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Lipophilicity and pharmacodynamics of cardiotonic steroids in guinea-pig isolated heart muscle preparations

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1 The inhibitory potency of cardiotonic steroids on myocardial Na^+-K^+-ATPase increases with increasing lipophilicity. Employing the inotropic action on guinea-pig isolated left atria, the relationship between lipophilicity, chemical structure and the pharmacodynamic properties of cardiac glycosides was further examined. Nineteen digitoxigenin- and digoxigenin-derivatives, whose lipophilic nature and inhibitory effects on the myocardial Na^+-K^+-ATPase have been previously investigated, were tested.

2 All steroids exhibited positive inotropic effects which varied with the lipophilicity of these drugs. The dependence of the relationship between these two parameters on structural transformations of the steroids showed qualitatively very close parallelism to that between lipophilicity and their inhibitory effects on myocardial Na^+-K^+-ATPase.

3 The positive inotropic effects and the inhibitory effects on myocardial Na^+-K^+-ATPase also correlated very well, exhibiting similar patterns in their respective correlations with the individual lipophilicity parameters.

4 It is inferred that (a) the positive inotropic effects of the cardiac glycosides vary with their lipophilicity, exhibiting trends similar to those shown for their inhibitory effects on myocardial Na^+-K^+-ATPase, (b) this interdependence is secondary to the influence of structural changes, particularly on the steroid nucleus, and (c) both the lipophilic nature and pharmacodynamic behaviour of the cardiac glycosides almost exclusively depend on the steroid nucleus itself.

Introduction

Despite extensive investigations during the past two decades, the relationship between the inhibitory activities of cardiotonic steroids on myocardial Na^+-K^+-ATPase (E.C. 3.6.1.3.) and their positive inotropic actions still remains a subject of controversy. Opinions of different investigators seem to be further polarizing rather than merging towards a common consensus.

The main pharmacodynamic property of the cardiac glycosides is their ability to increase the force of myocardial contraction. Although their therapeutic value in the treatment of congestive heart failure is still unchallenged, the toxicity of these drugs, resulting from their narrow therapeutic range, remains a serious clinical problem. Indeed, it is this outstanding character of the cardiac glycosides that has provided such an impetus to research on their pharmacodynamic properties and has provided more information on bioavailability for this class of drugs than for almost any other.

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Our present knowledge of the pharmacological activities of these steroids owes its roots mainly to the investigations on their sugar moieties (Thomas *et al.*, 1974a,b) and the lactone ring (Portius & Repke, 1964; Chen & Henderson, 1965; Matsui & Schwartz, 1968; Jones & Middleton, 1970; Saito *et al.*, 1970). Although these two components and their derivations profoundly affect the pharmacological behaviour of the steroids, they are known today to be dispensible for the activity of cardiac steroids (Jones & Middleton, 1970).

The few investigations so far carried out on the steroid nucleus indicate on the other hand, that it is virtually impossible to distort the rigid steroid structure without rendering the glycosides inactive (Zürcher *et al.*, 1969; Ishikawa *et al.*, 1974). The aglycone moiety has until now been considered, theoretically at least, to be largely responsible for the pharmacodynamic responses of these compounds. Thus, the prerequisite for the cardiotonic steroids to retain their pharmacodynamic activity is that the steroid nucleus

remains intact.

Until just over a decade ago, the hypothesis suggesting that myocardial Na^+-K^+-ATPase is the 'receptor' for the positive inotropic activity of the cardiotonic steroids still claimed widest support as an explanation for their mode of action (Repke & Portius, 1963; Glynn, 1964; Repke *et al.*, 1965; Schwartz *et al.*, 1974). However, with the use of more sophisticated and specific biochemical methods for receptor studies, profound scepticism has developed towards this hypothesis (Dutta & Marks, 1969; Okita *et al.*, 1973; Park & Vincenzi 1975; 1976; Fricke, 1978; Isenberg; 1984).

