

Choice reaction times for human head rotations are shortened by startling acoustic stimuli, irrespective of stimulus direction

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Auditory startle reflexes can accelerate simple voluntary reaction times (StartReact effect). To investigate the role of startle reflexes on more complex motor behaviour we formulated two questions: (1) can auditory startle reflexes shorten choice reaction times?; (2) is the StartReact effect differentially modulated when startling auditory stimuli are delivered ipsilaterally or contralaterally to an imperative ‘go’ signal? We instructed 16 healthy subjects to rotate their head as rapidly as possible to the left or to right in response to a guiding visual imperative stimulus (IS), in both a simple and choice reaction protocol. Startling acoustic stimuli (113 dB) were delivered simultaneously with the IS (from either the same or opposite side) to induce the StartReact effect. We recorded kinematics of head rotations and electromyographic responses. The StartReact effect was present during choice reaction tasks (56 ms onset reduction; $P < 0.001$). The presentation side of the startling stimulus (left/right) did not influence the effect in choice reaction tasks. We observed a directional effect in simple reaction tasks, but this probably occurred due to a flooring effect of reaction times. Onsets of EMG responses in neck muscles were not influenced by the direction of the acoustic startling stimulus. Startling acoustic stimuli decrease reaction times not only in simple but also in choice reaction time tasks, suggesting that startle reflexes can accelerate adequate human motor responses. The absence of a clear directional sensitivity of reaction times to startling acoustic stimuli suggests that the acceleration is not highly specific, but seems to provide a global preparatory effect upon which further tailored action can be undertaken more quickly.

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The auditory startle reflex is a fast response to a sudden, loud acoustic stimulus. It is characterized by a radiating pattern of muscular contractions travelling upward and downward from a presumed trigger centre within the brainstem (Davis *et al.* 1982; Wilkins *et al.* 1986; Brown *et al.* 1991; Brown, 1995; Bakker *et al.* 2006). The physiological role of this startle reflex for everyday motor behaviour remains largely unknown. Startle reflexes may well be part of the normal motor repertoire in the context of ‘fight-or-flight’ behaviour, allowing either attacking or defending more quickly. Even if this is the case, the integration with other movement programmes is unknown.

One of the earliest speculations on the functional relevance of startling in man concerned the resulting movement of the startle response itself. This was based

on kinematic analyses of body movements following a sudden loud noise. The general notion at that time was that startling responses might serve as a rather non-specific ‘protective response’, for example, to shield the head against external stimuli (Suhren *et al.* 1966). On the other hand, exaggerated startle reflexes can also have detrimental effects on motor performance, as illustrated by major type hyperekplexia. Startling can cause patients to drop objects or cause stiffness in these subjects. Voluntary movements become impossible, resulting in falls without subjects being able to break their fall (Tijssen *et al.* 2002; Bakker *et al.* 2006).

One indication that startle reactions are functional did not ensue from startle movements themselves, but resulted from observations on the influence of a startling stimulus upon other, planned, movements. Reaction

times in simple reaction time tasks are accelerated when the required ballistic movements coincide with startling acoustic stimuli: the StartReact effect (Valls-Solé *et al.* 1995, 1999). This has also been demonstrated for saccadic eye movements (Castellote *et al.* 2007).

Our first question here concerned the possible presence of the StartReact effect during choice reaction time tasks. We reasoned that the behavioural response to a startling stimulus may well depend on the circumstances and would thus involve a choice. Previous research has shown that startle reflexes can accelerate reaction times during a simple reaction time task (Valls-Solé *et al.* 1999; Siegmund *et al.* 2001; Carlsen *et al.* 2004b). It remains unclear whether the StartReact effect is also present when a choice between two or more tasks is offered (choice reaction time task) (Valls-Solé, 2004; Carlsen *et al.* 2004a; Kumru *et al.* 2006). If so, this would provide further evidence for a functional role of startle responses.

Our second research question concerned the possible directional dependence of the StartReact effect, which could also have functional relevance if present. It is currently unknown whether the reaction to a startling stimulus is in any way sensitive to the direction from which the stimulus reaches the subject.

We investigated involvement of the StartReact effect in choice reaction tasks, and a possible directional dependence by asking subjects to rotate their head as rapidly as possible to the left or right, while concurrently administering startling acoustic stimuli from either the left or the right side of the head.

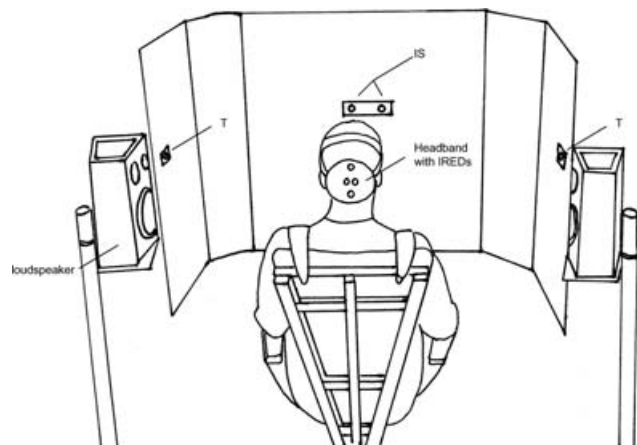


Figure 1. Experimental set-up

Posterior view of the experimental set-up showing a subject strapped in the chair, facing the screen. The screen holds two light-emitting diodes (LEDs) in front of the subjects that served as visual imperative stimuli (IS) and two LEDs to the side that served as targets (T) for the requested head rotation. The speakers that delivered the acoustic stimuli were positioned at the height of the subjects' ears at 0.5 m distance.

Methods

Subjects

Sixteen right-handed, healthy subjects (8 men; mean age 23.5 years, range 21.1–25.5 years) participated in the main experiment. Six of these subjects (2 men; mean age 24 years, range 22.7–25 years) also participated in a separate validating experiment. All subjects gave written informed consent prior to the experiment. The experiments conformed with the standards of the *Declaration of Helsinki* and in accordance with local ethical guidelines. Subjects were paid a nominal amount for their participation.

Reaction time task

Subjects sat in a dimly lit room surrounded by a custom-made screen with a height of 1.60 m at a distance of 0.65 m (Fig. 1). The head of the subject was centrally positioned in front of this screen. Four light-emitting diodes (LEDs) of 4 mm in diameter each were mounted on the screen. Two were positioned 10 cm apart and next to each other directly in front of the subject, and two others were positioned 60 deg away from the centre to the left and the right. We instructed subjects to rotate their head as rapidly as possible when either the left or the right LED directly in front of them was lit: this was the visual imperative stimulus (IS). Lighting the left LED indicated that subjects should turn their heads to the left and vice versa. The lateral LED at 60 deg on the same side as that indicating the IS was lit at the same time as the IS to serve as the target (T) for head rotation, thus ascertaining similar head rotations across subjects and across conditions. Subjects were instructed to rotate their head until they faced the target LED, and to keep that position until after the LED was switched off 2 s after stimulus onset.

