Gating of cutaneous input to cerebellar climbing fibres during a reaching task in the cat

Richard Apps*†, Melanie J. Atkins* and Martin Garwicz*‡

*Department of Physiology, University of Bristol, UK and ‡Department of Physiology and Neuroscience, Lund University, Sweden

1. Task-dependent modulation of cutaneous input to climbing fibres projecting to the C1, C2 and C3 zones in the cerebellar paravermal lobule V was investigated in awake cats during performance of a reaching task.

2. Climbing fibre responses resulting from low intensity (non-noxious) electrical stimulation of the ipsilateral superficial radial nerve were recorded as extracellular field potentials in the cerebellar cortex using chronically implanted microwires.

3. Response size, measured as the time–voltage integral of the evoked field potential, was assessed during three phases of the reaching movement, *reaction*, *reach* and *grasp*, and compared with the response size at rest.

4. At C1 and C3 zone recording sites response size was usually reduced during the task (7/10 sites). The reduction was most pronounced in the grasp phase, occasionally accompanied by a smaller reduction in the reach and reaction phases. In one case an enhancement was found in the reach phase.

5. Response size was also modulated during the task at four of six C2 zone recording sites. However, the results were mixed. In three cases the modulation resembled the pattern at C1/C3 sites with the responses being reduced in the grasp phase accompanied on occasion by a lesser reduction in the reach phase. In the remaining case there was an enhancement during grasp. In this case and one other there was also an enhancement during the reaction phase.

6. The findings indicate significant gating of cutaneous input to climbing fibres projecting to the C1, C2 and C3 zones during reaching movements, while the variability between recording sites suggests functional differences, both between and within zones.

The role of the cerebellum in motor control is well defined by the neurological symptoms associated with cerebellar lesions, which involve disturbances of motor co-ordination that affect posture, gait and voluntary movements. By contrast, the understanding of *how* the cerebellum accomplishes its functions is rather fragmentary and the specific roles of the major types of afferents, in particular the olivocerebellar climbing fibres, remain enigmatic (see Ito, 1984).

A key to unravelling the function of the climbing fibre system, and probably also the mode of operation of the cerebellar neuronal circuitry, lies in defining the type of information that is conveyed to the inferior olive (see recent review by Simpson, Wylie & De Zeeuw, 1996). To date, this has been difficult in the context of limb movement control, since there is an abundance of negative findings in the literature concerning the conditions for activation of climbing fibres during movement in awake animals (see Simpson *et al.* 1996), while positive findings are relatively scarce (see Discussion). In light of the high sensitivity of inferior olivary neurones and climbing fibres to peripheral stimulation in the awake but passive animal (Gellman, Gibson & Houk, 1985; Andersson & Armstrong, 1987) it seems likely that the transmission of peripheral input to the inferior olive is modulated by task-dependent mechanisms, and several recent studies indicate that this is indeed the case (Gellman *et al.* 1985; Andersson & Armstrong, 1987; Apps, Lidierth & Armstrong, 1990; Lidierth & Apps, 1990; Apps, Hartell & Armstrong, 1995*b*; Horn, van Kan & Gibson, 1996).

Given the high degree of multi-modal convergence on inferior olivary cells (e.g. Jörntell, Garwicz & Ekerot, 1996), a possible role for task-dependent modulation of input could be to allow the climbing fibre system to make selective use of information from different types of peripheral receptors

† Authors' names are in alphabetical order.

depending on the requirements of the particular motor task. A similar principle of modality-specific modulation of sensory transmission has previously been suggested for motor systems in general (Prochazka, 1989). Based on the assumption that defining the type of input that is selected in a given context will provide important clues to the function of the olivocerebellar system, the present study represents a step towards characterizing task-dependent modulation of input to climbing fibres projecting to the paravermal cerebellar cortex.

The paravermal cortex is divisible into three sagittal zones, from medial to lateral termed C1, C2 and C3 (for a review see Voogd & Bigaré, 1980). The C1 and C3 zones receive similar climbing fibre input from the ipsilateral limbs and both project to nucleus interpositus anterior, while the C2 zone receives bilateral climbing fibre input and projects to nucleus interpositus posterior (cf. Oscarsson, 1980; Trott & Armstrong, 1987). Both the paravermal cortex and its efferent nuclei have been implicated in the control of reaching movements (e.g. Harvey, Porter & Rawson, 1977; MacKay, 1988; Thach, Goodkin & Keating, 1992; Kitazawa, Goto & Urushihara, 1993; van Kan, Houk & Gibson, 1993) and therefore the behavioural task selected for the present study was a forelimb reaching movement. The input examined was mainly, if not exclusively, that of Aβ type cutaneous afferents within the superficial radial nerve which mediate low-threshold mechanoreceptive information from the forepaw.

Our findings indicate that cutaneous tactile input to climbing fibres projecting to the paravermal C1, C2 and C3 zones is substantially modulated during the performance of a reaching task. At most recording sites the modulation consisted mainly of a progressive reduction in the response size of evoked climbing fibre field potentials during the course of the reaching movement, demonstrating a gating of afferent input to cerebellar climbing fibres. However, variations on this theme were common. For example, at two of six C2 zone sites there was an increase in response size around onset of the movement.

Preliminary results have been reported (Apps, Atkins & Garwicz, 1995*a*).

METHODS

Animals and implants

Experiments were performed on three purpose-bred adult male cats (4–5 kg) in accordance with UK Home Office guidelines. Following behavioural training (see below) implantations were carried out at an aseptic operation under sodium pentobarbitone anaesthesia (Sagatal, BDH; 40 mg kg^{-1} I.P., maintenance doses of 1·5 mg kg^{-1} as required). Throughout the operation a surgical level of anaesthesia was maintained, characterized by general muscle atonia except in respiratory muscles, strongly or completely depressed withdrawal reflexes and slow, regular breathing. A single dose of atropine (0·5 ml S.C.) was given in order to prevent excessive secretion in respiratory passages and an antibiotic (Crystapen, Britannia Pharmaceuticals, Redhill, UK) was administered pre- and post-operatively (4 spaced doses of 75 mg kg^{-1}). Throughout the operation the temperature of the animal was kept within physiological limits.

A small craniotomy was made to expose the paravermal part of cerebellar lobule V and the boundaries of the C1, C2 and C3 zones on the cerebellar surface were identified electrophysiologically (e.g. Trott & Armstrong, 1987). An array of seventeen to twenty-one fine flexible platinum–iridium microwires, Teflon insulated except at the tip and 25 μm (Goodfellow, Cambridge, UK) or 75 μm (Advent, Halesworth, UK) in total diameter were inserted to a depth of 1–2 mm into the tips of the folia and the point of insertion of each wire was indicated on a scale drawing. Prior to sealing the wires in position by closing the skull defect with dental acrylic cement, the cerebellar surface was covered with gelfoam (Sterispon; Allen and Hanburys, London, UK). For further details of microwire implants and the advantages of microwires compared with conventional microelectrodes see Apps *et al.* (1995*b*).

