Effect of LPS administration on body colour of the carp

Number	Length (cm)	Weight (g)	LPS (μg)	Body colour change Latency (min)	Peak (mm, min)		50% recovery (min)	Visceral temperature (°C)
1	27.0	505.0	0.02	15.5	− 48.0	27.0	53.0	20.6
2	26.0	510.0	0.02	5.0	− 48.0	24.0	-	20.8
3	25.5	420.0	0.02	7.0	− 141.9	38.0	51.0	23.4
4	27.0	540.0	0.50	18.0	− 99.5	112.0	-	20.9
5	25.0	515.0	0.10	29.0	− 62.0	39.0	74.0	21.5
$\bar{x}$	26.1	498.0	0.13	14.9	− 79.9	48.0	59.3	21.4
$S\bar{x}$	± 0.4	± 1.8	±0.08	± 3.8	± 16.2	± 14.6	± 6.0	± 0.5

Body colour was evaluated semiquantitatively as mm of recorder deflexion. Negative values for body colour indicate lightening of body colour.

in 59.3 min. Application of only fish saline to the anterior brainstem induced no significant responses in body colour. Febrile properties of the LPS used was examined in five rabbits: i.v. injection of 0.2 μg/ml per kg LPS induced an increase in rectal temperature by over 2.0 °C within 3 h.

It is possible that 'excitatory paling' of body colour as an arousal reflex is induced by nonspecific effects on the telencephalon during LPS-treatment. Such an excitatory paling, however, can be discriminated from LPS-induced lightening, because the former change begins and recovers relatively quickly, and is smaller in amplitude. The activity of cutaneous sympathetic efferents were deduced to be increased during body colour lightening, according to the functional property of effector neurones for body colour. Likewise, in the mammal, the activity of cutaneous sympathetic efferents is thought to be increased during pyrogen-induced fever because there is vasoconstriction to reduce heat loss from the skin[16,17].

If LPS increases a set point temperature in the thermoregulatory centre, the response should be predictable, viz., like that caused by cold stimulation. In fact, cold stimulation of the skin[18,19] and the spinal cord[9] induces lightening of body colour in the fish. Therefore, the present results could be taken to indicate that an increase in set point temperature is responsible for increased cutaneous autonomic activity following LPS-administration. In conclusion, the present investigation has provided further evidence that the fish has the same central mechanism as that of homeotherms for temperature responses, both in behaviour and in autonomic functions.

1 We express thanks to Dr S. Kanoh, National Institute of Hygenie Sciences, Osaka, Japan, for suppling the LPS.
2 L.I. Crawshaw and H.T. Hammel, Life Sci. *10*, 1009 (1971).
3 L.I. Crawshaw and H.T. Hammel, Brain Behav. Evol. *7*, 447 (1973).
4 L.I. Crawshaw and H.T. Hammel, Comp. Biochem. Physiol. *47A*, 51 (1974).
5 S.B. Strømme, K. Myhre and H.T. Hammel, J. Physiol., Paris *63*, 433 (1971).
6 L.I. Crawshaw, H.T. Hammel and W.F. Garey, Science, N.Y. *181*, 579 (1973).
7 M. Iriki, S. Murata, M. Nagai and K. Tsuchiya, Comp. Biochem. Physiol. *53A*, 61 (1976).
8 M. Nagai and M. Iriki, Comp. Biochem. Physiol. *57A*, 417 (1977).
9 M. Nagai, M. Iriki and K.S. Iwata, J. exp. Biol. *68*, 89 (1977).
10 W.W. Reynolds, M.E. Casterlin and J.B. Covert, Nature *259*, 41 (1976).
11 J. Villablanca and R.D. Myers, Am. J. Physiol. *208*, 703 (1965).
12 A. Wit and S.C. Wang, Am. J. Physiol. *215*, 1160 (1968).
13 J.S. Eisenman, Am. J. Physiol. *216*, 330 (1969).
14 D. Myers, T.A. Rudy and T.L. Yaksh, Neuropharmacology *10*, 775 (1971).
15 K.S. Iwata and H. Fukuda, in: Response of Fish to Environmental Changes, p. 316. C.C. Thomas, Springfield 1973.
16 J.R.S. Hales, J.W. Bennett, J.A. Baird and A.A. Fawcett, Pflügers Arch. *339*, 125 (1973).
17 W. Feldberg and P.N. Saxena, J. Physiol., Lond. *249*, 601 (1975).
18 K. von Frisch, Biol. Zbl. *31*, 236 (1911).
19 D.C. Smith, J. exp. Zool. *52*, 183 (1928).

