CHAPTER II

STATE OF LITERATURE - II

A. INTRODUCTION

The validity of studying the profiles of carbohydrate metabolism using the isolated perfused rat heart has been amply justified from the reviewed literature. The following drugs were selected for study in this thesis:

1. Insulin hydrochloride
2. Tribenosine dihydrochloride
3. Fucopentol hydrochloride
4. Depaspir sulphate
5. Xylometazoline (Xylometazoline, Xyloxa) hydrochloride.

B. METHODS FOR CHOICE OF DRUG FOR STUDY

1. Insulin hydrochloride and Tribenosine dihydrochloride

Insulin hydrochloride, has been established over a long period, as the most specific and highly potent agent against intestinal and extraintestinal activities. With the high incidence of diabetes in India, 10 to 15% of the population being estimated to be suffering from chronic constipation (Jastkevici et al., 1966; Patel, 1965; Khan et al., 1960), the usage of this drug has been wide-spread and local.
FIGURE 6
STRUCTURAL FORMULAE OF
EMETINE AND DEHYDROEMETINE

Emetine

Dehydroemetine
However, the side effects caused by its high cardio-toxicity limited its use. The cardio-toxicity has been monitored with electrocardiographic observations which revealed prolongation of the Q-T interval, elevation of the S-T segment and inversion of the T wave (Canseco de Canseco, 1973) with the incidence of serious cardiovascular collapse.

This forced the investigators to search for new substances with equivalent efficacy, but without the side-effects of Lortabine. During the chemical research which led to the total industrial synthesis of Lortabine (Brown et al, 1970a), several substances exhibiting experimental anti-arrhythmic activity, and lesser toxicity than Lortabine were discovered (Brown et al, 1970) and 2,3-Dihydro-4-cinarine was chosen from these substances for human trials and its therapeutic efficacy was confirmed (Ortis de Castellano, 1981).

The apparent safety of 2,3-Dihydro-4-cinarine over Lortabine in relation to cardio-toxicity was confirmed by many subsequent reports in experimental animals (Schweitzer & Motzer, 1961a) and in clinical trials (Canseco de Canseco, 1973; Vaglio et al, 1973; Fainzilber et al, 1973; Merchant & Shlansky-Goldstein, 1961). The chemical structural differences between the two compounds are as shown in Fig. 6.

Several workers have reviewed the various advantages of 2,3-Dihydro-4-cinarine over Lortabine, as listed overleaf.
(1) The overall toxicity of Diethylcarbamazine is half, as much as with Bantone (Brown et al., 1938).

(2) Half-time period for the whole body (time in which half the amount of an injected dose disappears from the whole body) is 2.32 days as compared to 5.37 days for Bantone (Schurz & Mcker, 1962b).

(3) Half-time period for the liver (period in which half the maximal concentration in the liver disappears) is 1.72 days as compared to 6.16 days for Bantone (Schurz & Mcker, 1962b).

(4) Diethylcarbamazine cannot be detected in plasma after 12 days, as against 20 days for Bantone (Schurz & Mcker, 1962b).

(v) Electrocardiographic evidence of toxicity of a transient nature and to a milder degree has been reported in only 0-13% of cases using the conventional dose of Diethylcarbamazine, while similar dose of Bantone result in toxic manifestations in 25-50% of cases (Bart and Kingland, 1930; Conshel in Cacchetta, 1940; Szondal et al., 1943; Vahil et al., 1943).

A tabulation of the comparative electrocardiographic changes obtained with Bantone and Diethylcarbamazine for clinical trials with over 50 patients is presented in Fig.7.
<table>
<thead>
<tr>
<th>Parameter Measured</th>
<th>Electrolyte $\ell$ Excitons</th>
<th>Electrolyte $\ell$ Excitons</th>
</tr>
</thead>
<tbody>
<tr>
<td>Prolongation of O-O interval</td>
<td>10</td>
<td>70</td>
</tr>
<tr>
<td>Elongation of O-O segment</td>
<td>-</td>
<td>15</td>
</tr>
<tr>
<td>Lower voltage for T wave</td>
<td>90</td>
<td>200</td>
</tr>
<tr>
<td>Invasion of T wave</td>
<td>26</td>
<td>200</td>
</tr>
</tbody>
</table>