Two pairs of bipolar cuff electrodes, for stimulation and nerve volley recording, respectively, were implanted around each of the left and right superficial radial (SR) nerves. A unipolar lead was implanted subcutaneously into the distal forelimb ipsilateral to the craniotomy to allow monitoring of paw lift-off during reaching. Post-operative analgesia was maintained for 24 h with buprenorphine (Temgesic, Reckitt and Coleman; 10 mg kg^{-1} I.M.). No complications occurred post-operatively and the animals showed no signs of discomfort at any stage of the experiment. Upon termination of the experiment (*ca* 6 weeks after the initial operation) animals were killed by an overdose of anaesthetic, the cerebellum was removed and the folial location of the microwires (in lobule Va–c) was verified by inspection of the dorsal surface of the cerebellum. For further details of implants see Apps *et al.* (1990).

Behavioural task

The behavioural protocol was similar to that introduced by Sybirska & Górska (1980). Prior to the operation, cats were trained non-aversively (*ca* 6 weeks) to reach for and retrieve a piece of fish or turkey from a horizontally oriented Perspex tube (55 mm long, 30 mm wide) placed in front of the animal at approximately shoulder height, as shown in Fig. 1*A*. The cue to reach with the ipsilateral forelimb was the opening of a door (*a*) in front of the tube (*c*). Recording sessions usually started about 3–4 days after surgery with normally one each day (occasionally two). During each session, low-intensity electrical stimuli were applied to the ipsilateral SR nerve both during task performance (usually about 70 stimuli) and during periods of rest (usually about 50 stimuli). Data during rest were obtained either immediately before or after collection of task data. Throughout the sessions the animals were sitting down, lightly restrained by a loosely applied harness, and showed no signs of discomfort. At the end of the recording session the animal was returned to its pen (shared with other purpose-bred cats), where additional food was freely available.

Recordings and stimulation

As indicated in Fig. 1*B*, door opening (*a*), paw lift-off from the ground (*b*) and paw entry into/exit from the tube (*c*) were continuously monitored. The two latter records were obtained by using a high-frequency carrier signal applied to a contact plate (*b* in Fig. 1*A*) and a photo-electric switch at the tube mouth, respectively. Shocks to the ipsilateral SR nerve were delivered as a square pulse (100 μs) at intensities 2–4T, where T is the threshold for the most excitable fibres in the nerve. At the start of every recording session, T was defined as the lowest stimulus strength that gave rise to a consistent (> 50% of stimulus presentations) deflexion at the appropriate latency on the nerve volley trace, as determined by

visual inspection of single sweeps. Only one stimulus was delivered for each performance of the task. The timing of that stimulus was 'pseudo-randomized' so that, over a whole recording session, data were collected from stimuli that were distributed evenly throughout the task (cf. Fig. 5). At rest, stimuli were delivered at regular intervals (0·7 Hz). Throughout all recording sessions the afferent volley in the nerve was monitored. Stimulation of the nerve contralateral to the cerebellar recording sites was used to facilitate identification of the C2 zone, which receives bilateral climbing fibre input (cf. Oscarsson, 1980). Filter settings for recording of field potentials and the nerve volley were, respectively, 30 Hz to 2·5 kHz and 300 Hz to 10 kHz. All data were stored on digital audio tape (DAT) for off-line analysis.

Data analysis

The biological signals were digitized by customized trigger-sampled software running a CED1401 (Cambridge Electronic Design) A/D converter. The sampling rates for the field and nerve were 2·5 kHz and 20 kHz, respectively. The size of individual climbing fibre field potentials was measured by integration (mV ms; see Apps *et al.* 1990) of the initial component of the response (duration typically *ca* 5 ms), whereas the amplitude of individual nerve compound action potentials was measured peak to peak, as shown in Fig. 1*C*. The sizes of climbing fibre field potentials evoked during each of three phases of the task, *reaction* (1), *reach* (2) and *grasp* (3), were compared with the size of those evoked at rest and the ratio was taken as an index of modulation of afferent transmission to climbing fibres at the site under study. The phases were defined as the intervals between (1) door opening and paw lift-off, (2) paw lift-off and tube entry and (3) tube entry and tube exit (Fig. 1*B*). Only trials with a complete sequence of the three phases of the movement preceded by a minimum of approximately 3 s of foot contact were included in the analysis.

Note that in the present experimental set-up, motor activity of the forelimb starts during the reaction phase. Paw lift-off, which was used to indicate the transition between reaction and reach phases, is preceded by muscle contractions and movement, especially in proximal segments of the limb.

Statistical analysis

The climbing fibre responses for each recording session were displayed as bar charts showing response mean size (+ S.E.M.) for each phase of the task, normalized relative to rest (e.g. Figs 4 and 7). For the purpose of statistical analysis, comparisons were made between the size of climbing fibre responses at rest and each of the three phases of the task. Multiple regression (analysis of variance) was used so that any fluctuations in the size of the nerve volley were also taken into account. Linear (least-squares) regression analysis was also carried out to assess whether any trends in size of individual climbing fibre responses occurred within different phases of the task (e.g. Fig. 5).

RESULTS

Climbing fibre responses evoked on stimulation of the ipsilateral SR nerve were recorded as extracellular field potentials usually with a mainly positive wave configuration (i.e. with the microwire recording tip in the granular layer, e.g. Fig. 3), but sometimes with a mainly negative wave configuration (i.e. with the microwire recording tip in the molecular layer, e.g. Fig. 6). These potentials were readily distinguishable from potentials related to mossy fibre input based on their latencies and fluctuations in amplitude over time which were characteristic of climbing fibre input. At eleven of sixteen sites (see below), the potentials were also subjected to a 'paired pulse test', and a ~100 ms refractory period was used for identification of climbing fibre activity (Armstrong & Harvey, 1968). On the grounds of the stimulus intensities used (2–4*T*) and latencies of the evoked climbing fibre field potentials, it is concluded that the low-threshold cutaneous input described in the present report was mainly, if not exclusively, in the Aβ range (cf. Ekerot, Gustavsson, Oscarsson & Schouenborg 1987; Ekerot, Garwicz & Schouenborg, 1991*b*).

