Br. J. Pharmacol. (1988), **94**, 759–764

Flushing and haemodynamic responses to vasopressin peptides in the rhesus monkey

*,[1]David P. Brooks, *Paul F. Koster, **Frans L. Stassen, *Christine R. Albrightson, †William F. Huffman, *Martin A. Wasserman & *Lewis B. Kinter

Departments of *Pharmacology, **Molecular Pharmacology and †Peptide Chemistry, Smith Kline & French Laboratories, King of Prussia, PA, U.S.A.

1 The mechanism of the flushing, hypotension and tachycardia associated with i.v. administration of desGlyd$(CH_2)_5$D-Tyr(Et)VAVP (SK&F 101926; 25 $\mu g\,kg^{-1}$) and the selective V_2 antidiuretic agonist, desamino-8-D-arginine vasopressin (dDAVP; 3 $\mu g\,kg^{-1}$) was studied in ketamine-anaesthetized rhesus monkeys.

2 The flushing associated with SK&F 101926 was reduced by pretreatment with a mast cell stabilizer and by repeated administration of peptide (within 2–4 weeks). A similar desensitization to dDAVP-associated flushing was observed on repeated administration.

3 Treatment with dDAVP also resulted in reduced SK&F 101926-associated flushing.

4 The hypotension associated with SK&F 101926 was not affected by pretreatment with a mast cell stabilizer. A similar degree of hypotension was observed with repeated administration of either SK&F 101926 or dDAVP.

5 The tachycardia associated with SK&F 101926 was reduced by pretreatment with a mast cell stabilizer or repeated administration of SK&F 101926. Repeated administration of dDAVP, however, resulted in an enhanced tachycardia.

6 Indomethacin (5 mg kg^{-1} i.v.) did not alter the flushing or the hypotension associated with the administration of either SK&F 101926 or dDAVP, but resulted in an enhanced tachycardia to SK&F 101926.

7 Administration of a selective V_1 vasopressor antagonist did not result in flushing, hypotension or tachycardia.

8 It was concluded that the flushing response to vasopressin-like peptides in rhesus monkeys may be due to an action on mast cells, whereas the haemodynamic responses are not, but probably involve direct vasodilator actions.

Introduction

Facial flushing, hypotension and tachycardia in humans have been described in association with administration of desamino-8-D-arginine vasopressin (dDAVP; Manucci *et al.*, 1977; Jeffrys *et al.*, 1979; Ockelford *et al.*, 1980; Belch *et al.*, 1982; Yokoto *et al.*, 1982), Derkx *et al.*, 1983; Pigache, 1984; Williams *et al.*, 1986; Brink *et al.*, 1987; Bichet *et al.*, 1988), a potent antidiuretic agonist acting at renal vasopressin V_2 receptors; dDAVP exhibits little or no vasopressor activity by acting at vascular (V_1) receptors. The mechanism of flushing has been attributed to: (1) antagonism of endogenous vasopressin-induced vasoconstriction (Derkx *et al.*, 1983; Pigache, 1984); (2) production of vasodilator prostaglandins (Belch *et al.*, 1982); or (3) direct vasodilator activity (Brink *et al.*, 1981; Williams *et al.*, 1986). The vasopressin analogue, desGlyd$(CH_2)_5$D-Tyr(Et)VAVP (SK&F 101926), has been identified as a vasopressin antidiuretic antagonist in rats (Manning *et al.*, 1984; Stassen *et al.*, 1984), dogs (Kinter *et al.*, 1987a) and squirrel monkeys (Kinter *et al.*, 1987b), but was found to be a full antidiuretic agonist in man (Dubb *et al.*, 1987; Allison *et al.*, 1988) and rhesus monkeys (Brooks *et al.*, 1988). SK&F 101926 also caused

[1] Author for correspondence.

flushing in man, but was not associated with changes in systemic haemodynamics (Dubb *et al.*, 1987; Allison *et al.*, 1988). The flushing response to SK&F 101926 was reduced with repeated administration (Allison *et al.*, 1988). In the present study, we have determined that SK&F 101926 and dDAVP cause flushing, hypotension and tachycardia in rhesus monkeys, and that this animal model can be used to study the mechanisms involved in these responses.