(after Canadian da Conceicao, 1983)
While all these reports a she it quite clear that—

(a) Maneine is highly cardio-toxic and that (b) Dehydroemanine is relatively less toxic, the evidence is purely electrocardiographic, and is essentially a reflection of the behaviour of the junctional tissue and conduction mechanisms in the heart. No insight is available as to the exact mechanism of this altered conduction behaviour in the presence of these drugs. It was therefore considered germane to study changes in the carbohydrate metabolism of the heart in the presence of these drugs, carbohydrates being the usual energy substrate of the normal metabolic activity, if this could possibly throw some light on the reasons for—

(a) the cardiotoxicity of Maneine and Dehydroemanine and
(b) the relative safety of Dehydroemanine as compared to Maneine

(b) Procarnosin hydrochloride:

Procarnosin, a beta-adrenergic blocking agent is being increasingly employed in the treatment of angina pectoris and cardiac arrhythmias primarily, and a variety of clinical disorders like idiosyncratic hypertrophic sub-aortic stenosis, Torsades de Pointes, atrial shunts, Parkinsonism tremor etc. Like several other antagonists, the beta-adrenergic blocking agents are structurally similar to the beta-adrenergic antagonist, Ipracarnosin. (Fig. 8).
FIGURE 8

BETA-ADRENERGIC AGONIST & ANTAGONISTS

AGONIST

**Isoprenaline**

ANTAGONISTS.

**Dichloro-isoprenaline**

**Pronethalol**

**Propranolol**

Chemical structural similarities between the beta-adrenergic agonist, isoprenaline and the three beta-adrenergic blocking agents (Antagonists) Dichloro-isoprenaline, Pronethalol and Propranolol.
The treatment of angina pectoris was one of the earliest clinical applications of beta-adrenocorticosteroids and the first agent to be clinically tested was propranolol which produced significant symptomatic improvement, (Aldridge et al., 1965) but the finding that it produced coronary changes in mice, (Fayet, 1965) made it unsuitable for clinical use. Propranolol, the closely related chemical congener of propranolol, was developed and tested for the same condition, and was found to be very effective in decreasing the number of anginal attacks. (Cullen and Pritchard, 1965; Turner, 1966; Bechtle et al., 1966).

The utility of propranolol rapidly spread to the treatment of cardiac arrhythmias, when it was demonstrated to effectively antagonize arrhythmias induced by catecholamines (Brown et al., 1960; Rovan et al., 1962) and digitalis (Sobin et al., 1963; Inochon et al., 1966) in laboratory animals. Clinical reports closely followed these experimental findings, when arrhythmias induced during the course of digitalis therapy were either totally abolished or markedly reduced in frequency by propranolol (Boschley et al., 1966; Turner, 1966).

The role of propranolol in the treatment of non-digitalis induced arrhythmias particularly atrial fibrillation and atrial flutter has also been clearly demonstrated. Essentially, in every case with atrial fibrillation and a rapid ventricular rate, propranolol when administered has shown to decrease the
ventricular rate, and this response has been observed as early
as two minutes after an I.V. administration of Propranolol.
(Ravina et al., 1965; Readland et al., 1965; Schamroth, 1966).

While so much is known of the clinical role and the pharma-
co-dynamics of Propranolol's action in the treatment of cardiac
arrhythmia, as reviewed by Epstein and Brunwald (1966), so
little is known of any of the effects on the myocardial metabolism.
Griendling and Pastore (1987) using the unanesthetized vagotomized
dog studied the coronary saline flow and myocardial arteriovenous
difference for glucose, lactate, pyruvate and non-oxidized
fatty acid, before and during nor-adrenergine infusion into the
left circumflex coronary artery. The myocardial uptake of glucose,
fatty acid and lactate was significantly increased during the
nor-adrenergine infusion. Propranolol prior
to the nor-adrenergine infusion resulted in little change in
the consumption of glucose, pyruvate and lactate but a marked
decrease in the uptake of fatty acid suggesting that the regul-
ation of myocardial uptake of fatty acid was probably under the
control of beta-adrenergic receptors.