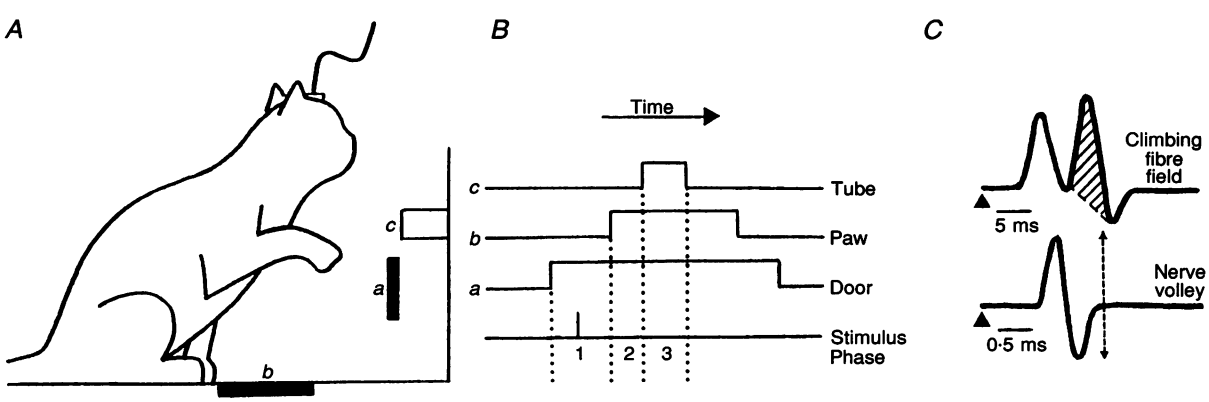

Figure 1. Schematic diagram of experimental arrangements

A, cat performing the reaching task. *a*, tube door; *b*, contact plate; *c*, tube. *B*, task cue records. Upward deflection of traces represents door opening (*a*), paw lift-off (*b*) and tube entry (*c*). Reaction (1), reach (2) and grasp (3) phases are defined. In this example, the stimulus was delivered during the reaction phase. *C*, typical form of biological signals. Top trace, cerebellar field potential. Climbing fibre response shaded to indicate area measured in each sweep. Lower trace, nerve volley. Dashed arrow indicates peak-to-peak measurement. Arrowheads indicate nerve stimulation. Note different time scales.

Although an array of up to twenty-one microwires was implanted in each animal (see Methods) only a total of twelve (~20%) yielded discriminable field potentials, presumably because the majority were not favourably placed relative to the cortical layers. Best results were obtained from microwires with an impedance of ~0·35 MΩ. Ipsilateral, short-latency (11·0–14·4 ms) responses and bilateral, long-latency (18·0–20·9 ms) responses were taken to be related to C1 or C3 and C2 zones, respectively (Oscarsson, 1980). The climbing fibre field potentials recorded from six of the wires were classified as exclusively C1 or C3 zone responses and from two of the wires as exclusively C2 zone responses, while both types of potentials were recorded from the remaining four wires, thus yielding a total of sixteen *recording sites*. Note that although C2 zone responses were sometimes recorded from the same microwires as C1 or C3 zone responses this should not be taken as evidence for an incomplete anatomical segregation of the zones. Most probably, this is simply a reflection of the rather low impedance of the microwires in combination with their placement close to zonal boundaries.

Since no systematic differences were found between C1 ($n = 7$) and C3 ($n = 3$) zone responses, they will be considered together. Furthermore, given the limited number of C1 and C3 zone recording sites, no attempt was made to seek differences between medial and lateral subzones (cf. Trott & Apps, 1991). Ten of the sixteen recording sites were studied on more than one occasion (on average, ~2 recording sessions per site), giving a total of thirty-four *recording sessions* (23 from the C1/C3 zones and 11 from the C2 zone). For any given recording site, the different recording sessions were performed on separate days (range 1–20 days apart) and in each session a complete set of data was collected, including rest and the three phases of the movement (cf. Figs 4 and 7). In accordance with a previous report by Apps *et al.* (1995*b*), at any given site the task-related changes in response size were found to be relatively stable between recording sessions, although the depth of modulation was influenced by differences in stimulus intensity between sessions (see later section).

Variations in size of afferent volley in relation to fluctuations of cerebellar responses

Fluctuations in response size on consecutive presentations of an unchanging stimulus are characteristic of climbing fibre field potentials and were observed when the animals were sitting quietly, in the resting condition (e.g. Fig. 5*A*). However, during performance of the task it is possible that

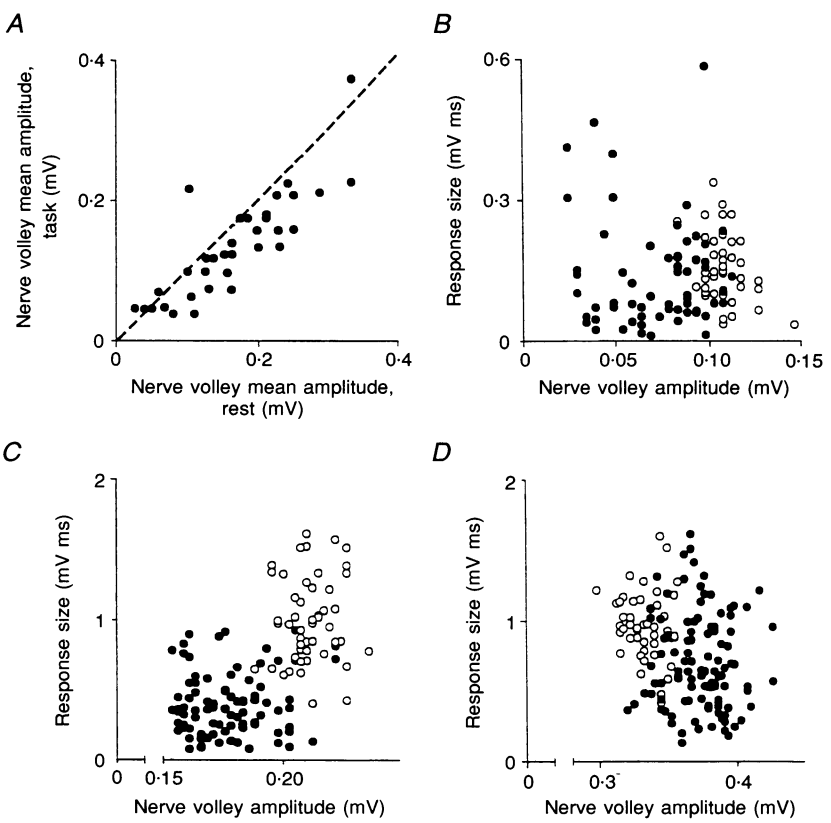

Figure 2. Variations in afferent nerve volley amplitude

A, nerve volley mean amplitude during task performance as compared with rest ($n = 34$). Dashed line indicates equal values. *B–D*, relationship between nerve volley amplitude and climbing fibre field potential size during three recording sessions. The recording sessions were selected as having the smallest (*B*) and largest positive (*C*), and largest negative (*D*) correlation coefficients r ($r = 0·00$, 0·60 and −0·28; $P > 0·05$, $P < 0·05$ and $P < 0·05$, respectively). ○, rest; ●, task.

movement of the stimulating electrodes might also occur causing changes in stimulus efficacy which could in turn induce additional fluctuations in size of the cerebellar responses. It was important therefore to establish if stimulus-evoked afferent input during task performance differed from that during rest and, if any variations were found, to what extent they were likely to contribute to fluctuations of the climbing fibre responses.