Although it is well established today that both their inhibitory actions on the myocardial Na^+-K^+-ATPase as well as their positive inotropic potencies are unique for these particular steroids, and despite numerous parallelisms drawn between these two properties of the cardiotonic steroids (Akera *et al.*, 1973; Allen *et al.*, 1975; Lindenmayer, 1976; Grupp *et al.*, 1985), there is no direct evidence that Na^+-K^+-ATPase inhibition is the underlying mechanism for the positive inotropic effects of these drugs. In fact, the detailed information being acquired indicates that their inhibitory actions on the only identified specific binding site, the Na^+-K^+-ATPase, may not necessarily be responsible for their positive inotropic effects (Fricke & Klaus, 1969; 1971a,b; Ten Eick *et al.*, 1973; Okita *et al.*, 1973; Brown & Erdmann, 1984; Erdmann *et al.*, 1984; Fricke, 1984; Werdan *et al.*, 1984).

It is, however, generally agreed that the toxic effects of the cardiotonic steroids are related to their inhibitory actions on myocardial Na^+-K^+-ATPase. From pharmacokinetic studies, these effects are also known to be dependent on their lipophilic nature (Schwartz *et al.*, 1974; Hougen & Smith, 1978). It is therefore surprising that, in view of our present understanding of their reaction mechanism, very little has been done so far to investigate the relationship between lipophilicity and the pharmacodynamic properties of the cardiac glycosides.

In a previous study, the relationship between chemical substitution, lipophilicity and the inhibitory effects of almost all commercially available cardiotonic steroids on myocardial Na^+-K^+-ATPase has been discussed (Dzimiri *et al.*, 1987). In this paper, a further attempt has been made to try and establish a broadly-based relationship between lipophilicity and the pharmacodynamic properties of the cardiac glycosides. For this purpose, nineteen digitalis derivatives have been selected from the foregoing study, paying particular attention to certain structural variations, such as hydrogenation of the lactone ring, substitution of the sugar moieties and the C_{12}-hydroxyl group of digoxin. The relationship between lipophilicity and inotropic effects of the cardiac glycosides on guinea-pig isolated left atria was then comparatively analysed taking into account previous investigations of their potencies as inhibitors of myocardial Na^+-K^+-ATPase.

Methods

Atria were obtained from guinea-pigs (200–300 g) of either sex, and were stimulated (twice threshold voltage) at a frequency of 2 Hz, with a pulse width of 6 ms and a resting tension of 9.8 mN (1.0 g). Contractile force was measured isometrically by means of a force-displacement transducer and recorded after amplification (Fleck, Mainz, F.R.G.) on a thermorecorder (Hellige Servomed, Freiburg, F.R.G.). After pre-incubation for about 30 min in an 80 ml double-walled tissue bath, the atria were transferred into a 10 ml bath (Dr Dinkelacker and Co., Mainz, F.R.G.) containing modified Tyrode solution (mmol l^{-1}: Na^+ 155.0, K^+ 5.4, Ca^{2+} 0.9, Mg^{2+} 1.1, Cl^- 152.4, HCO_3^- 11.9, $H_2PO_4^-$ 0.4 and glucose 10.0) which was equilibrated with 95% O_2: 5% CO_2 to give a pH of 7.4 at a temperature of 30°C.

The inotropic potencies of the cardiac steroids were determined according to the method of Reiter (1983) using dihydrouabain as reference. The atria were first stimulated at 1, 2, 3 and 4 Hz successively at regular intervals and then allowed to return to their original amplitudes at 2 Hz. Thereafter, cumulative doses of dihydro-ouabain were singly pipetted directly into the tissue bath until the atria showed clear toxic effects, appearing as arrhythmias, contracture or decreasing contractile force. After three washings, the atria were transferred again into the 10 ml bath, being allowed to stabilize to their original amplitudes. Cumulative doses of the cardiac glycosides to be studied were then applied until the muscles showed toxic effects as above.

The concentrations for half-maximal positive inotropic effects (ED_{50}) were calculated from the individual concentration-response curves as proposed by Hafner *et al.* (1977). pD_2-values were calculated as the negative logarithms of the concentrations required for half maximal positive inotropic effects of the respective cardiotonic steroids. The effective concentration range (ED_{10}–ED_{max}) is the range within which the cardiac steroid exerts its positive inotropic effect without exhibiting toxicity. All data were analysed by standard statistical methods (mean value and s.e. mean, regression analysis).

Materials

The cardiotonic steroids examined (cf. Table 1) were purchased from Merck (F.R.G.) or Roth (F.R.G.). All other reagents (analytic grade), unless otherwise stated, were obtained from Merck (F.R.G.).

