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RESEARCH ARTICLE

Multisensory integration for orienting responses in humans requires the activation of the superior colliculus

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Abstract Animal studies have shown that the superior colliculus (SC) is important for synthesising information from multiple senses into a unified map of space. Here, we tested whether the SC is a critical neural substrate for multisensory spatial integration in humans. To do so, we took advantage of neurophysiological findings revealing that the SC does not receive direct projections from short-wavelength-sensitive S cones. In a simple reaction-time task, participants responded more quickly to concurrent peripheral (extra-foveal) audiovisual (AV) stimuli than to an auditory or visual stimulus alone, a phenomenon known as the redundant target effect (RTE). We show that the nature of this RTE was dependent on the colour of the visual stimulus. When using purple short-wavelength stimuli, to which the SC is blind, RTE was simply explained by probability summation, indicating that the redundant auditory and visual channels are independent. Conversely, with red long-wavelength stimuli, visible to the SC, the RTE was related to nonlinear neural summation, which constitutes evidence of integration of different sensory information. We also demonstrate that when AV stimuli were presented at fixation, so that the spatial orienting component of the task was reduced, neural summation was possible regardless of stimulus colour. Together, these findings provide support for a pivotal role of the SC in mediating multisensory spatial integration in humans, when behaviour involves spatial orienting responses.

Introduction

Humans, as well as other higher organisms, monitor their spatial environments through several specialised senses. Neural integration of signals from all these different modalities is a prerequisite for a coherent and unified representation of space. Accordingly, many behavioural studies (Calvert et al. 2004) have revealed that combining multisensory information enhances the probability of detecting, localising or responding to external events. For example, normal adults are known to respond more rapidly to AV targets co-localised in space than to spatially dislocated multisensory stimuli (Spence and Driver 1997; Frassinetti et al. 2002; Bolognini et al. 2005).

In recent years, comparative research has identified several brain structures that receive and combine information from different sensory modalities, including the superior colliculus (SC) of the midbrain (Stein and Meredith 1993; Kadunce et al. 2001). It is widely held that the SC plays a central role in the integration of information from different sensory modalities and the generation of spatial orienting responses. The supporting evidence comes mainly from animal lesions, and, most compellingly, from single-unit recordings in the cat (Stein and Meredith 1993; Burnett et al. 2007). These latter studies suggested several simple principles or rules by which multisensory integration is achieved in the SC. Neurons in the SC exhibit *multisensory response enhancement* (i.e. the multisensory response exceeds the most effective unisensory component response) when visual and acoustical stimuli occur in close

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spatial and temporal proximity, whereas spatially and temporally disparate stimuli produce either depression or no change in neuronal response (the so-called spatial and temporal rules (Stein and Meredith 1993; Kadunce et al. 2001; Stanford et al. 2005).

Although the SC has provided an influential model for studies of multisensory integration in animals, as well as in brain-damaged patients (Frassinetti et al. 2005) and healthy individuals (Frassinetti et al. 2002; Bolognini et al. 2005), it remains still an open question whether in humans this region is necessary for the multisensory integration effects observed at the level of behaviour, or whether these effects critically depend on other multisensory areas of the brain. Consistent with this latter possibility, neuroimaging studies (Calvert 2001; Calvert and Thesen 2004) have revealed several cortical areas that are involved in the detection and integration of multisensory stimuli based on their shared spatial location. However, physiological evidence in the cat indicates that multisensory integration responses in neurons of cortical association areas (i.e. the anterior ectosylvian fissure and the lateral sulcus) are less restrained by the precise temporal and spatial congruency of the multisensory stimuli (Wallace et al. 1992), and therefore they may not directly mediate the behavioural consequences of multisensory integration.

Here, we directly tested the hypothesis that multisensory AV spatial integration is mediated by the SC in healthy humans. Towards this end, we exploited the fact that neurophysiological studies have reported that short-wave *S* cones in the retina do not send, or send very few projections to the SC (Marrocco and Li 1977; Schiller and Malpeli 1977; de Monasterio 1978; Sumner et al. 2002, 2004). There is evidence that the S cones mediate primarily colour perception (Mollon 1989), and their signals are carried by morphologically distinct types of retinal ganglion cells, which project to the koniocellular layers of the lateral geniculate nucleus and thence to layers 2 and 3 of the striate cortex (Dacey and Lee 1994; White et al. 1998). As well, *S*-cone stimuli cannot reach the SC via corticotectal projections (i.e. projections running from visual cortex to the SC), because *S*-cone stimuli are invisible also to the magnocellular pathway, which feeds these projections. However, even if an *S*-cone input to the magnocellular pathway, or even to the SC, exists (Stockman et al. 1991; Calkins 2001), it is small and, more importantly, it is not chromatically opponent and can therefore be masked with luminance noise (Mollon 1982; Birch et al. 1992). Therefore, stimuli uniquely detected by S cones, should be invisible to the SC.

