J. Physiol. (1987), **384**, *pp.* 467–477
With 6 text-figures
Printed in Great Britain

ISOPERIODIC BURSTING BY MAGNOCELLULAR NEUROENDOCRINE CELLS IN THE RAT HYPOTHALAMIC SLICE

BY R. DAVID ANDREW

From the Department of Anatomy, Queen's University, Kingston, Ontario K7L 3N6, Canada

(*Received 13 March 1986*)

SUMMARY

1. Recruitment of magnocellular neuroendocrine cells (m.n.c.s) to a repetitive burst pattern (phasic firing) is associated with increased vasopressin secretion from neurohypophysial terminals in the intact animal. Based on invertebrate studies, bursts of action potentials can arise in two distinct ways: as an intrinsic property of the recorded cell or as an emergent property of synaptic interactions.

2. The majority of phasic m.n.c.s in the hypothalamic slice preparation display an endogenous pace-maker mechanism underlying bursting. It is voltage dependent and varies considerably in periodicity and time course as described in the accompanying paper (Andrew, 1987).

3. In contrast to this intrinsic mechanism, the present study examined if cells might be driven by periodic synaptic input. Intracellular recordings from six of thirty-two phasic m.n.c.s in the supraoptic nucleus revealed an isoperiodic oscillation of the membrane potential, where each depolarizing phase could support a burst.

4. The oscillation had a smooth trajectory and fixed period (range, 5–17 s). The oscillatory frequency was not voltage dependent, i.e. periodicity was unaffected by steady current injection through the recording electrode.

5. The frequency and amplitude of the oscillation remained unaltered by action potential firing. The isoperiodic oscillation could abate spontaneously, leaving intact the endogenous ability to fire a triggered burst driven by an underlying plateau potential.

6. Perfusion with either 10 mM-Mg^{2+}–0·05 mM-Ca^{2+} or 0·5–2·0 μM-tetrodotoxin blocked both the oscillation and evoked post-synaptic potentials, indicating that the oscillation was synaptically generated. Given that both treatments could also block the intrinsic burst process and that the oscillation could spontaneously abate, the synaptic nature of the oscillation remains a tentative but reasonable conclusion.

7. In total, the evidence suggests that the isoperiodic oscillation has a synaptic origin independent of intrinsic mechanisms. It probably results from synaptic input generated within the slice but the source is not yet identified. This input could support phasic bursting in those m.n.c.s lacking a pace-maker ability and so promote the release of vasopressin in the intact animal.

INTRODUCTION

Magnocellular neuroendocrine cells (m.n.c.s) synthesize either vasopressin or oxytocin and store these hormones in axon terminals comprising the neurohypophysis. Systemic release of these hormones is increased during bursts of action potentials recorded from m.n.c. somata located in the paraventricular and supraoptic nuclei, as reviewed by Poulain & Wakerley (1982). In the rat, the oxytocinergic population fires a single, synchronous burst in response to suckling. This releases a pulse of oxytoxin which evokes milk ejection. During periods of haemorrhage or elevated blood osmolarity, each neurone of the vasopressinergic population is recruited in a non-synchronous manner to a repetitive bursting pattern (termed phasic firing). Burst and inter-burst periods can last from several seconds to more than a minute, indicating that the process underlying repetitive bursting activates and inactivates over relatively long time periods.

Repetitive bursting by neurones and neuronal networks has been studied in both vertebrates and invertebrates, particularly in systems where firing can be associated with a rhythmic motor output, such as during locomotion or respiration (Simmers, 1981; Jordan, 1983). Repetitive bursting can be endogenously generated, the 'pace-maker' mechanism underlying rhythmicity residing within the membrane of the recorded neurone. Alternatively, a burster may be strictly passive, firing only when synaptically driven past spike threshold and so behaving as a 'follower' (Johnston & Brown, 1984). As observed in this report, a neurone may display a combination of traits, possessing intrinsic burst ability which may reinforce or oppose a patterned synaptic input imposed on the cell.

In a majority of cultured supraoptic neurones, phasic activity results from patterned synaptic input (Gahwiler & Dreifuss, 1979). Alternatively, in the hypothalamic slice preparation, most phasic firing arises from an endogenous burst ability (Andrew & Dudek, 1983; Andrew, 1987). The experiments described here attempt to provide evidence that, in addition to an endogenous process, rhythmic synaptic input can drive repetitive bursting by some m.n.c.s in the slice preparation. In the intact animal this mechanism may aid the recruiting of endocrine neurones into a phasic firing pattern and so promote vasopressin release. A preliminary report of these findings has recently appeared (Andrew, 1985).

