

Associative learning with and without perceptual awareness

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Abstract

Background: The relationship between awareness and associative learning is a key controversial issue that remains to be elucidated. An experiment was designed to assess associative learning with and without perceptual awareness. **Method:** Participants received repeated trials of two compatible stimuli sequences ($S1_A \rightarrow S2_A$ and $S1_B \rightarrow S2_B$), where $S1$ was a masked stimulus, and $S2$ an imperative stimulus for a reaction time (RT) task. After the acquisition phase, some probe trials of incompatible stimuli sequences ($S1_A \rightarrow S2_B$ and $S1_B \rightarrow S2_A$) were inserted among the compatible sequence trials during two testing sessions. Subsequently, subjects were classified as perceptually aware or perceptually unaware by means of a forced-choice identification task that was administered at the beginning and end of the experiment. **Results:** The results showed perceptually unaware participants responded faster to compatible than to incompatible stimuli sequences. However, no priming effect was observed in aware participants. **Conclusions:** These results are discussed in terms of an S-R associative mechanism and provide strong evidence of unconscious associative learning.

Keywords: Associative learning, unconscious learning, masked priming.

Resumen

Aprendizaje asociativo con conciencia y sin conciencia perceptiva.

Antecedentes: la relación entre la conciencia y el aprendizaje es un importante problema aún no resuelto. A tal fin, fue diseñado un experimento para evaluar el aprendizaje asociativo con conciencia y sin conciencia perceptiva. **Método:** para ello, los participantes recibieron muchos ensayos repetidos de dos secuencias de estímulos compatibles ($E1_A \rightarrow E2_A$ y $E1_B \rightarrow E2_B$), donde $E1$ era un estímulo enmascarado y $E2$ un estímulo imperativo para una tarea de tiempo de reacción. Durante dos sesiones de prueba fueron insertados algunos ensayos de secuencias de estímulos incompatibles ($E1_A \rightarrow E2_B$ y $E1_B \rightarrow E2_A$) entre los ensayos de secuencias de estímulos compatibles. A continuación los participantes fueron clasificados como perceptivamente conscientes o inconscientes mediante una tarea de identificación de elección forzada, efectuada al principio y al final del experimento. **Resultados:** los resultados mostraron que los participantes perceptivamente inconscientes respondían más rápidamente ante las secuencias compatibles que ante las incompatibles. Sin embargo, no se observaron efectos de priming en los sujetos perceptivamente conscientes. **Conclusiones:** los resultados son discutidos en términos de un mecanismo asociativo E-R y constituyen una clara evidencia de aprendizaje asociativo inconsciente.

Palabras clave: aprendizaje asociativo, aprendizaje inconsciente, priming enmascarado.

The verification in the 1990s (Klotz & Neumann, 1999; Neumann & Klotz, 1994) that non-consciously presented stimuli could affect behaviour has raised a renewed and growing interest in the study of unconscious processes in associative learning. The relationship between associative learning and conscious awareness has been studied mainly in the classical conditioning paradigm by presenting the conditioned stimulus (CS) for a few milliseconds (ms) to render it unavailable to conscious perception (subthreshold techniques), or immediately followed by a mask that disrupts its processing (backward masking procedure). Studies have shown that unconscious learning can occur with threat-relevant CSs (Esteves, Parra, Dimberg, & Öhman, 1994), delayed eyeblink conditioning (Clark, Manns, & Squire, 2001; Manns, Clark, & Squire, 2002),

evaluative conditioning (De Houwer, Hendrix, & Baeyens, 1997), or when this learning was measured by event-related brain potentials (Wong, Bernat, Snodgrass, & Shevrin, 2004). However, other studies suggest that awareness of the stimulus contingency is required for conditioning to occur (e.g., Dawson & Furedy, 1976; Mitchell, De Houwer, & Lovibond, 2009).

A possible explanation for these contradictory results may be found in the procedures used to determine contingency awareness between CS and unconditioned stimulus (US). As Lovibond and Shanks (2002) have indicated, most of the studies providing evidence of unaware conditioning have used unreliable or invalid measures of awareness. Moreover, it is plausible to believe that the intensity of the CSs may not be strong enough to trigger a conditioned response (CR) when subthreshold techniques and backward masking procedures are used. Hence, alternative experimental procedures relying on behavioural measures such as spatial cueing (Raes, Koster, Van Damme, Fias, & De Raedt, 2010), motor behaviour (Destrebecqz, Perruchet, Cleeremans, Laureys, Maquet, & Peigneux, 2010; Perruchet, Cleeremans, & Destrebecqz, 2006) and associative priming (Marcos, 2007)

have been implemented in recent years in research on associative unconscious learning.

