Journal of Physiology (1993), **469**, *pp.* 525–534
With 4 figures
Printed in Great Britain

DOES ENDOGENOUS PERIPHERAL ARGININE VASOPRESSIN HAVE A ROLE IN THE FEBRILE RESPONSES OF CONSCIOUS RABBITS?

BY N. G. N. MILTON*†, E. W. HILLHOUSE* AND A. S. MILTON†

*From the *Department of Clinical Biochemistry, University of Newcastle upon Tyne, Framlington Place, Newcastle upon Tyne NE2 4HH and the †Division of Pharmacology, University of Aberdeen, Marischal College, Aberdeen AB9 1AS*

(*Received 8 October 1992*)

SUMMARY

1. The actions of peripheral arginine vasopressin (AVP) on the febrile responses of conscious rabbits induced by peripherally administered polyinosinic:polycytidylic acid (poly(I)·poly(C)) have been studied using an AVP V_1 receptor antagonist ([deamino-Pen[1], *O*-Me-Tyr[2], Arg[8]]-vasopressin).

2. Temperature responses were monitored continuously using rectal thermistor probes. Test substances were administered intravenously (I.V.). Blood samples were taken at timed intervals from a marginal ear vein and plasma PGE_2 and $PGF_{2\alpha}$ levels determined by radioimmunoassay.

3. Poly(I)·poly(C) (2·5 μg/kg) stimulated a reproducible biphasic rise in body temperature with a lag phase of 45–60 min and peaks at 90 and 225 min. The febrile response was accompanied by a 5-fold rise in circulating immunoreactive (ir) PGE_2, which peaked after 90 min and remained elevated up to 300 min. Poly(I)·poly(C) also stimulated a 2·5-fold rise in circulating ir$PGF_{2\alpha}$, which peaked after 150 min and was followed by a return to basal levels after 300 min.

4. The overall magnitude of the febrile response to poly(I)·poly(C) (2·5 μg/kg, I.V.) was significantly antagonized by the AVP V_1 receptor antagonist (250 μg/kg, I.V.) administered 5 min prior to the pyrogen.

5. The irPGE_2 response to poly(I)·poly(C) (2·5 μg/kg, I.V.) was significantly antagonized by the AVP V_1 receptor antagonist (250 μg/kg, I.V.) administered 5 min prior to the pyrogen. The ir$PGF_{2\alpha}$ response was only reduced at the peak 150 min time point measurement.

6. In conclusion, these results show a modulatory role for a peripherally administered AVP V_1 antagonist in the febrile responses to poly(I)·poly(C), suggesting a possible propyretic role for endogenous peripheral AVP. This modulatory role appears to be mediated via actions on prostaglandin E_2.

INTRODUCTION

The febrile responses to pyrogens are mediated via actions of endogenous cytokines (Kluger, 1991) and are thought to be regulated by hypothalamic thermoregulatory centres (Cooper, Cranston & Honour, 1967). However, peripherally

administered pyrogens produce fevers without apparently crossing the blood–brain barrier (Dinarello, Weiner & Wolff, 1978; Dascombe & Milton, 1979), suggesting that a second messenger is involved. The inhibition of prostaglandin (PG) generation by antipyretic drugs (Vane, 1971) and the induction of fever by centrally administered PGs (Milton & Wendlandt, 1971) suggests that PGs may be the endogenous second messengers in the febrile response. The observed increases in circulating blood PGE_2 levels during fever (Rotondo, Abul, Milton & Davidson, 1988) and pyrogen-induced PGE_2 release from blood monocytes and lymphocytes (Kozak, Milton, Abul, Davidson & Rotondo, 1989) has led to the suggestion that the febrile response may be mediated by peripheral production of PGs.

Previous studies have suggested that central arginine vasopressin (AVP) may be an endogenous antipyretic (Veale, Cooper & Ruwe, 1984). The central site of action is suggested by: (1) antipyresis induced by central injection of AVP (Cooper, Kasting, Lederis & Veale, 1979; Kovacs & De Weid, 1983; Naylor, Ruwe & Veale, 1986); (2) secretion of AVP into the septal region during fever (Cooper *et al.* 1979); and (3) an enhancement of fever by perfusion of the septum with an antiAVP antiserum to block the actions of septal AVP (Malkinson, Bridges, Lederis & Veale, 1987). The ability of a centrally administered AVP V_1 antagonist to potentiate fever indicates that the antipyretic actions of AVP require central V_1 receptors (Cooper, Naylor & Veale, 1987).