METHODS

The materials and methods were similar to those described in the accompanying paper (Andrew, 1987). The six supraoptic nucleus cells studied in detail here were distinguished as m.n.c.s based on criteria set down in the Methods section of Andrew & Dudek (1984). These cells were distinct from other m.n.c.s in that steady hyperpolarization revealed an oscillation in the membrane potential. Following some experiments, this oscillation was recorded on tape and then played out on a chart recorder. This recorded oscillation was simulated using a sine-wave generator also with output to a chart recorder. The recorded and simulated oscillations were visually aligned to confirm if the intracellular recording was indeed isoperiodic.

RESULTS

In the accompanying paper I examined an intrinsic process underlying repetitive bursting in m.n.c.s. The most important feature of this process is a plateau depolarization of the membrane potential that repetitively drives a burst and is independent of synaptic input (Andrew, 1987). Initiation of the plateau is at times discernable between bursts as a slow depolarization. Plateau activation accelerates following an action potential, apparent as the depolarizing after-potential (d.a.p.). Even without the slow activation, any brief depolarizing stimulus evoking one or a few action potentials can fully activate the plateau and a burst then ensues.

Activation of the plateau was necessary and sufficient to account for phasic bursting in the majority of m.n.c.s. As shown in Fig. 1, plateau potentials became

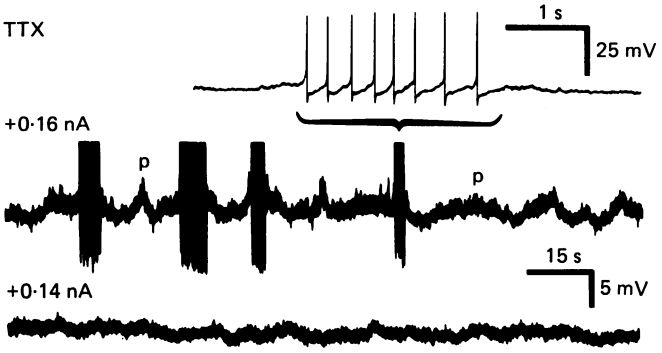

Fig. 1. Non-synaptic properties of intrinsic bursting. Treatment with 2 μM-TTX blocked evoked synaptic input (not shown) and low-threshold Na^+ spiking. This in turn revealed the plateau potential (p shows two examples) that drives intrinsic bursting in m.n.c.s, as detailed in the accompanying paper (Andrew, 1987). Unlike the oscillation described in the following Figures, plateau potentials were sporadic and variable in trajectory and duration. In TTX they were fully activated at membrane potentials near the Ca^{2+} spike threshold (+0·16 nA), and less apparent at slightly less polarized levels (+0·14 nA).

more apparent as the cell was steadily depolarized to a level near the Ca^{2+} spike threshold. Plateau amplitude decreased if the cell was held at a more hyperpolarized potential. Since evoked post-synaptic potentials were blocked in tetrodotoxin (TTX; Fig. 3 of Andrew, 1987), the plateau was independent of synaptic input. Most relevant to the findings below, the plateaux occurred sporadically and with highly variable rates of activation and inactivation (Fig. 1).

Six of thirty-two phasic cells in the supraoptic nucleus, while capable of generating d.a.p.s and triggered bursts, displayed a markedly different process underlying repetitive bursting. The membrane potential fluctuated as a sinusoidal wave form with smooth trajectory and fixed periodicity (Fig. 2). This oscillation maintained a constant periodicity at all levels of tonic current injection (Figs. 3 and 4*A*). Among the six cells, the oscillation period ranged from 5 to 17 s. In each cell, the oscillation could be simulated with the output of a sine-wave generator (Figs. 4 and 5), demonstrating the isoperiodicity of the wave form. In three cells tested, the

oscillation increased in amplitude as hyperpolarizing current injection was increased (Fig. 4*A*). In contrast, the amplitude of the plateau potential was greatest when the membrane potential was held just below spike threshold and further hyperpolarization inactivated this process (Fig. 1).