If the SC is a critical neural substrate for mediating multisensory integration for orienting responses, then we predicted an effect of spatial congruency (i.e. RTs faster for spatially coincident than for spatially disparate AV stimuli) when using bi-sensory stimulation composed by a red monochromatic stimulus (i.e. a stimulus visible to the SC) and a concurrent auditory stimulus, presented on the same or opposite hemifield as the visual stimulus. Conversely, multisensory spatial integration is not expected when using bi-sensory stimulation composed by a purple monochromatic stimulus (i.e. a stimulus invisible to the SC) accompanied by a spatially congruent or incongruent auditory stimulus.

To provide evidence for multisensory spatial integration we employed a redundant signals paradigm for simple reaction time, in which the observer must initiate a response as quickly as possible following the detection of any stimulus onset (auditory, visual or bi-sensory AV stimulus, see Fig. 1 for a schematic diagram of a typical trial). Since the pioneering study by Todd (1912), it is known that responses to bi-sensory stimuli are faster, on average, than responses to unisensory ones. This phenomenon is known as the *redundant target effect* (RTE), and the difference in RT between single and double-targets is called the *redundancy gain*. Many subsequent studies have replicated this effect not only for cross-modal stimuli (Miller 1982, 1986; Hughes et al. 1994; Plat et al. 2000; Maravita et al. 2005; Zampini et al. 2007), mostly with visual-auditory stimulus combinations, but also for unisensory stimulus combinations (Murray et al. 2001; Savazzi and Marzi 2002, 2004). Two alternative models

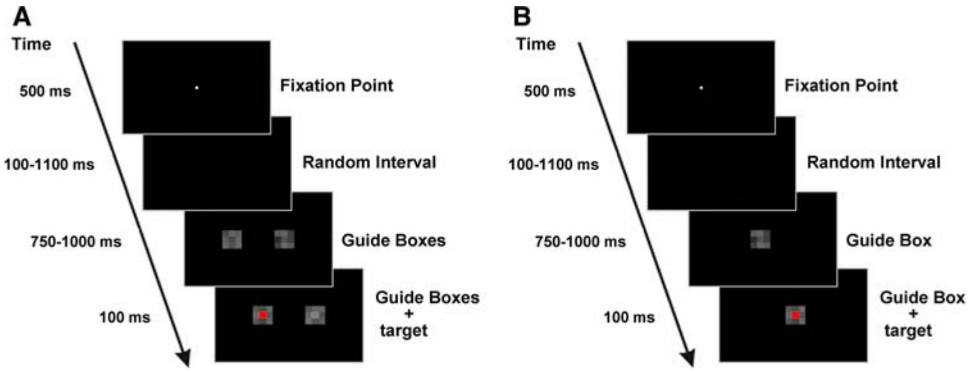

Fig. 1 Schematic diagram of a typical trial in Experiment 1 (Panel A) and Experiment 2 (Panel B). The visual target shown here is a long-wavelength stimulus (*red*). In both experiments, loudspeakers (not reported in the figure) were placed immediately below the guide boxes

have been suggested to explain the RTE: the *race model* (*statistical facilitation*) and the *neural coactivation model*. According to the race model, both elements of a bi-sensory stimulus are processed by independent channels; the one that reaches the output stage first triggers the response. This model suggests that RTE is generated by statistical facilitation: if detection latencies are interpreted as random variables, the time to detect the first of several redundant signals is faster, on average, than the detection time for any single signal (Raab 1962). A generalisation of Raab's model was recently developed in a study by Miller and Ulrich (2003). The maximal reaction time gain that can be explained by statistical facilitation is described by the *race model inequality* (Miller 1982, 1986):

$$p(t < t_0|\mathrm{AV}) = p(t < t_0|A) + p(t < t_0|V).$$

When comparing the cumulated reaction time distributions to unisensory and bi-sensory stimuli, the race model predicts that for reaction time bins (t) shorter than a particular reaction time (t_0), the probability (p) for the reaction time to a bi-sensory stimulus (audiovisual, AV) is smaller than or equal to the summed probabilities for the unisensory stimulus components (auditory and visual stimuli, A and V).

If the limit predicted by the race model inequality is violated, then a statistical facilitation explanation is no longer tenable, and the RTE can be ascribed to a mechanism that is commonly referred to as *neural coactivation*. This mechanism would transform by nonlinear summation the separate sensory inputs into an integrated product (multisensory integration).

The aim of the present study is to assess whether the SC plays a critical role in mediating multisensory spatial integration by studying RTE. Accordingly, when two simultaneous AV stimuli are presented in the same position, we should observe a violation of the race model (that is, multisensory integration) when the visual stimulus is red (i.e. visible to the SC); in contrast, when the visual stimulus is purple (i.e. invisible to the SC), a violation of the race model inequality is not expected, and the redundancy gain will be simply explained by statistical facilitation.

Experiment 1

Methods

Participants

Ten healthy subjects (age range: 21–26 years; two males and eight females) took part in the experiment. All had normal hearing and normal or corrected-to-normal vision and were naïve as to the purpose of the experiment. Participants received course credit for their participation, and gave informed consent prior to beginning. The experimental procedures were approved by the Ethical Committee of the Department of Psychology, University of Bologna. The experiment was carried out according to the principles laid out in the 1964 Declaration of Helsinki.