Many neuropeptides, plus their receptors, are located in both the central nervous system (CNS) and peripheral tissues. Consequently, during many responses neuropeptides may have multiple sites of action. Such site-specific actions can sometimes have opposing physiological consequences; for example, corticotrophin-releasing factor-41 (CRF-41) when administered centrally causes vasoconstriction whilst when administered peripherally CRF-41 causes vasodilatation (Gardiner, Compton & Bennett, 1990). Such contrasting physiological roles for CRF-41 are also seen during fever, with studies suggesting a central antipyretic action for CRF-41 (Bernardini, Richards & Lipton, 1984; Opp, Obal & Krueger, 1989) and a peripheral propyretic action for CRF-41 (Milton, Hillhouse & Milton, 1993*a*) in conscious rabbits. During the febrile response increases in both peripheral circulating and central cerebrospinal fluid AVP levels have been observed (Kasting, Carr, Martin, Blume & Bergland, 1983). Peripheral AVP appears to be unable to penetrate the blood–brain barrier (Wang, Share, Crofton & Kimura, 1981; Mens, Witter & Van Wimersma Greidanus, 1983) and may therefore have a different action to central AVP during the febrile response. The major actions of peripheral AVP include cardiovascular actions, mediated by V_1 receptors, and antidiuretic actions, mediated by V_2 receptors. AVP is also known to activate PG release from the kidney (Wuthrich & Vallotton, 1986) and pituitary (Vlaskovskav, Hertting & Knepel, 1984). The actions of AVP on PGE_2 production by the kidney appear to be mediated via actions on V_1 receptors (Wuthrich & Vallotton, 1986). Another peripheral action of AVP has been observed in the regulation of production of the pyrogenic cytokine γ-interferon by immune cells (Johnson, Farrar & Torres, 1982). As with the actions on PG generation these actions on immune cells appear to be mediated by V_1 receptors (Johnson & Torres, 1985). Thus peripheral AVP has

actions which could potentially influence the febrile response to interferon or interferon inducers. Polyinosinic:polycytidylic acid (poly(I)·poly(C)) is a potent interferon inducer (Stringfellow, 1984), which also activates febrile and prostaglandin responses (Rotondo *et al.* 1988). In this study we have therefore investigated the role of peripheral AVP V_1 receptors during febrile and prostaglandin responses produced by the poly(I)·poly(C).

METHODS

Animals

Male Dutch rabbits (1·7–2·3 kg), obtained from Hylyne Rabbits Ltd (Northwich, Cheshire) were used for all experiments. These animals had not been previously exposed to any agent or experimental manipulation. Animals were lightly restrained in conventional stocks throughout experimental procedures. All experiments were conducted at an ambient temperature of 22 ± 3 °C between 10.00 and 18.00 h.

Temperature response measurement

Temperature responses were measured using rectal thermistor probes (Yellow Springs Instruments 401 series) inserted to a depth of 9 cm and held in position with adhesive tape. Probes were connected to a multi-channel Jaquet chart recorder (Model KS 406) with responses recorded at 20 s intervals. The temperature of each animal was allowed to stabilize for at least 60 min prior to the onset of experimental procedures. Animals were initially placed in stocks at regular intervals to acclimate them to the experimental regime.

Administration of test substances

AVP V_1 antagonist [deamino-Pen[1], *O*-Me-Tyr[2], Arg[8]]-vasopressin (Bachem Ltd, Bubendorf, Switzerland) and polyinosinic:polycytidylic acid (Sigma Chemical Company Ltd, UK) were reconstituted in 0·9 % pyrogen-free saline and the solutions were filtered through a Millex GS 0·22 μm filter prior to administration. Test substances (250 μl/kg body weight) were administered intravenously (I.V.) into a marginal ear vein. V_1 antagonist or saline vehicle were injected 5 min before poly(I)·poly(C) (2·5 μg/kg, I.V.) or saline control.