The oscillation and action potentials

When action potentials overrode the depolarizing phase of the isoperiodic oscillation, their summed d.a.p.s contributed to a further reduction in membrane potential. These two independent mechanisms could be complementary in promoting a burst (Fig. 3). Strict 'following' persisted if the oscillation amplitude was large

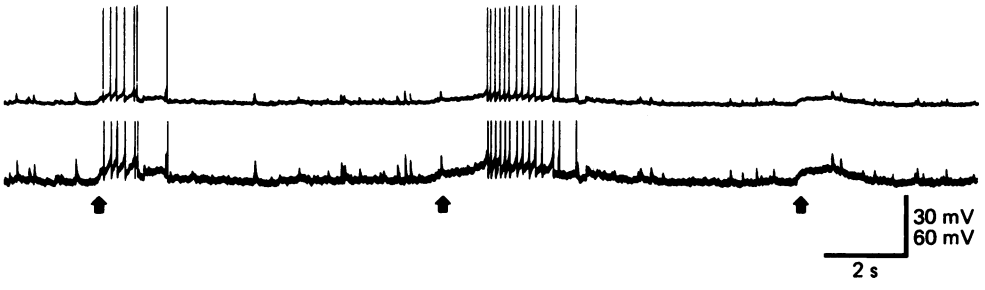

Fig. 2. Typical isoperiodic oscillation of the membrane potential in an m.n.c. The depolarizing phase (arrows) occurred at fixed intervals, were not altered by action potential discharge (right) and could spontaneously abate (not shown).

enough not only to drive the membrane potential to burst threshold, but then to hyperpolarize the cell and overcome the cell's intrinsic tendency to maintain bursting.

In all six cells, the oscillation was a process independent of the ability to fire a triggered burst (i.e. to generate a plateau potential). This is demonstrated in Fig. 3 where the oscillation spontaneously abated, resulting in a silent cell. However, if the cell was then briefly depolarized with a current pulse, a plateau potential was generated which drove a burst. Hence burst ability was maintained, but, without the isoperiodic oscillation, the cell could not spontaneously reach spike threshold.

Fig. 4 again illustrates the functional separation of the isoperiodic oscillation (Fig. 4*A*) from the plateau potential (Fig. 4*C*). Careful examination of the burst pattern when the oscillation was no longer detectable showed that it still influenced spike patterning. In Fig. 4*A*, the recorded cell was a 'faithful' follower in that spiking waxed and waned with the underlying rhythmic oscillation. The amount of spiking was reduced with increasingly hyperpolarized levels of steady current injection, and the oscillation amplitude increased. Several minutes later the cell began firing in the more typical phasic mode, i.e. burst and inter-burst periods were variable in duration. Apparently a diminution of the oscillatory amplitude caused the m.n.c. to follow less faithfully. Bursts initiated by the depolarizing phase of the oscillation now outlasted it (Fig. 4*B*). Further diminution of the oscillation over several minutes resulted in silence (Fig. 4*C*), although a burst could still be triggered. The oscillatory input was no longer detectable but its sculpting influence on the firing pattern was still apparent.

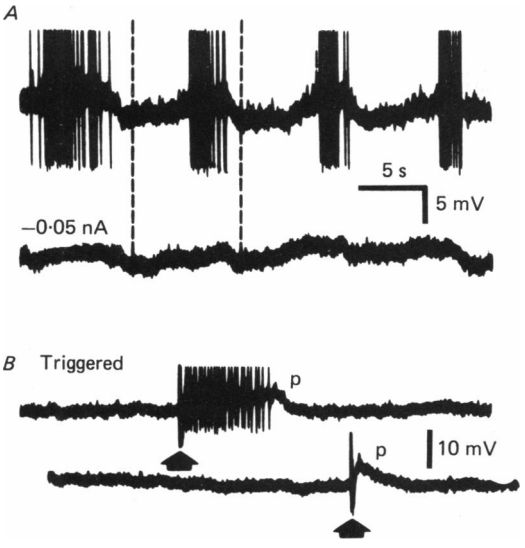

Fig. 3. The isoperiodic oscillation and intrinsic bursting are independent events. *A*, the cell normally fired rhythmic bursts (upper trace) driven by an oscillation of fixed periodicity, as was revealed with a low level of steady hyperpolarization (lower trace). The period (dashed lines) was neither activity dependent nor voltage sensitive. *B*, later during the same impalement, this continuous record shows that the oscillation abated but that triggered burst ability (evoked with a 150 ms depolarizing pulse; arrow in upper trace) remained. As seen with a second pulse injection (arrow in lower trace), such bursts were driven by activation of the plateau potential (p).