Methods

Male rhesus monkeys (*Macaca mulatta*; 6–13 kg) were housed, handled and maintained according to standard procedures. Monkeys were fed Purina Monkey Diet supplemented with fruit, and water was available *ad libitum*. Before each experiment, animals were deprived of food, but not water, for 16 h. Animals were anaesthetized with ketamine (75–100 mg i.m. plus 20 mg supplemented as necessary) and placed on a circulating water heating pad (Aquamatic Model K-20). A catheter (Abbocath 20G) was inserted into a saphenous vein and flushed with heparinized saline. Arterial pressure and heart rate were measured indirectly using a Dinamap vital signs monitor with the cuff placed around an upper arm. All drugs were administered i.v. and flushed with 1 ml isotonic saline. The following protocols were conducted.

Protocol 1

Baseline blood pressures and heart rates were obtained at 2 min intervals for at least 10 min. SK&F 101926 (25 $\mu g\,kg^{-1}$), dDAVP (3 $\mu g\,kg^{-1}$) or the selective vasopressin V_1 pressor antagonist, (1-(β-mercapto-β,β-cyclopentamethylenepropionic acid), 2-(O-methyl)tyrosine)arginine vasopressin, SK&F 100273; Kruszynski *et al.*, 1980; 25 $\mu g\,kg^{-1}$), were then administered to naïve monkeys, i.e., monkeys that had not received either SK&F 101926 or dDVAP for at least 4 months before the experiment. Threshold doses of SK&F 101926 and dDAVP associated with flushing, hypotension and tachycardia in the rhesus monkey were determined in preliminary studies. The dose of SK&F 100273 was chosen to equal that of SK&F 101926. Flushing was scored on a scale of 0 to 5 where 0 represented no response and 5 represented a bright red flush over at least the whole face and neck. Blood pressure and heart rate were monitored for at least 10 min following dosing. The maximum changes in blood pressure and heart rate always occurred within the first 10 min following administration of peptide. Baseline blood pressure and heart rate were calculated from the mean values obtained before drug treatment.

Protocol 2

Five naïve monkeys were administered a mast cell stabilizer, 5-acetyl-4-hydroxy-3-[1-[(3-amino-4-hydroxyphenyl)amino]ethylidene]2H-pyran-2,6(3H)-dione hydrochloride (SK&F 78729-A), at a dose shown to be effective in the rhesus monkey (5 $mg\,kg^{-1}$; Chakrin & Krell, 1978). Five min later, SK&F 101926 (25 $\mu g\,kg^{-1}$) was administered and flushing, blood pressure and heart rate responses were determined as in protocol 1. Baseline blood pressure and heart rate were determined for the period between the administration of the mast cell stabilizer and SK&F 101926.

Protocol 3

Four naïve monkeys were administered indomethacin (5 $mg\,kg^{-1}$). Thirty min later, SK&F 101926 (25 $\mu g\,kg^{-1}$) or dDAVP (3 $\mu g\,kg^{-1}$) was administered. Flushing, blood pressure and heart rate responses were determined as in protocol 1. Baseline blood pressure and heart rate were determined for the period between administration of indomethacin and SK&F 101926 or dDAVP.

Protocol 4

Four monkeys that had received SK&F 101926 (25 $\mu g\,kg^{-1}$) or dDAVP (3 $\mu g\,kg^{-1}$) 2 to 4 weeks earlier were re-administered the same drug and flushing, blood pressure and heart rate were determined.

Protocol 5

Four monkeys that were naïve with respect to SK&F 101926 but had received dDAVP (3 $\mu g\,kg^{-1}$) 2 weeks earlier were administered SK&F 101926 as in protocol 1. Flushing, blood pressure and heart rate responses were determined.