Acoustic stimuli

Acoustic stimuli were delivered simultaneously with the IS, to either the left side or the right side of the subject through loudspeakers, placed on the very left and right side of the screen (Fig. 1). Stimuli (50 ms, white noise) were generated using a 24-bit sound card, and presented with an intensity of 113 dB (startling) or 60 dB (non-startling) sound pressure level, measured at the position of the ears of the subject, using a Precision Sound Level Meter (Lutron SL 4001). The amplitude of the sound was calibrated using the same white noise used in the experiment, but with a pulse duration of 1000 ms. Timing of the experiment was arranged using Presentation software (Neurobehavioural Systems Inc., Albany, USA), which delivered the auditory and visual stimuli and triggered EMG and kinematic

recordings. The auditory stimuli were delivered at the same time as the IS, according to Valls-Solé *et al.* (1995).

Experiments

Subjects were tested under a range of different conditions, involving head rotations (HR) to both the left or right, startling or non-startling acoustic stimuli from either side, or combinations of HR and acoustic stimuli.

Validation experiment. The validation experiment consisted of four conditions: (1) 'startle only': 16 startling acoustic stimuli from the left or the right, without HR; (2) 'non-startle only': 16 non-startling acoustic stimuli from the left or the right, without HR; (3) 'HR only': 32 head rotations to the left or the right, as fast as possible towards the IS, without an accompanying acoustic stimulus; (4) 'HR–startle': 32 head rotations *only* to the right, combined with a startling acoustic stimulus from either the left or from the right. Trials were presented randomly and separated with a varying inter-trial interval of 10–20 s. Prior to the experiment, subjects received seven practice trials.

Main experiment. Subjects always had to rotate their head towards the IS, either to the left or the right. Simultaneously with the IS a startling (HR–startle) or a non-startling acoustic stimulus (HR–non-startle) was administered from the left or the right side. This resulted in 'ipsilateral trials', where the auditory stimulus came from the same side as the IS, or 'contralateral trials' in which the auditory stimulus came from the opposite side of the IS (Fig. 2). Subjects were instructed to focus on the IS and to disregard any other stimuli.

Subjects performed both a simple reaction time task (simple RT) and a choice reaction time task (choice RT).

In the simple RT, the IS was preceded by a visual warning stimulus, consisting of lighting one of the LEDs in front of the subjects indicating the required direction of head rotation. The warning stimulus was then turned off, and the IS followed with a random delay between 2 and 7 s. For the choice RT test the visual warning stimulus was left out.

Both the simple RT and choice RT were subdivided into four blocks of 24 trials each: 8 HR–startle trials and 16 HR–non-startle trials, with random intertrial intervals between 10 and 20 s. Within each block, trials were randomized for stimulus intensity (non-startling or startling) and stimulus direction (ipsilateral or contralateral). Before the actual experiment, subjects performed eight practice trials without acoustic stimuli.

Outcome measures

Head rotations were measured with a motion analysis system. The primary outcome measure was the kinematic analysis of the StartReact effect, defined as the difference in reaction time between the startle and non-startle conditions. Furthermore, as a secondary variable of interest, we measured electromyographic activity in four neck muscles to determine the presence of startle reflexes.

Motion analysis. Four infrared emitting diodes (IREDs) were mounted on a headband to record head movements (Fig. 1). The IREDs were positioned on a disc at the back of the subjects' head at ear height in a rhombic montage with maximal interspaces of 10 cm. An Optotrak 3020 motion analysis system (Northern Digital Inc., Waterloo, Canada) tracked IRED positions with an accuracy of 0.1 mm or better in all directions. The Optotrak system was mounted on the ceiling above the subject at a distance of approximately 2.5 m behind the seated subject, tilted downward at an angle of 30 deg relative to the ceiling.

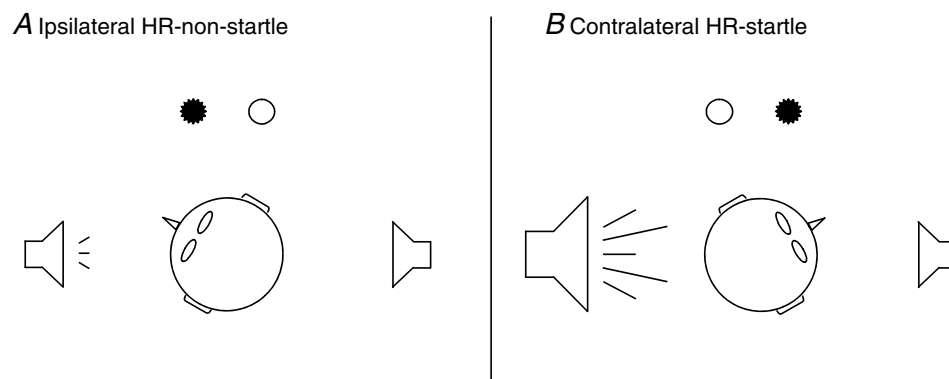


Figure 2. Experimental trial types

Two different experimental trial types schematically represented. *A*, trial in which a non-startling acoustic stimulus (HR–non-startle) was delivered ipsilateral to the direction of the imperative stimulus (IS). *B*, trial in which a startling acoustic stimulus (HR–startle) was delivered contralateral to the direction of the IS.

Prior to the experiment, three axes were defined in reference of the subject as *x*-axis (anterior–posterior), *y*-axis (left–right) and *z*-axis (up–down). Afterwards, all measured positions were referred to this coordinate system. Recordings started 100 ms pre-stimulus in sweeps of 1000 ms with a sample rate of 500 Hz, capable of detecting movement onsets in the *y*-direction with an accuracy of 2 ms.

EMG. Electromyographic activity was recorded from the left and right sternocleidomastoid (SCM) and cervical paraspinal muscles using pre-gelled surface electrodes (3M Red Dot 2258) in a belly tendon montage. EMG electrodes were attached over the sternal head of the SCM. We positioned one electrode four fingers below the mastoid process and the other electrode 3 cm lower (centre to centre). For the paraspinal muscles, we placed the electrodes directly over the splenius muscles, as these are mainly involved in head rotation: one electrode was placed two fingers below the occipital bone, inferior to the lateral one third of the superior nuchal line and the other electrode one finger beside the seventh cervical vertebrae. EMG signals were recorded from 100 ms before the onset of the auditory stimulus until 400 ms after it with a sample frequency of 2000 Hz. These signals were amplified and converted analog-to-digital using a multichannel electromyography system (Refa, TMSI Enschede). EMG signals were filtered using a 15–400 Hz bandpass filter prior to analysis.

Data analysis

Motion analysis. Both Optotrak and EMG recordings were analysed using customized Matlab programmes (The Mathworks Inc., Natick, MA, USA). Position data recorded with Optotrak were differentiated to obtain velocity values and smoothed using a first order Savitsky–Golay filter. We defined onset latencies of head movements as the point at which velocity signals in the direction of the head movements exceeded 15 mm s^{-1} . In trials in which one of the IREDs was out of the camera view, the remaining three were used to calculate the onset of head movement. In none of the trials more than one IRED was out of view. Onset latencies were confirmed visually. Trials, in which subjects initially rotated in the wrong direction, were reported as response errors and were excluded from further analysis. These trials were not repeated.

