J. Pharm. Pharmacol. 2000, 52: 333–340
Received October 18, 1999
Accepted October 26, 1999

Inhibitory Effects of Glibenclamide on the Contraction of Human Arterial Conduits used in Coronary Artery Bypass Surgery

ALAN E. CROSBIE, ALAIN VUYLSTEKE, ANDREW J. RITCHIE*, RAYMOND D. LATIMER AND BRIAN A. CALLINGHAM†

*Department of Anaesthesia and *Department of Surgery, Papworth Hospital, Papworth Everard, Cambridge, CB3 8RE and †Department of Pharmacology, University of Cambridge, Tennis Court Road, Cambridge, CB2 1QJ, UK*

Abstract

Glibenclamide has been shown to inhibit prostanoid-induced contraction in a number of blood vessel types. In this study, the effects of glibenclamide on the contraction of human peripheral arteries in response to both prostanoid and non-prostanoid agonists were compared and possible mechanisms of action were investigated.

Segments of left internal mammary artery (LIMA) and radial artery, taken from patients undergoing coronary artery bypass graft (CABG) surgery, were mounted in organ baths containing physiological saline solution aerated with 95% O_2/5% CO_2 at 37°C. Contractions were obtained by either the use of a thromboxane analogue (U46619), L-phenylephrine, KCl or $CaCl_2$. The effects of glibenclamide on these contractions were observed and pEC50 values were determined after manipulation of a logistic curve-fitting equation. Concentration-dependent relaxation of U46619-contracted LIMA and radial artery was observed in the presence of glibenclamide, with calculated pEC50 values of $4{\cdot}2 \pm 0{\cdot}17$ ($n = 7$) for LIMA and $3{\cdot}26 \pm 0{\cdot}48$ ($n = 5$) for radial artery. Incubation of both LIMA and radial artery with glibenclamide (50 μM) caused the concentration–response curves for U46619 and L-phenylephrine to shift significantly to the right. Similarly the KCl tension relationship was caused to shift to the right. Finally, glibenclamide (100 μM) also had an inhibitory effect on Ca^{2+}-induced tension in radial artery.

These results show that the inhibitory effects of glibenclamide on human peripheral blood vessels are not restricted to prostanoid-induced contractions. Furthermore, evidence has been provided to suggest that these effects might be mediated through an interaction with voltage-sensitive Ca^{2+} channels.

Activation of ATP-sensitive potassium (K_{ATP}) channels is known to cause vasodilatation by increasing the membrane permeability of vascular smooth muscle to potassium ions (Standen et al 1989). This effect is antagonized by the antidiabetic sulphonylurea, glibenclamide (Eltze 1989; Quast & Cook 1989; Winquist et al 1989), which blocks vascular K_{ATP} channels in a similar manner to the way that it blocks pancreatic K_{ATP} channels to cause the release of insulin (Schmid-Antomarchi et al 1987).

However, Cocks et al (1990) showed that glibenclamide, when added to isolated segments of canine coronary circumflex artery contracted with the synthetic thromboxane analogue, 9,11-dideoxy-9,11-epoxymethanoprostaglandin F_2. (U46619), caused a concentration-dependent reduction in tone. Similar results have also been found in other canine arteries as well as in rat aorta in-vitro (Zhang et al 1991). It is now apparent that this property is shared by a range of sulphonylurea compounds (Nielsen-Kudsk & Thirstrup 1991; Delaey & Van de Voorde 1997).

Cocks et al (1990) suggested that glibenclamide caused relaxation by competitive antagonism at the thromboxane A_2 receptor as the effect was not seen if the vessels had been contracted with endothelin-1, noradrenaline or potassium chloride. However, in pig coronary artery, the ability of glibenclamide and other amidoethylbenzenesulphonylureas to antagonize the vasoconstriction due to U46619,

Correspondence: A. Vuylsteke, Department of Anaesthesia, Papworth Hospital, Papworth Everard, Cambridge, CB3 8RE, UK.

was shown to correlate well with their ability to antagonize the effects of levcromakalim, a K_{ATP}-channel activator (McPherson et al 1997).

Some controversy also surrounds the idea that the inhibitory effects of glibenclamide on vascular contraction are observed only in blood vessels that have been pre-contracted with prostanoids. While it appears that the presence of glibenclamide and other sulphonylureas does not influence the contractions of rat aorta elicited by either noradrenaline, 5-hydroxytryptamine or high concentrations of potassium (Delaey & Van de Voorde 1997), Nielsen-Kudsk & Thirstrup (1991) have shown that glibenclamide could cause a relaxation of rabbit coronary artery contracted with high concentrations of potassium. In addition, Huang & Chan (1998) have found that glibenclamide reduces the contraction of the rat mesenteric artery induced by both L-phenylephrine and high concentrations of potassium. These observations suggest that not only are species differences important but also the choice of vessel as well.