Collection of blood samples and assay of prostaglandins

Blood samples (1 ml) were collected from a marginal ear vein into tubes containing 150 μl of 3 mM EDTA plus 53 mM ketoprofen. The plasma was immediately separated and aliquots of 500 μl dispensed into tubes containing 50 μl of 1 M HCl.

Prostaglandins were extracted from acidified rabbit plasma samples using Sep-Pak C_{18} columns (Rotondo *et al.* 1988). Columns were prewashed with 5 ml methanol followed by 10 ml distilled water. Acidified plasma was passed through the column and bound PGs eluted in 1·5 ml methanol. Radioimmunoassay kits were used to determine circulating immunoreactive PGE_2, $irPGE_2$ (Dupont, Stevenage, UK) and $irPGF_{2\alpha}$ (Baxter Healthcare, Cambridge, MA, USA) levels. The PGE_2 assay had a sensitivity of 4 pg/ml and showed cross-reactivity with PGE_1 (3·7 %), but no cross-reactivity with $PGF_{2\alpha}$. The $PGF_{2\alpha}$ assay had a sensitivity of 250 pg/ml and showed cross-reactivity with $PGF_{1\alpha}$ (28·2 %), but no cross-reactivity with PGE_2.

Statistics

Thermoregulatory indices (TRI_5) were determined by integration of the area under the curve for temperature responses during the 5 h following injection of pyrogen with 1 unit equivalent to a 1 °C rise for 1 h (Milton & Wendlandt, 1971). Data, expressed as the means of n experiments ± S.E.M., were analysed by either one-way analysis of variance or unpaired Student's t test and differences were considered significant when $P < 0·05$. Rate of temperature change was calculated over 15 min periods and the data expressed as change in temperature (°C) per hour. The rates of change were compared by one-way analysis of variance with saline-treated control animals and considered significant when $P < 0·05$.

RESULTS

Effect of V_1 antagonist on temperature responses to poly(I)·poly(C)

Polyinosinic:polycytidylic acid (poly(I)·poly(C)) at a dose of 2·5 μg/kg body weight produced a biphasic rise in core body temperature from a stable baseline of

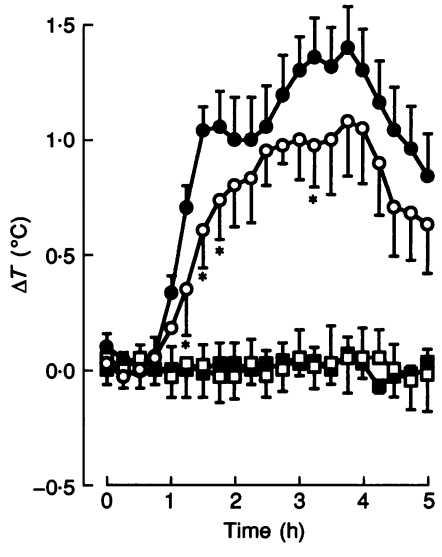

Fig. 1. Effect of V_1 antagonist on poly(I)·poly(C) fever in conscious rabbits. V_1 antagonist (250 μg/kg, I.V.) or saline vehicle was administered 5 min prior to poly(I)·poly(C) (2·5 μg/kg, I.V.) at time zero. Results, expressed as means ± S.E.M. change in core body temperature (ΔT °C) from a stable baseline from at least five measurements per point, are shown for saline vehicle plus poly(I)·poly(C) (●), V_1 antagonist plus poly(I)·poly(C) (○), saline vehicle plus saline vehicle (■) and V_1 antagonist plus saline vehicle (□). *$P < 0·05$ compared to poly(I)·poly(C) alone by Student's t test.

38·3 ± 0·2 °C ($n = 8$), with a lag phase of 60 min between injection and the first significant temperature rise, followed by peaks at 90 and 225 min after injection. When given 5 min prior to poly(I)·poly(C) (2·5 μg/kg, I.V.), the V_1 antagonist (250 μg/kg, I.V.) resulted in a fever with a lag phase of 75 min between injection and the first significant temperature rise and caused a significant reduction in the change in temperature at 75, 90, 105 and 195 min (Fig. 1). The V_1 antagonist alone (250 μg/kg, I.V.) or saline vehicle had no effect on body temperature when given prior to saline vehicle at time zero (Fig. 1).