EMG. Onset latencies of EMG bursts were calculated across each trial and muscle. We used a semi-automatic computer algorithm that determined when the signal deviated for the first time more than 2.5 s.d.s from the mean baseline EMG for more than 50 ms. All onset latencies were visually inspected and manually adjusted

when necessary by one person (blinded for muscle, direction of head rotation and the direction and type of auditory stimulus). Amplitudes were determined by calculating the area under the curve (AUC) over an interval of 50 ms following the onset latency. To illustrate the interaction between startle and voluntary activity, we calculated ratios and arithmetic differences between EMG amplitudes in the different conditions, as previously described by Siegmund *et al.* (2001). For this purpose, we only used head rotations to the right.

For each muscle, trials were rejected if muscle activity preceded stimulus onset, if onset of muscle activity could not be determined due to small EMG activity, or if the onset was ambiguous.

EMG-defined startle reflexes. In an attempt to discriminate between EMG activity as related to the startle reflex on the one hand, and EMG activity as related to the concurrent head movement in the HR–startle trials on the other hand, we used trials in which EMG onset latencies could be determined and a startle reaction had occurred. Therefore, muscle responses were included for analysis when onset latencies occurred before a pre-defined cut-off point in HR–startle trials. These cut-off points were based on the fastest reactions (1st percentile) to a non-startling stimulus in the particular muscle, to ensure the exclusion of non-reflexive voluntary movements. This meant that we only included trials for analysis when the onset latencies in HR–startle trials were shorter than the fastest reactions in the HR–non-startle trials (83 ms for the SCM and 80 ms for the cervical paraspinal muscles). The use of these cut-off points led to a probability of occurrence of startle reflexes ranging from 33% to 35% in the paraspinal muscles and 37% to 48% in the SCM. We calculated the probability of occurrence of a startle reflex for the paraspinal and the SCM in each subject by dividing the number of startle reflexes by the total number of trials in which an additional startling stimulus was administered.

Statistical analyses

We identified onsets of head movement in all experimental trials. EMG onset latencies were also determined in all trials for the SCM and paraspinal muscles at both sides. We applied a log-transformation to movement onsets and EMG onset latencies to correct for the observed skewed distributions of the data.

For analysis of movements, data values of all trials were incorporated in a linear mixed model with random intercept, to assess differences in onset of head movement related to: stimulus type (startle only, non-startle only, HR only or HR–startle in the validating experiment; HR–startle or HR–non-startle in the main experiment),

direction of the auditory stimulus (left or right), direction of head rotation (left or right), task (simple RT or choice RT), and their interactions. For analysis of the EMG-defined startle reactions (based on the previously described cut-off points), we performed additional linear mixed-model analyses to evaluate differences in EMG onset latencies and amplitudes, related to direction of the auditory stimulus (left or right), muscle side (left or right) and their interaction. The probability of occurrence of a startle reaction was analysed using a similar mixed model, but with a Bernoulli distribution function. Differences with a P value of less than 0.05 were considered significant. Values are given as mean \pm interquartile range for the kinematic data values and as mean \pm s.d. for EMG data values.

Results

Validating experiment

Motion analysis. When subjects received the low-intensity acoustic stimulus (non-startle only), any head movements were small and did not reach threshold for onset detection. Therefore, we never recorded onsets of head movement in left/right directions. When subjects only received the high-intensity acoustic stimulus (startle only), early head movements with an average onset of 166 ± 79 ms occurred in 27% of the trials. The top panel in Fig. 3A shows the data for the trial with the fastest recorded head movement (in the left–right direction). Note that the peak head velocity (about 0.1 m s^{-1}) is much smaller than the peak velocity for the conditions ‘HR only’ and ‘HR startle’. Linear head movements were never faster for up–down and forward–backward directions than for the left–right directions.

In the HR only condition, the average onset of the head movements was 403 ± 82 ms. Onsets could be determined in 97% of the trials (Fig. 3A). In the HR–startle condition, the average onset of head movement was 313 ± 114 ms and the probability of occurrence was 98%.

EMG. Startle only trials clearly induced startle responses in the EMG at latencies of about 60 ms in the neck muscles (Table 1). In contrast, ‘non-startle only’ trials did not evoke any detectable EMG activity in any neck muscle.

In the HR only condition, EMG latencies were over 300 ms. This is much *later* than the latencies of EMG activity in response to ‘startle only’ conditions (Table 1). Moreover, head movements to the left or right distinctly evoked lateralized EMG responses. The right SCM and the left paraspinal muscle showed significantly higher amplitudes for head rotation to the left (Table 1) and vice versa.

When head rotations were performed in combination with a startling acoustic stimulus (HR–startle), onset

latencies of EMG-defined startle reflexes were not different from the Startle only trials ($P = 0.77$; Table 1). In the startle conditions, there were no significant lateralized EMG responses to different HR directions ($P = 0.90$; interaction IS side \times muscle side in the HR–startle condition).

Main experiment

StartReact in simple and choice RTs. For the simple RT, head rotations started significantly earlier during HR–startle trials compared with HR–non-startle trials (Figs 4A and 5). The median reduction in onset of head rotation (i.e. the StartReact effect) was 45 ms ($P < 0.001$).

For the choice RT, head rotations also started significantly earlier during HR–startle trials compared with HR–non-startle trials (Fig. 5), resulting in a median StartReact effect of 56 ms ($P < 0.001$). As expected, head movements had, irrespective of stimulus type, a significantly later onset during the choice RT compared with the simple RT (mean difference, 88 ms; $P < 0.001$).

Directional sensitivity of the StartReact effect. The StartReact effect was present when the accompanying acoustic stimulus was administered from both the same side (ipsilateral) and the opposite side (contralateral) of the IS (solid lines in Fig. 6A and B).

Although the non-startling acoustic stimulus was not capable of eliciting startle responses, onsets of head movement in the HR–non-startle condition were significantly faster for ipsilateral stimuli compared with contralateral stimuli (Fig. 6; $P < 0.001$). This suggests that we recorded an effect of an acoustic stimulus in addition to a visual stimulus as in intersensory facilitation (Nickerson, 1973; Schmidt *et al.* 1984), with a more pronounced effect of the complementary stimulus in ipsilateral trials compared with the conflicting stimulus in contralateral trials.

For the choice RT, this StartReact effect was not significantly different for ipsilateral compared with contralateral acoustic stimuli (52 *versus* 50 ms, $P = 0.18$; Fig. 6B).

For the simple RT, the StartReact effect was significantly smaller ($P < 0.005$) for ipsilateral (median 38 ms) compared with contralateral (median reduction in onset of 49 ms) acoustic stimuli (Fig. 6A). This appeared to be caused by a floor effect for two reasons. First, median onsets for HR–startle trials were almost identical for ipsilateral (154 ms; interquartile range, 134–182) and contralateral (157 ms; interquartile range, 136–180) acoustic stimuli. This is remarkable because we would have expected the complementary stimuli in ipsilateral trials to lead to faster head movements compared with the conflicting stimuli in contralateral trials, as was observed in the HR–non-startle condition. Moreover, inspection of the distribution of response times for the ipsilateral acoustic stimuli showed

an asymmetry with positive skewing towards faster onset latencies, supporting the presence of a floor effect.