Stanke et al (1998) have shown that glibenclamide relaxes human left internal mammary artery (LIMA) and saphenous vein pre-contracted with U46619. Human LIMA and radial artery are used as arterial conduits in coronary artery bypass graft operations, with the radial artery having been recently re-introduced as a favourable graft, because earlier problems with its use have been largely circumvented by improvements in the technique for harvesting the artery and also by the use of calcium-channel antagonists during surgery (Acar et al 1992; Buxton et al 1997). These arteries are increasingly regarded as superior conduits to saphenous vein and provide the promise of long-term graft patency (Metcalfe et al 1994; Nwasokwa 1995).

In this investigation, the effects of glibenclamide have been examined on isolated segments of human LIMA and radial artery in an attempt to examine further its ability to oppose the actions of vasoconstrictor agents and the underlying mechanism of action behind these effects.

Materials and Methods

This study was approved by the Huntingdon Local Ethics Committee and patients gave their informed consent before surgery.

Experimental preparation

At elective coronary artery bypass graft (CABG) operations, left internal mammary artery (LIMA) and radial artery were harvested to provide the graft vessel in heparinized pre-cardiopulmonary bypass patients under normothermia. Segments of the arteries, surplus to the needs of the operation, were placed immediately into ice-cold physiological saline solution of the following composition (mM): NaCl 118, KCl 4·5, $CaCl_2$ 1·27, KH_2PO_4 1·19, $MgSO_4$ 1·19, $NaHCO_3$ 25 and M-glucose 5·55, which had been previously aerated by bubbling with 95% O_2 and 5% CO_2 and transported to the laboratory within an hour of dissection.

On arrival, the blood vessels were transferred to fresh aerated, ice-cold physiological saline solution. After careful removal of any surrounding connective tissue, segments of artery (3 mm in length) were mounted on two parallel hooks of 0·5-mm stainless steel wire in organ baths of 10 mL capacity. One hook was fixed, while the other was attached to a Biegestab K30 isometric strain gauge transducer (Hugo Sachs Elektronik, Germany) to allow continuous recordings of tension to be made via a Plugsys model 600 polygraph (Hugo Sachs Elektronik, Germany) and recorded on a Gould model TA4000 chart recorder (Gould Electronics Inc., Cleveland, OH). Each vessel was bathed in the physiological saline solution at 37°C and aerated with 95% O_2 and 5% CO_2.

At the start of each experiment, 3 g tension was applied to each vessel via a micrometer adjustment and 1 h (LIMA) or 2 h (radial artery) allowed for equilibration with the tension maintained at 3 g by adjustment if necessary.

Experimental procedure

To test for the presence of an intact endothelium after the equilibration period, each vessel was contracted with a submaximal concentration (approximately 80% of maximum) of L-phenylephrine (3 μM), determined by constructing a concentration–response curve to L-phenylephrine and when the response had reached a plateau, 10 μM carbachol was added. Vessels which relaxed to greater than 80% of the pre-contracted tension were deemed to have an intact endothelium. Those which relaxed to less than 20% of the pre-contracted tension were considered as being endothelium-denuded. In this study, 73% of the radial artery segments used had an intact endothelium compared with only 10% of the LIMA segments.

Cumulative concentration–relaxation curves to glibenclamide were determined in both LIMA and radial artery segments, which had been pre-contracted with a submaximal concentration of U46619 (30 nM). The effect of another sulphonylurea drug, tolbutamide, on the concentration–relaxation curves caused by glibenclamide in

LIMA vessels was determined by addition of tolbutamide (200 μM) 10 min before contracting the segment with the thromboxane analogue. Other experiments examined the relaxation to glibenclamide of LIMA in the presence of the cyclooxygenase inhibitor, indomethacin (10 μM), added to the saline solution. Such experiments were designed to see if prostanoid release was involved in the relaxation caused by glibenclamide. Cumulative concentration–response curves were also constructed for the contractions caused by U46619 and L-phenylephrine. The concentration–response curves were then repeated in the presence of glibenclamide (10 or 50 μM) added 10 min before.

Concentration–response data were also obtained for KCl in the absence and presence of glibenclamide. Physiological saline solutions containing increasing concentrations of KCl were prepared by equimolar substitution of Na^+ ions with K^+ ions. In experiments with glibenclamide, vessels were incubated for 10 min with 100 μM glibenclamide immediately before addition of the saline containing a high concentration of K^+ ions.

