J. Pharm. Pharmacol. 1997, 49: 1030–1035
Received April 3, 1997
Accepted June 19, 1997

Beneficial Effects of Clonidine in Streptozotocin-induced Diabetes and DOCA-hypertensive Rats

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Abstract

This investigation was undertaken to study the effects of chronic treatment with clonidine on cardiovascular complications in streptozotocin-induced diabetes and DOCA-hypertensive rats.

Injection of streptozotocin induced glucosuria, hyperglycaemia, hypoinsulinaemia, hypothyroidism, hypercholesterolaemia, hypertriglyceridaemia, bradycardia and a decrease in left ventricular developed pressure (LVDP). DOCA by itself did not induce any change in blood-glucose levels in non-diabetic animals. However, in diabetic animals DOCA significantly reduced blood-glucose levels. Treatment of diabetic and diabetic hypertensive animals with clonidine ($25\,\mu g\,kg^{-1}$ every day for six weeks) significantly prevented diabetes-induced loss of body weight, bradycardia, cardiac hypertrophy and hypothyroidism. It also partially, but significantly, prevented diabetes-induced hyperglycaemia and hypoinsulinaemia in both diabetic and diabetic-hypertensive animals. There was a significant reduction in diabetes-induced elevation of cholesterol and triglyceride levels and an improvement in LVDP at higher filling pressure in diabetic and diabetic hypertensive animals.

This investigation shows that chronic treatment with clonidine produces a number of beneficial effects such as prevention of hyperlipidaemia and hypothyroidism and improvement in cardiomyopathy and glycaemic control in diabetic and diabetic hypertensive rats.

The occurrence of hypertension with diabetes mellitus is critically important not only because of its increased prevalence but also because the microvascular and macrovascular complications of diabetes are greatly accelerated by the presence of hypertension (Fuller et al 1983). It has further been reported that hypertension might be causally related to primary diabetic cardiomyopathy (Factor et al 1980). Thus efficient antihypertensive therapy in diabetic patients would be positively beneficial in arresting the rate of progression of diabetic complications. The choice of antihypertensive agent is therefore crucial not only in the reduction of elevated blood pressure but also in the reversion of hypertension-induced cardiovascular changes and satisfactory control of metabolic disequilibrium. However, there are few if any comparative data available to guide the rational choice of an antihypertensive agent for diabetic hypertensive patients. Optimum antihypertensive drug therapy of such patients is based on limited experimental data, practical experience and educated guesswork, and therefore needs to be tailored to each drug.

It is reported that cardioselective β-blockers do not improve diabetes-induced cardiac dysfunctions, cardiomyopathy or other complications like hyperlipidaemia (Bangaru et al 1990). Atenolol is reported to worsen the condition of diabetic rats. Prazosin (Lakkad et al 1992) and hydralazine (Rodrigues et al 1986) were found to improve cardiac dysfunction; they also prevented diabetes-induced hyperlipidaemia and hypothyroidism. Angiotensin-converting enzyme inhibitors such as enalapril prevented diabetes-induced cardiomyopathy, cardiac dysfunctions and hypercholesterolaemia in rats (Lakkad et al 1992). The calcium-channel blocker nifedipine has been shown to prevent diabetes-induced cardiomyopathy, hypertension and hyperlipidaemia and also to increase insulin sensitivity by nifedipine treatment (Shah et al 1995). The centrally acting drugs are one of the several differing classes of drug available for antihypertensive therapy. Clonidine is reported to reduce blood pressure without inducing sodium retention (Weber 1989). A growing awareness of the importance of treatment-induced metabolic effects has served to re-awaken interest in centrally acting agents. Clonidine has been reported to induce either beneficial effects or neutral effects on lipid levels in patients with primary hypertension with (Satia et al 1995) or without (Houston et al 1990) diabetes. The current investigation was undertaken to study the effect of treatment with clonidine on cardiovascular complications and lipid levels in streptozotocin-induced diabetes and DOCA-induced hypertension in rats.

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Materials and Methods

Induction of diabetes mellitus and hypertension in rats

Healthy female albino Wistar rats, 180–220 g were made diabetic by single tail-vein injection of streptozotocin ($45\,mg\,kg^{-1}$) dissolved in citrate buffer (pH 4·5). Control rats were injected with citrate buffer alone. Hypertension in rats was induced by subcutaneous administration of deoxycorticosterone (DOCA) at a dose of $5\,mg\,kg^{-1}$ every day throughout the treatment period. DOCA-treated animals were fed with 2% salt solution in drinking water. Animals with blood pressure above 150 mm Hg after 10 days of DOCA treatment were considered as hypertensive.