The plot in Fig. 2*A* shows the relationship for all thirty-four recording sessions between nerve volley mean amplitude at rest and during task performance (all phases of the task pooled). There was a general tendency for the nerve volley mean amplitude to be smaller during the task compared with rest. For each recording session scatterplots of the size of individual climbing fibre field potentials plotted as a function of afferent nerve volley amplitude during rest and task performance were also constructed and in each case a linear regression correlation coefficient r was calculated (range for r, −0·28 to 0·60). In Fig. 2*B*–*D*, example scatterplots from recording sessions with the smallest (*B*) and the largest positive (*C*), and largest negative (*D*) correlations are shown. Filled and open circles represent responses resulting from individual stimulus presentations during task performance and rest, respectively. A similar analysis was carried out using only data obtained during task performance (not shown; range for r, −0·24 to −0·62).

Overall, the scatterplots suggest that the size of the nerve volley may, in some cases, differ between rest as compared with performance of the task. Moreover, there may also be a systematic variation in size of the nerve volley related to different phases of the task. In light of this possibility, in the following presentation of data in which the size of climbing fibre responses at rest and during task performance are compared, the size of the nerve volley is also taken into account when statistical comparisons are made (see Methods for details).

Modulation of climbing fibre response size during reaching movements

C1/C3 zone recording sites. Superimposed single sweeps and averaged waveforms from the resting condition and the three phases of the task for a C1/C3 zone recording site are shown in Fig. 3. In this example, the C1 zone climbing fibre

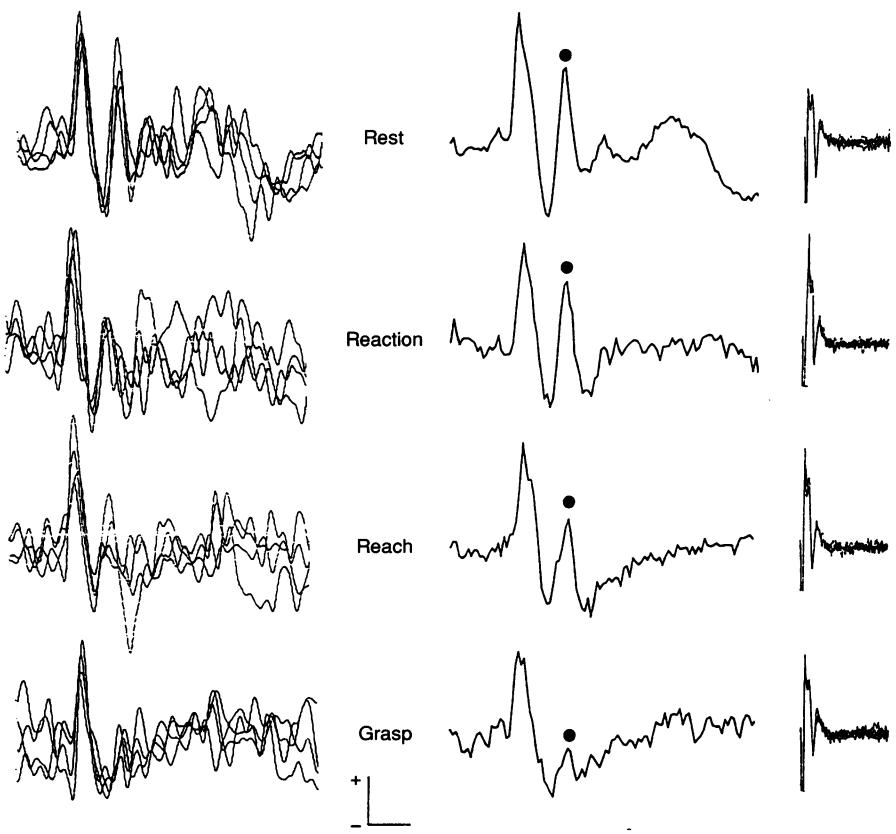

Figure 3. Examples from a recording session of C1 zone climbing fibre field potentials

From top to bottom, field potentials recorded in the granular layer during rest as compared with the three phases of the reaching task. Left panels, superimposed single sweeps ($n = 4$). Centre panels, averaged cerebellar waveforms ($n = 49$, 23, 14 and 34, respectively). Right panels, superimposed single sweeps of nerve volley ($n = 4$). ●, climbing fibre response. Note earlier positive-going potential attributable to input from short latency mossy fibre pathways. Scale bar: 0·1 mV, 5 ms for cerebellar responses and 0·04 mV, 1 ms for nerve volley records. Stimulus delivered at start of all records.

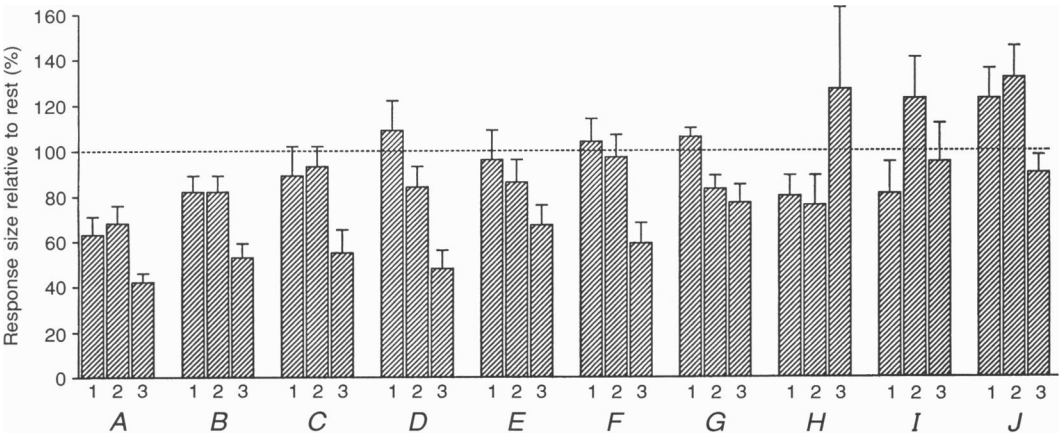

Figure 4. Summary of all C1 and C3 zone climbing fibre recording sites

A–J, for each of ten recording sites example histograms indicate response mean size relative to rest for the three phases of the reaching task (+ S.E.M.). 1, reaction; 2, reach; 3, grasp. Horizontal dashed line indicates response size at rest (100 %).