Two general observations could be made. First, the ability to fire a single burst (i.e. to generate a plateau potential) was independent of the oscillation. Secondly, these two mechanisms could interact to varying degrees in m.n.c.s to control the pattern of bursting. This second point is further illustrated in Fig. 5. The cell fired isoperiodically (left), but stopped following when burst initiation was delayed well into the depolarizing phase of the oscillation (bar); once under way it then failed to follow the hyperpolarizing phase (dashed line). Finally, the cell spontaneously fell silent (right), revealing the sinusoidal oscillation in membrane potential. Apparently, then, the intrinsic propensity to burst could upset faithful following of the oscillation.

Source of the oscillation

Was the isoperiodic oscillation generated endogenously by the recorded cell or did it result from synaptic input? One way to differentiate was to test if its periodicity was voltage sensitive. As shown in Figs. 3 and 4*A*, steady hyperpolarizing current injection did not alter the period of oscillation. Furthermore, injection or brief depolarizing or hyperpolarizing pulses neither changed the periodicity nor phase-shifted the oscillation (not shown). This is in sharp contrast to the plateau potential which was clearly voltage sensitive, remaining inactive when the cell was hyperpolarized only 2–3 mV below spike threshold (Fig. 1). The trajectory and time course of the oscillation were also independent of action potential firing, unlike the intrinsic

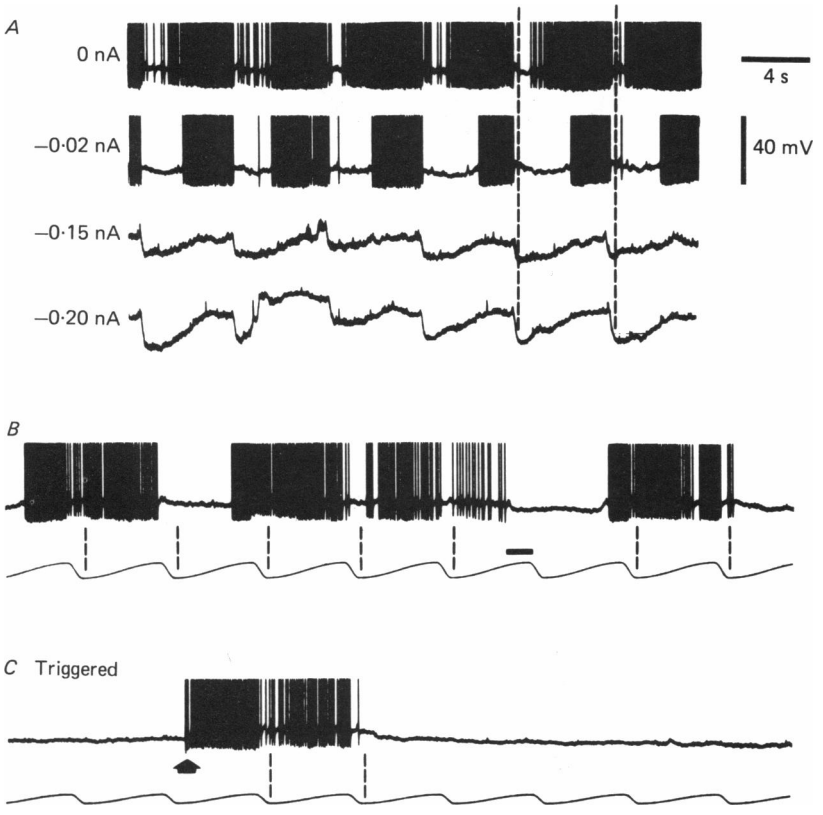

Fig. 4. The oscillatory influence on burst behaviour can gradually diminish with time. *A*, early in the recording period the cell faithfully followed the isoperiodic oscillation. Stronger hyperpolarizing current increased the amplitude but not the period of the oscillation (dashed lines). *B*, several minutes later the isoperiodic oscillation appeared lost and the cell no longer followed faithfully. However, when compared to an artificially generated sine wave matched to the oscillation in *A*, the oscillatory input still affected the firing pattern. Firing slowed during each hyperpolarizing phase (dashed lines) and, with one exception (bar), firing increased during each depolarizing phase. *C*, further into the impalement period, the m.n.c. was no longer driven to burst threshold, yet it could fire a burst when triggered with a 150 ms depolarizing pulse (arrow). Comparison with the hypothetical sine wave indicated that the underlying oscillation in membrane potential still influenced firing.

mechanism where an action potential could rapidly activate the plateau potential in the form of a depolarizing after potential.