Ca^{2+}-induced tension in depolarized arterial segments was examined by the use of a modification of the procedure outlined by Julou-Schaeffer & Freslon (1988). The vessels were washed for 30 min with Ca^{2+}-free depolarizing physiological saline solution containing 35 mM KCl and 1 mM EGTA and then for 20 min with the Ca^{2+}-free depolarizing solution without EGTA. Concentration–response curves to Ca^{2+} ions were obtained by adding increasing concentrations of $CaCl_2$ to the bath. The non-selective α-adrenoceptor antagonist, phentolamine (1 μM), was present in the bath to counteract any possible depolarization-induced neuronal release of noradrenaline. Where the effect of glibenclamide was studied, the vessels were incubated with the compound (100 μM) for 10 min after washing with Ca^{2+}-free depolarizing solution and immediately before addition of the $CaCl_2$.

Data and statistical analysis

Relaxations in response to added drugs are expressed as percentages of the contraction induced by U46619 in the same segment. Contractions are expressed as percentages of the maximum contraction caused by each agonist in each segment. All data are given as the mean ± standard error of the mean (s.e.m.). The n values are of the number of patients. When more than one vessel segment was used from the same patient, the results were pooled.

EC50 values are obtained from concentration–response curves by fitting the data to the following logistic equation by the use of KaleidaGraph software (Nbelbeck Software, version 3.05):

$$R = \frac{R_{max} \cdot C^{n_H}}{C^{n_H} + EC_{50}^{n_H}} \quad (1)$$

where R is the response, R_{max} is the maximum response, C is the concentration of the agonist and n_H is the Hill coefficient. Calculated EC50 values were then converted into negative logarithms (pEC50 values). pEC50 and R_{max} values are compared where appropriate with either Student's unpaired or paired *t*-test.

Drugs

The drugs used and their sources were as follows: L-phenylephrine hydrochloride, 9,11-dideoxy-9, 11- epoxymethanoprostaglandin F_2. (U46619), tolbutamide, indomethacin, ethylene glycol-bis-(β-aminoethyl ether)-*N*,*N*,*N*′,*N*′-tetraacetic acid (EGTA) and phentolamine methanesulphonate were all from Sigma Chemical Co., Poole, Dorset, UK; carbamylcholine chloride (carbachol) was from BDH, Poole, Dorset, UK; and glibenclamide was from Aldrich Chemical Co., Poole, Dorset, UK. All other reagents were obtained from Fisher Scientific UK Ltd, Loughborough, Leics, UK and were of analytical grade wherever possible.

Stock solutions of glibenclamide and tolbutamide were prepared in dimethylsulphoxide (Sigma, UK). The final bath concentration of dimethylsulphoxide was never allowed to exceed 0·1%. U46619 was initially dissolved in 70% (v/v) ethanol but all subsequent dilutions were made in water. Indomethacin was made up as a stock solution of 10 mM in 5% (w/v) $NaHCO_3$ solution. All other stock solutions were made in water.

Results

Effect of glibenclamide on the contractions of human LIMA and radial artery segments in response to U46619

Glibenclamide caused a concentration-dependent relaxation of both LIMA and radial artery, which had been pre-contracted with 30 nM U46619. The calculated pEC50 values were 4·2 ± 0·17 (n = 7) for LIMA (Figure 1A) and 3·26 ± 0·48; n = 5 for radial artery (Figure 1B).

Indomethacin (10 μM), when included in the bathing solution, had no significant effect on the responses to either U46619 or glibenclamide in LIMA (pEC50 value = 3·87 ± 0·01; n = 5). Pretreatment of the vessels for 10 min with 200 μM