Organization of the mammalian red nucleus and its interconnections with the cerebellum

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Summary. The red nucleus in monkeys and rats consists of a magnocellular, rubrospinal portion which receives its cerebellar information from the nucleus interpositus, and a parvocellular, rubroolivary portion which receives cerebellar afferents from the nucleus lateralis. Distinct interpositorubrospinal and dentatorubroolivary projections are therefore common to these 2 species.

Although the interconnections of the mammalian red nucleus (RN) and the cerebellum have been the subject of investigation for many years, certain details of their organization have only recently become apparent. In the cat, a somatotopically organized projection from the anterior interposed nucleus (NIA) terminates within all but the rostralmost portion of the RN[1,2] while the lateral (dentate) nucleus (NL) projects only to the rostral area which is devoid of NIA input[2,3]. The RN of the cat is not well suited to a study of the functional organization of this centre, however, because it is not clearly divided into magnocellular and parvocellular portions[4], i.e. large and small cells are interspersed throughout most of its rostrocaudal extent. It was therefore undertaken to study the organization and connections of the RN in the monkey and the rat in which this centre is more clearly polarized[5-8] and to compare their

organization with the same pathways in the cat. Discrete electrolytic lesions were placed selectively into the deep cerebellar nuclei and the distribution of degeneration was examined in Fink-Heimer[9] and Nauta-Laidlaw[10] preparations. In both species a similar pattern of organization emerged. The NL projected exclusively to the rostrally situated, small-celled (parvocellular) part of the nucleus (figure 1, a), while the NIA terminated within the caudally located, large-celled (magnocellular) region (figure 1, b). Lesions within the rostral portion of the NIA gave rise to degeneration within the ventrolateral part of the RN (hindlimb area), whereas lesions further caudally situated within the NIA caused degeneration within the central and medial (trunk and forelimb) parts of the RN. These findings were pursued further in the rat, using similar methods, to establish the efferent connections of the magnocellular and parvocellular RN respectively. Lesions within the caudally situated, magnocellular portion of the RN were followed by degeneration throughout the contralateral rubrospinal tract and in various structures at bulbar levels (e.g. facial nucleus, lateral reticular nucleus). Lesions of the rostral, parvocellular portion brought about degeneration ipsilaterally in the region of the central tegmental tract and in both dorsal and ventral lamellae of the principal inferior olive. This divergence of rostral and caudal projections was further substantiated by placing surgical lesions within the rubrospinal tract at cervical levels of the spinal cord. Following this procedure, the large neurons located throughout the magnocellular RN were found to undergo marked central chromatolysis, whereas the parvocellular cells remained unaffected. A similar pattern of organization in rubral efferents has been found in the monkey[11,12]. Thus, in both the rat and the monkey the NIA projects to the magnocellular rubrospinal part of the RN while the NL provides a synaptic input to the parvocellular, rubroolivary portion. Although the overall organization of the cat RN is somewhat different from that of the rat or the monkey, it is likely that a similar pattern exists in that species as well. No