Selective blockade of chemical synapses with a concomitant loss of the isoperiodic oscillation would provide strong evidence for the oscillation having a synaptic origin. In fact, in the one cell tested, it disappeared following perfusion with saline containing 10 mM-Mg^{2+}–0·05 mM-Ca^{2+}. However, it should be noted that in ten other phasic cells without the oscillation, intrinsic phasic firing was also blocked by such treatment. This is probably due to an inward Ca^{2+} current associated with plateau potential generation (Andrew, 1987). Therefore, low-Ca^{2+} solutions were not useful in differentiating between synaptically or intrinsically mediated bursting, because both

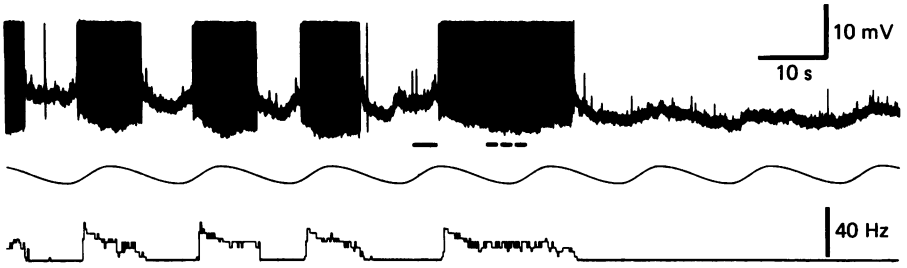

Fig. 5. Intrinsic properties affect an m.n.c.'s capacity to follow synaptic input faithfully. The isoperiodic oscillation was recorded intracellularly (upper trace) and was simulated with a sine-wave generator (middle trace). The cell faithfully followed the oscillation (left) but then spontaneously failed to initiate a burst (bar). The delayed burst then overrode the hyperpolarizing phase (dashed line). Finally, the cell fell silent (right) despite the oscillation. The spike rate-meter record is shown in the lower trace.

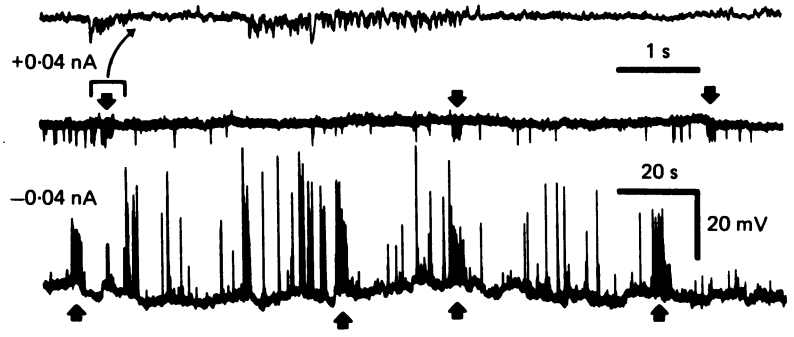

Fig. 6. An expected pattern of post-synaptic potentials in an m.n.c. that probably received synaptic input from a phasic m.n.c. Bursts of i.p.s.p.s (arrows, middle trace) were irregular in periodicity and duration, as is the repetitive burst pattern in most m.n.c.s. Upper trace shows an i.p.s.p. burst at faster chart speed. The i.p.s.p.s reversed with steady injection of hyperpolarizing current (lower trace). This pattern of synaptic input differs markedly from the smooth, isoperiodic oscillation shown in Figs. 2–5.

processes appeared Ca^{2+} dependent. It was fortunate that the oscillation could abate spontaneously, demonstrating that it was a process distinct from the plateau potential, which remained intact.

One possible source of rhythmic synaptic input to an m.n.c. is from another phasic m.n.c. However phasic firing in most m.n.c.s (twenty-four out of thirty-two in the previous study) was not characteristically isoperiodic. i.e. burst and inter-burst durations varied considerably. It follows that the synaptic input provided by a phasic m.n.c. would not usually be isoperiodic. In fact, the asynchronous pattern of post-synaptic potentials suggestive of a presynaptic m.n.c. has been observed in two recordings. At irregular intervals, a volley of i.p.s.p.s was recorded in one cell (Fig. 6, arrows), as shown in expanded form in the upper trace. During 0·40 nA of hyperpolarizing current, the i.p.s.p.s were reversed (lower trace). As would be expected if the presynaptic cell was a phasic m.n.c., the presynaptic input was not isoperiodic. This contrasted sharply with the smooth rhythmicity of the isoperiodic oscillation.