Treatment protocol and observation of animals

Induction of diabetes was checked 48 h after injection of streptozotocin by measuring the extent of glycosuria with

enzyme test-strips (Diastix Ames India). Rats with glycosuria greater than 2% were considered as diabetic for further experiments. Other animals (non-diabetics) administered citrate buffer were considered as control. Animals from the diabetic group were divided into four subgroups: diabetic, diabetic clonidine-treated, diabetic hypertensive and diabetic hypertensive-treated. Similarly the animals from the non-diabetic group were divided into another four subgroups: control, control clonidine-treated, hypertensive and hypertensive clonidine-treated. Clonidine hydrochloride ($25\,\mu g\,kg^{-1}$ dissolved in distilled water) was given orally daily for six weeks. All the groups were maintained for six weeks in a constant environment with food and water freely available. They were monitored throughout the six-week study period for water intake, food intake, changes in body weight, blood pressure, heart rate and mortality.

Measurement of blood pressure and heart rate

Blood pressure and heart rate were recorded, by use of the tail-cuff method (Haward blood pressure monitor, Kent, UK) attached to Student's oscillograph, before starting the clonidine therapy and on every 10th day until completion of the treatment.

Blood-sample collection and serum analysis

After six weeks of clonidine treatment, animals were fasted for 12 h and blood samples, approximately 4–5 mL, were collected from the retroorbital plexus of the eye into centrifuge tubes and left to clot for 30 min at room temperature. Serum was separated by centrifugation at $3000\,rev\,min^{-1}$ for 30 min. Supernatant clear serum was separated and transferred to Eppendorf tubes. The serum samples were stored at −20°C until analysis.

Serum immunoreactive insulin and triiodothyronine were assayed by means of a kit from Bhabha Atomic Research Centre (Bombay, India). Serum glucose, total cholesterol and triglycerides were estimated by use of standard diagnostic assay kits from Miles India Ltd.

Recording of cardiac functions

One day after collection of blood samples the animals were killed and their hearts quickly dissected out and placed in Chenoweth-Koelle buffer (composition (mM): NaCl 120·0, KCl 5·6, $CaCl_2$ 2·18, $MgCl_2$ 2·1, $NaHCO_3$ 19·2, glucose 10·0) maintained at 37 ± 1°C. They were mounted as in the modified Neely's working heart model (Vadlamudi & McNeill 1983). In the working heart model the perfusate entered the left ventricle through the left atrium and was pumped out through the aortic pump. Aortic outflow was subjected to an after-load of 45 cm H_2O. Hearts were left to stabilize for 10 min at 10 cm H_2O perfusion pressure. The LVDP was measured by changing the height of the left atrial filling reservoir from 2·5 cm to 20 cm in 2·5-cm increments. At each point pressure development was left to stabilize before it was recorded by means of a pressure transducer-polygraph system (Inco, India).

Statistical analysis

Statistical analysis was performed by one-way analysis of variance then Tucky's multivariance test (Bolton 1984). Tucky's multiple range test is a multiple comparison test based on keeping the error rate at 5% from an 'experiment-wise' view point. In the multiple range test different treatments can be compared by use of the formula $Q(S^2/N)^{\frac{1}{2}}$, where S^2 is the error variance from analysis of variance (within mean square for one-way analysis of variance) and N is the sample size. If the sample sizes are not equal in the two groups to be compared N is replaced by $2N_1N_2/(N_1+N_2)$ where N_1 and N_2 are the sample sizes of the two groups. Q is the value of the Studentized range at the 5% level. Any differences of treatment means which exceed the value given by the formula are considered to be significant.

Results

General features of the experimental animals

Treatment of animals with streptozotocin induced a significant loss of body weight. The maximum loss of body weight was observed during the first ten days after induction of diabetes. This loss in body weight was significantly prevented by clonidine treatment. Weight was gained by all other groups of animals (Table 1). There was an increase in food and water intake in diabetic and diabetic hypertensive animals. Clonidine treatment did lead to any significant change in water intake in any of the groups (Table 1).

Cardiovascular parameters

The mean blood pressure of diabetic animals, diabetic hypertensive animals and DOCA-hypertensive animals was found to be significantly higher than that of control animals after 20 and

Table 1. Effect of clonidine treatment on the general features of experimental animals.