The data are presented as individual data and means $\pm$s.e. Flushing scores were analysed statistically by use of a non-parametric test (Mann-Whitney) and the haemodynamic responses were analysed statistically by use of Student's t test. P values < 0.05 were considered statistically significant.

Results

Intravenous administration of SK&F 101926 (25 $\mu g\,kg^{-1}$) to naïve rhesus monkeys resulted in flushing (Figure 1), hypotension ($P < 0.01$; Figure 2)

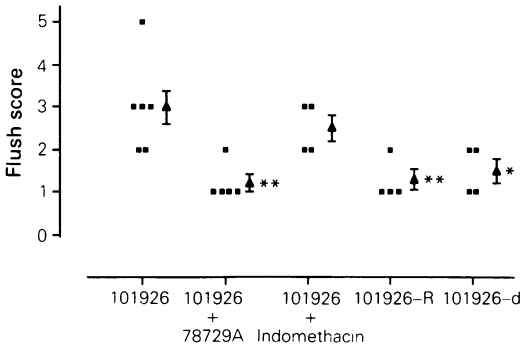

Figure 1 Individual (■) and mean (▲) flush scores in rhesus monkeys after administration of SK&F 101926 (25 $\mu g kg^{-1}$) in the presence and absence of SK&F 78729-A (5 $mg kg^{-1}$) or indomethacin (5 $mg kg^{-1}$) or after prior exposure to SK&F 101926 (101926-R) or dDAVP (101926-d). $*P < 0.05$; $**P < 0.01$ (vs SK&F 101926). Vertical lines indicate s.e. mean.

and tachycardia ($P < 0.01$; Figure 2). In two experiments using direct femoral arterial blood pressure measurements, SK&F 101926 was also shown to reduce blood pressure and increase heart rate (Figure 3). The time course of the response indicates that the maximum hypotension and tachycardia occurs within the first 10 min. The flush response and tachycardia were reduced significantly ($P < 0.01$) by a mast cell stabilizer (Figure 1). Indomethacin pretreatment did not alter the flushing (Figure 1) or hypotensive (Figure 2) response to SK&F 101926

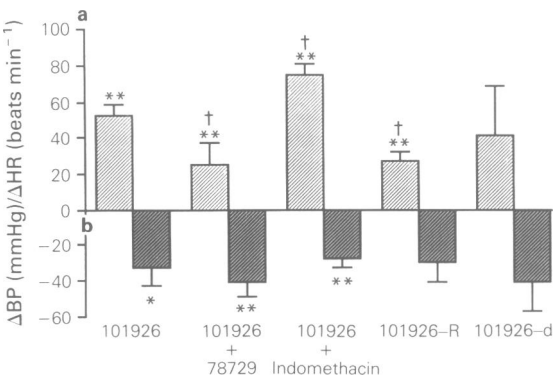

Figure 2 Mean changes (a) heart rate (Δ HR) and (b) blood pressure (Δ BP) in rhesus monkeys after administration of SK&F 101926 (25 $\mu g kg^{-1}$) in the presence and absence of SK&F 78729-A (5 $mg kg^{-1}$) or indomethacin (5 mg kg^{-1}) or after prior exposure to SK&F 101926 (101926-R) or dDAVP (101926-d). See Table 1 for baseline blood pressures and heart rates. $*P < 0.05$; $**P < 0.01$ (vs baseline); $\dagger P < 0.05$ (vs SK&F 101926).