DISCUSSION

Phasic firing by magnocellular neuroendocrine cells (m.n.c.s) is of interest not only because it is correlated with increased systemic release of vasopressin, but because it represents long, alternating periods of activation and inactivation in a mammalian neurone. 'Isoperiodicity' refers to a repetitive burst discharge recurring at a fixed interval, a situation not often observed in m.n.c. recordings *in vivo* or *in vitro*. This is because the plateau potential underlying repetitive bursting activates and inactivates at variable rates, resulting in an irregular phasic pattern (Andrew, 1987).

The m.n.c.s of this study display bursting of regular periodicity when they faithfully follow an isoperiodic oscillation of the membrane potential. If the depolarizing trajectory of the oscillation crosses spike threshold and if the hyperpolarizing trajectory overcomes the cell's tendency to continue bursting, then action potentials will follow faithfully. However, such following is often disrupted by the cell's endogenous ability to burst. Indeed, the oscillation and intrinsic burst events are completely separable. The oscillation continues when a cell is hyperpolarized below spike threshold and, conversely, a cell retains its ability to fire a triggered burst when the oscillation spontaneously abates. The isoperiodic oscillation provides a means of repetitively driving an otherwise silent cell across burst threshold.

The isoperiodic oscillation differs from the intrinsic plateau potential which drives phasic firing in a majority of m.n.c.s (Andrew & Dudek, 1983; Andrew, 1987) in the following respects: (1) its frequency is voltage independent; (2) it remains unaltered by action potential discharge; (3) its amplitude is a function of the membrane potential; (4) it can abate spontaneously, while bursting generated by the plateau remains intact; and (5) it cannot be promoted using either steady injection of depolarizing current or Ca^{2+} agonists, both of which enhance slow Ca^{2+}-activated events underlying intrinsic bursting.

It is possible to infer the synaptic nature of the oscillation by blocking it and showing a loss of evoked synaptic input concurrently. Unfortunately, as demonstrated in the accompanying paper (Andrew, 1987), the standard method of synaptic blockade using high-Mg^{2+}–low-Ca^{2+} saline also eliminates plateau-generated bursting, whether spontaneous or evoked. As pointed out by Johnston & Brown (1984), such solutions not only block synapses but also interfere with the intrinsic burst mechanism itself. Thus Ca^{2+} antagonists or low-Ca^{2+} solutions cannot distinguish between an extrinsic or intrinsic source for the isoperiodic oscillation.

Because of the small number of cells studied here ($n = 6$), it is difficult to say if the oscillation was comprised of periodic e.p.s.p.s, i.p.s.p.s or both. During steady hyperpolarizing current injection in three cells, the oscillation amplitude was undiminished at the levels where Cl^-- or K^+-mediated events reversed in m.n.c.s. However in one cell, steady depolarization increased the amplitude. Further experiments are required to determine the conductance changes responsible for the isoperiodic oscillation.

Rhythmic oscillation in other mammalian neurones

In m.n.c.s, the isoperiodic oscillation is qualitatively similar to the locomotor drive potentials recorded in motoneurones of the cat during fictive locomotion (Shefchyk

& Jordan, 1985). The drive potential source is probably an alternating series of e.p.s.p.s and i.p.s.p.s generated by spinal interneurones as part of a hypothetical central pattern generator for locomotion (reviewed by Jordan, 1983). Although slower in time course, the oscillation displayed by m.n.c.s is also similar in trajectory to the 6–10 Hz oscillation seen in thalamic relay neurones during a spindle sequence in the cat (Roy, Clercq, Steriade & Deschenes, 1984). A component of this oscillation is generated endogenously by individual thalamic cells (Jahnsen & Llinás, 1984) and is probably activated and supported by synaptically evoked changes in voltage (Roy *et al.* 1984). In the thalamic cells, the depolarizing trajectory of the oscillation normally initiates and drives a burst of action potentials. In m.n.c.s intrinsic firing often overwhelms the repolarizing phase of the oscillation and the burst continues. Alternatively, intrinsic inhibition following a burst can act to prevent renewed bursting. In neither case is there strict following of the oscillation as in the motor and thalamic neurones mentioned above. This may explain why m.n.c. bursting of fixed periodicity is rarely observed in intact animal studies.