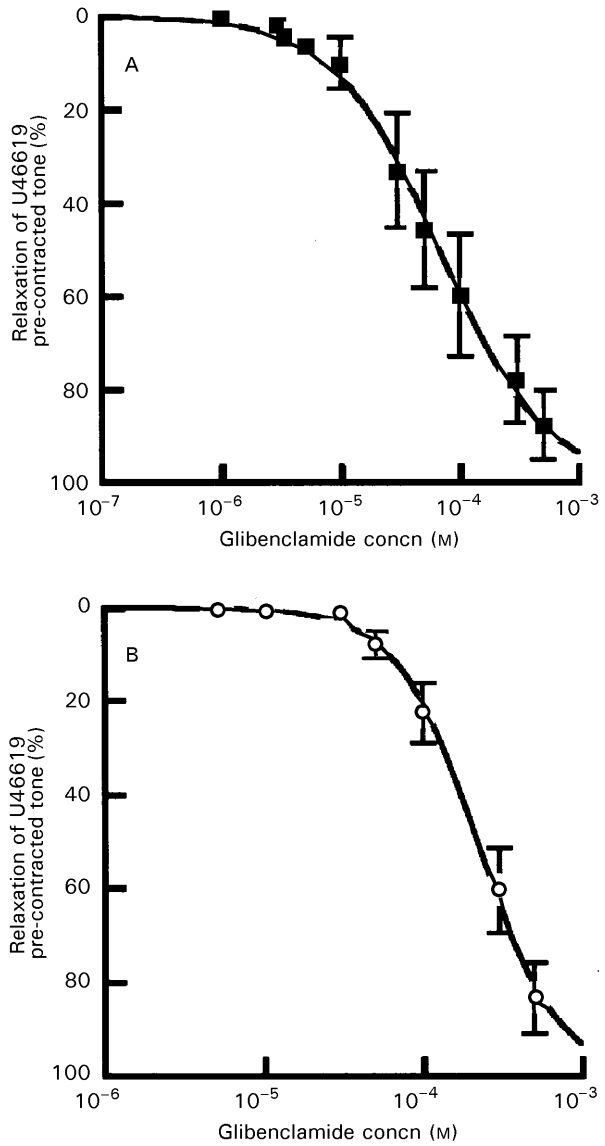

Figure 1. Relaxation of U46619 pre-contracted human LIMA (A) and radial artery (B) segments by glibenclamide. Blood vessel segments were pre-contracted with U46619 (30 nM) and then exposed to increasing concentrations of glibenclamide. Data represent mean values ± s.e.m for all segments tested from 7 patients (LIMA) and 3 patients (radial artery).

tolbutamide before contraction with 30 nM U46619 also had no effect (pEC50 value = 3·93 ± 0·31; n = 3). The control pEC50 value for these experiments was 4·14 ± 0·16 (n = 5).

Addition of glibenclamide (50 μM) 10 min before beginning the cumulative addition of increasing concentrations of U46619 to segments of radial artery or LIMA, antagonized the agonist effect of the U46619, resulting in significant shifts of the concentration–response curves to the right. With segments of LIMA and of radial artery, the changes in pEC50 were significant (Table 1; $P < 0.01$).

The effect of glibenclamide (10 μM) on segments of both arteries was not significant.

Effect of glibenclamide on the contraction of human LIMA and radial artery segments to L*-phenyl-ephrine and high concentrations of potassium*

Pre-treatment with glibenclamide reduced the contractions caused by the α_1-adrenoceptor selective agonist, L-phenylephrine, in both LIMA and radial artery segments. In the presence of 50 μM glibenclamide, the maximum responses of segments from both arteries to L-phenylephrine were significantly reduced (Table 1; LIMA, $P < 0.05$; radial artery $P < 0.001$).

Significant shifts in the concentration–response relationship for KCl were observed in both LIMA and radial artery vessels, which had been pre-treated with 100 μM glibenclamide compared with control results (Figures 2A and B) ($P < 0.05$–0·001, on comparison of control values with treated values at single concentrations of KCl).

Effect of glibenclamide on Ca^{2+}-induced tension in human radial artery

Figure 3 shows the effect of glibenclamide on Ca^{2+}-induced tension in depolarized radial artery. Increased contractions were obtained with concentrations of Ca^{2+} ions up to 3 mM and there was a reduction in tension as the concentration of Ca^{2+} ions was increased beyond this (n = 5).

After 10 min exposure to glibenclamide (100 μM), the contractions of radial artery segments, in response to added Ca^{2+} ions were reduced, with the concentration–response curve being shifted significantly to the right.

Discussion

In this investigation, the effects of glibenclamide on the contraction of both isolated LIMA and radial artery were studied. In LIMA and radial artery vessels, which had been pre-contracted with a submaximal concentration of the synthetic thromboxane A_2 receptor agonist, U46619, glibenclamide was shown to cause a concentration-dependent relaxation of the vessels (Figures 1A and B). Addition of 50μM glibenclamide to segments for 10 min before beginning to determine the concentration–response relationship was shown to inhibit U46619-induced contraction. These results are in agreement with previous findings in other species (Cocks et al 1990; Zhang et al 1991; Delaey & Van de Voorde 1997) and with a study

Table 1. Comparison of pEC50 and R_{max} values for the effects of glibenclamide on the contractions of LIMA and radial artery segments by U46619 and L-phenylephrine

Tissue	Contractile agent	Treatment	pEC50	R_{max}	n
LIMA	U46619	Control	8·21 ± 0·12	98·9 ± 0·88	7
		Glibenclamide (10 μM)	8·09 ± 0·13	95·5 ± 2·35	6
		Glibenclamide (50 μM)	7·25 ± 0·30**	96·5 ± 2·41	4
	L-Phenylephrine	Control	6·14 ± 0·14	102 ± 0·63	5
		Glibenclamide (10 μM)	5·73 ± 0·16	98·7 ± 1·51	5
		Glibenclamide (50 μM)	5·62 ± 0·18	62·7 ± 5·02***	3
Radial artery	U46619	Control	8·48 ± 0·17	99·0 ± 1·59	5
		Glibenclamide (10 μM)	8·09 ± 0·05	97·4 ± 2·91	4
		Glibenclamide (50 μM)	7·11 ± 0·25**	92·1 ± 4·74	3
	L-Phenylephrine	Control	5·99 ± 0·18	102 ± 2·18	5
		Glibenclamide (10 μM)	5·59 ± 0·16	95·2 ± 6·51	3
		Glibenclamide (50 μM)	5·47 ± 0·2	76·1 ± 9·48*	5

Data are expressed as mean values ± s.e.m. pEC50 and R_{max} values were derived by the curve fitting procedure described in Materials and Methods. Control and test values were compared by Student's unpaired *t*-test; $^*P < 0{\cdot}05$, $^{**}P < 0{\cdot}01$, $^{***}P < 0{\cdot}001$.

by Stanke et al (1998), which demonstrated similar effects with glibenclamide in human LIMA and saphenous vein. The effects of 10 μM glibenclamide were not significant.