Stimuli and procedure

Subjects sat in a completely dark room in front of a Samsung SyncMaster 153B 17 inch PC monitor at a distance of 57 cm. All stimuli were generated by a PC with MATLAB 6.1 and PSYCHTOOLBOX 2.50 (Brainard 1997; Pelli 1997). Subjects were required to hold constant fixation on a central white dot against a black background (0.05 cd/m2). In each trial (see Fig. 1a for the sequence of events in a trial), the fixation point lasted 500 ms and then, after an interval whose duration was randomised in a 100–1,100 ms time window, two 3° × 3° square "guide boxes" were displayed 6° to the right or the left of the fixation point with an exposure duration ranging between 750 and 1,000 ms. Each box was made up of nine small squares changing luminance every 67 ms to a value drawn randomly from the range 4.05–6.75 cd/m2. This luminance noise ensured that the colour changes described below could be detected only by a chromatic channel (Mollon 1982; Birch et al. 1992; Sumner et al. 2002; Savazzi and Marzi 2004).

The visual stimuli were 1° × 1° squares randomly displayed at the centre of one of the two guide boxes, 100 ms before guide boxes were extinguished. The visual stimuli could be either short-wavelength (purple; colorimetric values: $x = 0.1826$, $y = 0.0869$) or long-wavelength (red; colorimetric values: $x = 0.6194$, $y = 0.3455$). For all visual stimuli, exposure duration was 100 ms and the luminance was 5.4 cd/m2.

The auditory stimuli consisted of 2 kHz, pure-tone bursts emitted for 100 ms by one of two piezoelectric loudspeakers (0.4, 8 W) located in front of the PC monitor, ~4 cm below the visual stimuli. The intensity of the auditory stimuli was 66.3 dB SPL.

There were seven randomly intermingled stimulus conditions:

1. unisensory auditory condition: the auditory stimulus was presented alone;
2. unisensory visual condition: the visual stimulus (purple or red) was presented alone;
3. same-position audiovisual (SP-AV) condition: the visual (purple or red) and the auditory stimulus were presented simultaneously and in close spatial proximity;

4. different-position audiovisual (DP-AV) condition: the visual (purple or red) and the auditory stimulus were presented simultaneously and in different positions (i.e. opposite hemifields).

Chromaticities and luminances were calculated from the spectra of each stimulus, which was measured directly with a Minolta chromameter CL-200.

Subjects binocularly viewed the PC monitor where the stimuli were displayed. They were instructed to hold constant fixation on the central white dot, and to respond by pressing the space bar of the keyboard with the index finger of the dominant hand as quickly as possible to any unisensory (visual or auditory) or bi-sensory stimulus.

The experiment was subdivided into six blocks of trials with an overall number of 1,120 presentations for each participant. There were 120 trials for each type of stimulus condition, and 280 catch trials in which only the guide boxes were presented without the target stimuli. Catch trials were introduced to discourage participants from responding to the guide box rather than to the target stimulus. For each subject, a 5-min practice preceded data collection.

Statistical analysis

For each subject, the average RTs were calculated and their distributions were recorded for each stimulus condition. To test for the presence of an RTE, planned comparisons between each of the unisensory stimulus conditions and the bi-sensory stimulus conditions were performed.

To determine if a statistical facilitation explanation for the RTE was tenable, or if a coactivation explanation of the RTE was necessary, Miller's test of the race model was implemented by using RMITest software (Miller 1982; Ulrich et al. 2007). The program computes the estimated cumulative density functions (CDFs) of RT for each condition and the estimated race model inequality bound. It also computes a *t*-test at each percentile to see whether the race model violation is statistically significant. The difference between the cumulative probability for the double stimuli and the sum of the cumulative probability for the single stimuli indicates the magnitude of violation.

Results

Participants responded to less than 3% of catch trials. RTs faster than 140 ms and slower than 650 ms were considered as anticipations and delayed responses, respectively, and discarded (<1% of trials). Reaction time data were subsequently analysed with a two-way ANOVA with Colour (red versus purple stimuli) and Condition (unisensory visual, unisensory auditory, bi-sensory SP-AV, bi-sensory DP-AV) as within-subject factors. Pairwise comparisons were conducted using the Newman–Keuls test.

Overall, we found that responses to long-wavelength, red stimuli (301 ms) were faster than responses to short-wavelength, purple stimuli (306 ms), as evident from the significant effect of the main factor Colour [$F(1, 9) = 16.3$, $P < 0.003$].

The main factor Condition was also significant [$F(3, 27) = 98.9$, $P < 0.0001$]. Responses to unisensory visual stimuli (352 ms) were significantly slower than responses to AV stimuli both in the SP-AV condition (277 ms; $P < 0.0002$) and in the DP-AV condition (280 ms; $P < 0.0002$). Likewise, RTs in the unisensory auditory condition (304 ms) were significantly slower than RTs in both the SP-AV condition ($P < 0.0002$) and DP-AV condition ($P < 0.0002$). Finally, the redundancy gain (i.e. the RT difference between single and double stimulation) was similar in red and purple stimuli (see Table 1).

More important for the present purpose, there was a significant interaction between Colour and Condition [$F(3, 27) = 22.3$, $P < 0.00001$]. Thus, when the visual stimulus was red, RTs in the SP-AV condition were faster than RTs in the DP-AV condition (272 and 282 ms, respectively, $P < 0.0003$), whereas there was no significant difference between SP-AV (281 ms) and DP-AV (278 ms) conditions, when the visual stimulus was purple. Moreover, responses were significantly faster in the red SP-AV condition than in the purple SP-AV condition ($P < 0.0002$) (see Fig. 2).