Isoperiodic bursting can be induced in irregularly firing magnocellular neurones in culture following exposure to nicotine or γ-aminobutyric acid (GABA) antagonists (Gahwiler & Dreifuss, 1980). Each burst rides on a depolarizing envelope that appears to be synaptically generated (B. Gahwiler, personal communication). The fact that neurotransmitter agonists or antagonists can induce an isoperiodic bursting in cultured m.n.c.s supports the suggestion that phasic firing of fixed periodicity arises from chemical synaptic input. However, the unlikely possibility remains that an intrinsic isoperiodic oscillation, which is not voltage dependent, could be activated by such neuromodulators.

Possible sources of oscillation

Isoperiodic synaptic input requires a presynaptic cell or network of cells with a rhythmic output of fixed periodicity. Anterograde and retrograde tracer studies show that cells in the supraoptic and paraventricular nuclei receive input from the two circumventricular organs, the median preoptic nucleus, the nucleus of the solitary tract and the A1 region. Perinuclear areas receive input from limbic structures (Tribollet, Armstrong, Dubois-Dauphin & Dreifuss, 1985). Since none of these areas are present in our hypothalamic coronal slice, a local site must be responsible for the oscillation. One possible source is recurrent input from the phasic m.n.c.s themselves. M.n.c.s immunoreactive for oxytocin do receive oxytocin fibres (Theodosis, 1985), but the source has not been ascertained. In any case, phasic m.n.c. output is not characteristically isoperiodic, so a cell situated post-synaptically to a phasic m.n.c. would display bombardments of post-synaptic potentials at irregular intervals. In fact, this is occasionally observed in m.n.c. recordings (Fig. 6; see also Fig. 10 in Andrew & Dudek, 1984). Therefore some cells probably do receive input originating from other phasic m.n.c.s, either directly or possibly via a small population of interneurones within the supraoptic nucleus (Leng & Dyball, 1983). However this source still does not account for the isoperiodicity.

Another possible local source of input is a population of cholinergic neurones just dorsal to the supraoptic nucleus (Sofroniew, Eckenstein, Thoenen & Cuello, 1982; Hatton, Ho & Mason, 1983). Although m.n.c. axon collaterals enter this area (Mason,

Ho & Hatton, 1984), electrophysiological studies *in vivo* have not reported phasic cells in this location. Phasic cells have been reported in the lateral hypothalamus, but these do not fire with fixed periodicity (Leng, 1981).

The fact is that no neurones which burst isoperiodically have yet been reported *in vivo* within the region comprising the coronal hypothalamic slice. A possible isoperiodic source could be a non-spiking, pace-maker type of neurone existing within the locale of the supraoptic nucleus. Endogenous voltage swings in such cells would lead to graded transmitter release (Simmers, 1981). This type of interneurone would be missed with intracellular and extracellular recordings because action potential firing is the primary diagnostic feature of a neuronal recording. To date the existence of such a cell type in mammals is only speculative.

To summarize, then, m.n.c.s possess two mechanisms which can act in concert or independently to provide an envelope of depolarization that drives a burst. First, there is the plateau potential which is an active, voltage-dependent event not requiring synaptic integrity. It can be triggered following evoked spikes in most m.n.c.s and occurs spontaneously but irregularly in the majority of phasic m.n.c.s. Secondly, as described here, a few m.n.c.s display a sinusoidal oscillation in membrane potential which can drive repetitive bursting. Unlike intrinsic bursting, this process is voltage independent and isoperiodic. Its amplitude usually increases with increased hyperpolarization, whereas the plateau potential is inactivated.

Rhythmic synaptic input promotes phasic firing, but faithful following of the oscillating membrane potential can be obscured by the cell's intrinsic propensity to burst. The oscillation in membrane potential apparently arises from periodic synaptic input generated within the slice; the source is not yet identified. This input could promote and support phasic bursting in m.n.c.s lacking a pace-maker ability. It may be an important mechanism for recruiting phasic m.n.c.s and thus increasing vasopressin release in the intact animal.

Note added in proof. An identical sinusoidal oscillation (but of higher frequency) was recently described in inferior olivary neurones by Llinás & Yarom (*Journal of Physiology* **376**, 163–182 (1986)). It may result as a subthreshold property of a neuronal ensemble which is coupled by electrotonic junctions. Such a mechanism could be operative in m.n.c.s.

Thanks to Mr H. Verstappen for photographic work and Miss A. Doyle for secretarial assistance. This work was supported by a Medical Research Council of Canada Operating Grant (MA-7884), an M.R.C. Scholarship and the Botterell Foundation.