The associative masked priming paradigm can offer an innovative and more direct means of examining unconscious associative learning. In the response priming paradigm, participants are required to make a forced choice reaction time to a target stimulus preceded by a prime. In compatible pairings, the prime is mapped to the same response as the target. Priming is observed when the latencies of response to target are shortened by a previous presentation of the prime. In incompatible pairings, the prime and target are mapped to the opposite response. As for neutral pairings (where the prime is not mapped to any response), the RTs are expected to be slower and less accurate in incompatible pairings.

Neumann and Klotz (1994) reported several experiments showing response priming effects when the primes were masked, and therefore, inaccessible to perceptual awareness. These results have been substantiated by several recent studies (e. g., Barbot & Kouider, 2012; Mattler & Palmer, 2012; Schlaghecken, Bowman, & Eimer, 2006). Masked primes have been considered to operate through semantic and/or response processes (Kiesel, Kunde, & Hoffman, 2007a; Reynvoet, Gevers, & Caessens, 2005). From a semantic processing framework, it is assumed that the presentation of the masked prime automatically spreads activation in its semantic network, facilitating the response to the target if this stimulus belongs to the same semantic network (Marcel, 1983). Abrams and Greenwald (2000) offer an alternative explanation for response processes that postulates that an S-R link will be formed between a conscious stimulus and a motor response when they are repeatedly associated. Later presentation of this stimulus as a masked prime will activate its associated response and provide the responding if the target requires the same response (see Kiesel, Kunde, & Hoffman, 2007b). Both of the proposed mechanisms assume the previous acquisition of some sort of association between a conscious prime and a target (S-S association in semantic processes), or between a conscious prime and a motor response (S-R association in response processes) to account for the priming effects. Notwithstanding, to our knowledge, there is no mention in the literature as to whether any of these associations can be acquired when the prime is presented outside of awareness.

From this theoretical background, we can hypothesize that repeated pairings of a masked prime with a non-related imperative target stimulus to an RT task will yield associative learning. This learning will be manifest in facilitating the response to target by prime presentation. Facilitation will be detected as shorter RTs to target. If the target requires an opposite response, then the expected RTs to target will be slower and less accurate. Bearing in mind these hypotheses, an experiment was designed to assess associative S1-S2 learning, with and without awareness, using an associative masked priming (AMP) paradigm where S1 was the masked prime, and S2 the target.

Most of the studies on masked response priming have used interstimulus intervals (ISIs) that were under 100 ms. However, priming effects have also been observed with ISIs between 112 ms and 448 ms (Klapp and Greenberg, 2009). In the present experiment, a long ISI (S1 offset/mask onset and S2 onset) of 305 ms was used. With this ISI, the AMP exhibits some parallelism to other associative learning procedures, where S1 and S2 are normally separated by a brief ISI, as in S1-S2 motor learning and classical conditioning.

Method

Participants

Subjects were 60 undergraduate volunteers, aged 18-25 years. All had normal or corrected-to-normal visual acuity. An additional 2 subjects were rejected from later analyses for producing exceptionally high RT error rates, and another for laterality disorders. All participants were naïve as to the purpose of the experiment and gave their informed consent.

Instruments

The stimuli were similar to those already used in previous experiments by Marcos (Marcos, 2007, 2011): the letter 'o' for S1_A and the letter 'x' for S1_B, printed in Arial font, with a visual angle of approximately 0.95° (vertical) × 0.86° (horizontal). S2 consisted of a right-pointing arrow (>) for S2_A and a left-pointing arrow (<) for S2_B, subtending a visual angle of 1.05° (vertical) × 1.14° (horizontal). The stimuli were presented on a grey background (9.04 cd/m²) in the centre of a Sony VGA colour monitor, with 85 Hz refresh rate. S1 stimuli were drawn in grey (17.10 cd/m²), and S2 in white (24.54 cd/m²). The contrast of S1 was reduced to favour masking. The mask was a square, measuring 1.43° × 1.43°, filled with a random texture that was created in each trial to avoid perceptual learning of the mask, which could result in an increased ability to discriminate S1 stimuli. Stimulus presentation and data collection were controlled by Psych Toolbox software (Brainard, 1997), using an IBM-compatible personal computer.