Parameter	Control (n = 6)	Control treated (n = 6)	Diabetic control (n = 6)	Diabetic treated (n = 6)	Deoxycorti-costerone control (n = 6)	Deoxycorti-costerone treated (n = 7)	Streptozo-tocin-control (n = 6)	Streptozo-tocin-treated (n = 6)
Body weight (μg)								
Initial	220 ± 3·53	218 ± 6·90	214 ± 9·18	212 ± 5·20	202 ± 8·68	200 ± 6·61	205 ± 2·85	216 ± 3·83
After 20 days	244 ± 4·57	239 ± 8·40	172 ± 8·56	191 ± 4·57	224 ± 9·63	209 ± 4·32	169 ± 5·19	189 ± 6·61
After 40 days	264 ± 3·63	250 ± 9·22	160 ± 7·30*	183 ± 4·51†	234 ± 10·3	224 ± 4·58	158 ± 6·6*	184 ± 5·2†
Food intake (μg animal^{-1} day^{-1})								
Initial	16	14	16	16	15	15	15	15
Final	20	21	34*	32	25	26	35*	24
Water intake (mL animal^{-1} day^{-1})								
Initial	23–25	20–24	22–24	20–22	20–22	20–23	23–25	20–23
Final	23	25	76*	64	40	60	70*	60

*$P < 0·05$, significantly different from control. †$P < 0·05$, significantly different from diabetic control.

Table 2. Effect of clonidine treatment on cardiovascular parameters.

Parameter	Control (n=6)	Control treated (n=6)	Diabetic control (n=6)	Diabetic treated (n=6)	Deoxycorti-costerone control (n=6)	Deoxycorti-costerone treated (n=7)	Streptozo-tocin-costerone control (n=6)	Streptozo-tocin-costerone treated (n=6)
Blood pressure (mm Hg)								
Initial	104±1·1	104±3·5	105±2·4	105±2·5	104±3·8	100±4·0	105±2·4	103±2·7
After 20 days	104±1·4	101±2·7	130±2·5	106±6·0	134±2·2	112±3·4	135±5·3	100±3·1
After 40 days	104±1·5	100±4·0	140±1·9*	103±6·2†	157±4·7†	112±3·5‡	155±2·7†	112±4·3§
Heart rate (beats min^{-1})								
Initial	382±8·3	383±10·2	384±7·7	385±12·0	388±10·1	383±8·9	382±11·4	382±10·1
After 20 days	385±7·7	346±16·3	272±8·6	330±12·2	390±11·0	380±9·8	275±11·9	339±13·5
After 40 days	387±7·3	340±9·1*	305±8·4*	315±9·4	384±9·2	334±14·0‡	302±1·5	325±11·0
Cardiac hypertrophy (wet heart weight/body weight)	4·27±0·1	4·67±0·2	5·84±0·2*	5·14±0·1†	4·53±0·2	4·63±0·2	5·93±0·2‡	5·06±0·2§

*$P < 0·05$, significantly different from control. †$P < 0·05$, significantly different from diabetic control. ‡$P < 0·05$, significantly different from DOCA control. §$P < 0·05$, significantly different from DOCA diabetic control.

40 days of the treatment period. Administration of DOCA induced a greater rise in blood pressure in non-diabetic and diabetic animals. Streptozotocin did not cause a further increase in blood pressure in DOCA-hypertensive rats. Clonidine treatment significantly ($P < 0·05$) reduced streptozotocin- and DOCA-induced hypertension to a value comparable with that of control animals in all the diabetic, hypertensive and diabetic hypertensive animals (Table 2).

The initial heart rate of all the animals was in the normal range. After 20 days and at the end of the treatment period the heart rate of diabetic animals was found to be significantly ($P < 0·05$) lower than that of control animals. In diabetic hypertensive animals also significant ($P < 0·05$) bradycardia was observed. Clonidine treatment partially corrected bradycardia in diabetic and diabetic hypertensive animals. Clonidine treatment induced a significant ($P < 0·05$) reduction in heart rate in non-diabetic non-hypertensive (control) animals. However the bradycardia induced in diabetic animals was not further augmented in clonidine-treated diabetic and non-diabetic hypertensive animals (Table 2).

Increase in left atrial filling pressure from 2·5 to 20 cm water resulted in a gradual increase in LVDP. Hearts from diabetic animals showed significantly less LVDP, especially at higher filling pressure, compared with hearts from control animals. Clonidine treatment significantly ($P < 0·05$) prevented the depressed left ventricular function in diabetic animals (Fig. 1). Hearts from diabetic hypertensive animals also showed lower LVDP. Depression of cardiac function in diabetic hypertensive animals was also significantly ($P < 0·05$) prevented by clonidine treatment. The decrease in LVDP was insignificant for non-diabetic hypertensive hearts compared with control hearts. Treatment of non-diabetic hypertensive animals with clonidine induced a significant increase in LVDP compared with untreated animals (Fig. 2).