Response errors. A total of 119 response errors were made out of 3072 trials during the experiment. The number of errors varied with the type of reaction time test, stimulus intensity and side of the stimulus. Most errors arose in the choice RT, particularly in the contralateral HR–startle trials. In the choice RT, we found significantly more errors than in the simple RT (11%; $P < 0.001$). Furthermore, we observed significantly more errors in HR–startle trials compared with HR–non-startle trials (6%; $P = 0.001$), as well as in trials in which the stimulus was delivered contralateral to the side of head rotation compared with trials in which the stimulus was delivered ipsilateral to the side of head rotation (9%; $P < 0.001$).

Acceleration of EMG onset latencies. Onset latencies in SCM and paraspinal muscles occurred significantly earlier in HR–startle trials than in HR–non-startle trials. In the simple RT, the median onset latency for the HR–startle condition was 87.94 ms earlier in the SCM ($P < 0.001$) and 61.21 ms earlier in the paraspinals ($P < 0.001$), compared with the HR–non-startle condition (Fig. 3B). In the choice RT, the median onset latency for the HR–startle condition was 149.62 ms earlier in the SCM ($P < 0.001$) and 109.37 ms earlier in the paraspinal muscles ($P < 0.001$), compared with the HR–non-startle condition.

For the simple RT, EMG onset latencies could be determined in 93.8% of trials for the SCM and in 96.8% of trials for the paraspinal muscles. In choice RT trials the probability of occurrence was lower: 91.0% for the SCM

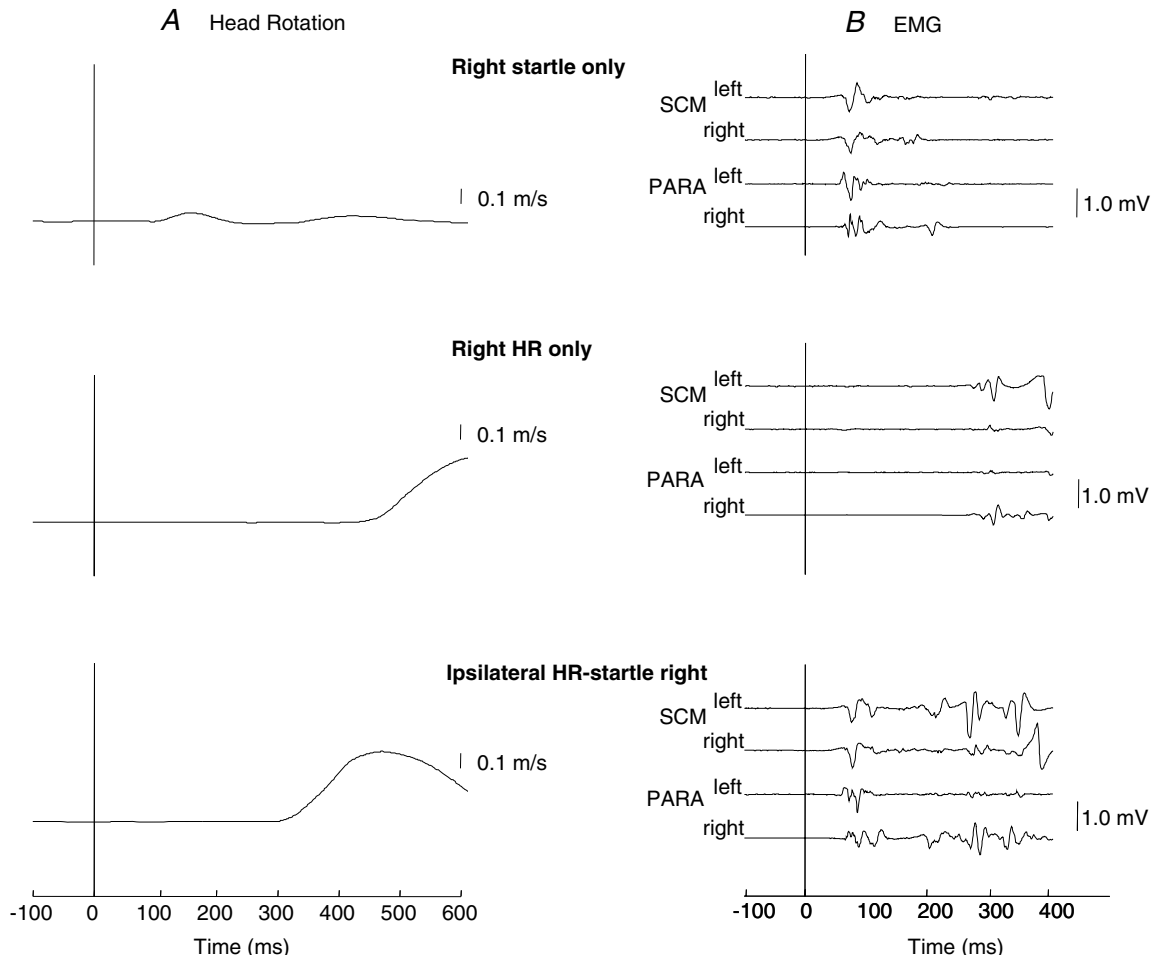


Figure 3. Examples of kinematic and EMG recordings in a startle only, HR only and HR–startle trial

Kinematic head movements (A), as well as EMG recordings of the left and right sternocleidomastoid and cervical paraspinal muscles (B) of a single subject in the validation experiment. From top to bottom, the conditions displayed are: ‘startle only’ delivered from the right side, a ‘HR only’ to the right side and a right (ipsilateral) HR–startle trial. The vertical lines through all traces at 0 ms indicate the onset of the imperative stimulus and/or the acoustic stimulus.

Table 1. Validating experiment: mean (s.d.) EMG onset latency (ms), amplitude (mV) and probability of occurrence (%) in the sternocleidomastoid and paraspinal muscles for startle only trials, head rotation only trials and trials with combined head rotation and startle (HR–startle)

		Latency	Amplitude	Probability
Startle only (from the right*)	SCM left	64.24 (8.74)	2.43 (2.94)	83%
	SCM right	64.28 (7.94)	1.80 (1.90)	81%
	PARA left	61.43 (6.33)	2.89 (3.19)	67%
	PARA right	62.70 (7.49)	2.01 (1.65)	71%
Non-startle only	—	—	—	—
HR only (to the right)*	SCM left	336.19 (40.88)	3.50 (1.50)	73%
	SCM right	333.95 (39.62)	0.77 (0.48)	65%
	PARA left	331.35 (51.12)	0.90 (0.42)	60%
	PARA right	320.27 (49.92)	2.63 (1.65)	68%
HR–startle right* (EMG-defined startles)	SCM left	65.45 (8.32)	3.08 (5.17)	92%
	SCM right	63.46 (10.18)	2.04 (3.10)	94%
	PARA left	62.87 (8.65)	2.37 (2.98)	81%
	PARA right	63.47 (9.05)	2.57 (2.56)	77%
Mixed model <i>P</i> values				
Stimulus (startle only/HR–startle)		—	—	—
Head only IS side × muscle side		—	***	—