Procedure

Participants were seated in front of the computer screen, at a distance of approximately 60 cms. No chin support device was used, but they were instructed not to change position during the experiment. The participants were told to look at the centre of the screen and press as fast and accurately as possible the "L" key when the arrow appeared pointed toward the right side (>) of the screen, and the "D" key if the arrow pointed to the left (<).

The experiment was carried out in four phases that were distributed into two experimental sessions:

- 1) Previous assessment of perceptual awareness. A forced-choice discrimination task was employed to assess perceptual awareness of the "o" and "x" masked stimuli (S1), with two objectives: (a) to classify each participant as perceptually aware or unaware of S1 stimuli during the following phase; and (b) to measure individual *d'* values. For this purpose, 100 trials of compatible and incompatible sequences were presented using the same parameters as those of the acquisition and testing phases. The participants were informed that immediately before the presentation of a square filled with a texture (mask), the letter "o" would appear in 50% of the trials and the letter "x" in the other 50%. The participants were to indicate which of the two letters appeared in each trial by pressing the "o" key or the "x" key. The instructions stressed that they should notice any minor component or differences in low level features for responding, without needing a complete discrimination between the letters.

2) Acquisition phase. After the discrimination task, the participants received 250 pairing trials, grouped into 5 blocks, of two compatible stimuli sequences ($S1_A \rightarrow S2_A$ and $S1_B \rightarrow S2_B$), where $S1$ was a masked prime stimulus, and $S2$ an imperative target stimulus to an RT task. To avoid contingency awareness of the $S1$ - $S2$ relationship, $S1$ was presented below the perceptual threshold, using a backward masking procedure. In each block, 50 trials of compatible stimuli sequences (25 trials of $S1_A \rightarrow S2_A$ and 25 of $S1_B \rightarrow S2_B$) were presented. The assignment of $S1$ - $S2$ was counterbalanced in compatible sequences: half of the participants were trained with “o/>” and “x/<”, and the other half with “o/<” and “x/>” stimulus sequences. The stimulus sequences were presented randomly.

As depicted in Figure 1, each trial began with the presentation of the fixation cross for 305 ms, followed by a 200 ms blank screen, after which $S1$ was presented for 23 ms (two refresh rates). When $S1$ was over, the mask appeared for 105 ms and was followed by a 200 ms blank screen. Subsequently, $S2$ was presented for 105 ms and the RT to this stimulus was recorded. The typing of the key indicated the end of the trial. The intertrial interval (ITI) oscillated randomly between 2 and 3 seconds. After each block, a pause of 1-2 minutes was introduced, and the experimental session ended on completion of the fifth block. One or two days later, the participant returned to the laboratory to begin the next phase.

3) The testing phase. To determine if the RT change was truly a result of AMP, some testing sequences of incompatible stimuli ($S1_A \rightarrow S2_B$ and $S1_B \rightarrow S2_A$) were inserted during the pairing trials. The incompatible sequences resulted from crossing the stimuli of the compatible sequences. Thus, this phase consisted of two testing sessions of 250 pairing trials each, distributed into 5 blocks. Each block consisted of 42 trials of compatible stimuli sequences (21 trials of $S1_A \rightarrow S2_A$ and 21 trials of $S1_B \rightarrow S2_B$), and 8 trials of incompatible sequences (4 trials of $S1_A \rightarrow S2_B$ and 4 trials of $S1_B \rightarrow S2_A$) that were inserted to test associative priming effects. All stimuli sequences were presented randomly, using the same parameters as those of the acquisition phase. After the first testing session, participants were allowed to leave the experimental cabin and rest for the 30 minutes before beginning the second testing session.

4) Final assessment of perceptual awareness. After finishing the second testing session, a new forced-choice discrimination task, identical to the previous discrimination task, was administered to assess perceptual awareness during the testing phase and to classify the subjects as perceptually aware or perceptually unaware of $S1$ presentation. According to this classification, the effect of awareness on AMP can be

assessed, as can any differences between unconscious and conscious learning.

Data analysis

According to a binomial distribution and $p = 0.5$, the chance level for 100 trials ranged from >40 to <60 correct responses. Thus, participants with a number of correct responses between these values in the two forced-choice discrimination tasks were considered to be perceptually unaware. The remaining participants were classified as perceptually aware.