A significant increase in the index of hypertrophy was observed for diabetic and diabetic hypertensive animals. Clonidine treatment significantly ($P < 0·05$) prevented this increase in the index of hypertrophy in both diabetic and diabetic hypertensive animals (Table 2).

Effect of clonidine on glucose and insulin levels

Injection of rats with streptozotocin induced a significant increase in blood-glucose levels in diabetic animals. Treatment with clonidine in diabetic animals significantly reduced this hyperglycaemia. Treatment of non-diabetic rats with DOCA alone did not cause significant alteration of glucose levels; however, in diabetic rats DOCA induced a significant reduction in glucose levels. Treatment with clonidine caused a further decrease in glucose levels in these animals. Treatment of DOCA-hypertensive animals with clonidine caused a significant increase in the blood glucose level. However, the increased glucose levels in these animals was significantly less than in untreated diabetic rats.

Injection of streptozotocin caused a state of hypoinsulinaemia in diabetic and diabetic hypertensive animals. Treatment with clonidine significantly ($P < 0·05$) prevented the diabetes-induced decrease in insulin levels both in diabetic and in diabetic hypertensive animals. Administration of DOCA in control and diabetic rats did not induce any significant change in insulin levels. Treatment of these groups with clonidine did not alter insulin levels compared with their respective controls.

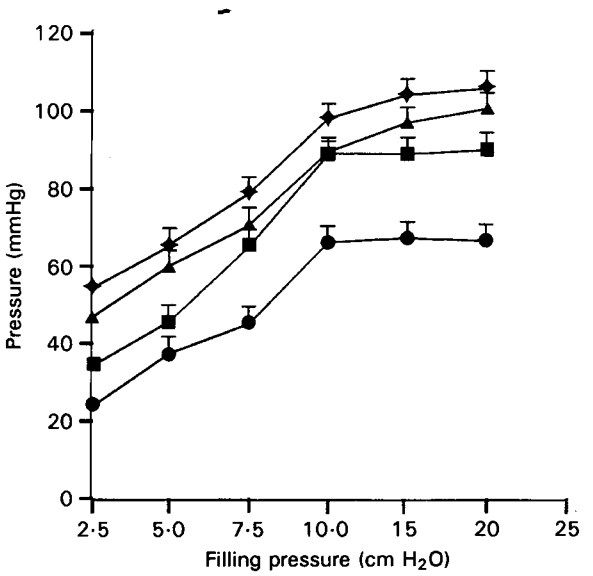

FIG. 1. Effect of six weeks of clonidine treatment on left ventricular developed pressure in control (◆), control treated (▲), diabetic (●) and diabetic treated (■) rats. Each point depicts the mean ± s.e.m. of results from six experiments.

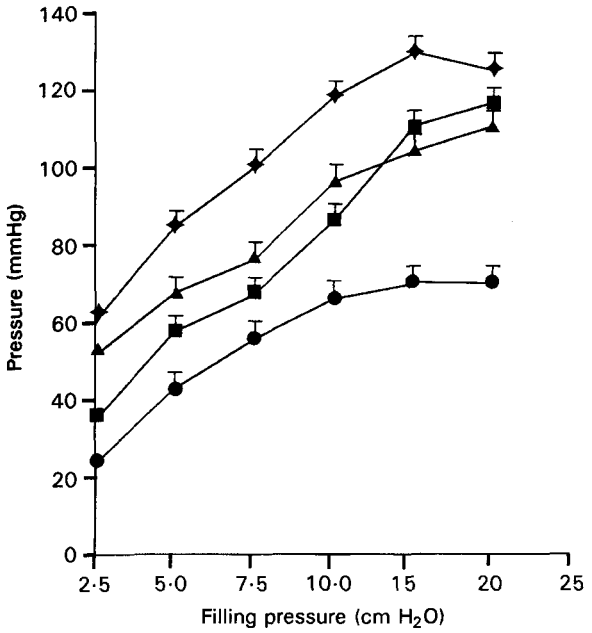

FIG. 2. Effect of six weeks of clonidine treatment on left ventricular developed pressure in hypertensive (◆), hypertensive treated (▲), diabetic hypertensive (●) and diabetic hypertensive treated (■) rats. Each point the depicts the mean ± s.e.m. of results from six experiments.

Effect of clonidine on other biochemical parameters

Serum triiodothyronine levels were found to be significantly ($P < 0.05$) less in the diabetic and diabetic hypertensive animals. Clonidine treatment of both the groups of animals significantly ($P < 0.05$) prevented diabetes-induced hypothyroidism. In all other groups no significant change in triiodothyronine level was observed.