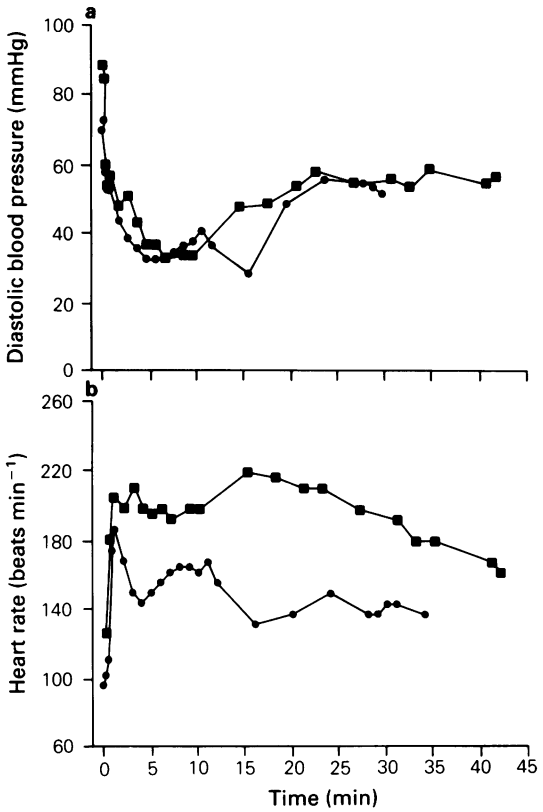

Figure 3 Time course of the diastolic blood pressure (a) and heart rate (b) after intravenous administration of SK&F 101926 10 $\mu g kg^{-1}$ (■) and 30 $\mu g kg^{-1}$ (●).

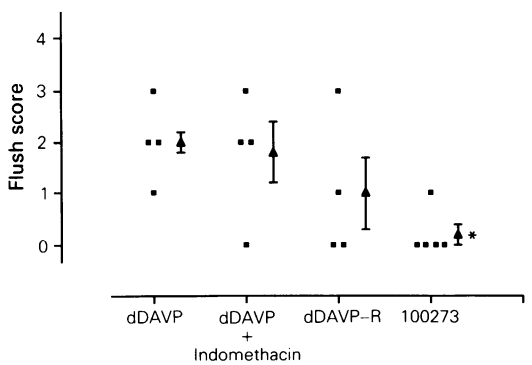

Figure 4 Individual (■) and mean (▲) flush scores in rhesus monkeys after administration of dDAVP (3 $\mu g kg^{-1}$) in the presence and absence of indomethacin (5 mg kg^{-1}) or after prior exposure to dDAVP (dDAVP-R) and SK&F 100273 (25 $\mu g kg^{-1}$). $*P < 0.01$ (vs dDAVP). Vertical lines indicate s.e. mean.

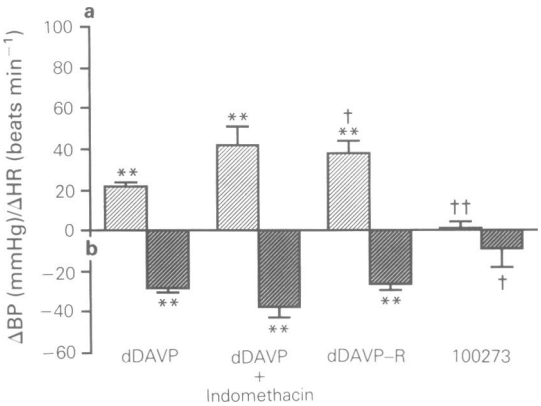

Figure 5 Mean changes in (a) heart rate (Δ HR) and (b) blood pressure (Δ BP) in rhesus monkeys after administration of dDAVP ($3\,\mu g\,kg^{-1}$) in the presence and absence of indomethacin ($5\,mg\,kg^{-1}$) or after prior exposure to dDAVP (dDAVP-R) and SK&F 100273 ($25\,\mu g\ kg^{-1}$). See Table 1 for baseline blood pressures and heart rates. $^{**}P < 0.01$ (vs baseline); $\dagger P < 0.05$; $\dagger\dagger P < 0.01$ (vs dDAVP).