Pastore and Griendling (1987) employed the same technique as
described above, to demonstrate a decrease in the myocardial
uptake of glucose, lactate, pyruvate and fatty acids, after an
infusion of Propranolol, accompanied by a fall of heart rate
from 175/min to 125/min with a slight decrease in sever
of constriction also, which seemed suggestive that the metabolic effects could have been initiated by a functional change created by Propranolol. To separate these two effects, they attached a pacing electrode to the left atrial appendage and stimulated the heart to beat at a rate near-normal, then glucose, lactate and pyruvate uptakes significantly increased while fatty acid uptake remained essentially unchanged (lower than normal) which added further evidence that fatty acid uptake was closely linked to beta-adrenergic receptor activity.

The studies cited above have been done on intact dog hearts estimating metabolic parameters from A-V differences, while, the studies undertaken in this series were on isolated hearts, wherein metabolic effects could be measured without other functional influences, and therefore Propranolol was included in the preview of this study.

(c) Trepant System

Trepant, the physiological anticoagulant is being used frequently and increasingly, in the prevention and treatment of thrombo-embolic disease, both venous and arterial, such as pulmonary occlusion, cerebral thrombosis, thromboangiitis, for the prevention of thrombo-embolic phenomena after vascular and cardiac surgery and after coronary thrombosis (admittedly controversial). It is a macromonomeride with a chemical structure as shown in Fig.9.
**FIGURE: 9**

**CHEMICAL STRUCTURE OF HEPARIN**

HEPARIN

Muco-polysaccharide structure showing the sulphated D-glucosamine and D-glucuronic acid the two hexose moieties present in equimolar amounts, appearing alternatingly along the polysaccharide chain.
While liberal use of heparin in I.V. infusions have been employed during and after coronary thrombolysis and after experimental ischemic episodes, with beneficial affects, its action pry on the myocardium and its metabolism have not been reported. Reports on the effects of heparin on metabolism in general are few. Rayfield et al (1964) reported an increase in the level of free fatty acids in the plasma, after heparin, at the height of alimentary lipasemia, with a fall in triglycerides probably by their hydrolysis, and concluded that this prevention or decrease in alimentary hypertriglyceridemia was probably beneficial.

Intravenous injections of heparin in rats increased lipase activity in plasma while it decreased lipase activity in the tissues (Pyea et al, 1967). Rat heart homogenate, in the presence of heparin revealed a 2 to 3 fold increase in lipolytic activity characterized by Carter & Valentine (1966) (using triolein-triolein-gel and differential purification with acetone fractionation) as an inactive form of lipoprotein lipase which was enzymatically activated in the presence of heparin.

The metabolism of fats and carbohydrates is intimately related in both normal and pathological conditions. A good example of the latter is diabetes, in which a growing importance is being ascribed to a disturbance of fat metabolism (Barnes & Lupt, 1952; Adorno & Miller, 1939) in addition to the classical
Disturbances of carbohydrate metabolism. The occurrence of atherosclerosis, a disease of advanced fat metabolism (Brenton-Stewart, 1981; Goldsmith, 1981) along with diabetes is extremely common (Lietor et al., 1973; Baylog & Bradley, 1957). The promising results with heparin in the management of generalized atherosclerosis (Hess & Moore, 1958) and in coronary atherosclerosis (Brenton et al., 1961) prompted this investigation to study possible effects of heparin on carbohydrate metabolism in the myocardium. The influence of heparin on carbohydrate metabolism per se or on the metabolic cycle in the myocardium has not been reported hitherto in literature.