Given that agents which cause activation of K_{ATP} channels, such as levcromakalim and pinacidil, are known to cause vasodilatation (Standen et al 1989), it would seem likely that the inhibitory effect of glibenclamide on vasoconstriction was mediated through an action distinct from its well characterized blocking effect on K_{ATP} channels. It is therefore unusual that there should be a correlation between the ability of glibenclamide and related compounds to antagonize the effects of U46619 on pig coronary artery and their ability to block K_{ATP} channels. One suggestion would be that glibenclamide at higher concentrations might somehow cause a paradoxical opening of the K_{ATP} channel to elicit the vascular effects. McPherson et al (1997) showed that tetraphenylphosphonium (which blocks the K_{ATP} channel in a different manner from that of glibenclamide) had no effect on U46619-induced contraction in pig coronary artery suggesting that glibenclamide is not mediating these effects through an interaction with K_{ATP} channels. This is supported our findings that treatment of LIMA segments with another sulphonylurea compound, tolbutamide (at a concentration which would cause a block of K_{ATP} channels but not affect vascular contraction), was shown not to affect the relaxation effects of glibenclamide.

As glibenclamide binds to a sulphonylurea receptor on the K_{ATP} channel, it has been suggested that the compound might bind to a similar site at the thromboxane A_2 receptor. However, recent cloning studies, which indicate that the sulphonylurea and thromboxane A_2 receptors are structurally dissimilar, tend to dispel this theory (Coleman et al 1994; Aguilar-Bryan et al 1995).

Glibenclamide does not appear to inhibit vasoconstriction through the release of vasodilator prostanoids as the actions of the drug on LIMA vessels pre-contracted with U46619 were unaffected when 10 μM indomethacin was present in the physiological saline solution. This supports the findings of previous studies, which showed that indomethacin had no effect (Delaey & Van de Voorde 1997; Huang & Chan 1998).

In this study, the effects of glibenclamide were not restricted to vessels contracted by prostanoids but were also seen when other agonists were used. Glibenclamide exerted an inhibitory effect on the contraction caused by the α_1-adrenoceptor selective agonist, L-phenylephrine, causing a rightward shift in the concentration–response relation in both LIMA and radial artery. These effects would not be explained by the previously proposed interaction of glibenclamide with prostanoid receptors or their second messenger systems (Delaey & Van de Voorde 1997). Glibenclamide also had effects on the contractions caused by a high extracellular K^+ concentration. Here, the normal contribution of potassium currents to the resting membrane potential across the vascular membrane is dissipated and voltage-sensitive Ca^{2+} channels are opened (Weiss 1977). Figures 2A and 2B show that the concentration–response relationship was shifted to the right in the presence of 100 μm glibenclamide in both LIMA and radial artery. The fact that glibenclamide can affect the response to high concentrations of K^+ ions provides further support for the idea that it is not causing these effects through an action on K_{ATP} channels.

The pharmacology of human peripheral arteries used for coronary artery surgery is important because of their inherent spasmogenic tendency. This is particularly true for the radial artery, which has been recently re-introduced as an arterial graft

