probenecid, probably because of the variability of 5-HIAA values in their experiments (see Whitton et al 1983). Timing between valproic acid and pargyline application and decapitation could be the reason why Chung Hwang & Van Woet (1979) were unable to detect an increase of 5-HT turnover in rat brain after valproic acid (see Whitton et al 1983). Dipropylacetamide, the primary amide of valproic acid also increases the turnover of 5-HT in the brain of rat (Whitton et al 1983).

Lazarova et al (1983) have observed that treatments which reduce 5-HT transmission decrease the effectiveness of valproic acid against pentetrazol-induced convulsions. Our finding that the drug increased 5-HT turnover would appear to be consistent with assumption that the anti-convulsive effect of valproic acid is mediated by 5-HT at least in part. However, it has been observed (Horton et al 1977) that depletion of cerebral 5-HT did not affect the anticonvulsant action of valproic acid in mice exposed to audiogenic convulsions produced by 109 dB.

It has been postulated that a cerebral defficiency of 5-HT may be involved in the aetiology of some types of myoclonic epilepsy (Chadwick et al 1975), a condition in which valproic acid appears to be therapeutically useful (Pinder et al 1977). Fahn (1978) described a patient with severe post-anoxic intention myoclonus in whom 5-HIAA was undetectable in cerebrospinal fluid (CSF); valproic acid improved the patient during which time the CSF 5-HIAA level rose to normal. Current evidence suggests that 5-HT may play a role in protective effect of valproic acid in some types of seizure.

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The effect of indoramin on plasma lipids in hypercholesterolemic patas monkeys

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In patas monkeys fed a high cholesterol diet, chronic dosing with the antihypertensive agent indoramin increases the cholesterol ratio by raising the plasma HDL-C concentration. Epidemiological evidence suggests that increasing HDL-C reduces the incidence of coronary heart disease (CHD). If, therefore, indoramin is able to evoke these changes in man it may be expected to favourably influence the progress of CHD.

Epidemiological evidence from studies such as the Framingham Heart Study suggests that certain physiological parameters can be used to assess the risk of developing coronary heart disease (CHD) (Castelli 1984). Those physiological parameters of primary importance, known as risk factors, have been identified to be blood pressure and plasma lipid content. Elevation of either of these above the normal range predisposes towards the development of CHD.

Treatments to lower blood pressure have had beneficial effects upon the incidence of stroke and congestive heart failure in hypertensive patients, but despite this success it has proved difficult to show a concomitant reduction in CHD after treatment for hypertension. In some studies the treatment has been shown to increase the incidence of CHD. The reason for this finding is thought to be that current treatments aggravate one or more risk factors thereby negating the beneficial effect of lowering blood pressure (Ames 1983). One risk factor affected by drug therapy is the level and constituents of plasma lipids. The major plasma lipids, including cholesterol, are transported in the plasma in three main lipoprotein complexes: very low density (VLDL), low density (LDL) and high density (HDL).

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There is a positive correlation between the concentration of VLDL + LDL, and a negative correlation between the concentration of HDL, and the incidence of CHD.

The effect of the antihypertensive agent indoramin, a selective α_1-adrenoceptor antagonist (Archibald 1980), on plasma lipoproteins in the patas monkey is now reported.

Methods

Six female patas monkeys were fed a commercially prepared pelleted diet (CRM, Labsure) and orally dosed with lactose-containing capsules twice daily (9 am and 5 pm). Throughout the study blood samples (5 ml) were taken from the femoral vein at weekly intervals following an overnight fast.

After a 3 week 'run in' period, the diet was changed to a high cholesterol mixture (skimmed milk powder, 30%; tallow, 15%; nutramol 30, 17·5%; casein, 13%; ground whole wheat, 19·8%; essential vitamins and minerals, 4·2%; cholesterol, 0·5%). After a further three weeks three of the monkeys continued to receive the placebo and the other three were dosed with indoramin (2·5 mg kg^{-1} p.o. twice daily). After eight weeks of this regime the treatments were crossed over. Thus after the eleventh week on the modified diet monkeys previously receiving indoramin were given lactose and vice versa.

