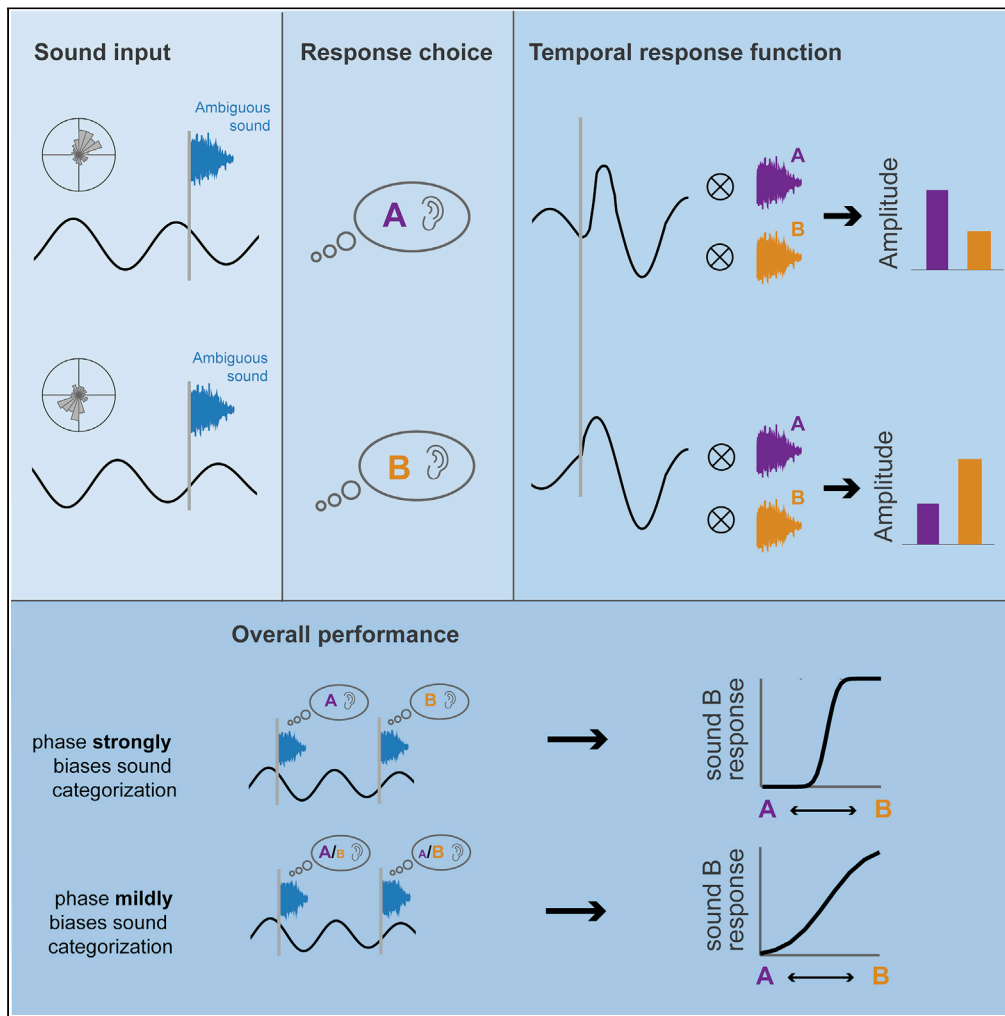# iScience

Article

Phase-Coded Oscillatory Ordering Promotes the Separation of Closely Matched Representations to Optimize Perceptual Discrimination

Sanne Ten Oever, Tobias Meierdierks, Felix Duecker, Tom A. De Graaf, Alexander T. Sack

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HIGHLIGHTS

Pre-stimulus theta/alpha phase co-determines how we perceive ambiguous sounds

Phase influences to which sound envelope evoked potentials fit better

Neural separation through phase clustering promotes sound discrimination

Article

Phase-Coded Oscillatory Ordering Promotes the Separation of Closely Matched Representations to Optimize Perceptual Discrimination

Sanne Ten Oever,[1,2,3,6,*] Tobias Meierdierks,[1] Felix Duecker,[1,4] Tom A. De Graaf,[1,4] and Alexander T. Sack[1,4,5]

SUMMARY

Low-frequency oscillations are proposed to be involved in separating neuronal representations belonging to different items. Although item-specific neuronal activity was found to cluster on different oscillatory phases, the influence of this mechanism on perception is unknown. Here, we investigated the perceptual consequences of neuronal item separation through oscillatory clustering. In an electroencephalographic experiment, participants categorized sounds parametrically varying in pitch, relative to an arbitrary pitch boundary. Pre-stimulus theta and alpha phase biased near-boundary sound categorization to one category or the other. Phase also modulated whether evoked neuronal responses contributed stronger to the fit of the sound envelope of one or another category. Intriguingly, participants with stronger oscillatory clustering (phase strongly biasing sound categorization) in the theta, but not alpha, range had steeper perceptual psychometric slopes (sharper sound category discrimination). These results indicate that neuronal sorting by phase directly influences subsequent perception and has a positive impact on discrimination performance.