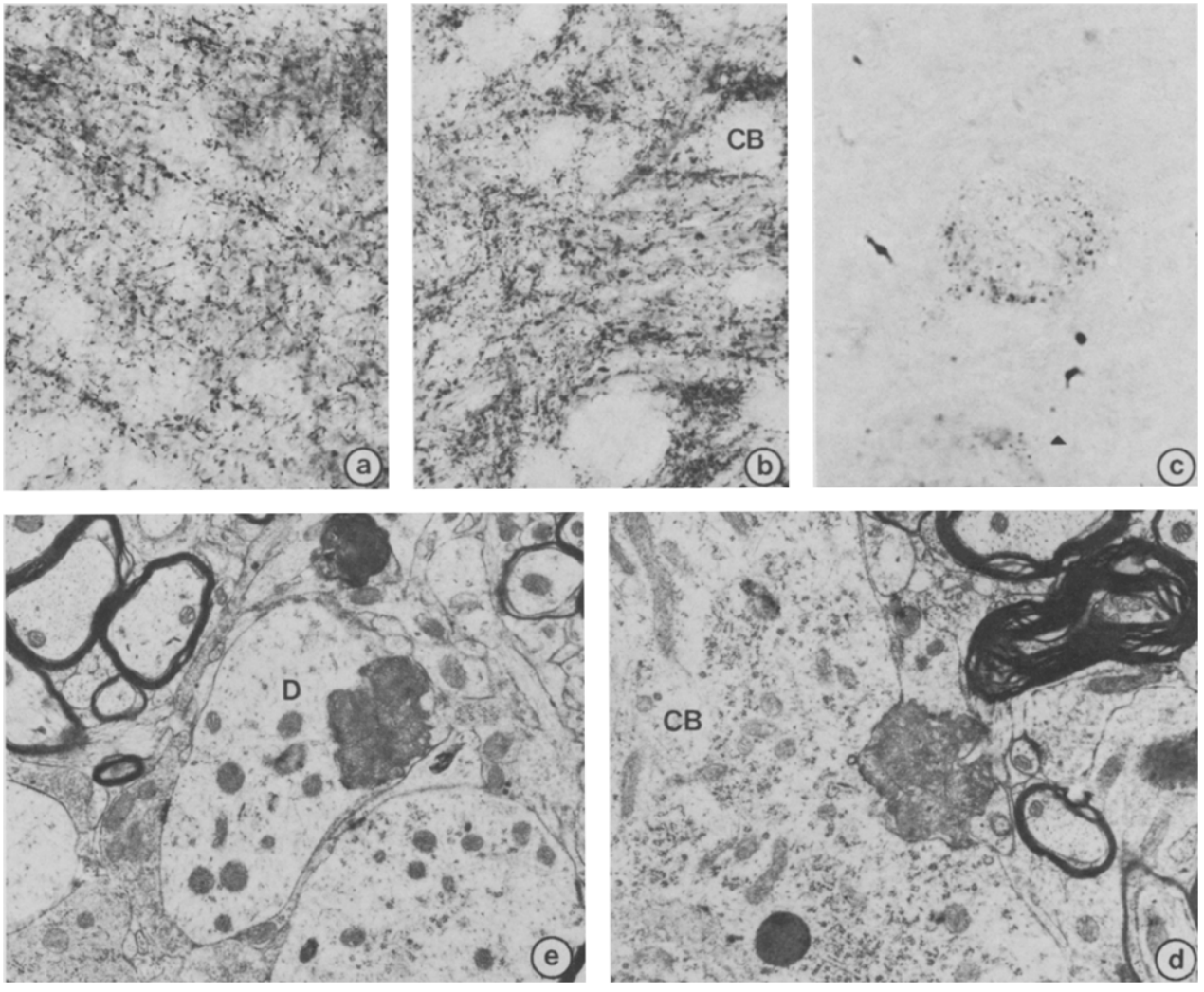

Fig. 1. *a* Parvocellular red nucleus containing fine preterminal and terminal degeneration throughout the neuropil following a lesion within the nucleus lateralis of the cerebellum. Fink-Heimer technique. ×0. *b* Magnocellular red nucleus containing abundant axonal debris around the cell bodies (CB) and proximal dendrites and throughout the neuropil following a lesion within the nucleus interpositus anterior. ×0. *c* Retrogradely labelled cell body of a large neuron in the nucleus interpositus anterior following injection of HRP into the red nucleus. ×0. *d* Synaptic terminal undergoing dark degeneration in the magnocellular red nucleus following interruption of the brachium conjunctivum. The degenerating terminal is comparable in size to that in *c* but lies in contact with a cell body (CB). ×,000. *e* Synaptic terminal undergoing dark degeneration in the parvocellular red nucleus following interruption of the brachium conjunctivum. The degenerating terminal is small in size (1–2 µm diameter) and is in contact with a small dendrite (D). ×,000.

well defined magnocellular red nucleus occurs in the cat, but it has been established that the portion of the RN which gives rise to the rubrospinal tract is in receipt of NIA terminals and is devoid of input from the NL[2-4].

Similarly, the rubroolivary projection arises from the rostralmost region of the RN which is the site of termination of NL fibres[2,3,13]. The only other species for which the details of rubral connections are well known is the North American opossum. The work of Martin and King and coworkers has shown that the RN in that species also possesses an underlying organization which agrees favourably with the data on the animals described above[14,15]. This apparent difference in the source of cerebellar input to magnocellular and parvocellular parts of the RN was investigated further by electron microscopy following placement of electrolytic lesions within the brachium conjunctivum of the rat. Degenerating synaptic terminals within the magnocellular RN were observed primarily on cell bodies and proximal dendrites (figure 1, d) whereas degenerative terminals in the parvocellular RN were found only on smaller dendritic profiles (figure 1, e). This evidence indicates that the mode of input from the cerebellum is different in the magnocellular and parvocellular RN. Such a difference may be interpreted as further evidence