The overall magnitude of the febrile response, as determined by TRI_5, was significantly ($P < 0·05$, Student's t test) reduced from 4·50 ± 0·31 for poly(I)·poly(C) (2·5 μg/kg, I.V.) plus saline vehicle-treated animals, to 3·37 ± 0·13 for poly(I)·poly(C) (2·5 μg/kg, I.V.) plus V_1 antagonist (250 μg/kg, I.V.) treated animals.

Rate-of-change analysis confirmed that temperature changes observed in poly(I)·poly(C) treated animals were biphasic (Fig. 2). After a lag phase of 45 min

the rates of change were significantly greater than zero between 45 and 90 min, significantly less than zero between 105 and 120 min, and again significantly greater than zero between 135 and 225 min. Pretreatment with the V_1 antagonist resulted in a monophasic pattern with significantly positive rates of change between 45 and 150 min. In both cases the rates of change in body temperature were negative after the peak rise at 225 min.

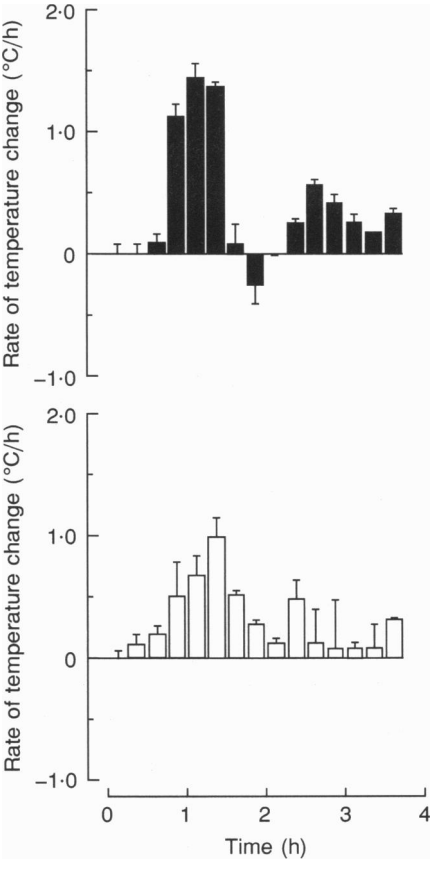

Fig. 2. Effect of poly(I)·poly(C) plus V_1 antagonist on the rate of body temperature change in conscious rabbits. Poly(I)·poly(C) (2·5 μg/kg) was administered at time zero. V_1 antagonist (250 μg/kg, I.V.) or saline vehicle was administered 5 min prior to poly(I)·poly(C). Results for poly(I)·poly(C) (upper panel) or poly(I)·poly(C) plus V_1 antagonist (lower panel) are expressed as means ± S.E.M. rate of change in core body temperature during each 15 min period with at least four measurements per point.

Effects on circulating irPGE$_2$ and irPGF$_{2\alpha}$

Poly(I)·poly(C) (2·5 μg/kg, I.V.) stimulated a 5-fold increase in circulating irPGE$_2$ levels, from a pretreatment level of 50 ± 17 pg/ml. The V_1 antagonist (250 μg/kg, I.V.) pretreatment significantly reduced the circulating irPGE$_2$ response stimulated by poly(I)·poly(C) (2·5 μg/kg), to the levels seen in saline-treated controls, at all time points measured (Fig. 3). The V_1 antagonist alone (250 μg/kg, I.V.) had no effect on circulating irPGE$_2$ when compared with saline vehicle-treated animals.

Poly(I)·poly(C) (2·5 μg/kg, I.V.) stimulated a 2·5-fold increase in circulating irPGF$_{2\alpha}$ levels, from a pretreatment level of 870 ± 100 pg/ml. The V_1 antagonist (250 μg/kg, I.V.) pretreatment significantly reduced the circulating irPGF$_{2\alpha}$ response to poly(I)·poly(C) (2·5 μg/kg) only at the 2·5 h time point (Fig. 4). The V_1 antagonist alone (250 μg/kg, I.V.) had no effect on circulating irPGF$_{2\alpha}$ when compared with saline vehicle-treated animals.