(d) Xylocaine (Lignocaine, Novocaine) hydrochloride

Xylocaine, which is structurally similar to procaine, is an established cardiac anti-arrhythmic agent (Fig. 20) was synthesized in 1943, by Udenfriend and subsequently investigated extensively as a local anaesthetic agent (Wadding, 1986). Southworth et al. (1990) were the first to report the successful use in ventricular fibrillation following during cardiac arrhythmisation. Subsequent workers have confirmed the value of Xylocaine in abolishing or preventing ventricular arrhythmias in animals under a variety of experimental conditions (Frederickson & North, 1993; Gordon & Shulman, 1955; Hitchcock & Brown, 1959; Auster & Rees, 1994).
FIGURE 10

CHEMICAL STRUCTURE OF XYLOCAINE AND ITS SIMILARITY TO PROCAINE-AMIDE

\[
\begin{align*}
\text{H}_2\text{N} & - \text{C} - \text{NH} - \text{CH}_2\cdot\text{CH}_2\cdot\text{N}\left(\text{C}_2\text{H}_5\right)_2 \\
\text{PROCAINE - AMIDE}
\end{align*}
\]

\[
\begin{align*}
\text{CH}_3 & \quad \text{NH} - \text{C} - \text{CH}_2 - \text{N}\left(\text{C}_2\text{H}_5\right)_2 \\
\text{XYLOCAINE} \quad \text{[Lignocaine, Lidocaine]}
\end{align*}
\]
Haus & Stull (1966) have shown the superiority of Xylocaine over quinidine, and over direct-current shocks in restoring slow rhythms in dogs with digitoxin-induced ventricular arrhythmias. Elshof (1959) and Wells (1960) have successfully used Xylocaine for the control of various arrhythmias arising during surgery. To Samotic (1965) and Bedenek et al. (1966) reported the use of Xylocaine in the treatment of ventricular ectopic beats and ventricular tachycardia of non-surgical origin. Reschuk et al. (1980) found Xylocaine to be particularly successful in the treatment of acute ventricular arrhythmias, notably those occurring after myocardial infarction, during and after cardiac surgery, and in the treatment of atrial ectopic beats, though of less value in sustained atrial arrhythmias.

Anti-arrhythmic agents in general, produce refractory period and decrease conduction velocity in the beating heart. Although these changes are probably the result of a decrease in sodium and potassium carriage across cell membrane (from & Inzel, 1966), it is not certain whether both, the electrophysiological and ischemic changes represent a myocardial action of the anti-arrhythmic agents or are a result of an intra-cellular metabolic effect.

Based on this speculation, studies on the metabolic effects of cardiac anti-arrhythmic agents on the myocardial metabolism,
particularly of antihypertensive have been reported. Using glucose as substrate, quinidine has been reported to decrease oxygen consumption of cardiac muscle by one (Hnes & Hugard, 1976; Grossman & Ship, 1968) and to have no effect on oxygen consumption by others (Takah et al., 1973; Levy & Richards, 1965). However, Grossman & Ship (1968) reported a decrease in uptake and utilization of glucose or pyruvate, in the presence of quinidine, by the dog heart. But the dose employed was extremely high, well beyond the therapeutic range.

Studies on the effects of ylloena, the anti-arrhythmic agent on cardiac metabolism was included in this series utilizing doses compatible with therapeutic utility. Keiffy & Grehke (1963), also using ylloena in the therapeutic dose range, reported no observable increase in the uptake or utilization of glucose or pyruvate in rabbit heart ventricular slices, suggesting that the mechanism of its anti-arrhythmic action was not related to changes in carbohydrate metabolism in the cardiac cell. Studies in this series, on the isolated perfused beating hearts, being on actively functioning tissues, was deemed more closely similar to natural conditions.

Besides these drugs described so far, chosen for this study, preliminary studies were carried out with theca and tyrode, which served a two-fold purpose.
(1) To test out the methodology established here and to assess the validity and acceptability of the preparation.

(2) To evaluate the methods of estimations employed in this study to measure the parameters of carbohydrate metabolism.

The parameters of carbohydrate metabolism, as measured in this series have already been reported in literature for both Insulin and Cortisone, and therefore comparison of the results obtained here with those of earlier studies already at record, facilitated a critical appraisal of the procedure adopted in this study.