Total serum cholesterol, low-density lipoprotein cholesterol and triglyceride levels were found to be significantly higher in diabetic and diabetic hypertensive animals; this increase was significantly prevented by treatment with clonidine. In control animals and non-diabetic hypertensive animals treated with clonidine total cholesterol and low-density lipoprotein cholesterol levels were not significantly different from those of their respective controls (Table 3). Triglyceride levels in non-diabetic hypertensive animals were found to be lower than those of control animal. Levels were further reduced by clonidine treatment. Clonidine by itself also reduced triglyceride levels in control treated animals (Table 3).

Discussion

Streptozotocin is reported to induce dose-dependent diabetogenic effects in rats ranging from mild diabetes to the severe ketotic stage at higher dose (Hofteizer & Carpenter 1973). Different laboratories have used different doses of streptozotocin ranging from 30 mg kg^{-1} to 80 mg kg^{-1}. In our laboratory a dose of 35 mg kg^{-1} or less failed to induce diabetes mellitus whereas a dose above 50 mg kg^{-1} was found to cause high mortality (> 80%). The dose of streptozotocin used in this study (45 mg kg^{-1}) not only caused a significant increase in blood-glucose levels but also significant hypoinsulinaemia and glycosuria (> 2%) in diabetic and diabetic hypertensive animals. These animals showed significant loss of body weight, polyuria and polydipsia. Clonidine treatment significantly prevented the loss of body weight and polydipsia in diabetic and diabetic hypertensive animals, compared with their respective controls.

The hyperglycaemic state is reported to cause a series of vascular changes (Haller et al 1991). Thus treatment of hypertension in diabetes must not worsen glycaemic control. In the current study blood-glucose levels in diabetic animals were found to be significantly higher than in controls. Treatment of non-diabetic animals with DOCA did not induce any significant change in blood glucose or insulin levels. However, administration of DOCA in diabetic animals caused a significant reduction in blood-glucose levels without significant alteration in insulin levels. It is thus possible that DOCA increases insulin sensitivity in diabetes. These results are in accordance with results previously reported after treatment of streptozotocin-diabetic rats with DOCA (Hebden et al 1990).

It was found in the current study that treatment with clonidine in control and DOCA-hypertensive rats caused an increase in blood-sugar levels without affecting the insulin levels compared with those of their respective control. It has been reported that acute or chronic treatment of normal or spontaneously hypertensive rats with clonidine induces an increase in glucose levels via an increase in hepatic glycogenolysis (Lewis et al 1989). Treatment of diabetic or diabetic hypertensive rats with clonidine reduced blood-glucose levels with a simultaneous increase in insulin levels compared with those of their respective controls. The presence of α_2-adreno-

Table 3. Effect of clonidine treatment on biochemical parameters of diabetic and diabetic hypertensive rats.

Parameter	Control (n = 6)	Control treated (n = 6)	Diabetic control (n = 6)	Diabetic treated (n = 6)	Deoxycorticosterone control (n = 6)	Deoxycorticosterone treated (n = 7)	Streptozotocin-corticosterone control (n = 6)	Streptozotocin-corticosterone treated (n = 6)
Glucose (mg dL^{-1})	115 ± 4·8	101 ± 5·9	359 ± 13·9*	241 ± 21·3†	113 ± 5·8	146 ± 11·6‡	233 ± 20·6†	219 ± 19·0
Insulin (I units mL^{-1})	28·8 ± 0·5	28·8 ± 1·1	14·7 ± 0·3*	21·3 ± 0·7†	24·4 ± 1·4	23·2 ± 1·3	18·7 ± 0·4‡	21·8 ± 0·8§
Triiodothyronine (ng dL^{-1})	1·4 ± 0·74	1·13 ± 0·1	0·7 ± 0·03*	1·1 ± 0·05†	1·1 ± 0·7	1·2 ± 0·05	0·8 ± 0·04‡	1·0 ± 0·07§
Cholesterol (mg dL^{-1})	75·5 ± 6·1	88·4 ± 9·6	163 ± 14·6*	118 ± 13·5†	66·0 ± 4·0	87·3 ± 7·0	144 ± 7·9‡	96·2 ± 4·3§
Triglyceride (mg dL^{-1})	77 ± 1·6	49 ± 4·2*	142 ± 5·0*	102 ± 8·4†	30 ± 2·1*	20 ± 2·8‡	130 ± 1·3*‡	62·1 ± 2·8§
Low-density lipoprotein (mg dL^{-1})	23·2 ± 4·6	33·2 ± 5·5	74 ± 8·4*	41 ± 1·7†	30·7 ± 2·8	29·5 ± 4·9	77 ± 14·6*‡	43·5 ± 4·9§