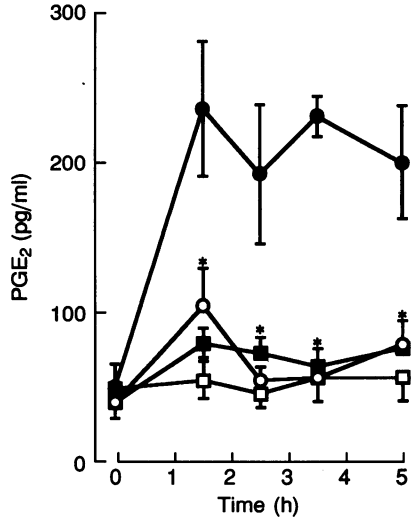

Fig. 3. Effect of V_1 antagonist on poly(I)·poly(C)-stimulated circulating PGE_2 in conscious rabbits. V_1 antagonist (250 μg/kg, I.V.) or saline vehicle was administered 5 min prior to poly(I)·poly(C) (2·5 μg/kg, I.V.) or saline vehicle at time zero. Circulating PGE_2 levels were determined by radioimmunoassay. Results, expressed as means ± S.E.M. from at least five measurements per point, are shown for saline vehicle plus poly(I)·poly(C) (●), V_1 antagonist plus poly(I)·poly(C) (○), saline vehicle plus saline vehicle (■) and V_1 antagonist plus saline vehicle (□). *$P < 0·05$ compared to poly(I)·poly(C) alone by one-way analysis of variance.

DISCUSSION

In agreement with previous studies (Rotondo *et al.* 1988) poly(I)·poly(C) stimulated a biphasic rise in body temperature and elevated circulating PG levels. In this study the febrile response to poly(I)·poly(C) was partially reduced by peripheral administration of an AVP V_1 antagonist, suggesting that endogenous AVP may have a modulatory propyretic role during the febrile response. The failure of the V_1 antagonist to alter resting body temperature itself suggests that the V_1 antagonist is interfering with a poly(I)·poly(C)-activated response and not some thermoregulatory mechanism. The potent interferon-inducing activities of poly(I)·poly(C) (Stringfellow, 1984) and the pyrogenicity of interferons (Dinarello *et al.* 1984) has led to suggestions that the febrile response to poly(I)·poly(C) may be mediated via endogenous interferon. AVP has been shown to stimulate γ-interferon production via actions on V_1 receptors (Johnson *et al.* 1982; Johnson & Torres, 1985) and a possible mechanism for the propyretic actions of AVP may therefore be via the stimulation of interferon production.

The reduction in the febrile response to poly(I)·poly(C) caused by administration of the V_1 antagonist was accompanied by an abolition of the circulating irPGE$_2$ response. The reduction in the irPGF$_{2\alpha}$ response to poly(I)·poly(C) caused by administration of the V_1 antagonist was less marked and only significant at the peak 2·5 h measurement. Poly(I)·poly(C)-induced temperature rises are PG dependent (Rotondo *et al.* 1988) and the observed antipyresis caused by the V_1 antagonist may