The solvent used was 5% dimethylsulphoxide (DMSO) in propanediol-1.2.

Results

The positive inotropic potencies of the digitalis compounds studied, are depicted as their median effective doses (ED_{50} values) in /Table 1. All the steroids exhibited concentration-dependent positive inotropic effects, their maxima ranging between 90 and 106% of that produced by dihydro-ouabain. Two distinct types of positive inotropic effective dose-ranges (piE-ranges) could be evaluated, namely the high concentration ranges for the cardanolides and the C_{12}-OH derivatives on the one hand, and the comparatively low ranges for the rest of the cardenolides on the other. The respective ED_{50} values comprise a concentration-range of about four decades, the aglycones after the dihydro-derivatives being the least potent cardiotonic steroids.

Attachment of a sugar moiety to the steroid produces a highly significant shift of the concentration-response curves to the left (Figures 1 and 2), except with 12-acetyldigoxin which showed an even weaker effect than digoxigenin (Figure 3). However, further increasing the number of the sugar moieties gave rather variable results, depending on the genin studied. As indicated by the respective affinities (taken from the ED_{50} values), among the digitoxigenin derivatives, increasing the number of sugar moieties leads to a nearly four-fold weakening of the positive inotropic potency (Table 1, Figure 1). In contrast, the digoxigenin-derivatives exhibited a group shift rather in the same order as their inotropic potencies, the 12-acetyl derivatives being the least and the non-substituted sugar derivatives the most active. The alkyl (acyl) digoxin derivatives occupied an intermediate position.

Digitoxigenin-mono-digitoxoside exhibited the highest potency of all compounds studied, the three sugar derivatives of digitoxigenin showing much more marked differences among themselves than those of digoxigenin (Figures 1 and 2). With regard to the three pharmacological entities of the cardiac glycosides, i.e. the steroid nucleus, the sugar moieties and the lactone ring, the following observations were made: (a) Based on the ED_{50} values, the more lipophilic digitoxigenin derivatives exhibited higher positive inotropic potencies than their corresponding digoxigenin analogues.

Table 1 Inotropic effect of digitalis compounds in guinea-pig isolated left atria

Cardenolides	R_m	*piE-range* ($\times 10^{-7}$ mol l^{-1})	*piE-max* (% DHO)	ED_{50} ($\times 10^{-7}$ mol l^{-1})	n
1 Digitoxigenin	1.03	0.89–19.2	102.3 ± 7.6	5.27 ± 0.67	12
2 Digitoxigenin-mono-digitoxoside	1.09	0.09–1.0	94.7 ± 12.3	0.47 ± 0.01	9
3 Digitoxigenin-bis-digitoxoside	1.28	0.22–3.4	89.9 ± 9.3	1.11 ± 0.10	10
4 Digitoxin	1.37	0.40–5.5	89.5 ± 9.1	1.47 ± 0.31	12
5 Dihydro-Digitoxin	1.42	3.80–220.0	99.1 ± 15.5	73.34 ± 6.41	9
6 β-Methyldigitoxin	1.71	0.30–3.6	91.0 ± 11.6	1.08 ± 0.13	12
7 Digoxigenin	−0.02	7.01–200.0	96.2 ± 11.5	35.79 ± 5.62	10
8 Digoxigenin-mono-digitoxoside	0.01	0.93–12.2	96.1 ± 13.7	2.60 ± 0.54	14
9 Digoxigenin-bis-digitoxoside	0.15	0.79–7.3	99.1 ± 14.7	2.40 ± 0.23	12
10 Digoxin	0.25	0.89–9.1	100.1 ± 11.0	3.40 ± 0.37	11
11 Dihydro-digoxin	0.24	10.90–400.0	96.8 ± 16.3	487.70 ± 54.95	7
12 α-Methyldigoxin	0.54	0.29–16.8	106.3 ± 12.1	4.42 ± 0.63	12
13 β-Methyldigoxin	0.59	0.98–18.5	95.9 ± 16.3	5.10 ± 0.71	14
14 α.β-Dimethyldigoxin	0.94	1.10–13.4	100.3 ± 9.1	3.47 ± 0.44	14
15 α-Acetyldigoxin	0.50	1.00–17.4	97.7 ± 14.3	5.15 ± 0.68	13
16 β-Acetyldigoxin	0.70	1.29–18.0	96.4 ± 14.4	5.11 ± 0.74	14
17 α.β-Diacetyldigoxin	1.16	1.00–16.6	94.4 ± 12.3	4.63 ± 0.74	15
18 12-Acetyldigoxin	0.89	24.30–310.0	103.5 ± 13.1	77.38 ± 7.43	9
19 12-Acetyl-β-methyldigoxin	1.26	13.10–280.0	105.2 ± 14.5	83.63 ± 7.03	11
32 Dihydro-ouabain	−0.71	20.65–190.0	100.0 ± 0.0	61.92 ± 0.61	10