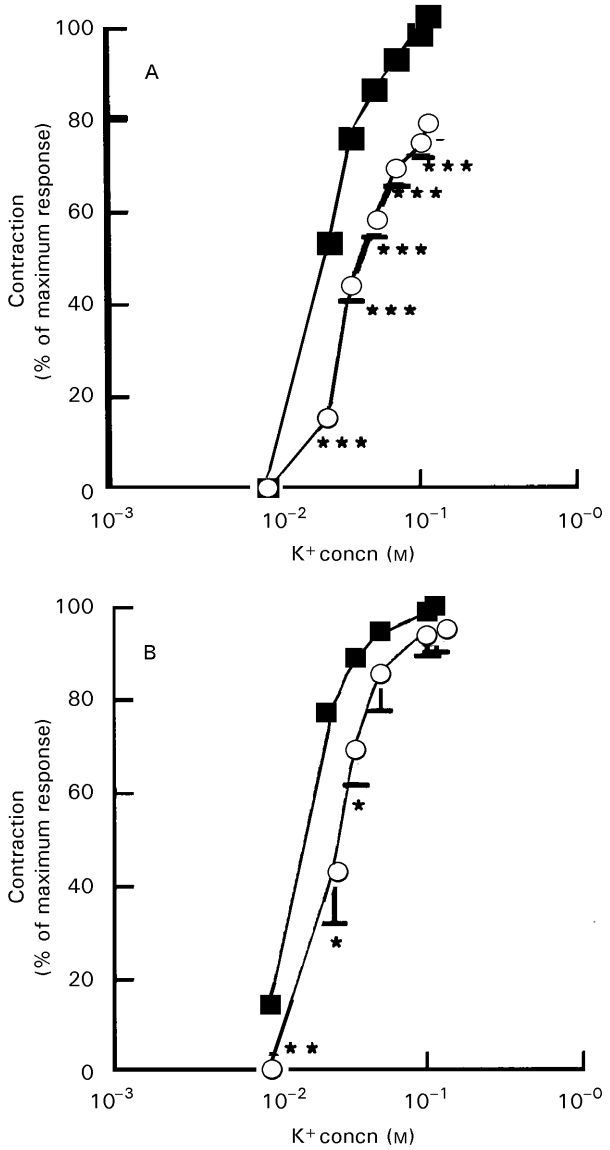

Figure 2. Effect of glibenclamide on K^+-induced tension in human LIMA (A) and radial artery (B). Contractions to increasing concentrations of K^+ depolarizing solutions were obtained in the absence (■) and presence (○) of 100 μM glibenclamide. Data represent mean values ± s.e.m for all segments tested from 4 patients. Statistical comparisons were made by Student's paired *t*-test at individual concentrations. $*P < 0{\cdot}05$, $**P < 0{\cdot}01$, $***P < 0{\cdot}001$ compared with control values.

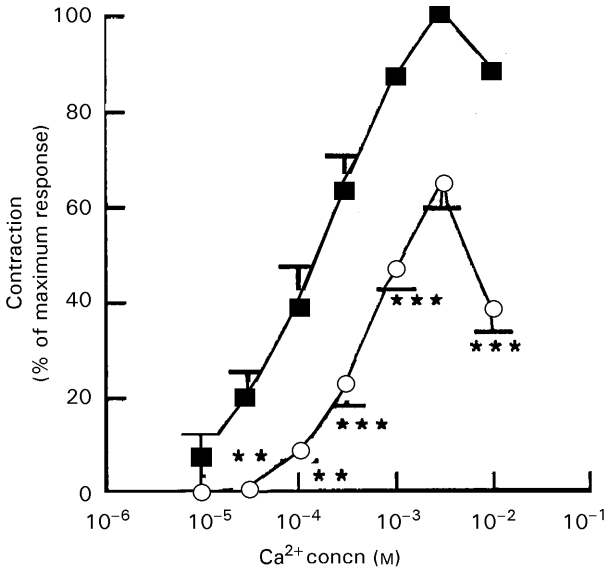

Figure 3. Effect of glibenclamide on Ca^{2+}-induced tension in human radial artery. Control Ca^{2+} response relationships (■) were obtained in the presence of 35 mM K^+ depolarizing solution in the absence of glibenclamide. After a pre-incubation period of 30 min with 100 μM glibenclamide, contractions to Ca^{2+} were recorded again in 35 mM K^+ depolarizing solution (○). All experiments were carried out in the presence of 1 μM phentolamine. Data represent mean values ± s.e.m. for all segments tested from 5 patients. Statistical comparisons were made by Student's paired *t*-test at individual concentrations. $**P < 0{\cdot}01$, $***P < 0{\cdot}001$ compared with control values.

(Acar et al 1992; Buxton et al 1997), although there have also been reports of spasm in LIMA (Sarabu et al 1987). The underlying cause of the spasm remains unknown but one of the contributory factors may be the release of endogenous thromboxanes and prostaglandins. Elucidation of the mechanisms involved can contribute to the design of successful anti-vasospastic strategies, which have enormous clinical and economic importance through maintaining the long-term patency of bypass conduits and freedom from further myocardial events.

To date, prevention of radial artery spasm has been achieved by the use of calcium channel antagonists (Acar et al 1992). In-vitro, verapamil can cause a relaxation of KCl-contracted radial artery segments (He & Yang 1996), while the vasoselective, ultrashort-acting dihydropyridine calcium-channel antagonist, clevidipine, relaxes both radial artery (Ferguson, unpublished data) and LIMA (Huraux et al 1997) contracted by U46619. Given that glibenclamide can affect the contraction of isolated blood vessels to a number of different agents, an interference with the processes which allow intracellular Ca^{2+} to increase, is a suggested mechanism of action.