The data of the forced-choice measures of perceptually unaware and aware participants were also analysed separately, using d' indices of signal detection theory. These measures were obtained from the second discrimination task that assessed awareness after the testing sessions. The d' measures were calculated by treating one level of the response (i.e., letter “o”) as signal and the other level (i.e., letter “x”) as noise. A mean d' score not significantly different from zero indicated chance performance, and therefore, no evidence of conscious discrimination between the two $S1$ stimuli.

RTs to target of compatible stimuli sequences were compared with the RTs of incompatible sequences. RTs with standard deviations 2.5 above or below the mean, and the RTs of incorrect keypress responses were discarded from later analyses. Analyses were performed on 95.57% of the trials.

Results

Data of the forced-choice discrimination task indicated that 28 participants were perceptually unaware and 32 perceptually aware. Mean d' values were 0.07 for unaware participants, and 1.65 for aware participants. Perceptual awareness was significantly higher for perceptually aware participants [$t(58) = 4.36, p < 0.01$]. The t -tests against the null mean showed that the mean d' value obtained for the perceptually unaware participants was not significantly different from 0 [$t(27) = 1.88, p > 0.05$], indicating no evidence of conscious discrimination between $S1$ stimuli. Mean d' value of perceptually aware participants showed correct choice above chance [$t(31) = 4.92, p < 0.01$], indicating conscious discrimination between the stimuli.

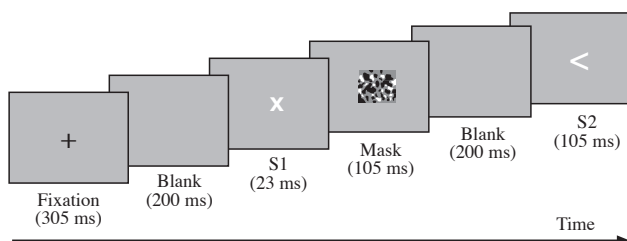


Figure 1. Sequence of displayed events for a standard trial

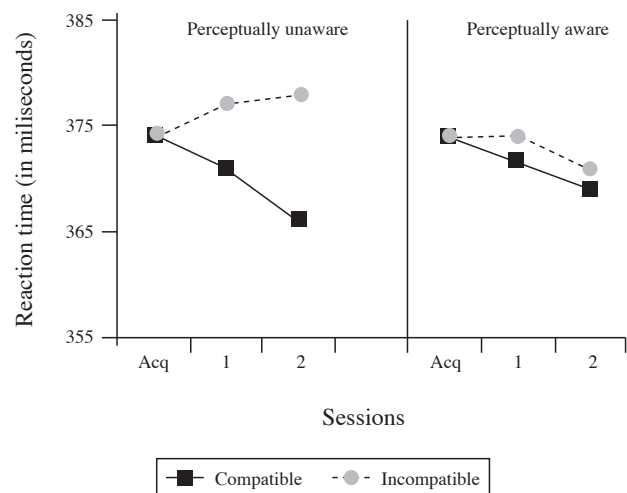


Figure 2. Mean reaction time to $S2$ in acquisition (acq) and testing sessions (1 and 2) for perceptually unaware and perceptually aware participants

Mean RT of the trial blocks of the testing sessions were subjected to a mixed-model ANOVA, with perceptual awareness (aware - unaware), compatibility (compatible - incompatible), testing session (Session 1 and Session 2), and blocks (5) as factors. The analysis revealed that the main effect of compatibility was significant [$F(1/58) = 30.55, p < 0.01, \eta_p^2 = 0.34$], showing faster RTs in compatible ($M = 369, SD = 39$) than incompatible ($M = 375, SD = 36$) trials. Moreover, the interaction between awareness and compatibility reached significance [$F(1/58) = 10.37, p < 0.01, \eta_p^2 = 0.15$]. Comparisons between the RTs of aware and unaware participants to compatible [$t(58) = 0.27, p > 0.05$] and incompatible stimuli sequences [$t(58) = 0.28, p > 0.05$] showed no significant differences. However, this interaction can be observed best by considering only the RTs of Session 2, because these results represent the accumulative effect of learning along the acquisition and testing phases. As shown in Fig. 3, the RTs of aware participants to compatible sequences ($M = 369; SD = 38$) were slightly longer than the RTs of unaware participants ($M = 366; SD = 45$). In contrast, the RTs to incompatible sequences were shorter in aware ($M = 371; SD = 29$) than unaware participants ($M = 378; SD = 43$). This underscores that the effects of compatibility (facilitation), and incompatibility (interference) were greater in unaware participants. Though independently, neither of these effects showed significant differences, the effects of both combined explained the interaction obtained. The remaining factors and interactions were not significant.