Shown are (1) the positive inotropic effective dose-ranges (piE-range = $ED_{10} - ED_{max}$), (2) maximum percentage increase of contractile force (piE-max) based on maximal inotropy obtained with dihydro-ouabain (% DHO) ± s.e.mean and (3) the half maximal effective doses (ED_{50}) ± s.e. of *n* individual determinations. For comparison, lipophilicity as expressed as R_m values is given (data obtained from Dzimiri *et al.*, 1987).

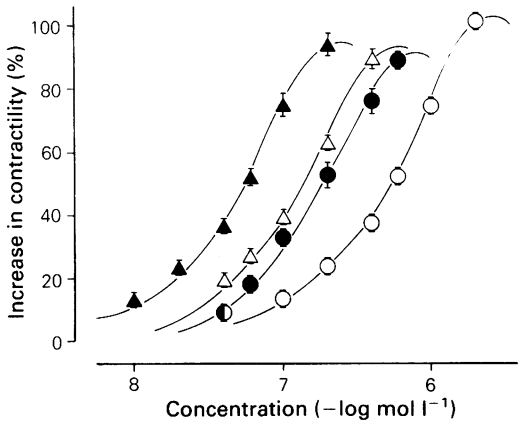

Figure 1 Positive inotropic effects of digitoxigenin (○) and its non-acylated sugar derivatives digitoxigenin-mono-digitoxoside (▲), digitoaxigenin-bis-digitoxoside (△) and digitoxin (●) in guinea-pig isolated left atria. Plotted is the increase in contractile force as percentage of the maximum contractility induced by dihydro-ouabain. Drugs were added cumulatively until toxic effects were obtained. Mean values and the standard deviation of the mean of 9–12 individual experiments are given. Contractile force before adding the drugs (control) was in the range of 2.96 and 3.56 mN.

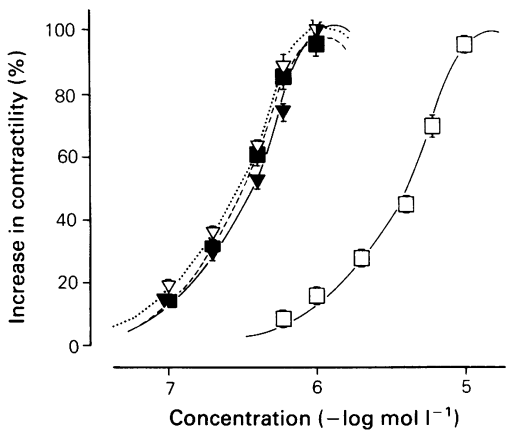

Figure 2 Positive inotropic effects of digoxigenin (□) and its non-acylated sugar derivatives digoxigenin-mono-digitoxoside (■), digoxigenin-bis-digitoxoside (▽) and digoxin (▼) in guinea-pig isolated left atria. Plotted is the increase in contractile force as percentage of the maximum contractility induced by dihydro-ouabain. Drugs were added cumulatively until toxic effects were obtained. Mean values and the standard deviation of the mean of 12–14 individual experiments are given. Contractile force before adding the drugs (control) was in the range of 2.94 and 3.36 mN.