field potential (see filled circles in average middle traces) is preceded by a large potential with an onset latency of ~7 ms, which is attributable to mossy fibre input. Potentials related to mossy fibre input were not analysed in detail, although a preliminary assessment indicates that they generally exhibited a task-related modulation in size that was broadly similar in pattern to the modulation of the accompanying climbing fibre response. However, the depth of the modulation was usually less pronounced (particularly in the example illustrated in Fig. 3).

By comparison with the mossy fibre-related potential, the size of the climbing fibre field potential was clearly reduced during the task as compared with rest, notably during reach and especially during grasp. In fact, single climbing fibre responses during grasp (see superimposed sweeps at left of Fig. 3) were often reduced below the level of detection, despite the relative constancy of the nerve volley as shown to the right.

To assess task-dependent modulation of afferent transmission to climbing fibres the mean size of the response during the different phases of the task was displayed relative to the mean size of the response during rest. In Fig. 4, histograms representing the example case (Fig. 4*D*) and all other C1 and C3 zone responses during reaction, reach and grasp are shown. For recording sites studied on more than one occasion the recording session with the least variation in nerve volley size is presented, while further reference to other recording sessions is made later. In order to facilitate comparison between histograms the response sizes were normalized with respect to the mean rest value for each recording session (100 %; dashed line) and are presented in a sequence from most to least substantial modulation. The number of stimulus presentations per phase in the ten illustrated recording sessions ranged from 7 to 87, 8 to 36 and 6 to 34, with mean values of 28, 23 and 21 for reaction, reach and grasp, respectively. In the resting condition the range was 16–71 and the mean was 53 stimulus presentations.

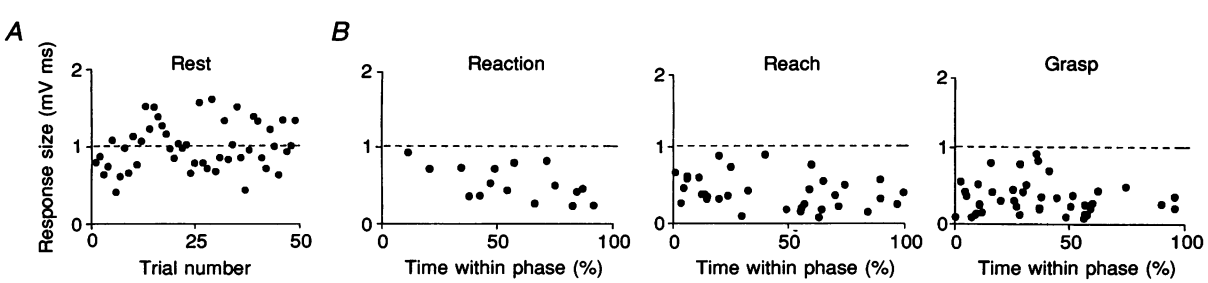

Figure 5. Distribution of response size resulting from individual stimulus presentations

From left to right for one example recording session. *A*, response size as a function of time during rest and *B*, during the three phases of the reaching movement. Due to variability in the duration of different phases, time scales in scatterplots representing task performance were normalized. Horizontal dashed lines indicate the mean size of response at rest (~1 mV ms).

During one or more phases of task performance, a statistically significant reduction of response size relative to rest was observed for seven of the ten C1/C3 zone recording sites investigated (Fig. 4*A–G*, also the case shown in *J* in the grasp phase, but not in the session selected for illustration; multiple regression, $P < 0{\cdot}05$). C1 zone ($n = 7$) and C3 zone ($n = 3$) recording sites did not differ systematically with respect to the presence or absence of modulation. Whenever reduction of response size was observed it was most pronounced in the grasp phase. Also, when all recording sessions are considered, smaller but statistically significant reductions during the reaction and reach phases were evident for three of ten and four of ten recording sites, respectively (multiple regression, $P < 0{\cdot}05$; see also Fig. 8). In one case (a C3 site) a statistically significant increase was also observed in the reach phase (Fig. 4*J*, multiple regression, $P < 0{\cdot}05$).

Although the tendency for a progressive reduction in response size during the course of the task was often clear from inspection of the mean values for the three phases, there was substantial variability of response size evoked by individual stimulus presentations during task performance. Scatterplots of individual response size as a function of time during reaction, reach and grasp, with an equivalent plot for the resting condition were constructed for each site. Scatterplots derived from an example recording session and typical of the material as a whole, are illustrated in Fig. 5. Linear regression analysis was carried out in each case in order to assess possible trends in size variation *within* each of the three phases of the task. However, the correlation coefficients thus obtained were usually low and in almost all cases not statistically significant ($P > 0{\cdot}05$). More importantly, no transient changes in response size were systematically detected within phases. In addition, note in Fig. 5*A* the fluctuations in response size throughout the rest sample with no systematic change in size from the first to the last stimulus presentation, showing that repeated stimulation did not lead to any progressive increase or decrease in response amplitude. Such findings were typical and provide reassurance that there was unlikely to have been any progressive shift in microwire tip position during the recording sessions.