RT data were then analysed with Miller's race inequality test to determine whether the observed redundancy gains could be explained in terms of a race or a neural coactivation model (Miller 1982, 1986; Ulrich et al. 2007). As shown in Fig. 3, a significant violation of race inequality was found only for the red, in the spatially coincident AV

Table 1 Redundancy gain (*RG*)

Red			Purple		
Stimulus	M	SD	Stimulus	M	SD
V	345	51	V	360	50
A	304	68			
AV	277	54	AV	280	55
RG	47		RG	52	

Mean RTs (*M*) and standard deviations (*SD*) in milliseconds for each stimulation condition, separately for the red visual stimulus and purple visual stimulus. RG is the difference between mean RTs in unisensory conditions and mean RTs in bi-sensory conditions. Note that the same RT data from unisensory auditory (*A*) condition were used to calculate RG for red and purple visual stimuli. Abbreviations: *V* visual alone, *A* auditory alone, *AV* audiovisual bi-sensory condition

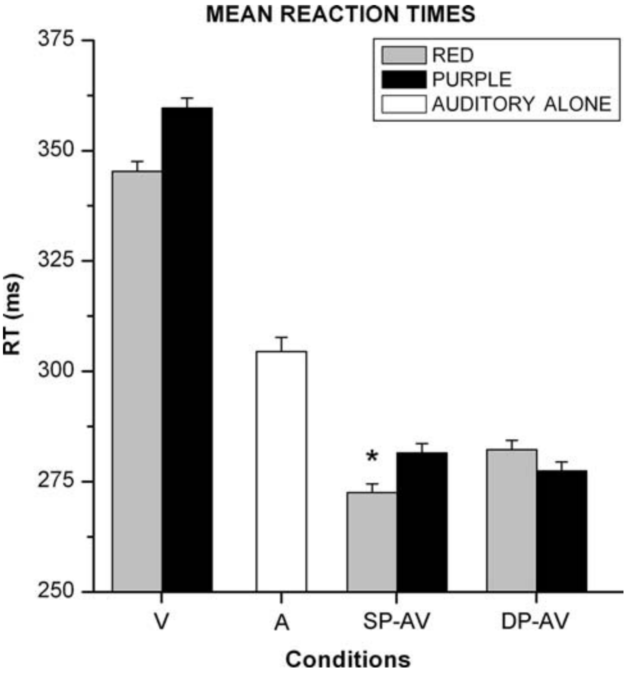

Fig. 2 Mean RTs (*SEM* indicated) for each condition of Experiment 1 (*V* visual alone, *A* auditory alone, *SP-AV* same-position audiovisual condition, *DP-AV* different-position audiovisual condition). *Grey bars* represent the conditions in which the visual stimulus was *red*; *white bar* represents the auditory-alone condition; *black bars* represent the conditions in which the visual stimulus was *purple*. A significant difference (denoted by an *asterisk*) was found between RTs in SP-AV and DP-AV conditions when the visual stimulus was *red*, but not when it was *purple*

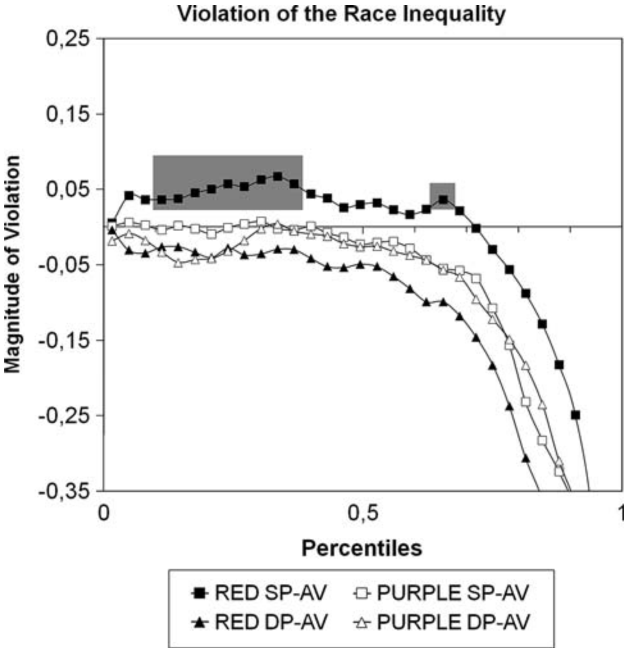

Fig. 3 Violation of the race inequality test for the four bi-sensory AV conditions of Experiment 1. The *grey rectangles* mark the areas in which the violation is significantly different from zero, as assessed by one-sample *t*-test

condition. The grey rectangles in Fig. 3 indicate the percentiles in which the violation was significantly different from zero, as assessed by a *t*-test. Therefore, the RTE with red SP-AV stimuli violated the race inequality and can be explained by neural coactivation, while with the red DP-AV or purple (either SP-AV or DP-AV) stimuli there was no violation, and thus RTE in this case can be related to statistical facilitation.