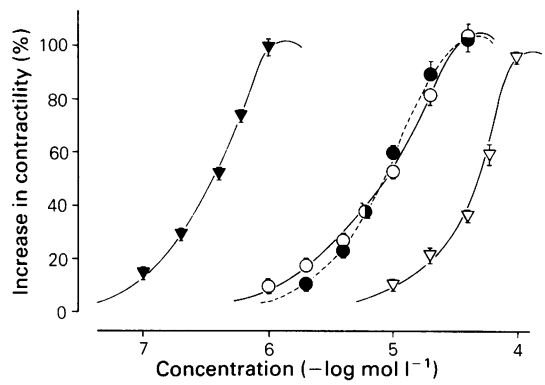

Figure 3 Positive inotropic effects of digoxin (▼) and its dihydro-(▽) and C_{12}-OH derivatives, 12-acetyldigoxin (●) and 12-acetyl-β-methyldigoxin (○) in guinea-pig isolated left atria. Plotted is the increase in contractile force as percentage of the maximum contractility induced by dihydro-ouabain. Drugs were added cumulatively until toxic effects were obtained. Mean values and the standard deviation of the mean of 7–14 individual experiments are given. Contractile force before adding the drugs (control) was in the range of 2.94 and 3.91 mN.

Acylation of the C_{12}-hydroxyl group (digoxin) markedly reduced the positive inotropic effects. Annexation of sugar moieties onto the genin increased the potencies by a factor of about ten, but further addition of sugar moieties showed neither a significant change nor a definite pattern. (b) Derivation of the sugar moieties had a slightly weakening effect on the inotropic potencies of digoxin, the effect of acetylation being more marked than that of methylation. On the other hand, β-methyldigitoxin and digitoxin were equipotent. (c) Hydrogenation of the lactone ring of both digitoxin and digoxin decreased the positive inotropic potencies of the parent cardenolides by a factor of ten.

Discussion

The correlation between the R_m values of the cardiotonic steroids, as depicted from Dzimiri *et al.* (1987), and the pD_2 values of their inotropic potencies in general is very weak. This is mainly due to the positions occupied by the cardenolides and the C_{12}-OH (digoxin) derivatives, since omission of these compounds transformed this correlation into a significant one (Figure 4), following a pattern similar to that established for the relationship between lipophilicity and the inhibitory potencies of the cardiotonic steroids

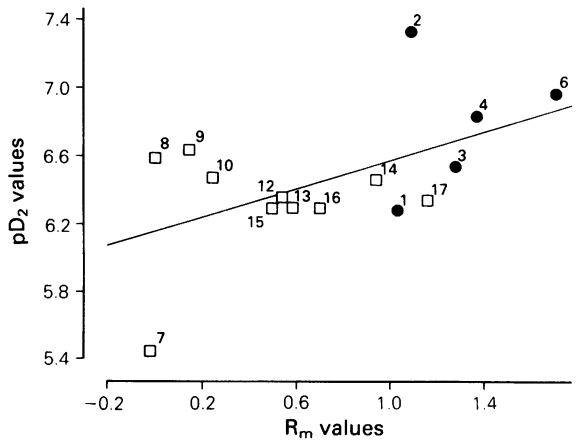

Figure 4 Correlation of lipophilicity and the positive inotropic action of the cardiac glycosides studied. Plotted are the pD_2 values against the R_m values. For code numbers given beside each point see Table 1. $y = 0.42x + 6.16$, $r = 0.54$ (0.20), $n = 15$ (19). The values in parenthesis represent the regression coefficient including the cardanolides (dihydro-derivatives) and the 12-acetylderivatives of digoxin.

on the myocardial Na^+-K^+-ATPase (Dzimiri *et al.*, 1987).

The roles and magnitudes of the influence of the three entities quoted under results on both the physicochemical properties as well as the pharmacodynamic responses of the cardiotonic steroids are qualitatively and quantitatively different from one another, but are consistently identical for each component. Thus, for example, whereas changes in the sugar moieties was accompanied by minimal variations in the respective pharmacodynamic responses, any slight modification of the steroid nucleus was always associated with massive loss or gain in lipophilicity and considerable reduction of the positive inotropism. Furthermore, although hydrogenation of the lactone ring does not influence lipophilicity of the steroids, it drastically reduced both pharmacodynamic properties.

According to the model of Thomas *et al.* (1980), it is particularly the sugar moiety closest to the steroid nucleus that is directly involved in the interactions of the cardiac glycosides with their receptor counterpart. The marked difference between the potencies of the aglycones and their mono-digitoxosides suggests that the attachment of the sugar molecule may indeed greatly influence these interactions. It is likely though, that their influence is of a pharmacokinetic nature, for example, enhancing the bioavailability of the steroids. They are known to play a major role in the pharmacokinetics of the cardiac glycosides (Saito *et al.*, 1970; Repke, 1972; Yoda & Yoda, 1977). Irrespective of the way in which the moieties may contribute towards the activities of the steroids, acylation weakens their influence, probably by impeding hydrogen bond forces of the hydroxyl group.