*$P < 0.05$, significantly different from control. †$P < 0.05$, significantly different from diabetic control. ‡$P < 0.05$, significantly different from DOCA control. §$P < 0.05$, significantly different from DOCA diabetic control.

ceptors has been reported in the beta-cells of islets of Langerhans and stimulation of these receptors causes inhibition of insulin secretion and hyperglycaemia (Nakaki et al 1981). A marked enhancement of glucose-induced insulin secretion has been reported in the pancreatic islets isolated from rats treated with clonidine for 10 days compared with islets obtained from control rats (Ishii et al 1985). It has been reported that clonidine increases blood growth-hormone levels in various animal species (Ruch et al 1976; Lancranjan & Marbach 1977) and that prolonged exposure to this hormone enhances the responsiveness of the pancreatic islets to glucose (Malaisse et al 1968; Martin et al 1968). Thus it is possible that the enhancement of serum insulin levels in diabetic rats after clonidine treatment could be attributed to the hyper-responsiveness developed in the pancreatic cells.

Increase in blood pressure after treatment with alloxan or streptozotocin has been reported previously by several workers (Cavaliere et al 1980; Bunag et al 1982). In our study also blood pressure of diabetic animals was found to be significantly higher than that of control animals. DOCA treatment of animals with increased sodium supplementation caused the development of hypertension after ten days. However, DOCA treatment of diabetic rats did not induce development of severe hypertension. Thus a sort of counteraction to diabetes-induced hypertension was observed. Clonidine treatment significantly prevented the development of hypertension in diabetic and hypertensive rats. Clonidine is reported to reduce blood pressure by inhibiting increased sympathetic activity and hence elevated levels of catecholamines at the point of origin within the central nervous system (Haeusier & Finch 1972).

Bradycardia and hypothyroidism have frequently been observed in diabetic rats (Savarese & Berkowitz 1979; Rodrigues et al 1985). The development of bradycardia has been attributed to down-regulation of myocardial β-adrenoceptors and increase in circulating and heart catecholamine levels (Savarese & Berkowitz 1979). The hypothyroidism induced by diabetes might be another factor responsible for changes in myocardial adrenoceptors (Vadlamudi & McNeill 1983) because treatment of diabetic rats with triiodothyronine is reported to prevent bradycardia (Goyal & McNeill 1985). In the current study we also found bradycardia and hypothyroidism associated with diabetes. DOCA treatment did not induce any significant change in heart rate in any groups. Clonidine treatment did not further augment the diabetes-induced bradycardia and prevent the diabetes-induced decrease in triiodothyronine levels. Our data support the hypothesis that hypothyroidism might be one of the causes of diabetes-induced bradycardia. Clonidine treatment also resulted in a significant decrease in heart rate in control and non-diabetic hypertensive animals. The hypotensive and sympathoinhibitory effects of clonidine were reported to be associated with bradycardia which is primarily attributable to central inhibition of cardiac sympathetic tone; however, central activation of cardiac vegal activity also appears to contribute to bradycardia produced by clonidine (Scriabine et al 1968).

Increase in left atrial filling pressure from 2·5 cm to 25 cm H_2O caused a gradual increase in LVDP in control animals. LVDP was found to be significantly lower in diabetic animals. It has been reported that streptozotocin-induced diabetes causes a decrease in cardiac myosin ATPase activity because of a shift from the more active V_1 myosin isoenzyme to the less active V_3 form (Banerjee 1986). The contractile function is thought to be related to myosin ATPase activity and thus it is possible that this shift might contribute to the diminished cardiac contractility of diabetic rats. Streptozotocin treatment has also been reported to reduce the levels of creatine kinase activity and mRNA levels, limiting the availability of the substance for myosin ATPase (Popovich 1989). Depression of myosin ATPase could also in part be the result of hypothyroidism because chronic administration of triiodothyronine to rats after streptozotocin treatment prevented the reduction in myosin ATPase activity and the shift in isoenzyme distribution (Dillman 1982). Hypothyroidism also depresses calcium transport by the sarcoplasmic reticulum (Suko 1971). Results from our investigation support the view that hypothyroidism is one of the factors responsible for cardiac depression because the combination of hypothyroidism and depressed cardiac contractility was seen in all diabetic animals. In the current investigation clonidine prevented the development of LVDP and also improved the index of hypertrophy. Clonidine is reported to affect the myocardial oxygen supply-demand ratio favourably by minimally altering coronary perfusion pressure, increasing diastolic perfusion time and reducing the inotropic state of ventricles in patients with congestive heart failure (Hermiller et al 1983).