In order to statistically validate this result, we carried out a three-way ANOVA for repeated measures with Colour (red versus purple), Condition (SP-AV versus DP-AV) and Percentile (those included in the violation area, from 0.09 to 0.66) as factors. A significant interaction between Colour and Condition [$F(1, 9) = 25.9$, $P < 0.0007$] was evident. Newman–Keuls post hoc showed that the violation of red SP-AV stimuli was reliably different from that of red DP-AV stimuli ($P < 0.0003$) and purple stimuli (both SP-AV and DP-AV, $P < 0.0007$ in all comparisons).

Thus, the crucial result of this experiment is that an RTE was obtained with all bi-sensory AV stimuli, but the nature of this redundancy gain, namely statistical facilitation versus neural coactivation, changed with the stimulus colour, purple versus red, respectively. Because the short wavelength stimuli do not send, or send very few inputs to the SC, we conclude that this midbrain region is required for neural integration of multisensory signals across space.

Experiment 2

Having shown that SC activation is critical for the integration of AV stimuli in the spatial domain, in Experiment 2 we tested the possibility that SC involvement in multisensory integration strictly depends on whether the perceptual task requires spatial orienting responses. Thus, participants responded to both unisensory and bi-sensory AV stimuli presented at fixation. If the SC integrates multisensory stimuli for the purpose of guiding gaze or attention shifts across space, then presenting AV stimuli at fixation should reduce the contribution of the SC to the task, because orienting responses are minimised. As a consequence, the differential effects of visual stimuli, visible or invisible to the SC, should be significantly reduced or abolished.

In order to measure multisensory integration, bi-sensory AV stimuli either occurred at the same time, or at different times, with the auditory stimulus always preceding the visual one; indeed, previous studies have shown that temporally coincident AV stimuli produce enhanced performance (faster RTs and increased perceptual sensitivity) relative to temporally disparate stimuli (Bolognini et al. 2005). Thus, when two central AV stimuli are presented at approximately the same time, we should observe a violation of the race model inequality (that is, multisensory integration), whereas this effect is not expected for temporally disparate AV stimuli. Unlike Experiment 1, however, the race violation should not depend on the colour of the visual stimulus as the task does not require orienting responses.

Methods

Participants

A new group of ten healthy subjects (age range: 21–29 years; three males and seven females) took part in the experiment. All had normal hearing and normal or corrected-to-normal vision, and were naïve as to the purpose of the experiment. They received course credit, and all gave informed consent prior to beginning.

Stimuli and procedure

The experimental apparatus was identical to the one used in Experiment 1, except that stimuli were always presented at central fixation, so that spatial orienting responses were substantially reduced.

There were 11 randomly intermingled stimulus conditions:

1. unisensory auditory condition: the auditory stimulus was presented alone;
2. unisensory visual condition: the visual stimulus (purple or red) was presented alone;
3. bi-sensory AV conditions: the auditory and visual stimulus (purple or red) were presented at four different separation intervals (0, 50, 100 and 200 ms), with the auditory stimulus always preceding the visual one.

The experiment was subdivided into six blocks of trials with an overall number of 1,760 presentations for each participant. There were 120 trials for each type of stimulus condition, and 440 catch trials in which only the guide boxes were presented without the target stimuli.

Statistical analysis

The statistical analysis was identical to the one performed in Experiment 1.

Results

Participants responded to <3% of catch trials. Data trimming, performed as in Experiment 1, involved ~1% of the trials. Reaction time data were analysed with a two-way ANOVA with Colour (red and purple stimuli) and Condition (unisensory visual, unisensory auditory, bi-sensory AV-0 ms, bi-sensory AV-50 ms, bi-sensory AV-100 ms and bi-sensory AV-200 ms) as the within-subject factors. Pairwise comparisons were conducted using the Newman–Keuls test.

As in Experiment 1, responses to red long wavelength stimuli (303 ms) were faster than responses to purple short wavelength stimuli (307 ms), as indicated by the significant effect of the main factor Colour [$F(1, 9) = 5.1$, $P < 0.05$]. There was a significant effect of Condition [$F\ (5, 45) = 66.2$, $P < 0.0001$], with RTs to the AV, bi-sensory stimuli (276, 281, 291 and 299 ms, for AV-0, AV-50, AV-100, AV-200 ms, respectively) being faster than responses to both unisensory auditory (321 ms) and unisensory visual stimuli (364 ms; $P < 0.0006$ in all comparisons). In addition, responses in the AV-0 and AV-50 ms conditions were faster than responses in the AV-100 and AV-200 ms conditions ($P < 0.05$ in all cases). The interaction between Colour and Condition was significant [$F(5.45) = 5.66$, $P < 0.0004$], due to faster RTs for red unisensory visual stimuli than purple unisensory visual stimuli. No other comparisons (see Fig. 4) reached a significant level.

As in Experiment 1, the redundancy gain (i.e. the RT difference between single and double stimulation) was similar for red and purple stimuli (see Table 2).