Table 1 Baseline cardiovascular parameters in rhesus monkeys

Treatment	n	*Blood pressure* (mmHg)	*Heart rate* (beats min^{-1})
101926	6	67 ± 9	115 ± 7
101926 + indomethacin	4	78 ± 3	109 ± 4
101926 + 78729-A	6	90 ± 4	109 ± 18
101926-R	4	73 ± 11	136 ± 16
dDAVP	4	78 ± 9	142 ± 2
dDAVP + indomethacin	4	93 ± 8	113 ± 18
dDAVP-R	4	87 ± 4	141 ± 14
100273	5	68 ± 6	107 ± 5
101926-d	3	80 ± 14	112 ± 2

See Figures 1 and 2 for explanation of treatments. Values are mean ± s.e.

but enhanced the tachycardia ($P < 0.05$; Figure 2). Repeated administration of SK&F 101926 between 2 and 4 weeks later reduced the flushing ($P < 0.01$; Figure 1) and tachycardia, but not the hypotension (Figure 2).

The antidiuretic agonist, dDAVP, caused similar flushing (Figure 4) and haemodynamic (Figure 5) responses as those obtained with SK&F 101926 (Figures 1 and 2). Once again, indomethacin pretreatment enhanced tachycardia (Figure 5) but did not affect flushing (Figure 4) or hypotension (Figure 5). Repeated administration of dDAVP reduced the flushing response in 3 of 4 animals (Figure 4), and caused a similar hypotensive response and an enhanced tachycardia ($P < 0.05$; Figure 5). Administration of SK&F 101926 to monkeys that had previously been treated with dDAVP showed the same diminished flushing response as that observed after SK&F 101926 (Figure 1).

The selective vasopressin V_1 pressor antagonist was not associated with intrinsic flushing or cardiovascular actions. Baseline blood pressures and heart rates (Table 1) were not different between the groups.

Discussion

Flushing and hypotension are commonly associated with administration or release of peptides. Clinical investigations of dDAVP in diabetes insipidus and some bleeding disorders have led to a number of reports of flushing, hypotension and tachycardia induced by this peptide in man (see Introduction). Furthermore, another vasopressin analogue (SK&F 101926) has also been associated with flushing in man (Allison *et al.*, 1988). Our observation that both dDAVP and SK&F 101926 also cause flushing, hypotension and tachycardia in the rhesus monkey provided an excellent model to study the mechanisms involved. Thus, by using a mast cell stabilizer, a cyclo-oxygenase inhibitor, a selective vasopressin pressor antagonist and administering repeated doses of peptide, we have been able to improve our understanding of the haemodynamic and flushing responses to one group of peptides, the vasopressin analogues.

The first conclusion from the present study is that the mechanism of the flushing response is clearly separable from that of the haemodynamic responses. Flushing associated with SK&F 101926 was attenuated by a mast cell stabilizer, whereas the hypotension was not. Furthermore, while the flush response associated with either dDAVP or SK&F 101926 was reduced with repeated administration of either peptide, the cardiovascular responses were unchanged. These observations suggest that the flushing mechanism involves mast cell degranulation and not a direct peptide-associated vasodilatation at the peripheral microvasculature. This conclusion contrasts with results from previous studies. Pigache (1984) and Derkx *et al.* (1983) concluded that flushing induced by dDAVP was due to an action of the peptide at V_1 receptors to block endogenous vasopressin-induced vasoconstriction or potentiation of other endogenous vasoconstrictors. While it is clear that high physiological levels of vasopressin can, indeed, reduce skin blood flow (Wiles *et al.*,

1986), it is apparent from recent studies in the pithed rat (Brink *et al.*, 1987) that dDAVP lacks significant vasopressin V_1 antagonist activity. In the present study, SK&F 101926, which does possess V_1 antagonist activity (Ohlstein & Berkowitz, 1986), was associated with a flush response. However, a selective vasopressin V_1 antagonist was not. In human studies in which SK&F 100273 has been administered in doses effective in blocking the haemodynamic effects of endogenous vasopressin, no flushing has been described (Gavras *et al.*, 1984; Creager *et al.*, 1986). Furthermore, dDAVP is also associated with flushing in patients with cranial diabetes insipidus who have minimal circulating vasopressin levels (Williams *et al.*, 1986). Thus, it appears that antagonism of endogenous vasopressin-induced vasoconstriction is not involved in the flush response.