Separate analyses were carried out to assess the effect of compatibility on each perceptual awareness group. The factors included in the analyses were compatibility (2), testing session (2), and blocks (5), with repeated measures on the three factors. The results of these ANOVAs showed that compatibility had a highly significant effect on the perceptually unaware group [$F(1/27) = 83.02, p < 0.01, \eta_p^2 = 0.75$], indicating faster RTs to compatible ($M = 369, SD = 41$) than to incompatible ($M = 378, SD = 39$) stimuli sequences. Notwithstanding, compatibility had no effect on the RTs of perceptually aware participants [$F(1/31) = 1.91, p > 0.05$]. Neither the remaining factors nor any of the interactions reached significance in either of the two groups.

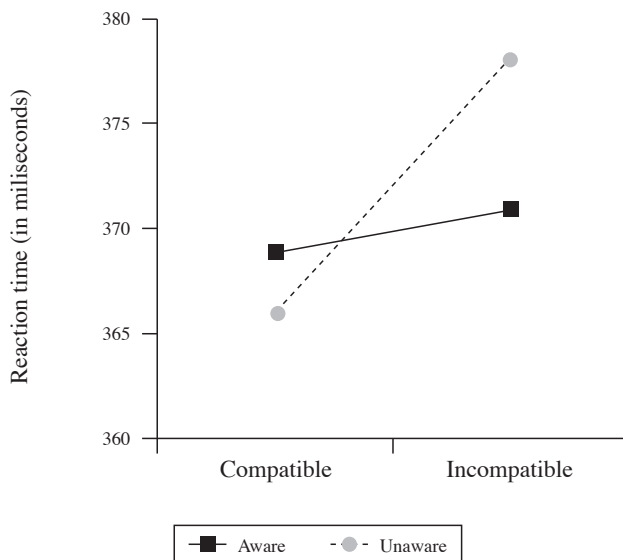


Figure 3. Mean reaction time to S2 in testing session 2 as a function of perceptual awareness and compatibility

| | | Compatible | | Incompatible | |
|------------|--------|------------|------|--------------|------|
| | | M | Sd | M | Sd |
| P. aware | Ses. 1 | 2.87 | 2.59 | 3.59 | 6.34 |
| | Ses. 2 | 2.73 | 2.66 | 3.59 | 5.23 |
| P. unaware | Ses. 1 | 2.61 | 2.38 | 3.04 | 3.22 |
| | Ses. 2 | 2.89 | 3.22 | 4.64 | 5.30 |

The rates of incorrect responses (in %) were subjected to a $2 \times 2 \times 2$ ANOVA (Perceptual awareness \times Compatibility \times Testing session). The results revealed that none of the factors affected the error rate, and the interactions between these factors were not significant. It is worth noting, however, that the main effect of compatibility was close to significance [$F(1/58) = 3.53, p = 0.06$], with a higher error rate in incompatible ($M = 3.75, SD = 4.65$) than compatible ($M = 2.78, SD = 2.53$) trials.

Discussion

The results of this study suggest that unconscious associative learning had occurred. The evidence for this learning is based on a simple dissociation between awareness and the RT measures. As indicated by the t -tests on the d' sensitivity measures, perceptually unaware participants were unable to consciously discriminate between S1_A and S1_B stimuli. However, they responded faster to S2 in compatible sequences than in the incompatible ones. This means that perceptually unaware participants had acquired associative learning through the AMP procedure, given that the presentation of S1 facilitated the response to S2 in the compatible stimuli sequences.

On the whole, the results of this experiment substantiate the findings of several studies that suggest unconscious associative learning (e.g., Clark et al., 2001; Manns et al., 2002; Raes et al., 2010; Wong et al., 2004). Likewise, our findings agree with the results obtained by studies on response masked priming (e.g., Barbot & Kouider, 2012; Mattler & Palmer, 2012; Neumann & Klotz, 1994; Schlaghecken, Bowman, & Eimer, 2006), which justify the use of the experimental data and the theoretical contributions of these studies for AMP research.