that the 2 subdivisions of the RN receive afferents from functionally distinct parts of the cerebellum. The posterior portion of the interposed complex (NIP) has also been proposed as the origin of a small pathway to the RN[16]. To investigate the presence of such a pathway in the rat, the horseradish peroxidase (HRP) tracing method was used[6,17]. Following injection of HRP into the red nucleus, cell bodies labelled with retrogradely transported enzyme were observed throughout the contralateral NL and NIA (figure 1, c). A small proportion of cells within the NIP were also found to possess granular reaction product characteristic of HRP activity. These results indicate that a projection from the NIP to the RN does occur in the rat but that it is much less extensive than the NIA-RN pathway. A projection from the RN back to the NIA, arising as collaterals of the rubrospinal tract, has also been described[18]. However, Eccles et al. have shown that only 5% of the rubrospinal axons studied in their investigation actually sent collaterals to NIA[19]. It is therefore unlikely that this pathway plays a significant role in cerebellorubral interaction.

The findings of the experiments described herein reveal a pattern of organization within the RN of the monkey and the rat that is very likely common to mammals generally (figure 2). The RN participates in 2 spatially and functionally distinct projections, an interpositorubrospinal pathway and a dentatorubroolivary pathway. Physiological investigations have shown that the rubrospinal tract has an excitatory action on contralateral flexor muscles and an opposite and inhibitory effect on extensor muscles[20]. The magnocellular RN therefore plays a role in adjusting the level of activity of different flexor muscles and in facilitating muscle tone. The parvocellular portion, through its olivary connections, most likely plays a role in feedback control of the neocerebellar system. This difference in the function of magnocellular and parvocellular RN is also reflected in the differing roles played by their afferent cerebellar systems. NL is believed to function in the preprogramming of movement whereas NIA is involved in the updating of ongoing movement. It is not surprising, therefore, that the magnocellular, rubrospinal portion of RN is in receipt of NIA fibres, whereas the parvocellular part, whose connections are suprasegmental, receives its cerebellar information from NL.