Yoshitake et al (1991) showed that pretreatment of rat aortic strips with 100 μM glibenclamide partially inhibited the elevation of intracellular Ca^{2+} ($[Ca^{2+}]_i$) and force development produced by both noradrenaline and high concentrations of K^+ ions. These authors postulated that glibenclamide was acting to decrease $[Ca^{2+}]_i$. However, as it did not affect the $[Ca^{2+}]_i$ force relationship, they suggested that it was unlikely that glibenclamide was exerting its effects by affecting the sensitivity of the contractile apparatus to $[Ca^{2+}]_i$ in the vascular strips. More recently, Huang & Chan (1998) have suggested that glibenclamide might interfere with Ca^{2+} influx across the vascular membrane. This

would be a plausible explanation since high extracellular K^+ concentration results in activation of voltage-gated Ca^{2+} channels and hence an increase in $[Ca^{2+}]_i$ while Ca^{2+} entry through voltage-sensitive Ca^{2+} channels is also stimulated in the presence of α_1-receptor agonists and thromboxanes, in addition to release of Ca^{2+} ions from intracellular stores.

To investigate this possibility in human blood vessels, the effect of glibenclamide on Ca^{2+}-induced tension in depolarized radial artery was determined. Here, vascular contraction is initiated by the entry of extracellular calcium through voltage-sensitive calcium channels with a negligible release of Ca^{2+} ions from intracellular stores (Julou-Schaeffer & Freslon 1988). On the addition of increasing concentrations of $CaCl_2$, a characteristic bell-shaped concentration–response relationship is obtained (see Figure 3). Once the concentration of $CaCl_2$ exceeded 3 ± 10^{-3} M, Ca^{2+}-induced relaxation is observed. A possible explanation for this has been suggested by Mupanomunda et al (1998), who suggest that at these high concentrations, Ca^{2+} ions activate calcium receptors on capsaicin-sensitive perivascular sensory neurones to cause them to release vasodilator transmitters. In the presence of 100 μM glibenclamide, the Ca^{2+}-induced contractions were significantly reduced. These findings suggest that glibenclamide affects vascular smooth muscle contraction by blocking voltage-gated Ca^{2+}
channels.

Additional studies have shown that glibenclamide can block other ion channels as well as K_{ATP} channels, such as the cystic fibrosis transmembrane conductance regulator (CTFR) chloride channel (Yamazaki & Hume 1997) and also a cyclic AMP-activated chloride channel in guinea-pig ventricular myocytes (Tominaga et al 1995). However, patch-clamp techniques would need to be employed to confirm whether glibenclamide was affecting voltage-sensitive calcium channels in these human blood vessel segments.

In this investigation, no evidence has been provided to suggest that the effects of glibenclamide were endothelium-dependent. Here, similar effects were obtained with glibenclamide in both endothelium-intact and -denuded vessels. In rat mesenteric artery, Huang & Chan (1998) showed that glibenclamide was more potent at causing relaxation of L-phenylephrine pre-contracted vessels with an intact endothelium. The significance of these findings remains to be determined.

In conclusion, this study has shown that high concentrations of glibenclamide can inhibit the contraction of human peripheral blood vessels in response to a number of vasoconstrictor agents. In addition, further evidence has been provided to support the theory that the drug may be interfering with entry of Ca^{2+} ions through voltage-sensitive calcium channels across the vascular membrane.

References

Acar, C., Jebara, V. A., Portoghèse, M., Beyssen, B., Pagny, J. Y., Grare, P., Chachques, J. C., Fabiani, J. N., Deloche, A., Guermonprez, J. L., Carpentier, A. (1992) Revival of the radial artery for coronary artery bypass grafting. Ann. Thorac. Surg. 54: 652–660

Aguilar-Bryan, L., Nichols, C. G., Wechsler, S. W., Clement, J. P., Boyd, A. E., Gonzalez, G., Herrera-Sosa, H., Nguy, K., Bryan, J., Nelson, D. A. (1995) Cloning of the β cell high-affinity sulfonylurea receptor: a regulator of insulin secretion. Science 268: 423–426

Buxton, B., Windsor, M., Komeda, M., Gaer, J., Fuller, J., Liu, J. (1997) How good is the radial artery as a bypass graft? Coronary Artery Dis. 8: 225–233

Cocks, T. M., King, S. J., Angus, J. A. (1990) Glibenclamide is a competitive antagonist of the thromboxane A_2 receptor in dog coronary artery in vitro. Br. J. Pharmacol. 100: 375–378

Coleman, R. A., Smith, W. L., Narumiya, S. (1994) VIII International Union of Pharmacology classification of prostanoid receptors: properties, distribution and structure of the receptors and their subtypes. Pharmacol. Rev. 46: 205–229

Delaey, C., Van de Voorde, J. (1997) Heterogeneity of the inhibitory influence of sulfonylureas on prostanoid-induced smooth muscle contraction. Eur. J. Pharmacol. 325: 41–46

Eltze, M. (1989) Glibenclamide is a competitive antagonist of cromakalim, pinacidil and RP 49356 in guinea-pig pulmonary artery. Eur. J. Pharmacol. 165: 231–239