Indoramin was prepared as the hydrochloride salt by the Dept of Chemistry, Wyeth Laboratories.

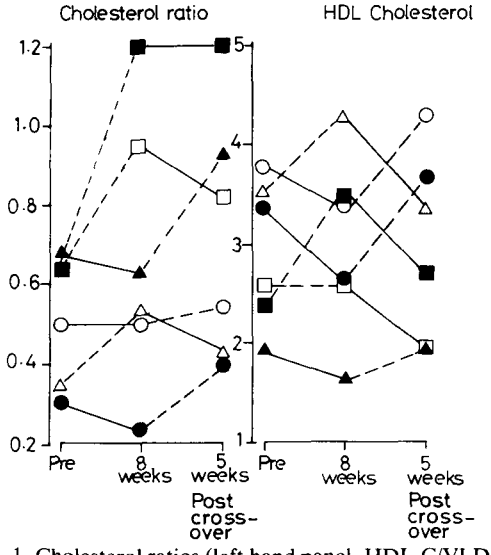

FIG. 1. Cholesterol ratios (left hand panel, HDL-C/VLDL-C+LDL-C), and HDL-C concentrations (right hand panel, mmol litre^{-1}) of individual patas monkeys fed a high cholesterol diet. The values given are those obtained before dosing (PRE); after eight weeks dosing with either placebo or indoramin (2·5 mg kg^{-1} p.o. twice daily); and five weeks after crossing over the treatments. The period for which each monkey was given indoramin is represented as a broken line and the period on placebo as a solid line.

Analysis of blood samples. The blood samples were collected into lithium-heparin tubes and then centrifuged at 3000*g* for 10 min. The plasma was then decanted and frozen until lipid analysis. When thawed, the samples were defibrinated and divided into three.

The lipoprotein fractions were separated by electrophoresis on cellulose acetate membranes in Tris-buffer (pH 8·8) at 180 V for 25 min. The cholesterol content of each fraction was visualized with a chromogen, using the CHOD-PAP method (Helena HDL kit, super ZX 5471) of Allain et al (1974). Quantification of the cholesterol content of each fraction was by densitometry at 505 nm (Gelman ACD18 Densitometer).

A second sample of plasma was taken to measure the total cholesterol content of each sample using the CHOD-PAP method with a Boehringer Kit (BCL cholesterol C-system, no. 290319).

The third sample was used to measure the triglyceride content of the sample by the enzymic hydrolysis method of Bucolo & David (1973), (Coulter Kit 996682).

The cholesterol ratio was calculated as HDL-C/VLDL-C+LDL-C (Johnson 1982; Leren et al 1982). Changes in this ratio, the HDL-C, VLDL-C+LDL-C concentrations, and the triglyceride concentration following indoramin treatment were compared statistically with those changes occuring after placebo treatment using an unpaired *t*-test.

Results

Throughout the text data are given as mean ± s.e.m.

After the monkeys had been on the cholesterol-containing diet for three weeks, their total plasma cholesterol concentration reached a steady state, rising from 2·52 ± 0·11 to 9·39 ± 1·7 mmol litre^{-1}. The cholestrol ratios ranged from 0·30 to 0·68, the HDL-C concentrations ranged from 1·93 to 3·79 mmol litre^{-1} and the VLDL-C+LDL-C concentrations ranged from 2·84 to 11·28 mmol litre^{-1}.

The cholesterol ratio and the HDL-C concentrations for individual monkeys are illustrated in Fig. 1. These values were obtained from blood samples taken before dosing with indoramin (pre), at the point where the treatments were crossed over (8 weeks after dosing commenced), and five weeks after the crossover.

After treatment with placebo for eight weeks, decreases in both the cholesterol ratio and HDL-C concentration compared with predose values were observed. The mean falls in these parameters were 0·04 ± 0·02 and 0·49 ± 0·14 mmol litre^{-1} respectively (Fig. 1). In contrast, in those monkeys receiving indoramin, both the cholesterol ratio and HDL-C concentration rose (mean increases, 0·36 ± 0·11 and 0·63 ± 0·31 mmol litre^{-1} respectively, Fig. 1). These rises were significantly different from the falls observed in the placebo group. ($P < 0·05$).