The existence of lipophilic interactions between the steroid skeleton and the receptor assumed in the model of Thomas *et al.* (1980), which has until now lacked experimental evidence, seems likely on the basis of the present studies. We consider that a number of our observations support the suggestion that the steroid skeleton is actually the important factor for both lipophilicity and pharmacodynamic behaviour of these steroids. First, the parent digitoxigenin and its derivatives were found to be the most lipophilic and pharmacodynamically the most active of the aglycones under consideration, responses of all the other compounds remaining much lower than these drugs. It was therefore virtually impossible to improve any of these characteristics by chemical transformation of the parent digitoxigenin analogues. Secondly, whereas structural variations on the sugar moieties produced no significant change and, as a result, had minimal influence on the correlations between lipophilicity and their respective pharmacodynamic properties, on the one hand, variations on C_{16}-OH fell within or even slightly improved the correlation (Dzimiri *et al.*, 1987) and acylation of C_{12}-OH was without significant effect. It can be inferred, therefore, that drastic changes of any of these characteristics can be obtained by derivation of the steroid nucleus itself, as for example, by attaching a hydoxyl group or sugar moiety or acylation of the C_{12}-OH (digoxin) but not of the other two components. Thirdly, not only did the two pharmacodynamic properties (i.e. increase in contractility and inhibition of Na^+-K^+-ATPase) exhibit an excellent correlation (Figure 5), but several parallelisms were observed on the way in which these pharmacodynamic effects are influenced by lipophilicity. These derivatives generally produced pharmacodynamic responses which exhibited similar trends for both inhibition of Na^+-K^+-ATPase and positive inotropic actions. In contrast to the hardly discernible pharmacodynamic effects resulting from variations of the sugar moieties, changes in the steroid skeleton produced marked changes in activity as demonstrated by the C_{12}-OH and C_{16}-OH derivatives of the two isomers, digoxin and gitoxin. The above points may easily be understood by comparing the properties of digitoxigenin with digitoxin, β-methyldigitoxin, β-methyldigoxin and 12-acetyl, β-methyldigoxin (see Table 1).

Our recent studies on the inhibitory effects of these cardiotonic steroids Na^+-K^+-ATPase have also shown that gitoxin exhibits slightly less inhibitory

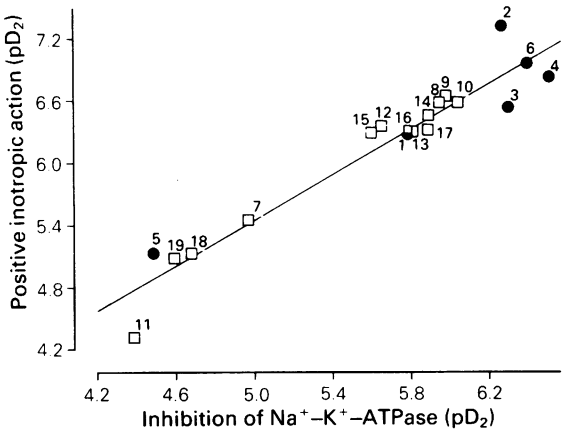

Figure 5 Correlation of the positive inotropic action and the inhibitory effects on the myocardial Na^+-K^+-ATPase of the cardiotonic steroids studied. Plotted are the pD_2 values. For code numbers given beside each point see Table 1. The drug concentrations for half maximal inhibition of the Na^+-K^+-ATPase are obtained from Dzimiri *et al.* (1987). (●) = digitoxigenin- (□) = digoxigenin-derivatives. $y = 1.10x + 0.06$, $r = 0.96$, $n = 19$.