In addition to hypothyroidism, alteration in lipid metabolism might also be involved in cardiac depression by modifying the structure of the cardiac plasma membrane and the subcellular membrane. In this investigation elevation of total cholesterol triglycerides and low-density lipoprotein-cholesterol levels were observed in diabetic and diabetic hypertensive rats. These results support previous reports which suggested an association between diabetes and alteration in lipid metabolism (Albrink et al 1963; Sosenko et al 1980). Elevation of serum lipids in the diabetic state indicates either the defective removal or overproduction, or both, of one or more lipoproteins. Insulin plays a role in both production and removal of triglyceride-rich proteins which might be the major cause of lipid disorders of diabetes. Insulin has an inhibitory action on HMG coenzyme A reductase which is the key rate-limiting enzyme in the cholesterol-rich low-density lipoprotein particles. Hypoinsulinaemia could, therefore, be responsible for the elevation of cholesterol levels. Treatment of diabetic rats with clonidine significantly reduced total cholesterol and triglyceride levels. It also reduced low-density lipoprotein-cholesterol levels in diabetic and diabetic hypertensive rats. The possible mechanism involved in the above changes by clonidine might be the improvement in hypoinsulinaemic state in diabetic and diabetic hypertensive animals.

Our data suggest that DOCA treatment does not aggravate the severity of cardiac dysfunction and hypertension observed in streptozotocin diabetic rats. Further chronic treatment with clonidine induces a number of beneficial effects such as prevention of hyperlipidaemia and hypothyroidism and improvement in cardiomyopathy and glycaemic control in diabetic and diabetic hypertensive rats.

Acknowledgement

The work was supported by a research grant from the Council of Scientific and Industrial Research, New Delhi, India.

References

Albrink, M. J., Lavietes, P. H., Man, E. B. (1963) Vascular disease and serum lipids in diabetes mellitus: observations over thirty years (1931–1961). Arch. Intern. Med. 58: 305–323

Banerjee, S. K. (1983) Comparative studies of atrial and ventricular myosin from normal thyrotoxic and thyroidectomized rabbits. Circ. Res. 52: 131–136

Bangaru, R. A., Lakkad, N. B., Goyal, R. K. (1990) Effects of chronic atenolol treatment on diabetes-induced cardiac depression and other complications. Indian J. Pharmacol. 23: 33–34

Bolton, S. (1984) In: Pharmaceutical Statistics: Practical and Clinical Application. 4th edn, Marcel Dekker, New York, pp 218–232

Bunag, R. D., Tomita, T., Saski, S. (1982) Streptozotocin diabetic rats are hypertensive despite reduced hypothalamic responsiveness. Hypertension 4: 556–565

Cavaliere, T. A., Taylor, D. G., Kerwin, L. J., Antonaccia, M. J., (1980) Cardiovascular effects of alloxan diabetes in normotensive and spontaneously hypertensive rats. Pharmacology 20: 211–223

Dillman, W. H. (1982) Influence of thyroid hormone administration on myosin ATPase activity and myosin isoenzyme distribution in the heart of diabetic rats. Metabolism 31: 199–204

Factor, S. M., Minase, T., Sonnenblick, E. H. (1980) Clinical and morphological features of human hypertensive diabetic cardiomyopathy. Am. Heart J. 99: 446–458

Fuller, J. H., Shipley, M. J., Rose, G., Jerrtt, R. J., Keen, H. (1983) Mortality from coronary disease and stroke in relation to degree of glycaemia. The Whitehall study. Br. Med. J. 287: 867–878

Goyal, R. K., McNeill, J. H. (1985) Effect of triiodo-1-thyronine treatment on cardiac responsiveness to methoxamine in streptozotocin-induced diabetic rats. Fed. Proc. 44: 7241

Haeusler, G., Finch, L. (1972) On the nature of the central hypotensive effect of clonidine and methyldopa. J. Pharmacol. 3: 544–545

Haller, H., Lindschau, C., Quass, P., Distler, A. (1991) High glucose concentration(s) increase protein kinase C activity in vascular smooth muscle cells. J. Hypertens. 10: 145A

Hebden, R. A., Todd, H. E., Tang, C., Goven, B., McNeill, J. H. (1990) Association of DOCA hypertension with induction of atherosclerosis in rats with short-term diabetes mellitus. Am. J. Physiol. 258: R1042–R1050

Hermiller, J. B., Magorien, R. D., Leithe, M. E., Unverferth, D. V., Leier, C. V. (1983) Clonidine in congestive heart failure: a vasodilator with negative inotropic effects. Am. J. Cardiol. 51: 791–795