He, G.-W., Yang, C.-Q. (1996) Use of verapamil and nitroglycerin solution in preparation of radial artery for coronary grafting. Ann. Thorac. Surg. 61: 610–614

Huang, Y., Chan, N. W. K. (1998) Involvement of endothelium in relaxant action of glibenclamide on the rat mesenteric artery. Eur. J. Pharmacol. 343: 27–33

Huraux, C., Makita, T., Szlam, F., Nordlander, M., Levy, J. H. (1997) The vasodilator effects of clevidipine on human internal mammary artery. Anesth. Analg. 85: 1000–1004

Julou-Schaeffer, G., Freslon, J. L. (1988) Effects of ryanodine on tension development in rat aorta and mesenteric resistance vessels. Br. J. Pharmacol. 95: 605–613

McPherson, G. A., Choi, R.T., Kong, D. C. M., Iskander, M. N. (1997) The thromboxane A_2 and the K_{ATP} channel antagonist actions of a series of sulphonylurea derivatives in the pig coronary artery. Eur. J. Pharmacol. 324: 193–200

Metcalfe, M. J., Lip, G. Y. H., Dargie, H. J. (1994) Factors influencing coronary artery bypass graft patency. Cardiovasc. Surg. 2: 679–685

Mupanomunda, M. M., Wang, Y., Bukoski, R. D. (1998) Effect of chronic sensory denervation on Ca^{2+}-induced relaxation of isolated mesenteric resistance arteries. Am. J. Physiol. 274: H1655–H1661

Nielsen-Kudsk, J. E., Thirstrup, S. (1991) Antidiabetic sulfonylureas relax isolated rabbit coronary arteries. Eur. J. Pharmacol. 209: 273–275

Nwasokwa, O. N. (1995) Coronary artery bypass graft disease. Ann. Intern. Med. 123: 528–545

Quast, U., Cook, N. S. (1989) *In vitro* and *in vivo* comparison of two K^+ channel openers, diazoxide and cromakalim, and their inhibition by glibenclamide. J. Pharmacol. Exp. Ther. 250: 262–271

Sarabu, M. R., McClung, J. A., Fass, A., Reed, G. E. (1987) Early postoperative spasm in left internal mammary artery bypass grafts. Ann. Thorac. Surg. 44: 199–200

Schmid-Antomarchi, H., De Weille, J., Fosset, M., Ladzunski, M. (1987) The receptor for antidiabetic sulfonylureas controls the activity of the ATP-modulated K^+ channel in insulin-secreting cells. J. Biol. Chem. 262: 15840–15844

Standen, N. B., Quayle, J. M., Davies, N. W., Brayden, J. E., Huang, Y., Nelson, M. T. (1989) Hyperpolarizing vasodilators activate ATP-sensitive K^+ channels in arterial smooth muscle. Science 245: 177–180

Stanke, F., Cracowski, J.-L., Chavanon, O., Magne, J.-L., Blin, D., Bessard, G., Devillier, P. (1998) Glibenclamide inhibits thromboxane A_2-induced contraction in human internal mammary artery and saphenous vein. Eur. J. Pharmacol. 341: 65–71

Tominaga, M., Horie, M., Sasayama, S., Okada, Y. (1995) Glibenclamide, an ATP-sensitive K^+ channel blocker, inhibits cardiac cAMP-activated Cl^- conductance. Circ. Res. 77: 417–423

Weiss, G. B. (1977) Approaches to delineation of differing calcium binding sites in smooth muscle. In: Casteels, R., Godfraind, T., Ruegg, J. (eds) Excitation-Contraction Coupling in Smooth Muscle. Elsevier, Amsterdam, pp 253–260

Winquist, R. J., Heaney, L. A., Wallace, A. A., Baskin, E. P., Stein, R. B., Garcia, M. L., Kaczorowski, J. (1989) Glyburide blocks the relaxation response to BRL 34915 (cromakalim), minoxidil sulfate and diazoxide in vascular smooth muscle. J. Pharmacol. Exp. Ther. 248: 149–156

Yamazaki, J., Hume, J. R. (1997) Inhibitory effects of glibenclamide on cystic fibrosis transmembrane regulator, swelling activated and Ca^{2+}-activated Cl-channels in mammalian cardiac myocytes. Circulation Res. 81: 101–109

Yoshitake, K., Hirano, K., Kanaide, H. (1991) Effects of glibenclamide on cytosolic calcium concentrations and on contraction of the rabbit aorta. Br. J. Pharmacol. 102: 113–118

Zhang H., Stockbridge, N., Weir, B., Krueger, C., Cook, D. (1991) Glibenclamide relaxes vascular smooth muscle constriction produced by prostaglandin $F_{2\alpha}$. Eur. J. Pharmacol. 195: 27–35