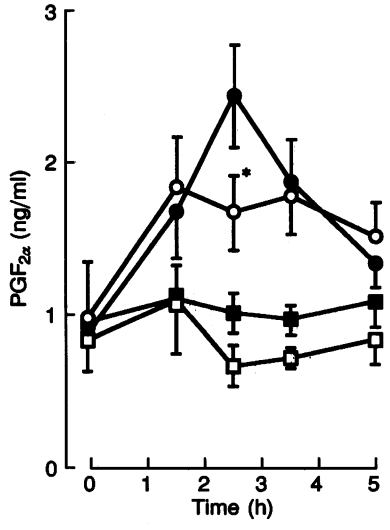

Fig. 4. Effect of V_1 antagonist on poly(I)·poly(C)-stimulated circulating PGF$_{2\alpha}$ in conscious rabbits. V_1 antagonist (250 μg/kg, I.V.) or saline vehicle were administered 5 min prior to poly(I)·poly(C) (2·5 μg/kg, I.V.) or saline vehicle at time zero. Circulating PGF$_{2\alpha}$ levels were determined by radioimmunoassay. Results, expressed as means ± S.E.M. from at least five measurements per point, are shown for saline vehicle plus poly(I)·poly(C) (●), V_1 antagonist plus poly(I)·poly(C) (○), saline vehicle plus saline vehicle (■) and V_1 antagonist plus saline vehicle (□). *$P < 0·05$ compared to poly(I)·poly(C) alone by one-way analysis of variance.

therefore be the result of inhibition of PGs. The specificity of the effects of the V_1 antagonist on poly(I)·poly(C)-stimulated PGs, with effects on PGE$_2$ but not PGF$_{2\alpha}$, may provide an explanation for the partial inhibition of the febrile response. These differential effects of the AVP V_1 antagonist on PGE$_2$ and PGF$_{2\alpha}$ responses may reflect different sites of PG production or an action of AVP which specifically modifies the PGE$_2$ response. Both PGE$_2$ and PGF$_{2\alpha}$ are pyrogenic when administered centrally (Milton & Wendlandt, 1971) and antipyretic drugs act by inhibition of arachidonic acid conversion to PGs (Vane, 1971). The results from this study suggest that the observed antipyresis with a V_1 antagonist may be due to specific inhibition of PGE$_2$, which is the most potent fever-inducing prostaglandin (Milton & Wendlandt, 1971). The observation that the V_1 antagonist pretreatment resulted in only a partially reduced febrile response to poly(I)·poly(C) and a complete blockade of the observed PGE$_2$ response, suggests that another PG, possibly PGF$_{2\alpha}$, may be responsible for the observed rises in body temperature.

Whilst the original studies by Milton & Wendlandt (1971) demonstrated that PGE_2 was the most potent prostaglandin, with a potency 27 times that of $PGF_{2\alpha}$ the results from this study show poly(I)·poly(C)-stimulated levels of circulating $PGF_{2\alpha}$, are 10–15 times those of circulating PGE_2, suggesting that $PGF_{2\alpha}$ may be present at sufficient concentrations to elicit effects.

The suggestion that the propyretic actions of AVP could be mediated via stimulatory actions on interferon production (Johnson *et al.* 1982; Johnson & Torres, 1985) are not incompatible with the results suggesting an involvement with PGE_2 since interferon fevers are PG dependent (Dinarello *et al.* 1984). This leads to the suggestion that poly(I)·poly(C) induces an interferon, which in turn stimulates PGE_2, and that production of this interferon is modulated by peripheral AVP.

The significant reduction in the thermoregulatory index (TRI_5) for the response to poly(I)·poly(C) when animals were pretreated with an AVP V_1 antagonist was accompanied by a change in the pattern of poly(I)·poly(C)-stimulated temperature rises. This change in the pattern of poly(I)·poly(C) fever, from biphasic to monophasic, when the AVP V_1 antagonist was administered peripherally, suggests either that AVP modulates the initial rises in body temperature or that the drop in body temperature could be due to stimulation of AVP, which is producing a transient antipyretic effect. The previous observations of AVP-induced antipyresis mediated via central AVP V_1 receptors (Cooper *et al.* 1987) is consistent with the suggestion that the biphasic temperature change pattern could be due to transient AVP-induced antipyresis.

Previous studies have suggested that biphasic fevers stimulated by exogenous pyrogens, including poly(I)·poly(C), are the result of induction of two or more endogenous pyrogens with different lag phases (Kluger, 1991). The possibility exists that the AVP V_1 antagonist inhibits the production, secretion or actions of one such endogenous pyrogen. The lag phases of the PGE_2 and $PGF_{2\alpha}$ responses to poly(I)·poly(C) have been shown to differ (Milton, Hillhouse & Milton 1993*b*) with PGE_2 having a lag phase of 1 h and $PGF_{2\alpha}$ a lag phase of 1·5 h. The AVP V_1 antagonist increases the lag phase between poly(I)·poly(C) injection and significant temperature increases and may therefore act by inhibiting the endogenous pyrogen responsible for the initial rise in body temperature, and also the PGE_2 response. The maintenance of a febrile response was accompanied by a rise in $PGF_{2\alpha}$ and could be the result of a second endogenous pyrogen activated by poly(I)·poly(C) independently of AVP.