RT data were then analysed with Miller's race inequality test to determine whether the observed redundancy gains could be explained in terms of a race or a neural coactivation model. As shown in Fig. 5a and b, a significant violation of race inequality was found both for the red and the purple stimuli, in the bi-sensory AV-0 ms and bi-sensory AV-50 ms conditions. The grey rectangles in the figures indicate the percentiles in which the violation was significantly different from 0, as assessed by a *t*-test; in contrast, no violation of race inequality was found for bi-sensory AV-100 ms, and bi-sensory AV-200 ms conditions (see Fig. 5c, d). Thus, the RTE found for short AV intervals (0 and 50 ms) can be explained by neural coactivation, while the RTE of all the other bi-sensory conditions can be related to statistical facilitation.

To statistically confirm this result, a three-way ANOVA for repeated measures with Colour (red versus purple), Condition (AV-0, AV-50, AV-100, AV-200 ms) and Percentile (those included in the violation area, from 0.1 to 0.7) as within-subject factors, was performed on the data.

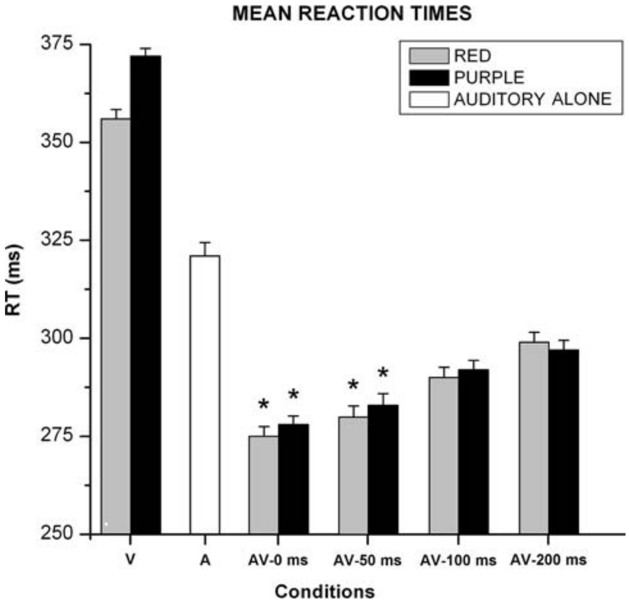

Fig. 4 Mean RTs (*SEM* indicated) for each condition of Experiment 2 (*V* visual alone, *A* auditory alone, AV-0 ms audiovisual condition, 0 ms of audio lead; AV-50 ms audiovisual condition, 50 ms of audio lead; AV-100 ms = audiovisual condition, 100 ms of audio lead; AV-200 ms = audiovisual condition, 200 ms of audio lead). *Grey bars* represent the conditions in which the visual stimulus was *red*; *white bar* represents the auditory alone condition; *black bars* represent the conditions in which the visual stimulus was *purple*. RTs in the AV-0 and AV-50 ms were significantly faster (denoted by an *asterisk*) than those in the AV-100 and AV-200 ms conditions, for both *red* and *purple* visual stimuli

Differently from Experiment 1, the interaction between Colour and Condition was not significant ($P = 0.73$), suggesting that the race model violation did not depend on the colour of the visual stimulus that was paired with the auditory stimulus.

Discussion

Physiological recordings in animals (Stein and Meredith 1993) suggest that the SC plays an important role in multisensory integration. When auditory and visual stimuli appear concurrently from the same spatial location, neural activity in this midbrain region is strongly enhanced. Importantly, multisensory enhancement in the SC parallels the striking changes in behavioural performance resulting from the simultaneous stimulation of two or more modalities with shared spatial location (Bolognini et al. 2005). Thus, the SC has been repeatedly used as a model system for how the brain integrates unisensory modalities to produce a unified multisensory map of the external environment (Stein and Meredith 1993).

In recent years, neuroimaging studies in humans have identified several cortical sites [among others, the insula, the superior temporal sulcus, the intraparietal sulcus, and several frontal regions (Calvert 2001; Bushara et al. 2003; Calvert and Thesen 2004; Macaluso and Driver 2005)] associated with multisensory spatial integration, thereby suggesting that this process may not necessarily involve the SC. However, functional imaging results cannot definitively establish that a brain area is necessary for a particular cognitive process.

In the present study, the hypothesis that the SC may be a critical neural structure for multisensory spatial processing involving nonlinear response summation was directly tested in Experiment 1. To test this hypothesis, we used visual stimuli visible and invisible to both the retino-collicular and magnocellular pathways. These stimuli were colour changes visible only to the short-wavelength-sensitive *S* cones of the retina, exploiting the fact that neurophysiological studies have consistently reported that there are no projections to the SC from *S* cones (Marrocco and Li 1977; Schiller and Malpeli 1977; de Monasterio 1978). This

Table 2 Redundancy gain (*RG*)

Red				Purple			
Stimulus	M	SD	RG	Stimulus	M	SD	RG
V	356	54		V	372	44	
A	321	77					
AV-0 ms	275	56	64	AV-0 ms	278	49	68
AV-50 ms	280	63	59	AV-50 ms	283	66	63
AV-100 ms	290	58	49	AV-100 ms	292	54	54
AV-200 ms	299	57	40	AV-200 ms	297	56	49