potency than its isomer digoxin. On the other hand, whereas acylation of the hydroxyl group at position C_{16} (gitoxin) increases the inhibitory activities on Na^+-K^+-ATPase, derivation at the position C_{12} (digoxin) diminishes these effects. This inconsistency is in excellent agreement with the assumptions that the sterical configuration adopted by the cardiac steroids during their interactions with their receptor is such that position C_{12}-OH is exposed to and undergoes interaction with the receptor face, while the sterically obstructed C_{16}-OH does not. Any change at the former position interferes accordingly with this interaction. The augmentation of these properties due to acylation of the sterically hindered position at C_{16}-OH is not likely to result from interactions of this position with the receptor. We regard it rather as a direct product of the resultant increase of lipophilicity. Moreover, from the above observations, it is evident that the hydroxyl group on the steroid nucleus lessens both the lipophilicity and the pharmacodynamic properties of the cardiotonic steroids. If this group were involved in the interactions, its acylation would further diminish the inhibitory effects of the gitoxigenin derivatives as it impedes those of digoxigenin from exerting their hydrogen bond forces. The involvement of the C_{16}-OH group in the drug-receptor interactions of the steroids has also recently been disputed by Griffin *et al.* (1986). The hydroxyl group appears, therefore, to exercise a double function, its existence on the steroid nucleus reducing the lipophilicity and consequently the Na^+-K^+-ATPase inhibitory activities as well as positive inotropic effects of the cardiotonic steroids on the one hand, and its sterical position determining the probability and extent of its direct involvement in the interactions with their binding sites.

Our postulation that both lipophilicity and the pharmacodynamic behaviour of the cardiotonic steroids are almost wholly dependent upon the steroid nucleus itself prompts the question as to whether this physiochemical property is not actually the major determinant binding force for the interactions between the steroids and their receptor, resulting in the inhibition of the Na^+-K^+-ATPase as well as positive inotropic potencies of the cardiotonic steroids. Although the mode of action responsible for the inhibitory effects of the cardiac glycosides does not explain adequately the mechanism for their positive inotropism, it has not been possible, as yet, to find conclusive evidence for the existence of different mechanisms for these two pharmacodynamic properties. Our observations indicate that these two parameters are influenced in a similar way and generally to the same extent by the lipophilic nature of the steroids. Furthermore, unlike many other drug classes whose physiological targets are situated intracellularly, the catalytic face of the only yet identified receptor for the cardiac glycosides, the myocardial Na^+-K^+-ATPase, is, according to the majority of investigators, on the outside of the cell membrane (Schwartz *et al.*, 1975; Albers, 1976; Erdmann, 1977). This enzyme itself is intimately associated with the cell membrane, which is a mainly lipophilic network comprising lipids and proteins that are partially embedded in or penetrate through the lipid matrix (Singer, 1977). Thus, for the inhibitory potency of the steroids at least, we have two essentially lipophilic components, i.e. the steroid and the Na^+-K^+-ATPase itself, interacting with one another. Lipophilicity is nothing other than an expression of the tendency of a compound to associate itself with hydrophobic components, when exposed to the choice between hydrophilic and lipophilic milieus. Increasing lipophilicity of the cardiotonic steroids might therefore be expected to augment inhibition of the enzyme. This is in excellent agreement with the observations made in our recent studies (Dzimiri *et al.*, 1987).

Our findings have not only demonstrated that both the physicochemical properties and the pharmacodynamic behaviour of these cardiotonic steroids are embedded in the steroid nucleus itself, but also that at least a causal relationship does exist among the three investigated parameters. It should be apparent from this, that the lipophilic nature of this rigid steroidal sterical configuration provides one of, if not the most determinant, of all the binding forces that are involved

in the interactions with their receptor. Other binding forces, such as, hydrogen bonding or electrostatic forces, certainly do play an important role. However, taking into consideration the hypothesis that the components which supply these binding forces are actually dispensible for the positive inotropic effects of the glycosides, their contribution may be secondary to their lipophilic interactions between the steroid nucleus and the receptor-area. This raises the question whether or not the cardiac steroids may be non-specific in character, since it is well established that lipophilicity is the predominant physico-chemical property governing most non-specific interactions.

We can conclude that lipophilicity of the steroid nucleus is at least one of the predominant components in the drug-receptor interactions of the cardiac steroids for both their inhibitory activities on the myocardial Na^+-K^+-ATPase as well as their positive inotropic actions. Its influence on these pharmacodynamic properties of the cardiac steroids is governed by structural modifications, particularly on the steroid nucleus itself. Whether or not the relationship between lipophilicity, inhibitory effects on myocardial Na^+-K^+-ATPase and the positive inotropic action of cardiotonic steroids is only causal still remains to be answered.

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