The table shows the data as function of stimulus type (Startle only, Non-startle only, HR only and HR–startle), stimulus side (*only the right side is depicted here, as data values for the left side were comparable) and muscle side (left, right). No increase in EMG activity was recorded to the non-startle only stimulus. In the HR–startle condition only the EMG-defined startles, trials in which the onset latency preceded 83 ms for the sternocleidomastoid (SCM) or 80 ms for the paraspinal muscles (PARA), were included. The lower portion of the table summarizes results of the mixed model analyses for the log-transformed data as function of the type of stimulus, comparing the startle only condition with the HR–startle condition. Furthermore, results of log-transformed data as function of direction of the head rotation and muscle side are shown for the HR only condition. *** $P < 0.001$.

and 92.8% for the paraspinal muscles. A typical reason for rejection of trials (6.4% of all trials) was an absence of a response within 400 ms of stimulus onset or pre-stimulus EMG activity, which was more likely to occur in the later responses in choice RT.

Directional sensitivity of the EMG-defined startle reflex.

When using cut-off points to try and discriminate the startle reflex from the activity due to the voluntary head movements, onset latencies and probabilities of responses did not significantly differ for muscle side, direction of the startling acoustic stimulus and their interaction (Tables 2 and 3). Onset latencies in the left and right muscles were clearly independent of the direction of the startling acoustic stimulus, as we found no significant interaction effect between muscle side and side of the acoustic stimulus in both the SCM ($P = 0.64$) and paraspinal muscles ($P = 0.85$). This was also found for the probability of occurrence in the SCM ($P = 0.78$) and paraspinals ($P = 0.59$). Separate analyses of the HR–startle trials in the simple RT and in the choice RT resulted in similar findings. Furthermore, the use of different cut-off points between 70 and 100 ms for the inclusion of startle reflexes

in both the SCM and paraspinal muscles did not affect the results.

Use of the cut-off points naturally led to lower probabilities of occurrence for HR–startle trials in the SCM (44%) and in the paraspinals (31%). However, all subjects demonstrated startle responses in the SCM, paraspinals or both, in at least one HR–startle trial.

Muscle response amplitudes in both simple RT and choice RT were significantly higher for HR–startle trials compared with HR–non-startle trials (Table 1) in both SCM ($P < 0.001$) and paraspinal ($P < 0.001$) muscles. However, response amplitudes in HR–startle trials were not related to the side of the startling acoustic stimulus. For example, the right SCM displayed higher activity compared with the left SCM for startling stimuli from the left, whereas the exact opposite occurred for the paraspinal muscles. Although these differences were not significant for the simple RT in SCM ($P = 0.73$) and paraspinals ($P = 0.53$) as well as for the choice RT in SCM ($P = 0.70$) and paraspinals ($P = 0.69$), the recorded amplitudes suggest a relation to the side of the head rotation. Differentiation between the startle reflex itself and the concurrent fast head rotation is, however, not easy to be made using EMG. We calculated amplitudes over an

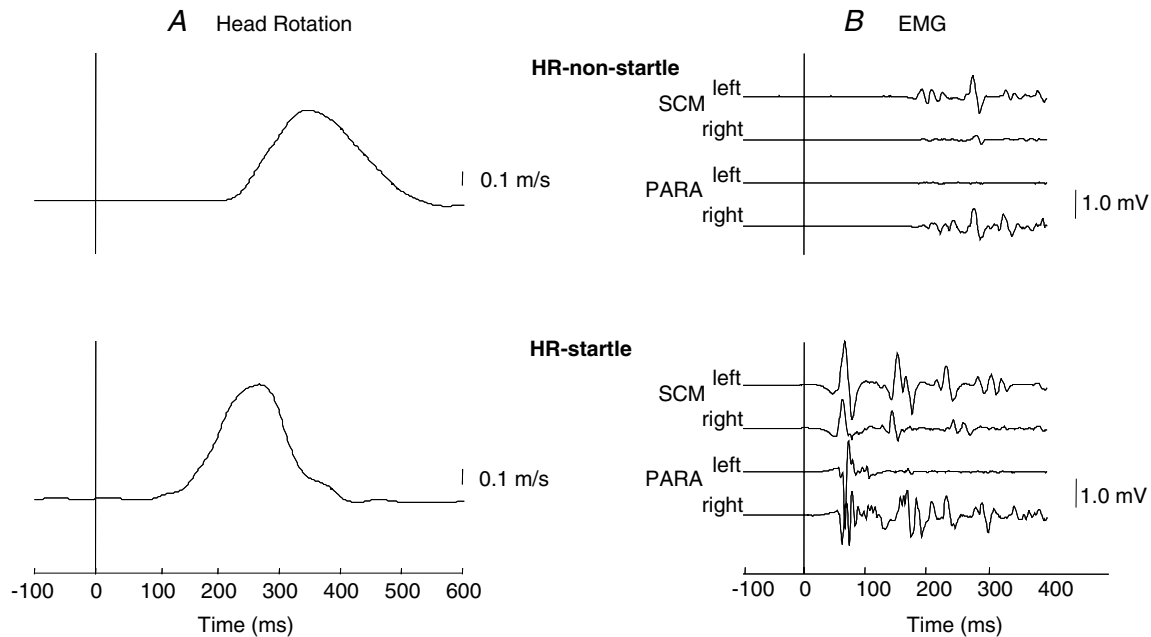


Figure 4. Examples of kinematic and EMG recordings from a HR–non-startle and HR–startle trial

Kinematic head movements (A), as well as EMG recordings of the left and right sternocleidomastoid and paraspinal muscles (B) of a single subject in the main experiment. In the upper illustrations a head rotation to the right in combination with a non-startling acoustic stimulus (HR–non-startle) from the left side is displayed in the simple reaction time protocol. In the bottom illustrations a similar rotation in combination with a startling acoustic stimulus (HR–startle) from the left is displayed. The vertical lines through all traces at 0 ms indicate the onset of the imperative stimulus.

interval of 50 ms after onset, which in some cases might not only have led to capturing activity of the startle reflex, but also part of the activity of the concurrent fast head rotation.

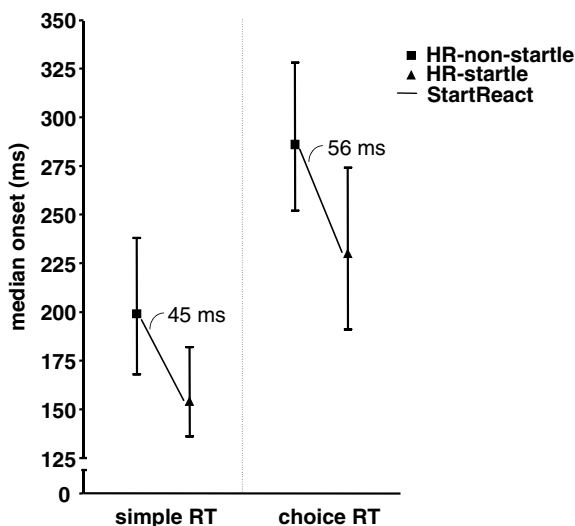


Figure 5. The StartReact effect in simple and choice RT

Onsets of head in movements HR–startle trials, compared with onsets when head movements were combined with non-startle (HR–non-startle) in a simple reaction time task (on the left) and in a choice reaction time task (on the right). Bars represent median onsets and interquartile ranges.