Mean RTs (*M*) and standard deviations (*SD*) in milliseconds for each stimulation condition, separately for the red visual stimulus and purple visual stimulus. RG is the difference between mean RTs in unisensory conditions and mean RTs in bi-sensory conditions. Note that the same RT data from unisensory auditory (*A*) condition were used to calculate RG for red and purple visual stimuli. Abbreviations: *V* visual alone, *A* auditory alone, *AV* audiovisual bi-sensory condition

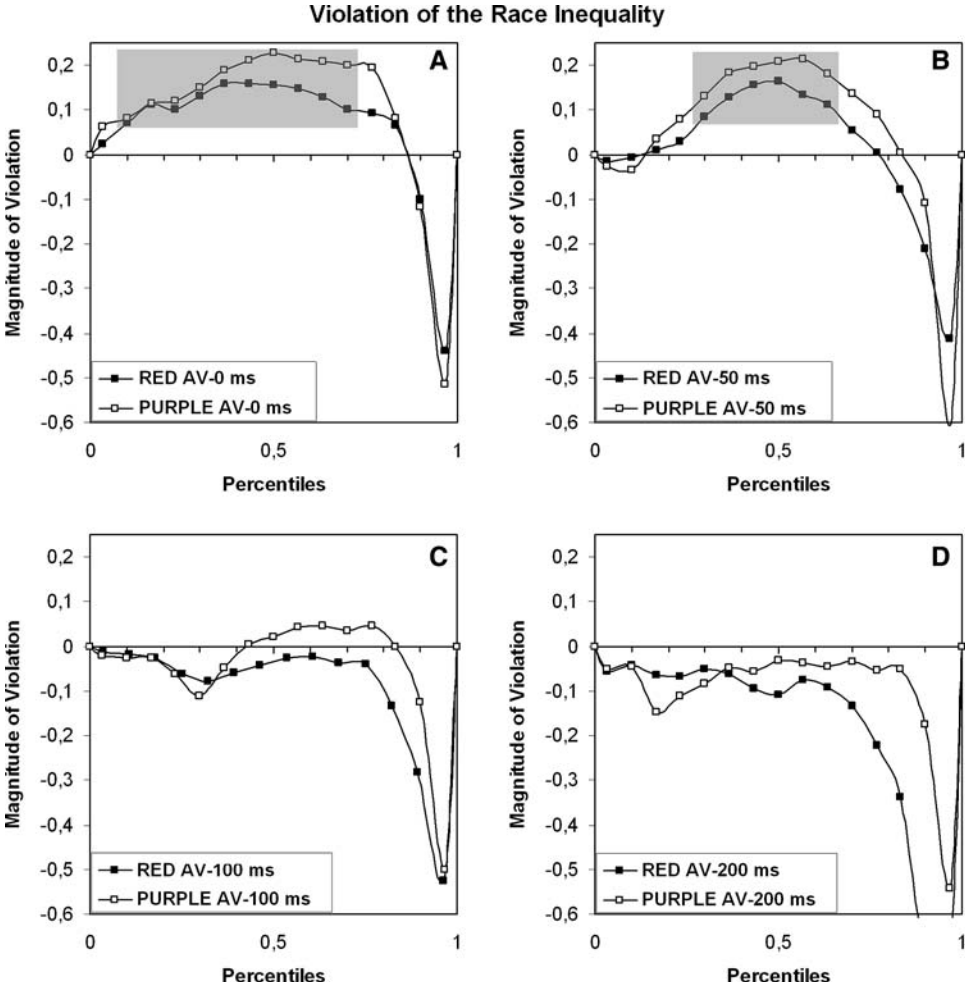

Fig. 5 Violation of the race inequality test for the eight bisensory AV conditions of Experiment 2. The *grey rectangles* in Panel A and Panel B mark the areas in which the violation is significantly different from zero, as assessed by one-sample *t*-test. Abbreviations as in Fig. 4

novel technique has already proved fruitful for understanding the functioning of the SC in visual attention orienting (Sumner et al. 2002, 2004, 2005; Savazzi and Marzi 2004). In Experiment 1, peripheral AV stimuli were simultaneously presented in the same or different spatial location of the visual field. Because the SC (and magnocellular pathway) is blind to short-wavelength stimuli, multisensory spatial integration mediated by this region should not occur with this type of visual stimuli. Results indeed supported this prediction.

We demonstrated that behavioural responses in the bisensory AV conditions depended on the colour of visual stimuli. Particularly, RTs to spatially coincident AV stimuli were significantly faster than RTs to the spatially disparate AV stimuli when red long-wavelength stimuli were used, but not when purple short-wavelength were presented. This finding shows a specific spatial AV integration effect for the red but not for purple visual stimuli, suggesting that multisensory spatial interactions may not occur for signals invisible both to the SC and magnocellular pathway. To gather further, more stringent evidence of the role played by the SC in multisensory spatial integration, we carried out a second analysis based on Miller's race inequality model (Miller 1982). According to this model, the redundancy gain (i.e. the RT advantage with double relative to single stimuli) may be due either to a probabilistic race between signals transmitted along different neural channels (statistical facilitation), or to a neural coactivation (multisensory integration) mechanism resulting from the convergence of the incoming signals onto one or more brain centres. In Experiment 1, this analysis showed a significant violation of the race model for spatially coincident AV stimuli, but only when a red long wavelength stimulus was used.