C2 zone recording sites. In Figs 6 and 7, C2 zone responses are presented in the same format as the C1/C3 zone responses in Figs 3 and 4. Superimposed single sweeps and averaged waveforms from the resting condition and the

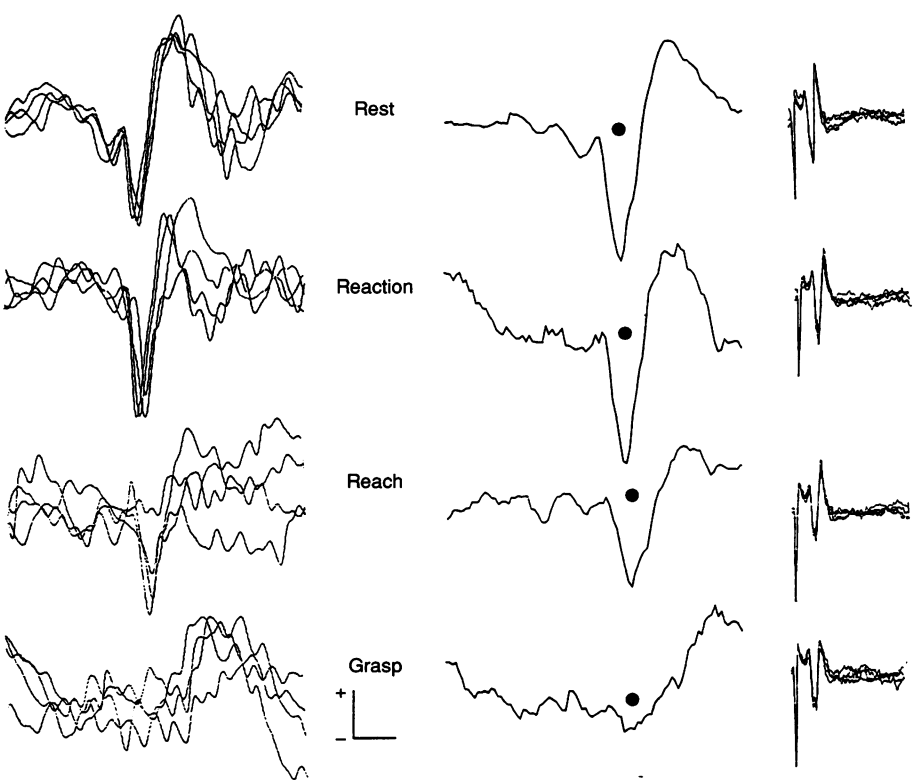

Figure 6. Examples from a recording session of C2 zone climbing fibre field potentials
From top to bottom, field potentials recorded in the molecular layer during rest as compared with the three phases of the reaching task. Left panel, superimposed single sweeps ($n = 4$). Centre panel, averaged cerebellar waveforms ($n = 49$, 7, 14 and 6, respectively). Right panel, superimposed sweeps of nerve volley ($n = 4$). ●, climbing fibre response. Scale bar 0·1 mV, 5 ms for cerebellar responses and 0·05 mV, 1 ms for nerve volley records. Stimulus delivered at start of all records.

three phases of the task for an example recording session are shown in Fig. 6. In this case, the size of the field potential (see filled circles in average middle traces) was similar in the rest and reaction phases and then decreased progressively in reach and grasp. As in the C1/C3 zone example recording session (Fig. 3), single responses during grasp were often reduced below the level of detection, as seen in the superimposed sweeps (again, despite relative constancy of the nerve volley as shown to the right). In Fig. 7 histograms of mean response size representing the example case (Fig. 7*A*) and all other C2 zone recording sites (total $n = 6$) during reaching movements are presented. For recording sites studied on more than one occasion the recording session with the least variation in nerve volley size is shown. The number of stimulus presentations per phase in the six illustrated recording sessions ranged from 7 to 56, 8 to 37 and 6 to 39, with mean values of 25, 23 and 18 for reaction, reach and grasp, respectively. In the resting condition the range was 49–66 and the mean was 52 stimulus presentations.

During task performance, statistically significant modulation of response size was observed for four of the six C2 zone recording sites investigated (multiple regression, $P < 0{\cdot}05$). In three of these cases (Fig. 7*B–D*) there was a significant reduction of response size in the grasp phase and in the one remaining case (Fig. 7*E*) a significant increase (but not in the session selected for illustration). When all recording sessions are considered, a usually smaller but statistically significant reduction in the reach phase was also observed at two of these sites (multiple regression, $P < 0{\cdot}05$). At the two other sites there was a significant *increase* during the reaction phase (in one case associated during grasp with a significant reduction and in the second by a significant increase; multiple regression, $P < 0{\cdot}05$).

Pooled data and comparisons between C1/C3 and C2 zone responses. Frequency distributions of mean response sizes for each phase, expressed as percentages of the corresponding rest value, are shown in Fig. 8*A* and *B* for all C1/C3 ($n = 23$) and all C2 ($n = 11$) zone recording sessions, respectively. Filled bars in the histograms indicate recording sessions selected for illustration in Figs 4 and 7, which were clearly representative of the material as a whole. For C1/C3 zone recording sites the distribution of response size was distinctly shifted from a modal value of 80–99% in reaction to 40–59% in grasp phase, with reach as an intermediate distribution more similar to reaction than to grasp. In addition, note that response size was smallest in the grasp phase for most (21/23) individual recording sessions. C2 zone recording sites had distributions rather similar to those of C1/C3 zone recording sites in reach and grasp. By contrast, in the reaction phase the majority (8/11) of recording sessions had responses above 100% of rest. Response size was smallest in grasp for the majority (6/11) of C2 zone recording sessions, whereas the largest responses occurred in most recording sessions (7/11) in the reaction phase.

Patterns of modulation in relation to stimulus intensity. Although the most common pattern of modulation of climbing fibre field potentials was such that response size was progressively reduced during the course of the reaching movement, variations were clearly not uncommon (Figs 4 and 7). This heterogeneity could be genuine, reflecting functional differences between recording sites, or alternatively due to other factors including differences in the intensity of nerve stimulation used for different recording sessions (2–4*T*; see Methods). Pooling of data from all C1/C3 zone recording sessions ($n = 23$) indicated that the present findings are compatible both with genuine differences between sites and with the existence of stimulus-dependent effects. On the one hand, plots of normalized mean response

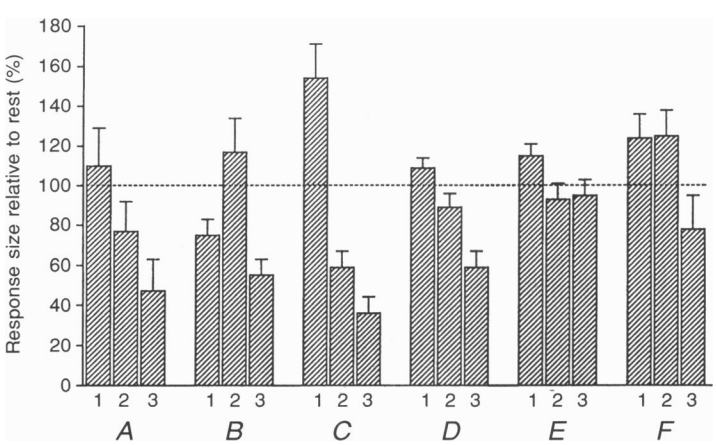

Figure 7. Summary of all C2 zone climbing fibre recording sites

A–F, for each of six recording sites example histograms indicate response mean size relative to rest for the three phases of the reaching task (+ S.E.M.). 1, reaction; 2, reach; 3, grasp. Horizontal dashed line indicates response size at rest (100%).

sizes as a function of stimulus intensity for the reaction, reach and grasp phases (not illustrated) indicated that some of the heterogeneity between recording sites remains, even when comparing the degree of modulation for the same phase and the same stimulus intensity, e.g. responses during the grasp phase evoked by a stimulus intensity of $3T$. On the other hand, some of the heterogeneity between sites was probably due to the experimental parameters used on different recording sessions, as there appeared to be an overall tendency for less reduction of response size relative to rest with increasing stimulus intensity. Specifically, when the mean values of data from all sessions at a given stimulus intensity were pooled (irrespective of phase), a rather strong stimulus intensity dependence of the modulation was evident. For T values increasing from $2T$ to $3T$ the modulation became progressively less marked, with a levelling out at values above $3T$ (not shown).