As Lovibond and Shanks (2002) point out, it is not an indispensable requisite for the participant to identify the masked stimuli for differential associative learning to have occurred. Detection of only some minimal components of masked S1 stimuli may be sufficient for the correct discrimination between these stimuli. Thus, the essential condition for determining whether unconscious learning had indeed occurred was that participants were unable to consciously discriminate between the masked stimuli. In the forced-choice discrimination task used in this study, minimal low-level features or any small difference detected between the S1 stimuli (letters "o" and "x") were sufficient to obtain a number of correct responses above chance, providing evidence of conscious discrimination between these stimuli. Furthermore, participants were classified as perceptually unaware according to

the absence of awareness in the two forced-choice discrimination tasks. Therefore, these results suggest that the priming effect of perceptually unaware participants can be considered proof of true unconscious learning.

In addition, the results of the separate ANOVAs, and the interaction observed between awareness and compatibility, indicated a double dissociation between perceptual awareness and AMP: only perceptually unaware participants showed priming effects. This suggests the expression of the associative learning generated by AMP requires the absence of perceptual awareness, and provides further evidence of the unconscious learning of discrimination.

These results support the existence of an associative mechanism of automatic response activation that operates outside of awareness. Though the concept of automaticity is complex and remains to be fully understood, it appears to encompass certain characteristics such as unintentional, uncontrolled/uncontrollable, fast and unconscious (for a review, see Moors & De Houwer, 2006). A type of automaticity that may shed some light on these results is explained in terms of direct stimulus-response (S-R) associations (Schneider, Dumais, & Shiffrin, 1984). Two direct S-R associative links are acquired and strengthened by the repeated S1-S2 pairings: an associative link between low-level prime features and motor response (RT) features, and another link between the target features and the same response. As a result of these associations, the presentation of the prime triggers automatic activation of its associate response, facilitating the RT to S2 if this stimulus requires the same response. Conversely, interference will occur if S2 requires an opposite response. As Klapp and Greenberg (2009) have shown, these associations are effective with both awareness and without awareness of the prime presentation. Notwithstanding, two crucial considerations concerning the mechanisms of automaticity based on S-R associations should be borne in mind: (a) the associative S-R mechanism does not exclude the involvement of other mechanisms based on semantic processing, and (b) the S-R associations as an explanatory principle of unconscious learning are restricted in this study to the automatic activation of motor responses; thus, they should not be generalized to other types of responses.

Nonetheless, perceptually aware participants showed no significant priming effects, even though the associative mechanism of automatic response activation is effective with both awareness and without awareness of the prime presentation (Klapp & Greenberg, 2009). A plausible explanation for these results may lie in the conflicting data on participants in incompatible trials.

As demonstrated by Ansorge, Fuchs, Khalid, and Kunde (2011), the priming effects obtained in a given trial diminish when the preceding trial consists of an incompatible stimuli sequence than a compatible stimulus sequence. But this effect occurs only if the participant is aware of the visual prime. Likewise, other researchers (e.g., Gratton, Coles, & Donchin, 1992; Greenwald, Draine, & Abrams, 1996; Kunde, 2003) have found that when participants saw an incompatible prime in a specific trial, they then focused the attention on the target and excluded prime processing in the subsequent trial, reducing the priming effects.

The parallelism between the associative mechanism of automatic response activation and the low-level conditioning process is evident. As postulated by the dual-process models (Furedy & Riley, 1987; Lovibond & Shanks, 2002), the changes arising from repeated CS-US pairings are the result of two independent learning processes: a propositional learning that leads to conscious knowledge of association between the stimuli, and a lower level, non-propositional conditioning process which directly triggers an automatic activation of a conditioned response by an associative mechanism, such as an excitatory link between CS and US nodes or between the CS and RC. This process does not depend on contingency awareness of CS-US relationship, but rather reflects the previous experience with the association between these stimuli. These properties of the low-level conditioning process exhibit certain similarity with the capacity acquired by S1 to elicit automatic activation as a result of AMP.

Differences in the RTs obtained in this AMP study were relatively small, compared to the differences observed in most studies on response priming and semantic priming. A plausible explanation is that in the response priming paradigm, the same stimulus was used as prime and target. Therefore, the capacity of the prime to activate the motor response will presumably be greater in these studies than in AMP, where two non-related stimuli are presented by a large number of pairings during one or two experimental sessions. One can also assume that the differences in RT observed in semantic priming experiments are greater because the associations produced by the natural use of the language between the prime and target words are much more numerous and frequent. Nonetheless, the findings of this experiment suggest that AMP is a promising procedure for assessing unconscious learning due to its sensitivity to the effects of S1-S2 associations when the participants were perceptually unaware of S1. Further studies on AMP are necessary to determine the effects of different ISIs, and the specific conditions that favour the manifestation of this unconscious process.

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