REFERENCES

ANDREW, R. D. (1985). Phasic magnocellular neuroendocrine cells can be 'pacemakers' or 'followers'. *Neuroscience Abstracts* **11**, 509.

ANDREW, R. D. (1987). Endogenous bursting by rat supraoptic neuroendocrine cells is calcium dependent. *Journal of Physiology* **384**, 451–465.

ANDREW, R. D. & DUDEK, F. E. (1983). Burst discharge in mammalian neuroendocrine cells involves an intrinsic regenerative mechanism. *Science* **221**, 1050–1052.

ANDREW, R. D. & DUDEK, F. E. (1984). Analysis of intracellularly recorded phasic bursting by mammalian neuroendocrine cells. *Journal of Neurophysiology* **51**, 552–566.

GAHWILER, B. H. & DREIFUSS, J. J. (1979). Phasically firing neurons in long-term cultures of the rat hypothalamic supraoptic area: pacemaker and follower cells. *Brain Research* **177**, 95–103.

GAHWILER, B. H. & DREIFUSS, J. J. (1980). Transition from random to phasic firing induced in neurons cultured from the hypothalamic supraoptic area. *Brain Research* **193**, 415–425.

HATTON, G. I., HO, Y. W. & MASON, W. T. (1983). Synaptic activation of phasic bursting in rat supraoptic nucleus recorded in hypothalamic slices. *Journal of Physiology* **345**, 297–317.

JAHNSEN, H. & LLINÁS, R. (1984). Ionic basis for the electroresponsiveness and oscillatory properties of guinea-pig thalamic neurones *in vitro*. *Journal of Physiology* **349**, 227–247.

JOHNSTON, D. & BROWN, T. H. (1984). The synaptic nature of the paroxysmal depolarizing shift in hippocampal neurons. *Annals of Neurology* **16**, S65–71.

JORDAN, L. M. (1983). Factors determining motoneuron rhythmicity during fictive locomotion. In *Neural Origin of Rhythmic Movements*, ed. ROBERTS, A. & ROBERTS, B. pp. 421–427. U.K.: Society for Experimental Biology.

LENG, G. (1981). Phasically firing neurones in the lateral hypothlamus of anaesthetized rats. *Brain Research* **230**, 390–393.

LENG, G. & DYBALL, R. E. J. (1983). Intercommunication in the rat supraoptic nucleus. *Quarterly Journal of Experimental Physiology* **68**, 493–504.

MASON, W. T., HO, Y. W. & HATTON, G. I. (1984). Axon collaterals of supraoptic neurones: anatomical and electrophysiological evidence for their existence in the lateral hypothalamus. *Neuroscience* **11**, 169–182.

POULAIN, D. A. & WAKERLEY, J. B. (1982). Electrophysiology of hypothalamic magnocellular neurones secreting oxytocin and vasopressin. *Neuroscience* **7**, 773–808.

ROY, J. P., CLERCQ, M., STERIADE, M. & DESCHENES, M. (1984). Electrophysiology of neurons of lateral thalamic nuclei in cat: mechanisms of long-lasting hyperpolarizations. *Journal of Neurophysiology* **51**, 1220–1235.

SHEFCHYK, S. J. & JORDAN, L. M. (1985). Motoneuron input-resistance changes during fictive locomotion produced by stimulation of the mesencephalic locomotor region. *Journal of Neurophysiology* **54**, 1101–1108.

SIMMERS, J. A. (1981). Non-spiking interactions in crustacean rhythmic motor systems. In *Neurones Without Impulses*, ed. ROBERTS, A. & BUSH, B. M. H. pp. 171–186. U.K.: Society for Experimental Biology.

SOFRONIEW, M. V., ECKENSTEIN, F., THOENEN, H. & CUELLO, A. C. (1982). Topograph of choline acetyltransferase-containing neurons in the forebrain of the rat. *Neuroscience Letters* **33**, 7–12.

THEODOSIS, D. T. (1985). Oxytocin-immunoreactive terminals synapse on oxytocin neurones in the supraoptic nucleus. *Nature* **313**, 682–684.

TRIBOLLET, E., ARMSTRONG, W. E., DUBOIS-DAUPHIN, M. & DREIFUSS, J. J. (1985). Extra-hypothalamic afferent inputs to the supraoptic nucleus area of the rat as determined by retrograde and anterograde tracing techniques. *Neuroscience* **15**, 135–148.