We additionally calculated ratios and arithmetic differences in EMG amplitude in the different conditions, and found similar results as described by Siegmund *et al.* (2001) (Fig. 7). This points to a summation of startle responses upon the preserved voluntary activation, and not towards a multiplicative effect.

Discussion

We demonstrated that startling acoustic stimuli advanced the onset latency of head rotations, not only in simple, but also in choice reaction time tasks. However, the direction of the startling acoustic stimulus did not influence the StartReact effect or the startle reflex.

The StartReact effect is present in choice reaction time tests

Previous research demonstrated that acoustic startling stimuli could accelerate the onset of simple ballistic movements by advancing the onset of EMG activity in the prime movers, while preserving the characteristic triphasic agonist–antagonist relationship. This finding is best explained by a direct effect of the startle response on reticulospinal pathways within the brainstem, where motor programs that have been prepared in advance

Table 2. Main experiment: mean (S.D.) EMG onset latency (ms), amplitude (mV) and probability of occurrence (%) for the simple RT in the sternocleidomastoid and paraspinal muscles

			Latency	Amplitude	Probability
All trials	HR–non-startle	SCM	166.83 (53.95)	1.98 (2.27)	n.a.
		PARA	165.95 (55.99)	2.59 (3.00)	n.a.
	HR–startle	SCM	102.62 (46.66)	3.03 (4.01)	n.a.
		PARA	104.83 (44.74)	4.09 (4.59)	n.a.
EMG-defined startles	HR–startle from the right*	SCM left	62.02 (11.39)	2.85 (3.24)	41%
		SCM right	62.53 (9.93)	3.15 (4.09)	37%
		PARA left	61.35 (8.54)	4.66 (4.76)	35%
		PARA right	63.76 (9.10)	5.56 (4.90)	33%
Mixed model analyses					
All trials	Stimulus (HR–non-startle/ HR–startle)	SCM	***	—	n.a.
		PARA	***	—	n.a.
EMG-defined startles	Startle side (left/right)	SCM	—	—	—
		PARA	—	—	—
	Muscle side (left/right)	SCM	—	—	—
		PARA	—	—	—
	Startle side × muscle side	SCM	—	—	—
		PARA	—	—	—

The table summarizes data as function of stimulus (HR–non-startle and HR–startle) and muscle (SCM and PARA) in all trials. This is followed by data as function of side of the startling stimulus (*only the right side is depicted here, as data values for the left side were comparable) and muscle side (left, right) in EMG-defined startles. EMG-defined startles included trials in which the onset latency preceded 83 ms for the SCM or 80 ms for the paraspinal muscles (PARA). Results of the mixed model analyses are shown for the log-transformed data as function of type of stimulus and muscle in all trials, followed by results of log-transformed data as function of the side of the startling stimulus and muscle side in the EMG-defined startles. ****P* < 0.001; n.a., not applicable.

are being stored or transmitted. (Valls-Solé *et al.* 1999; Siegmund *et al.* 2001; Carlsen *et al.* 2003, 2004a,b). Our results indicate that such an interaction is also present for planned movements involving choices.

For choice reaction time tasks, conflicting findings have been reported, perhaps due to differences in experimental design. Carlsen *et al.* instructed their subjects to actively flex or extend the wrist of either the left or the right

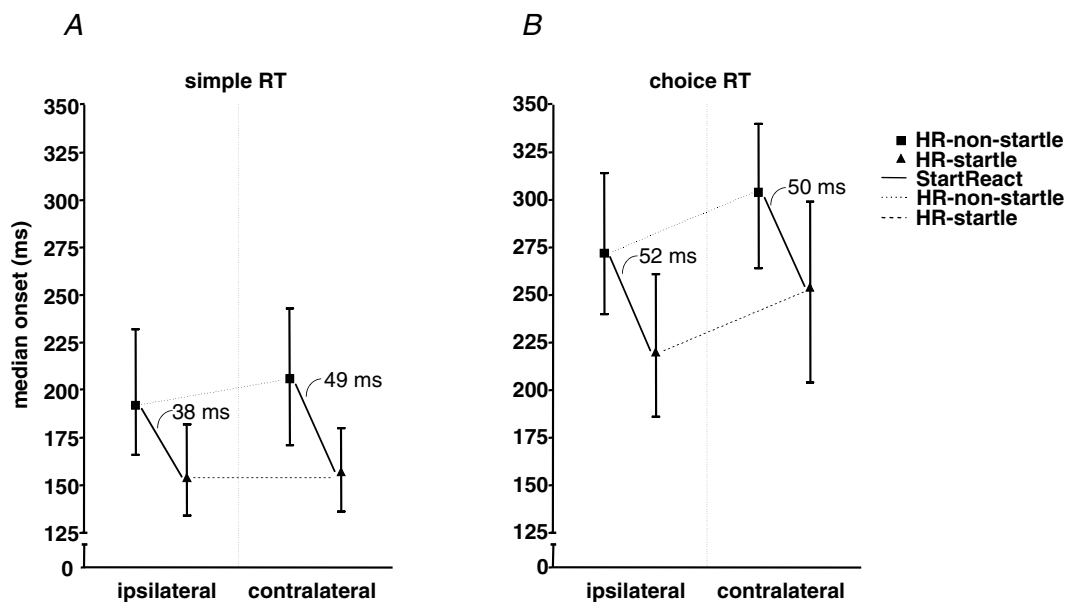


Figure 6. The StartReact effect for acoustic stimuli ipsilateral and contralateral to the head rotation
Onsets of head movements in HR–startle trials and HR–non-startle trials in which the auditory stimulus was administered ipsilateral or contralateral to the side of the imperative stimulus in a simple reaction time task (A) and in a choice reaction time task (B). Bars represent median onsets and interquartile ranges.