In conclusion, endogenous peripheral AVP may have a propyretic action, during fever stimulated by the interferon inducer poly(I)·poly(C), mediated via actions on V_1 receptors and PGE_2. AVP may also have a role in the generation of a biphasic febrile response to poly(I)·poly(C), either by stimulating a transient antipyresis or by modulating the release of the endogenous pyrogen responsible for the initial rises in body temperature stimulated by poly(I)·poly(C).

N.G.N.M. is a Sir Halley Stewart Trust Fellow. This work was supported by a grant from the Nuffield Foundation to A.S.M.

REFERENCES

BERNARDINI, G. L., RICHARDS, D. B. & LIPTON, J. M. (1984). Antipyretic effect of centrally administered CRF. *Peptides* **5**, 57–59.

COOPER, K. E., CRANSTON, W. I. & HONOUR, A. M. (1967). Observations on the site and mode of action of pyrogens in the rabbit brain. *Journal of Physiology* **191**, 325–337.

COOPER, K. E., KASTING, N. W., LEDERIS, K. & VEALE, W. L. (1979). Evidence supporting a role for endogenous vasopressin in natural suppression of fever in sheep. *Journal of Physiology* **295**, 33–45.

COOPER, K. E., NAYLOR, A. M. & VEALE, W. L. (1987). Evidence supporting a role for endogenous vasopressin in fever suppression in the rat. *Journal of Physiology* **387**, 163–172.

DASCOMBE, M. J. & MILTON, A. S. (1979). Study on the possible entry of bacterial Endotoxin and prostaglandin E_2 into the central nervous system from the blood. *British Journal of Pharmacology* **66**, 565–572.

DINARELLO, C. A., BERNHEIM, H. A., DUFF, G. W., LE, H. V., NAGABHUSHAN, T. L., HAMILTON, N. C. & COCEANI, F. (1984). Mechanisms of fever induced by recombinant human interferon. *Journal of Clinical Investigation* **74**, 906–913.

DINARELLO, C. A., WEINER, P. & WOLFF, S. M. (1978). Radiolabelling and deposition in rabbits of purified human leukocytic pyrogen. *Clinical Research* **26**, 522A.

GARDINER, S. M., COMPTON, A. M. & BENNETT, T. (1990). Cardiovascular instability induced by intracerebroventricular administration of vasopressin or corticotrophin-releasing factor in conscious Long Evans and Brattleboro rats. *Journal of Neuroendocrinology* **2**, 39–43.

JOHNSON, H. M., FARRAR, W. L. & TORRES, B. A. (1982). Vasopressin replacement of Interleukin-2 requirement in gamma interferon production: lymphokine activity of a neuroendocrine hormone. *Journal of Immunology* **129**, 983–986.

JOHNSON, H. M. & TORRES, B. A. (1985). Regulation of lymphokine production by arginine vasopressin and oxytocin: modulation of lymphocyte function by neurohypophyseal hormones. *Journal of Immunology* **135**, 743–745s.

KASTING, N. W., CARR, D. B., MARTIN, J. B., BLUME, H. & BERGLAND, R. (1983). Changes in cerebrospinal fluid and plasma vasopressin in the febrile sheep. *Canadian Journal of Physiology and Pharmacology* **61**, 427–431.

KLUGER, M. J. (1991). Fever: role of pyrogens and cryogens. *Physiological Reviews* **71**, 93–127.

KOVACS, G. L. & DE WEID, D. (1983). Hormonally active vasopressin suppresses endotoxin-induced fevers in rats. Lack of effect of oxytocin and a behaviourally active vasopressin fragment. *Neuroendocrinology* **37**, 258–261.