DISCUSSION

This is the first report on gating of cutaneous input to climbing fibres terminating within different paravermal cortical zones during the performance of a well-rehearsed reaching task, adding to previous studies concerned with modulation of input to the inferior olive (Horn *et al.* 1996; see also Gellman *et al.* 1985) and modulation of input to climbing fibres during locomotion (Lidierth & Apps, 1990; Apps *et al.* 1990, 1995*b*). Although not without exceptions, response size was usually progressively reduced during the course of the reaching movement, being smallest when the animal was grasping the food with the limb relatively extended. Our findings therefore indicate that transmission of input to climbing fibres projecting to the paravermal cerebellar zones is significantly modulated.

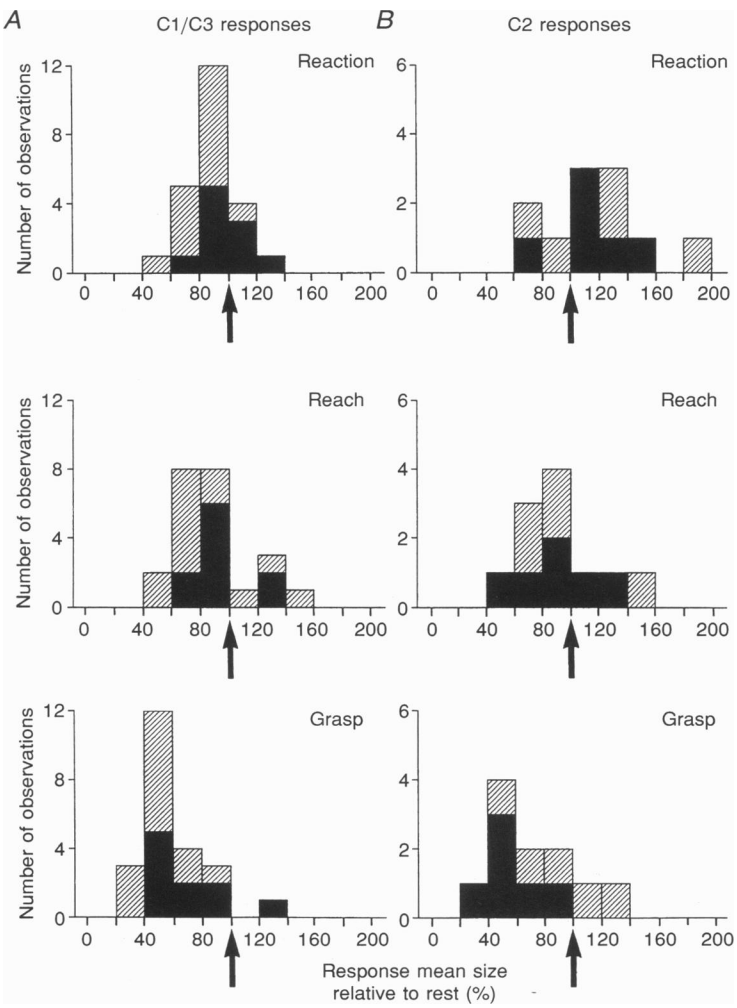

Figure 8. Frequency distribution of response size relative to rest for all recording sessions

Number of recording sessions *versus* response mean size relative to rest for the three phases of the reaching task. Note different scaling of y-axis in *A* and *B*. Arrows indicate mean size at rest (100%). *A*, responses from recording sessions ($n = 23$ in each histogram) at C1 and C3 zone sites. *B*, responses from recording sessions ($n = 11$ in each histogram) at C2 zone sites. Filled bars, data from example histograms shown in Figs 4 and 7.

Importance of monitoring the nerve volley

In the present study the nerve volley was monitored to provide an assessment of the constancy of afferent input during rest and performance of the task. A comparison between the multiple regression analysis used and an alternative analysis not taking fluctuations of nerve volley size into account (one way ANOVA) showed that changes in nerve volley size could indeed explain some of the changes in the size of the climbing fibre field potentials. This comparison indicates that the fluctuations of the nerve volley size at least partly reflected real changes in afferent input rather than simply being due to movement of the nerve recording electrode. These findings therefore highlight the importance of monitoring afferent input in behavioural studies of this kind.

Heterogeneity within zones

The C1 and C3 zones have an intricate microzonal organization, determined by highly specific olivocerebellar (Garwicz, Apps & Trott, 1996; see also Ekerot & Larson, 1979) and corticonuclear connections (cf. Garwicz & Ekerot, 1994). Different microzones have different climbing fibre receptive fields (Ekerot, Garwicz & Schouenborg, 1991*a*; Jörntell *et al.* 1996) and appear to control different groups of muscles in the limb (Ekerot, Jörntell & Garwicz, 1995). This detailed organization implies that, within the C1/C3 zones, precise function is site specific. Interpreted in this light, the heterogeneity of patterns and degree of gating observed in the present and previous studies (e.g. Apps *et al.* 1995*b*) would suggest that the gating may be tailored according to the specific role of a given microzone. Although the topography of climbing fibre input to the C2 zone has not been studied extensively, relatively detailed but restricted studies suggest that similar arguments could also hold true for this zone (cf. Garwicz, Ekerot & Schouenborg, 1992).

In a study of input to the inferior olive during a rather similar reaching task, Horn *et al.* (1996) described modulation of sensory transmission to the rostral dorsal accessory olive, which is the main source of climbing fibre input to the C1 and C3 zones. However, it is noteworthy that in their study no clear evidence was found for the existence of a microzone-specific pattern of gating.