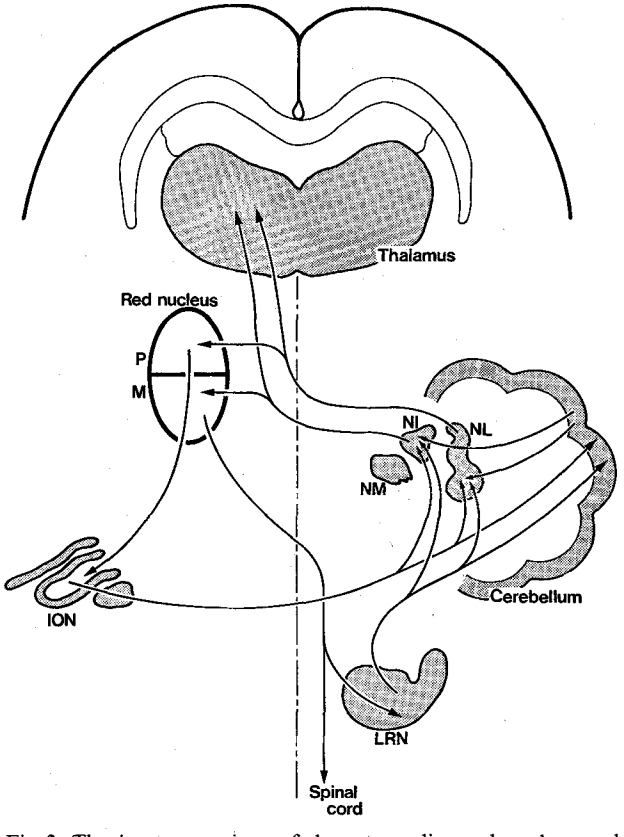

Fig. 2. The interconnections of the mammalian red nucleus and associated structures. P, parvocellular; M, magnocellular; NL, nucleus lateralis; NI, nucleus interpositus; NM, nucleus medialis; ION, inferior olivary nucleus; LRN, lateral reticular nucleus.

1 J. Courville, Exp. Brain Res. *2*, 191 (1966).
2 L.P. Renaud and J. Courville, Anat. Rec. *163*, 249 (1969).
3 F. Walberg, J. comp. Neurol. *104*, 77 (1956).
4 O. Pompeiano and A. Brodal, J. comp. neurol. *108*, 225 (1957).
5 K.A. Caughell and B.A. Flumerfelt, J. comp. Neurol. *176*, 295 (1977).
6 B.A. Flumerfelt and K.A. Caughell, Exp. Neurol. *58*, 95 (1978).
7 B.A. Flumerfelt, S. Otabe and J. Courville, Brain Res. *50*, 408 (1973).
8 J.M. Reid, D.G. Gwyn and B.A. Flumerfelt, J. comp. Neurol. *162*, 337 (1975).
9 R.P. Fink and L. Heimer, Brain Res. *4*, 369 (1967).
10 W.J.H. Nauta, in New Research Techniques of Neuroanatomy, p. 17. Ed. W.F. Windle. Thomas, Springfield, 111. 1957.
11 J. Courville and S. Otabe, J. comp. Neurol. *158*, 479 (1974).
12 R.A. Miller and N.L. Strominger, J. comp. Neurol. *152*, 327 (1973).
13 F. Walberg, J. comp. Neurol. *104*, 77 (1956).
14 J.S. King, R.M. Dom, J.B. Conner and G.F. Martin, Brain Res. *52*, 61 (1973).
15 G.F. Martin, R. Dom, S. Katz and J.S. King, Brain Res. *78*, 17 (1974).
16 P. Angaut, Brain Res. *24*, 377 (1970).
17 J.H. LaVail, K.R. Winston and A. Tish, Brain Res. *58*, 470 (1973).
18 J. Courville and A. Brodal, J. comp. Neurol. *126*, 471 (1966).
19 C. Eccles, P. Scheid and H. Táborikovà, J. Neurophysiol. *38*, 947 (1975).
20 J. Massion, Physiol. Rev. *47*, 383 (1967).