Another attractive hypothesis for the mechanism of dDAVP-induced flushing was proposed by Belch *et al.* (1982). These investigators suggested that the flushing was due to release of vasodilator prostaglandins since dDAVP caused endothelial release of prostacyclin from rabbit aortic rings, and aspirin could abolish facial flushing in humans who had previously demonstrated a response to dDAVP. Our observation that indomethacin, at a dose that was effective in enhancing the tachycardic effects of both dDAVP and SK&F 101926, failed to attenuate flushing in the rhesus monkey does not support this hypothesis. Furthermore, other investigators (Brommer *et al.*, 1984) observed that aspirin did not block dDAVP-induced flushing in man. It is possible that in the studies of Belch *et al.* (1982), the reduction in dDAVP-induced flushing in man attributed to aspirin may have been due to a desensitization similar to that which we observed in the rhesus monkey. Consistent with this idea is the observation in man that an allergic reaction to dDAVP could be reduced by administering the drug in gradually increasing doses (Yokoto *et al.*, 1982). Thus, it is likely that flushing induced by vasopressin-like peptides is not a direct haemodynamic event, but rather an action on mast cells. The observation that prior treatment with dDAVP also resulted in a desensitization to SK&F 101926 might suggest that the same receptor is involved. Since dDAVP and SK&F 101926 are both vasopressin V_2 antidiuretic agonists in both man (Dubb *et al.*, 1987; Allison *et al.*, 1988) and rhesus monkeys (Brooks *et al.*, 1988), one might speculate that stimulation of the vasopressin V_2-receptor is involved. Consistent with this observation is that high doses of vasopressin are also associated with facial flushing (Gavras *et al.*, 1984).

Our studies do not provide convincing evidence as to the mechanisms of hypotension and tachycardia associated with dDAVP and SK&F 101926 in rhesus monkeys. Certainly, indomethacin does not attenuate the haemodynamic responses to either peptide, thereby indicating that vasodilator prostaglandins are not involved. Furthermore, the observation that administration of a mast cell stabilizer, at a dose effective in reducing a flushing response to SK&F 101926, had no significant effect on the hypotension, would suggest that mediators released from mast cells during the flush response are also not involved. Similarly the V_1 pressor antagonist had no effect on blood pressure or heart rate, thus showing that these responses are not caused by blockade of the vasoconstrictor actions of endogenous vasopressin. It is possible that the cardiovascular responses to dDAVP and SK&F 101926 involve direct effects on vascular V_2-receptors to induce vasodilatation, thereby resulting in hypotension and reflex tachycardia. This would be consistent with the recent observations of Liard (1986) that vascular vasodilator V_2-receptors do exist and are functional. dDAVP-induced flushing and hypotension (and release of Factor VIII c and von Willebrand Factor) is observed in patients with central diabetes insipidus (vasopressin deficiency) but is not observed in patients with congenital nephrogenic diabetes insipidus (CNDI; Bichet *et al.*, 1988). Patients with CNDI have a defective renal V_2-receptor/signal transduction mechanism; thus, the lack of haemodynamic responses associated with dDAVP in these patients suggests the existence of extrarenal V_2-receptors that are possibly also defective.

The tachycardia associated with SK&F 101926 was reduced by the mast cell stabilizer and repeated administration of this peptide but enhanced by indomethacin. Conversely, repeated administration of dDAVP resulted in an enhanced tachycardia. The mechanisms involved in these responses are not clear, but may be related to a differential effect of SK&F 101926 and dDAVP on vasopressin-induced enhancement of baroreflex activity.

In summary, our studies have demonstrated that the rhesus monkey is an excellent model for studying peptide-induced flushing, hypotension and reflex tachycardia. We have also provided evidence that the flushing response to vasopressin-like peptides may be due to an action on mast cells, whereas the haemodynamic responses are not, but probably involve direct vasodilator actions.