Dependence on stimulus intensity

Our findings suggest that between nerve stimulus intensities $2T$ and $3T$, recruitment of afferent fibres can reduce the effectiveness of the gating mechanism. When present, the effect was clearest in the reaction and reach phases, while gating in the grasp phase was less sensitive to increases in stimulus intensity. A similar correlation between stimulus intensity and the degree of gating was previously reported for recording sites studied during locomotion (Apps *et al.* 1990, 1995*b*). In the present report responses at some recording sites were studied only at relatively 'high' (3–$4T$) stimulus intensities and it is possible that consistent use of weaker electrical stimuli, or indeed physiologically more appropriate natural activation of peripheral receptors, would have yielded larger relative reductions, especially in the reaction and reach phases.

The reduction of olive responses during reaching described by Horn *et al.* (1996) is in general agreement with ours. However, the degree of gating differed somewhat between the two studies in that they reported, on average, a much greater reduction of response size during task performance. In addition, for most of their sites there was a step-wise change between rest and task performance, whereas in the present study a progressive reduction in response size was the more common finding. In view of the stimulus intensity dependence of gating indicated by the present findings it is possible that the differences between the two studies may be explained by the use of somewhat different stimulus intensities. Furthermore, the possibility exists that the afferent nerve volley in their study varied in amplitude in a fashion similar to that reported here and thus contributed to the fluctuations in inferior olivary responses. However, since in the study by Horn *et al.* (1996) the afferent nerve volley was not monitored, neither of these two points can be commented upon further. As an alternative explanation it is worth noting that the differences between the two studies could instead reflect non-linearity in the input–output relationship of the inferior olive.

C1/C3 *versus* C2 recording sites

Studies of gating during locomotion have revealed substantial differences between C1/C3 zone (Apps *et al.* 1990, 1995*b*) and C2 zone climbing fibre field potentials (Lidierth & Apps, 1990) with respect to patterns of modulation in relation to the step cycle. During reaching, however, differences between patterns of modulation of C1/C3 and C2 zone responses were less pronounced. In the pooled data they were mainly restricted to the reaction phase, when responses at two of six C2 zone recording sites were significantly facilitated, while statistically significant facilitation was not found for C1/C3 zone responses (with the exception of one case during reach). By contrast, during locomotion facilitation was often found at C1/C3 sites, but only rarely at C2 sites (Apps & Hartell, 1995).

In the present study response sizes in the reach and grasp phases were rather similar in the C1/C3 and C2 zones. These similarities are not, however, necessarily incompatible with the differences previously found during locomotion. Lidierth & Apps (1990) suggested that common influences, such as a modulatory drive related to the movement of the limb(s) may act on the spino-olivocerebellar pathways innervating the two zones, and that the differences in modulation during locomotion can be explained by the fact that the C1 zone receives input from the ipsilateral forelimb only, whereas input from both forelimbs would summate for the C2 zone. In the reaching task in the present study, the exclusive use of the ipsilateral forelimb might well result in the absence of any modulatory drive on spino-olivocerebellar pathways to the C2 zone from the contralateral forelimb, and this in turn might result in a similar overall pattern of modulation for the two zones during this type of movement.

Sites and mechanisms of gating

The gating described in the present report could involve descending and/or ascending control mechanisms and could take place at any of the relays, or in fact at any combination of relays along the ascending spino-olivocerebellar pathways to the paravermal cerebellar cortex (Oscarsson, 1980; Ekerot *et al.* 1991*b*). However, the lack of evidence in the literature for movement-related reduction of 'spontaneous' climbing fibre activity (cf. Simpson *et al.* 1996; see also Horn *et al.* 1996) argues against an involvement of any inhibitory mechanism acting directly on the olive (e.g. Andersson, Garwicz & Hesslow, 1988) and would thus favour mechanisms acting at pre-olivary levels, for example at the level of the cuneate nucleus.

In fact, in a recent study aimed specifically at pinpointing possible sites and mechanisms involved in the gating of transmission through spino-olivocerebellar pathways (Lidierth, 1991), evidence was obtained for powerful 'lateral', mainly inhibitory interactions in the cuneate nucleus between input from the ulnar, median and SR nerves. By analogy, although not necessarily due to the same mechanism, inhibitory receptive fields of single climbing fibres have been described by Leicht, Rowe & Schmidt (1973). Both observations demonstrate the existence of afferent inhibitory control of transmission in the spino-olivocerebellar pathways and it is an interesting possibility that the movement-related gating is an expression of increased afferent inhibition subserving mechanisms for enhanced spatial contrast. An additional type of afferent modulation could be due to interactions between input from different modalities, e.g. skin and muscle afferents, which have been shown to have convergent input on single climbing fibres projecting to the C3 zone (Jörntell *et al.* 1996).

Since modulation of climbing fibre response size occurred during a reaching movement and similar modulation has previously been described during locomotion (Apps *et al.* 1990, 1995*b*; Lidierth & Apps, 1990), the data suggest that gating is a movement-dependent phenomenon. However, it cannot be excluded that the modulation is instead, or at least in part, simply dependent on the position of the limb (see also Horn *et al.* 1996). In fact, the most pronounced modulation was observed in the grasp phase, when there is little gross limb movement compared with that during the reach phase (in terms of velocity), or compared with that during the transition between reaction and early reach (in terms of acceleration).

In addition to movement and/or position dependence, the patterns of gating may be specific to other aspects of the task, e.g. the level of voluntary control and skill required and therefore probably the degree of involvement of supraspinal mechanisms. By analogy, movement-related modulation of sensory transmission to motor systems other than the cerebellum seems to be highly dependent on whether the behaviour is stereotypical and well rehearsed or exploratory (Prochazka, 1989). Task specificity of gating of peripheral input to the inferior olive would therefore not be unexpected and in fact several studies suggest that climbing fibres are activated during exploratory movements (Gellman *et al.* 1985) or when the specific task involves unpredictable external interference with the movement (Gilbert & Thach, 1977; Andersson & Armstrong, 1987).

Modality selectivity and task-specific gating

Climbing fibres projecting to the C1 and C3 zones have been shown to receive convergent input from a variety of cutaneous (e.g. Gellman, Houk & Gibson, 1983; Ekerot *et al.* 1987, 1991*a*) and muscle afferents (e.g. Murphy, MacKay & Johnson, 1973; Ekerot & Larson, 1979; Gellman *et al.* 1983; Jörntell *et al.* 1996). In the present investigation only low-threshold cutaneous input was tested and it is an open question whether or not input from other modalities is gated during reaching movements. Furthermore, although highly speculative, it is suggested that patterns of gating may be not only task specific, but in addition modality specific, such that the climbing fibre system could make selective use of different peripheral input depending on the demands of any particular motor task. This remains to be explored in future studies.

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Author's email address

R. Apps: r.apps@bristol.ac.uk

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