Together, our present findings reveal that short wavelength stimuli, to which the SC is blind, do not produce AV integration across corresponding locations in space, indicating that the SC may play a key role in integrating stimuli from separate unisensory modalities into a unified representation of space for the purpose of spatial orienting. Similar results have been reported recently by Bolognini et al. (2007).

Specifically, we found that the neural coactivation mechanism is observable when two conditions are satisfied:

first, visual information must reach the SC (as is the case of red long-wavelength visual stimuli); second, concurrent auditory and visual stimuli must be presented in the same spatial position (hemifield). Indeed, the violation of the race model occurred only when auditory stimuli were accompanied by red, spatially coincident visual stimuli. These results are consistent with neurophysiological findings revealing that neurons in the deeper layers of the SC respond to multiple sensory inputs from the same area in space (Wallace et al. 1993). Additionally, such neurons exhibit a strongly nonlinear response which is greater than the sum of their unisensory responses, when driven by multisensory stimulation which is coincident in time and space (Stein and Meredith 1993; Stanford et al. 2005). It is therefore justified to conclude that nonlinear, multisensory integration for spatial orienting (indexed here by the violation of the race inequality model) cannot be reliably found with visual inputs to which the SC is blind.

While our findings demonstrate that the SC is an important neural substrate underlying multisensory spatial integration in humans, they do not rule out the possibility that other brain areas in the magnocellular system (including frontal or parietal eye fields which may also be blind to short wavelength stimuli) may have a role in the ability to synthesise spatial AV stimuli. The present findings cannot allow us to adjudicate between these two alternative accounts. Note, however, that lesion studies in humans have shown that cortical damage affecting the fronto-temporo-parietal areas (in neglect patients) leaves unaltered the ability to synthesise spatially coincident AV stimuli [Frassinetti et al. 2005; but see Valenza et al. (2004) for different results concerning spatial interaction between vision and touch], thereby lending some support to the hypothesis that these cortical areas may be not necessary for mediating the behavioural effects of AV spatial integration.

Nevertheless, evidence in the cat suggests that deactivation of cortical association cortex eliminates the characteristic multisensory response enhancement in the SC (Jiang et al. 2002, 2007). Thus, although the SC is a critical neural substrate for enhanced orienting responses towards spatially coincident multisensory stimuli, as we report here, it may also require modulatory influences from other cortical regions. Additional studies are needed to clarify the specific influence played by cortical areas in modulating the activity of the SC in humans.

Another important point for discussion concerns the results of Experiment 2. In this experiment, AV stimuli occurred at the same or different time at central fixation, thus reducing the involvement of spatial orienting movements to perform the task. Under this condition, we showed that responses did not vary as a function of the colour of the visual stimuli. Indeed, RTs to temporally coincident (i.e. separation interval <100 ms) AV stimuli were significantly faster than RTs to temporally disparate AV stimuli, regardless of whether red long-wavelength or purple short-wavelength visual stimuli were presented. Moreover, a significant violation of race inequality (i.e. neural coactivation) was observed for temporally coincident versus temporally disparate central AV stimuli. More importantly, this effect did not depend on whether auditory stimuli were paired with long or short-wavelength visual stimuli.

Neurophysiological evidence in animals and humans supports the hypothesis that the SC is critically involved in coding spatial locations for the purpose of guiding saccades and shifts of attention towards them (Kustov and Robinson 1996; Ignashchenkova et al. 2004). Our findings are perfectly consistent with this view. They demonstrated that when reorienting of gaze or attention is minimised, because AV targets are consistently presented at fixation, multisensory spatial integration could occur without direct access to the SC or magnocellular pathway. Furthermore, the fact that short-wavelength stimuli were quite capable of eliciting multisensory integration under certain conditions rules out the possible objection that these stimuli are unsuited to mediate AV integrative effects only because their slower transmission speed relative to auditory stimuli. Although blind to the SC, short-wavelength visual stimuli are visible to several cortical areas with strong parvocellular inputs (Mullen et al. 2007), therefore suggesting that (purely temporal) AV integration effects observed at fixation may be mediated by multisensory cortical areas. This hypothesis is in line with the fact that a well-known example of multisensory integration, the McGurk illusion (McGurk and MacDonald 1976) is not affected by the relative location of audio and visual speech signals (Spence and Driver 2004), and is most likely implemented in cortical temporal areas (Miller and D'Esposito 2005; Holmes and Spence 2005). Overall, these effects underline the multiplicity and flexibility of integrative processes, mediated by several cortical and subcortical regions in the brain.

To sum up, our data provide novel evidence that the SC has an important role in mediating multisensory spatial integration effects in humans. By using a new behavioural method (i.e. short-wavelength monochromatic stimuli) that reduces, or even abolishes, visual inputs to the SC and magnocellular pathway, we demonstrated that the enhanced performance in response to spatially coincident AV stimuli no longer occurs, at least for those behaviours involving gaze and/or attention orienting in space. Indeed, when visual inputs had no access to the SC, subjects' responses to combinations of multisensory stimuli were simply determined by the faster of two sensory inputs (race mechanism), rather than by nonlinear summation of the auditory and visual inputs (neural coactivation mechanism).

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