Table 3. Main experiment: mean (s.d.) EMG onset latency (ms), amplitude (mV) and probability of occurrence (%) for the choice RT in the sternocleidomastoid and paraspinal muscles

			Latency	Amplitude	Probability
All trials	HR–non-startle	SCM	244.87 (57.44)	1.65 (1.92)	n.a.
		PARA	239.57 (57.58)	2.20 (2.38)	n.a.
	HR–startle	SCM	132.38 (88.69)	2.07 (2.68)	n.a.
		PARA	130.20 (74.09)	2.78 (3.38)	n.a.
EMG-defined startles	HR–startle from the right*	SCM left	57.77 (10.92)	2.02 (2.52)	48%
		SCM right	60.83 (10.31)	2.02 (2.81)	45%
		PARA left	63.07 (7.79)	3.82 (4.61)	35%
		PARA right	64.22 (7.72)	3.28 (3.62)	33%
Mixed model analyses					
All trials	Stimulus (HR–non-startle/ HR–startle)	SCM	***	—	n.a.
		PARA	***	—	n.a.
EMG-defined startles	Startle side (left/right)	SCM	—	—	—
		PARA	—	—	—
	Muscle side (left/right)	SCM	—	—	—
		PARA	—	—	—
	Startle side × muscle side	SCM	—	—	—
		PARA	—	—	—

The table summarizes data as function of stimulus (HR–non-startle and HR–startle) and muscle (SCM and paraspinals) in all trials. This is followed by data as function of side of the startling stimulus (*only the right side is depicted here, as data values for the left side were comparable) and muscle side (left, right) in EMG-defined startles. EMG-defined startles included trials in which the onset latency preceded 83 ms for the SCM or 80 ms for the PARA muscles. Results of the mixed model analyses are shown for the log-transformed data as function of type of stimulus and muscle in all trials, followed by results of log-transformed data as function of the side of the startling stimulus and muscle side in the EMG-defined startles. *** $P < 0.001$; n.a., not applicable.

hand (Carlsen *et al.* 2004a). The results showed that simple reaction time tasks were clearly accelerated during startle trials compared with non-startle trials (indicating a StartReact effect), but this effect was completely absent during the two conditions that involved either two or four different choices. One possible drawback was the use of rather loud (intensity of 80 dB) auditory stimuli in non-startle trials, which might have elicited startle reflexes (Blumenthal, 1988; Carlsen *et al.* 2007). Furthermore, no results of kinematic recordings were reported, so it remains possible that existing effects on the movement itself were missed. Consistent with Carlsen *et al.* (2004a), we used a visual imperative stimulus. However, in the literature the use of an acoustic ‘go’ signal, which is replaced by the startling acoustic stimulus, has also been reported (Siegmund *et al.* 2001; Carlsen *et al.* 2003, 2007). Note that this might have resulted in different results.

In a comparable study, Valls-Solé (2004) measured both EMG responses and kinematic profiles. The task in this experiment consisted of a movement with either the left or the right wrist, depending on the side where an imperative stimulus appeared. This was done for both simple and choice reaction time tasks. The main finding was that reaction times were similarly shortened by concurrent acoustic startling stimuli for simple and choice reaction

time tasks. The shortening of reaction times was also reported for the EMG activity with an advancement of the well-known triphasic agonist–antagonist activation pattern. The presence of this StartReact effect for a choice RT might be explained by a globally increased neural excitability and decreased neural thresholds, taking place at the cortical level where movement decisions are made (Carlsen *et al.* 2004a). However, one argument against this hypothesis is that simple stimulus intensity effects are different from onset latencies produced by startle (Carlsen *et al.* 2007). Another argument is that subjects made movement errors in about one third of the trials. This was interpreted as subjects actually solving the task by suppressing one of two movement options that were waiting in place within the reticulospinal pathway. Several movement programs may simultaneously be ‘stored’ in the brainstem where they can be accessed by startling acoustic stimuli. However, one possible critique is that mirror movements were observed in 92% of the trials. This could be taken as further evidence for the presence of two parallel and pre-programmed movement options within the brainstem, which are both released by the acoustic startling stimulus, but to a lesser extent for the unwanted movement. However, these mirror movements may also have occurred because the imposed task was perhaps too

simple, as there was no ‘penalty’ for making an incorrect choice (the desired wrist movement would not be harmed by co-movement of the contralateral arm).

We ruled out the latter option in our present study, where subjects were forced to make an axial rotation

movement with the head to either the left or the right. Furthermore, we used neck muscles because startle reflexes could be elicited reliably in these muscles (Brown *et al.* 1991; Siegmund *et al.* 2001). Neck muscles were also asymmetrically active during rotations to the left or the

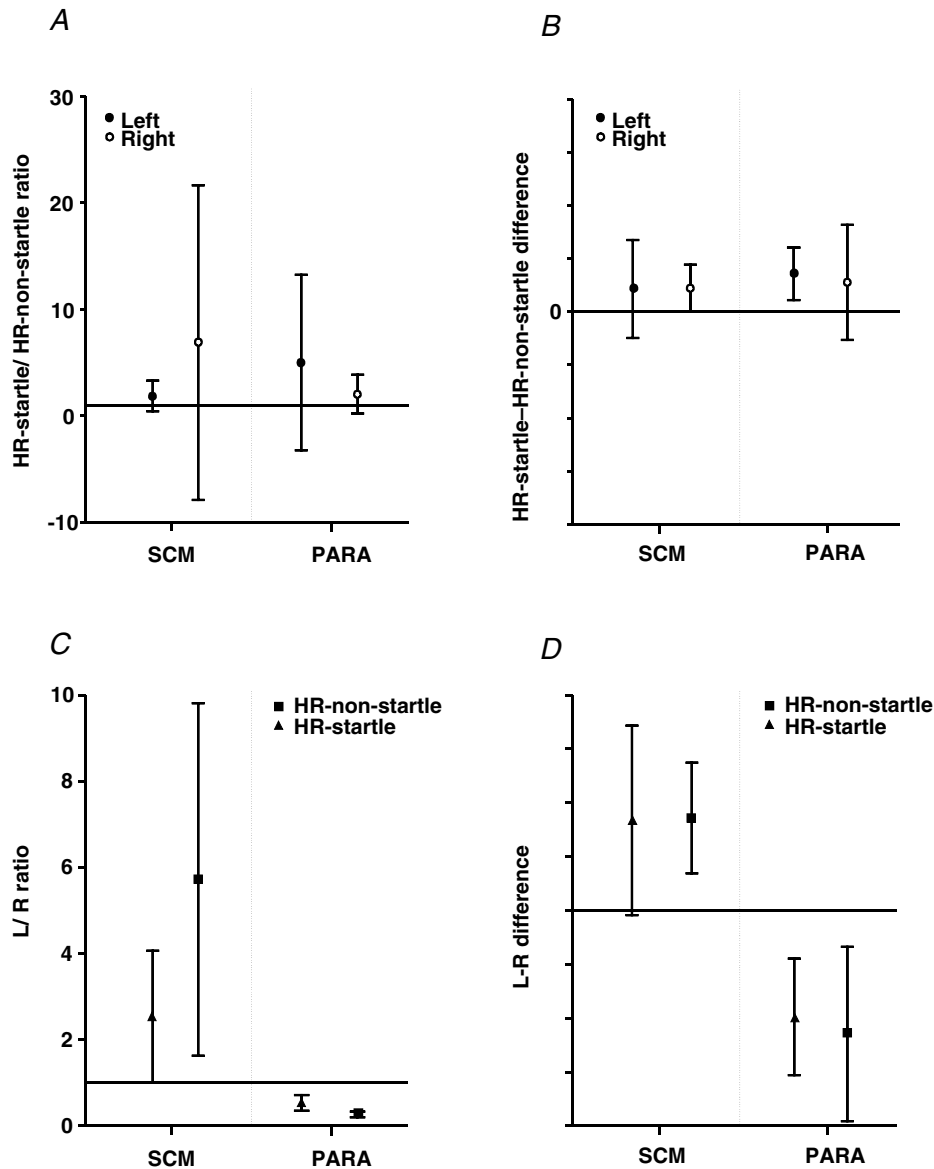


Figure 7. Ratios and differences for the neck muscles of all subjects in the choice RT