We gratefully acknowledge S. Campbell, E. Jenkins, C. Kenney, and J. Kissinger for excellent animal handling, veterinary care and assistance in setting up the experiments; and S. Tirri and L. Contino for assistance in preparing the manuscript.

References

ALLISON, N., ALBRIGHTSON-WINSLOW, C.R., BROOKS, D.P., STASSEN, F.L., HUFFMAN, W.F., STOTE, R. & KINTER, L.B. (1988). Species heterogeneity and antidiuretic hormone antagonists: What are the predictors? In *Proceedings of the 2nd International Vasopressin Conference*, ed. Cowley, A.W., Liard, J.F., & Aussielo, D. New York: Raven Press.

BELCH, J.J.F., SMALL, M., McKENZIE, F., HILL, P.A., LOWE, G.D.O., McINTYRE, D.E., FORBES, C.D. & PRENTICE, C.R.M. (1982). DDAVP stimulates prostacyclin production. *Thromb. Haemost.*, **47**, 122–123.

BICHET, D., RAZI, M., LONERGAN, M. & ARTHUS, M.F. (1988). Factor VIII c (F VIII c), Von Willebrand Factor (VWF), mean arterial blood pressure (MAP), pulse, and renin (PRA) responses to dDAVP infusion in patients (pts) with congenital nephrogenic diabetes insipidus (CNDI). *Kid. Int.* **33**, 182.

BRINK, H., DERKX, F., KOLSTEE, H., MERKUS, P., SMITS, J., MAN IN'T VELD, A. & SCHALEKAMP, M. (1987). Vasopressin V_2-receptor-mediated hypotensive response in humans. *Pharm. Weekblad. Ned.*, **9**, 187.

BROMMER, E.J.P., DERKX, F.H.H., BARRETT-BERGSHOEFF, M.M. & SCHALEKAMP, M.A.D.H. (1984). The inability of propranolol and aspirin to inhibit the response of fibrinolytic activity and Factor VIII-antigen to infusion of DDAVP. *Thromb. Haemost.*, **51**, 42–44.

BROOKS, D.P., KOSTER, P.F., ALBRIGHTSON-WINSLOW, C.R., STASSEN, F.L., HUFFMAN, W.F. & KINTER, L.B. (1988). SK&F 105494 is a potent vasopressin antidiuretic antagonist in the rhesus monkey (*Macaca mulatta*). *J. Pharmacol. Exp. Ther.*, (in press).

CHAKRIN, L.W. & KRELL, R.D. (1978). Pharmacologic characterization of the antiallergic activity of SK&F 78729-A. *J. Pharmacol. Exp. Ther.*, **207**, 756–766.

CREAGER, M.A., FAXON, D.P., CUTLER, S.S., KOHLMAN, O., RYAN, T.J. & GAVRAS, H. (1986). Contribution of vasopressin to vasoconstriction in patients with congestive heart failure: comparison with the renin-angiotensin system and the sympathetic nervous system. *J. Am. Coll. Cardiol.*, **7**, 758–765.

DERKX, F.H., MAN IN'T VELD, A.J., JONES, R., REID, J.L. & SCHALEKAMP, M.A.D.H. (1983). (1-desamino-8-D-arginine vasopressin): an antagonist of the pressor action of endogenous vasopressin. *J. Hypertens.*, **1** (Suppl. 2), 58–61.

DUBB, J., ALLISON, N., TATOIAN, D., BLUMBERG, A., LEE, K. & STOTE, R. (1987). SK&F 101926 is antidiuretic in man. *Kid. Int.*, **31**, 267.

GAVRAS, H., RIBEIRO, A.B., KOHLMANN, O., SARAGOCA, M., MULINARI, R.A., RAMOS, O. & GAVRAS, I. (1984). Effects of a specific inhibitor of the vascular actions of vasopressin in humans. *J. Hypertens.*, **6** (Suppl. 1), I156-I160.