Hofteizer, V., Carpenter, A. M. (1973) Comparison of streptozotocin-induced diabetes in the rat inducing volumetric quantitation of the pancreatic islets. Diabetologia 9: 178–184

Houston, M. C., Burger, C., Hays, J. T., Nadean, J., Swift, L., Olafsson, L. (1990) Effect of clonidine hydrochloride versus atenolol monotherapy on serum lipids lipid subfractions and apolipoproteins in mild hypertension. Am. Heart J. 120: 172–179

Ishii, K., Yamamoto, S., Kato, R. (1985) Increase in insulin response to glucose in rat chronically treated with clonidine. Naunyn Schmiedebergs Arch. Pharmacol. 328: 253–257

Lakkad, N. B., Bangaru, R. A., Rao, M. V., Goyal, R. K. (1992) Studies on the chronic treatment of enalapril and prazosin on diabetes-induced cardiac depression and other complications. Indian J. Pharmacol. 24: 62

Lancranjan, I., Marbach, P. (1977) New evidence for growth hormone modulation by the alpha-adrenergic system in man. Metabolism 26: 1225–1230

Lewis, S. J., Dunlop, M., Jarrott, B. (1989) Serum glucose and insulin levels in normotensive (WKY) and spontaneously hypertensive (SH) rats during and after the cessation of continuous (10 day) clonidine infusion. J. Pharm. Pharmacol. 41: 353–355

Malaisse, W. J., Malaisse-Large, F., King, S., Wright, P. H. (1968) Effect of growth hormone on insulin secretion. Am. J. Physiol. 215: 423–428

Martin, J. M., Akerblom, H. K., Garay, G. (1968) Insulin secretion in rats with elevated levels of circulating growth hormone due to MT-W15 tumor. Diabetes 17: 661–667

Nakaki, T., Nakadate, T., Ishii, K., Kato, R.(1981) Postsynaptic alpha-2 adrenergic receptors in isolated rat islets of Langerhans: inhibition of insulin release and cyclic 3′:5′-adenosine monophosphate accumulation. J. Pharmacol. Exp. Ther. 216: 607–612

Popovich, B. K. (1989) Diabetes decreases creatine kinase enzyme activity and mRNA level in the rat heart. Am. J. Physiol. 257: E573–E577

Rodrigues, B., Agrawal, D. K., McNeill, J. H. (1985) Are elevated plasma lipid and diabetic cardiomyopathy related? Fed. Proc. 44: 7291

Rodrigues, B., Goyal, R. K., McNeill, J. H. (1986) Effects of hydralazine on streptozotocin-induced diabetic rats: prevention of hyperlipidaemia and improvement of cardiac function. J. Pharmacol. Exp. Ther. 23: 292–299

Ruch, W., Jaton, A. L., Bucher, B., Marbach, P., Doepfner, W. (1976) Alpha adrenergic control of growth hormone in adult rats. Experientia 32: 520–530

Satia, M. C., Damani, R. H., Shukla, M. L., Gandhi, T. P., Goyal, R. K. (1995) Clonidine: still a better option for the treatment of diabetic hypertensive patients. Indian J. Pharmacol. 27: 64

Savarese, J. J., Berkowitz, B. A. (1979) Beta-adrenergic receptors decrease in diabetic rat hearts. Life. Sci. 25: 2075–2058

Scriabine, A., Stone, C. A., Stavorski, J. M. (1968) Studies on the mechanism of St 155-induced cardiac slowing in dogs. Pharmacologist 10: 156

Shah, T. S., Satia, M. C., Gandhi, T. P., Bangaru, R. A., Goyal, R. K. (1995) Effects of chronic nifedipine treatment on streptozotocin-induced diabetic rats. J. Cardiovasc. Pharmacol. 26: 6–12

Sosenko, J. M., Breshw, J. L., Miettimen, Q. S., Gabbay, K. H. (1980) Hyperglycaemia and plasma lipid levels: a prospective study of young insulin-dependent diabetic patients. N. Engl. J. Med. 302: 650–654

Suko, J. (1971) Alterations of Ca^{+2} uptake and Ca^{+2} activated ATPase of cardiac sarcoplasmic reticulum in hyper- and hypothyroidism. Biochem. Biophys. Acta. 252: 324–327

Vadlamudi, R. V. S. V., McNeill, J. H. (1983) Effect of experimental diabetes on rat cardiac cyclic AMP phosphodiesterase and inotropy. J. Pharmacol. Exp. Ther. 244: H844–H851

Weber, M. A. (1989) Clinical pharmacology of centrally acting antihypertensive agents. J. Clin. Pharmacol. 29: 598–602