KOZAK, W., MILTON, A. S., ABUL, H., DAVIDSON, J. & ROTONDO, D. (1989). Lipopolysaccharide, muamyl dipeptide and polyinosinic:polycytidylic acid induce the accumulation of inositol phosphates in blood monocytes and lymphocytes. *Cellular Signalling* **1**, 345–356.

MALKINSON, T. J., BRIDGES, T. E., LEDERIS, K. & VEALE, W. L. (1987). Perfusion of the septum of the rabbit with vasopressin antiserum enhances endotoxin fever. *Peptides* **8**, 385–389.

MENS, W. J. B., WITTER, A. & VAN WIMERSMA GREIDANUS, T. B. (1983). Penetration of neurohypophyseal hormones from plasma into cerebrospinal fluid (CSF): half times of disappearance of these neuropeptides from CSF. *Brain Research* **262**, 143–149.

MILTON, N. G. N., HILLHOUSE, E. W. & MILTON, A. S. (1992). Activation of the hypothalamo–pituitary–adrenocortical axis in the conscious rabbit by the pyrogen polyinosinic:polycytidylic acid is dependent on corticotrophin releasing factor-41. *Journal of Endocrinology* **135**, 69–75.

MILTON, N. G. N., HILLHOUSE, E. W. & MILTON, A. S. (1993*a*). A possible role for endogenous peripheral corticotrophin-releasing factor-41 in the febrile response of conscious rabbits. *Journal of Physiology* **465**, 415–425.

MILTON, N. G. N., HILLHOUSE, E. W. & MILTON, A. S. (1993*b*). Modulation of the prostaglandin responses of conscious rabbits to the pyrogen polyinosinic:polycytidylic acid by corticotrophin-releasing factor-41. *Journal of Endocrinology* **137** (in the Press).

MILTON, A. S. & WENDLANDT, S. (1971). Effects on body temperature of prostaglandins of the A, E and F series on injection into the third ventricle of unanaesthetized cats and rabbits. *Journal of Physiology* **218**, 325–336.

NAYLOR, A. M., RUWE, W. D. & VEALE, W. L. (1986). Antipyretic action of centrally administered arginine vasopressin but not oxytocin in the cat. *Brain Research* **385**, 156–160.

OPP, M., OBAL, F. JR & KRUEGER, J. M. (1989). Corticotropin-releasing factor attenuates interleukin 1-induced sleep and fever in rabbits. *American Journal of Physiology* **257**, R528–535.

ROTONDO, D., ABUL, H. T., MILTON, A. S. & DAVIDSON, J. (1988). Pyrogenic immunomodulators increase the level of prostaglandin E_2 in the blood simultaneously with the onset of fever. *European Journal of Pharmacology* **154**, 145–152.

STRINGFELLOW, D. A. (1984). Nonpolynucleotide inducers of interferon. In *Handbook of Experimental Pharmacology*, vol. 71, *Interferons and their Applications*, ed. CAME, P.E. & CARTER, W.A., pp. 371–383. Springer-Verlag, Berlin.

VANE, J. R. (1971). Inhibition of prostaglandin synthesis as a mechanism of action for aspirin-like drugs. *Nature* **231**, 415–417.

VEALE, W. L., COOPER, K. E. & RUWE, W. D. (1984). Vasopressin: its role in antipyresis and febrile convulsion. *Brain Research Bulletin* **12**, 161–165.

VLASKOVSKA, M. A., HERTTING, G. & KNEPEL, W. (1984). Adrenocorticotropin and β-endorphin release from the rat adenohypophysis *in vitro*: inhibition by prostaglandin E_2 formed locally in response to vasopressin and corticotropin-releasing factor. *Endocrinology* **115**, 895–903.

WANG, B. C., SHARE, L., CROFTON, J. T. & KIMURA, T. (1981). Changes in vasopressin concentration in plasma and cerebrospinal fluid in response to hemorrhage in anesthetized dogs. *Neuroendocrinology* **33**, 61–66.

WUTHRICH, R. P. & VALLOTTON, M. B. (1986). Prostaglandin E_2 and cyclic AMP response to vasopressin in renal medullary tubular cells. *American Journal of Physiology* **251**, F499–505.