JEFFRYS, D.B., STRAKOSCH, C.R. & KEEN, H. (1979). Facial flushing in diabetes. *Lancet*, **ii**, 1195.

KINTER, L.B., CHURCHILL, S., STASSEN, F.L., MOORE, M. & HUFFMAN, W. (1987a). Characterization and mechanism of action of vasopressin antagonists in squirrel monkeys (*Saimiri sciureus*). *J. Pharmacol. Exp. Ther.*, **241**, 797–803.

KINTER, L.B., MANN, W.A., WOODWARD, P., DEPALMA, D. & BRENNAN, F. (1987b). The vasopressin antagonist SK&F 101926 is a selective aquaretic agent in dogs. In *Diuretics: Basic, Pharmacological, and Clinical, Proc. International Meeting on Diuretics*, ed. Andreucci, V.E. & Dal, A. pp. 218–221. Boston: Canton, Martinus Nijhoff.

KRUSZYNSKI, M., LAMMEK, B., MANNING, M., SETO, J., HALDAR, J. & SAWYER, W.H. (1980). [1-(β-mercapto-β, β-cyclopentamethylenepropionic acid), 2-(0-methyl)tyrosine]arginine-vasopressin and [1-(β-mercapto-β,β-cyclopentamethylene-propionic acid)]arginine-vasopressin, two highly potent antagonists of the vasopressor response to arginine-vasopressin. *J. Med. Chem.*, **23**, 364–368.

LIARD, J.F. (1986). Cardiovascular effects associated with antidiuretic activity of vasopressin after blockade of its vasoconstrictor action in dehydrated dogs. *Circ. Res.*, **58**, 631–640.

MANNING, M., OLMA, A., KLIS, W., KOLODZIEJCZYK, A., NAWROCKA, E., MISICKA, A. SETO, J. & SAWYER, W. H. (1984). Carboxy terminus of vasopressin required for activity but not binding. *Nature*, **308**, 652–653.

MANUCCI, P.M., RUGGERI, Z.M., PARETI, F.I. & CAPITANIO, A. (1977). 1-deamino-8-D-arginine vasopresssin: a new pharmacological approach to the management of haemophilia and von Willebrand's disease. *Lancet*, **i**, 869–872.

OCKELFORD, P.A., MENON, N.C. & BERRY, E.W. (1980). Clinical experience with arginine vasopressin (DDAVP) in von Willebrand's disease and mild haemophilia. *N.Z. Med. J.*, **92**, 375–378.

OHLSTEIN, E.H. & BERKOWITZ, B.A. (1986). Human vascular vasopressin receptors: analysis with selective vasopressin receptor antagonists. *J. Pharmacol. Exp. Ther.*, **239**, 737–741.

PIGACHE, R.M. (1984). Facial flushing induced by vasopressin-like peptides lacking pressor activity. *Br. J. Clin. Pharmacol.*, **17**, 369–371.

STASSEN, F., BERKOWITZ, B., HUFFMAN, W., WIEBELHAUS, V. & KINTER, L.B. (1984). Molecular pharmacology and mechanisms of action of aquaretic agents. In *Diuretics: Chemistry, Pharmacology and Clinical Applications*, ed. Puschett, J. & Greenberg, A., pp. 64–71. New York: Elsevier.

WILES, P.G., GRANT, P.J. & DAVIES, J.A. (1986). The differential effect of arginine vasopressin on skin blood flow in man. *Clin. Sci.*, **71**, 633–638.

WILLIAMS, T.D.M., LIGHTMAN, S.L. & LEADBEATER, M.J. (1986). Hormonal and cardiovascular responses to DDAVP in man. *Clin. Endocrinol.*, **24**, 89–96.

YOKOTO, M., MATSUKURA, S., KAJI, H., TAMINATO, T. & FUJITA, T. (1982) Allergic reaction to DDAVP in diabetes insipidus successful treatment with its graded doses. *Endocrinol. Jap.*, **29**, 475–477.

(Received December 8, 1987
Revised January 25, 1988
Accepted February 1, 1988)