Decreases in the VLDL-C+LDL-C concentrations were observed following either placebo or indoramin treatments. Statistical evaluation of this data revealed

that the falls following indoramin (1·1 ± 0·12 mmol litre^{-1}) were significantly greater ($P < 0·05$) than those observed in the placebo group (0·38 ± 0·19 mmol litre^{-1}).

Following the crossover of the treatments the changes described above were reversed. The cholesterol ratios of two of the monkeys previously receiving indoramin fell when the monkeys were given placebo (the ratio of the remaining monkey did not change, Fig. 1). Conversely, the monkeys now given indoramin exhibited increasing ratios which rose by 0·13 ± 0·06 after five weeks (Fig. 1). Comparison of the data from these two groups reveals that this is a significant positive change compared with the placebo group ($P < 0·05$). The HDL-C concentrations fell in all those monkeys now given placebo (mean decrease = 0·80 ± 0·07 mmol litre^{-1}), and rose in all those given indoramin (mean increase = 0·72 ± 0·21 mmol litre^{-1} Fig. 1). The rise evoked by indoramin was significantly different from the changes observed in the placebo group. ($P < 0·05$).

The changes in the VLDL-C+LDL-C concentrations following the crossover were more variable than those seen in the other parameters. No significant difference was found between the changes in the two groups of animals.

The plasma triglyceride concentrations of the two groups did not significantly change relative to each other at any stage of the experiment.

Discussion

Indoramin is an effective and well-tolerated antihypertensive agent in man (Archibald 1980). Previous work has demonstrated that this compound lowers blood pressure in other species including the patas monkey (Archibald 1980). We have now demonstrated in the patas monkey that indoramin evokes significant rises in the cholesterol ratio (HDL-C/VLDL-C+LDL-C) and furthermore that this rise is almost entirely due to a rise in the HDL-C concentration.

Several studies have demonstrated that in addition to hypertension and smoking, elevated plasma cholesterol concentrations are an independent risk factor for the development of CHD (Castelli 1984). In recent years it has become evident that measurement of serum total cholesterol and triglycerides provides only limited information about circulating serum lipoproteins. It is now known that total serum cholesterol is the sum of three major fractions known as VLDL-C, LDL-C and HDL-C. The available evidence seems to suggest that VLDL-C and LDL-C promote CHD and HDL-C exerts a protective effect on the arterial wall (Miller & Miller 1975; Castelli et al 1977). As a consequence of these relations there may be an increased risk of CHD when the total serum cholesterol is within normal limits if a high proportion of LDL and/or VLDL is present. Consideration of these findings suggests that ideally in order to decrease the risk of CHD as much as possible an antihypertensive agent should, in addition to decreasing blood pressure, also lower the concentration of VLDL-C+LDL-C and raise the HDL-C concentration.

In preliminary experiments carried out to select a model for this study, electrophoretic separation of the plasma lipid fractions revealed that the patas monkey had a lipid profile similar to man, with a clear separation between the HDL and LDL fractions. A similar finding has previously been reported by Mahley et al (1976). In order to make our model approximate to the expected disease state in man the plasma cholesterol concentration of the monkeys was raised to 9·39 ± 1·7 mmol litre^{-1} by feeding them a high cholesterol diet. In man it is common to consider concentrations above 7 mmol litre^{-1} as hypercholesterolemic (Miller 1983).

This study has demonstrated that in monkeys fed a high cholesterol diet, indoramin significantly raises the cholesterol ratio by increasing the HDL-C concentration and lowering the VLDL-C+LDL-C concentration after eight weeks of treatment. Furthermore, animals which had been receiving the cholesterol diet for 11 weeks tended to exhibit potentially detrimental falls in the cholesterol ratio due to a reduction in the HDL-C concentration. These falls were reversed by subsequent indoramin treatment. Clearly, if indoramin were to exert these effects in man it would closely resemble the profile of an ideal antihypertensive agent as outlined above.

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