A, ratios ± 1 s.d. of the mean area under the curve (AUC) of the EMG amplitude of the HR-startle to the HR-non-startle response (HR-startle divided by HR-non-startle) for each muscle (sternocleidomastoid (SCM) and paraspinal muscles (PARA)), expressed as a function of muscle side (left/right). The EMG amplitudes were taken from the EMG-defined startles for head rotations to the right. B, mean arithmetic difference ± 1 s.d. of the HR-startle and HR-non-startle amplitudes (HR-non-startle minus HR-startle) for each muscle, as a function of muscle side (left/right). Note the consistent additive effect (difference scores close to zero) of startle in the HR-startle condition. C, mean ratio ± 1 s.d. of the left to right amplitudes (L/R) for each functional muscle pair as a function of stimulus. D, mean arithmetic difference ± 1 s.d. of the left and right amplitudes (L-R) for each functional pair as a function of stimulus. The consistent, within-muscle L-R difference indicated that the activity was preserved over the effect of the startle (Siegmund *et al.* 2001).

right (Mazzini & Schieppati, 1992), as confirmed by the EMG activity in the pure head rotations condition. This enabled us to compare left-sided with right-sided muscle activity. However, when looking at the EMG response pattern of startle trials, we recorded similar onset latencies in both agonist and antagonist muscles compared with pure startle reflexes. Furthermore, the amplitudes did not differ significantly, nor were they specific for the direction of head rotation. This suggests that startle responses are perhaps not specific reactions, but simply lead to a fast and generalized increase in muscle tone.

An important issue when interpreting the current data is that the ensuing fast increase in muscle activity makes it difficult to separate the startle reflex from the synchronous voluntary muscle responses in HR–startle trials. On the one hand, the remaining increase in muscle activity suggests that a generalized increase in muscle tone serves as a basis upon which further action may be undertaken. On the other, simultaneous activation of agonist and antagonist muscles (co-contraction) might negatively affect the movement requested by the imperative stimulus. Apparently this was not the case as we found that the requested voluntary head rotations were accelerated in both simple and choice reaction time tasks. Our data suggest that the remaining increase in muscle tone due to startle was relatively small compared with the activity due to the head movement (Table 2) and that the startle reflex therefore acted more upon a fast increase of muscle tone, serving as a basis upon which further action can be undertaken, instead of leading to an inability to move due to co-contraction. Indeed, we found evidence for a summation between startle and voluntary response amplitudes (Fig. 7), as previously described (Siegmond *et al.* 2001). These findings underscore how difficult it can be to separate the early startle reflex from concurrent movement activity when investigating the effect of startle responses on reaction times.

Assuming that the process of response selection is a cortical event, our finding that the StartReact effect is present in choice tasks might be explained by an increase of neural excitability and decreased neural thresholds (Carlsen *et al.* 2004a). The increase in muscle tone would then serve as a basis upon which choices can be executed faster. However, more recent evidence suggests that such stimulus intensity effects are distinct from the early response latencies produced by startle (Carlsen *et al.* 2007). Thus, the effect may be similar, but the mechanism different. The startle reflex led to a quick increase in muscle activity compared with the non-startle trials. As a certain muscle tone is necessary for movement, the rapid increase in muscle activity due to the startle reflex may also lead to the faster concurrent specific movement, explaining the incongruent evidence compared with Carlsen *et al.* (2004a).

The StartReact effect is not directionally dependent

No prior study examined the possible directional dependence of the StartReact effect. We reasoned that a directional dependence of the StartReact effect would provide further support for the hypothesis that the startle reflex has a functional relevance. For example, such directional dependence could assist subjects in the protection against an unpleasant lateralized stimulus (e.g. to move away from perceived danger, or to initiate a targeted protective response with the arms).

We addressed this issue by asking subjects to rotate their head as fast as possible into the direction of an imperative stimulus accompanied by a startling acoustic stimulus from either the ipsilateral side or the contralateral side. The results provided no clear evidence that the StartReact effect was directionally sensitive in either the simple RT or the choice RT. Kinematic analyses of head rotations during the choice RT showed that startling acoustic stimuli induced a very comparable reduction in movement onset, irrespective of the direction of the acoustic startling stimulus. That is, the magnitude of the StartReact effect was comparable even though startling acoustic stimuli were delivered from opposite sides. However, we did observe a difference in magnitude of the StartReact effect for the simple RT task, such that the reduction in movement onset was greatest when the acoustic startling stimulus was delivered from the contralateral side. Onsets of head rotation were reduced less than expected when the task was to move the head towards the side where the startling acoustic stimulus came from (ipsilateral trials). At first sight, this finding could be interpreted as evidence for a directional dependence of the StartReact effect, perhaps because subjects defensively tended to avoid the loud acoustic stimulus by moving less fast into its direction. However, several observations argue against this interpretation. Firstly, we would have expected a similar directional dependence for the choice RT task – where the StartReact effect itself was prominently present – but this was not the case. Secondly, a genuine effect on the kinematic responses should ideally be supported by concurrent reductions in onset of EMG activity (Valls-Solé *et al.* 1999), but this was not observed. Indeed, onsets of EMG responses in the sternocleidomastoid muscle were nearly identical, irrespective of the direction of the accompanying startling stimulus. Furthermore, onset latencies were comparable to the startle only condition in the validating experiment, where startling stimuli were administered without requesting rotation of the head. We therefore favour an alternative explanation, namely that during the simple RT tasks, the fast and similar movement onsets for ipsilateral and contralateral startling stimuli were caused by a floor effect. This explanation is supported by the positively skewed distribution of the reaction times in the startle trials for ipsilateral stimuli. In other words, a

further reduction of movement onsets due to the startling stimulus is likely to be physiologically impossible in the simple RT tasks in which movement onsets were already very fast. In the choice RT tasks, however, where movement onsets were longer, the startling stimulus could further accelerate responses.

Conclusion

We have demonstrated that startling acoustic stimuli can accelerate head movements in a choice reaction time protocol. However, we did not find any effects of the direction of the startling stimulus on the direction of the response. One conjectural reason for the lack of such an effect is that the movement had no direct functional bearing on the stimulus, nor any emotional load. It is conceivable that emotionally laden stimuli, or those that combine a threat with a direction, do affect directional responses. In this respect we cannot exclude the possibility that a directional dependence of the StartReact effect might have been found if the task had included a higher emotional content, as is typically the case in real-life fight-or-flight behaviour (Bradley *et al.* 2005). Such considerations may well form the basis for future experiments to assess the behavioural impact of startling.

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