement when added to anti-failure treatment. Enalapril now has been recommended for patients with mild, moderate or severe congestive heart failure. Enalapril is now been used in children with congestive heart failure, the overall findings are comparable with studies in adults, its safety dosage has been tested and investigation showed no severe side effects to compare with other vasodilators and captopril. Free informed consent will be obtained from the patient's parents before be enroled in this study. This study cautiously monitor the possible side effects, if there is any enalapril related severe side effect, the treatment will be discontinued.

3.11 Limitations:

This study has limited the end point at two weeks after trial started, it is a short term period to observe the efficacy of a drug. Therefor the long term effect should be carried out in the future study. The range of age in this study is below 3 years old, and sample were based on hospitalized patients. So the results may limited to be generalized to a wide range of childhood. However a study based on a hospitalized patients and focusing on a group of subjects who relatively had a similar characteristics may have a fair strong validity and reliability in the other hand.

3.12 Benefits of this study:

If this study can show that adding ACE inhibitors in the management child with congestive heart failure in conjunction with conventional therapy has a significant beneficial effects on hemodynamic and clinical changes, it might help to confirm the role of enalapril in the treatment of congestive heart failure in child and offer a new anti-failure therapeutic method.

3.13 Obstacles and strategies to solve the problems:

This clinical trial was conducted at pediatric hospital setting. The experience for this kind of study is limited in that institute. In order to successfully complete the study, we had taken a necessary measure to win a fully understand from the parents and also nurses. Parents had some concerns regarding the term of "clinical trial", a good relationship between physicians and parents is crucial , on daily regular base, the specialists always committed to communicate and answer any questions from the parents, this has successfully solved some problems. To get full cooperation from nurses, was another key issue. Obtaining the involvement of nurses by conducting short-course lecture and practice, had a remarkable benefit to this study.

3.14 Administration and time table

Before the trial started, permission had been obtained from Ethical Committee at Children's Hospital of Shanghai Medical University. Two meetings had been hold in

cardiac unit, all the staff were invited to join, the proposal had been introduced to all the members of unit at those meetings. One senior pediatric cardiologist and one medical resident were included as a team of this study. Regular meeting had been held for research team, corresponding medical staff, nurse and technician attended. Special record form was prepared.

Time schedule

Mar. 98 Apr. May June July Aug. Sep. Oct. Nov. Dec. Jan(99) Feb Mar

preparation

Data collection

Data analysis

writing final thesis