INTRODUCTION

Many everyday tasks require the online tracking of various types of information. For example, we need to keep track of a shopping list or of the voice identities of multiple speakers in a room. This type of tracking not only demands the active maintenance of different item or location representations but also requires these item representations to be strictly separated such that the representations do not entangle. These separated neuronal representations could thereby optimize the perception of future incoming sensory inputs.

Separating multiple item representation could be funneled through low-frequency oscillations (Lisman and Jensen, 2013; Jensen et al., 2012). Neuronal oscillations are time-varying fluctuations in membrane potentials of a group of neighboring neurons (Buzsáki and Draguhn, 2004). It has been shown that the partial depolarization and hyperpolarization at different phases of the oscillations relate to different probabilities of neuronal firing (Haegens et al., 2011; Skaggs and McNaughton, 1996; Volgushev et al., 1998). Therefore, oscillations naturally reflect distinct periods (phases) of optimal and non-optimal communication (Fries, 2005; Schroeder and Lakatos, 2009; Giraud and Poeppel, 2012). However, oscillations seem to be involved in more than just gating when information can be communicated. For example, different studies show that the preferred phase of firing changes from neuron to neuron (O'Keefe and Recce, 1993; Lee et al., 2005). In addition, gamma power has been shown to cluster on unique low-frequency phases (Heusser et al., 2016; Canolty et al., 2006; Lakatos et al., 2005). This suggests that within one fluctuation, also known as cycle, different types of information, and therefore different item representations, might be represented at distinct phases (O'Keefe and Recce, 1993; Lisman, 2005; Lisman and Idiart, 1995). Indeed, when extracting an item category from neuronal data, adding phase improves category identification above what can be extracted by spike counts alone (Kayser et al., 2012; Kayser et al., 2009; Montemurro et al., 2008; Masquelier et al., 2009). Phase-coded representation was also identified in an electrocorticography study (Bahramisharif et al., 2018). Bahramisharif, Jensen, Jacobs and Lisman (2018) presented multiple letters sequentially and instructed patients to memorize the letters. In the retention period, letter-selective electrocorticography (EcoG) channels displayed high gamma activity at different theta phases dependent on the serial position of the letter. Combined, these studies suggest that different items are represented by the clustering of information to different low-frequency oscillatory phases.

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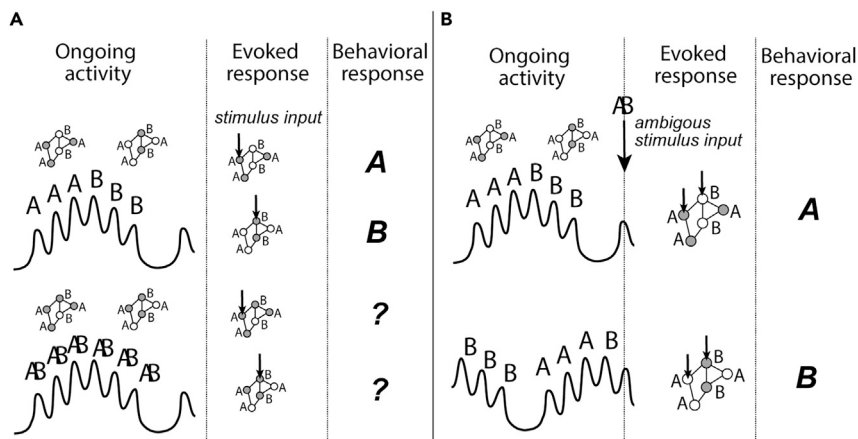
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Figure 1. Theoretical Background and Hypothesis

(A) During successful separation different neuronal populations are active at different phases during ongoing activity (top). During unsuccessful separation activation patterns are mixed (bottom). When a stimulus input activates a node in the network it will automatically spread to the connected nodes (middle). During successful separation this spread will be within the separated stimuli, but during unsuccessful separation this spread is more random (right).

(B) When an ambiguous stimulus is presented in a network that is successfully separating item representations the phase at which the information about this stimulus arrives in the population influences where the activity will spread to as populations are differentially pre-activated. This leads to different behavioral response choices. For unsuccessful separation there would be no such phase response bias for ambiguous stimulus as there are no item-specific pre-activations.

Although these studies indicate how phase may order item representations, it is currently unknown whether this representational mechanism has any consequences on perception and behavior (but see Kayser et al., 2016). Indeed, the goal of remembering items in a shopping list is to be able to identify them when one is in the store. Therefore, we need to understand whether and how the neuronal mechanisms for information representation influence perception. If we assume that during ongoing activity neuronal representations are separated through phase-specific sequential activations of these representations it has direct perceptual consequences. On the one hand, overall perceptual discrimination is expected to be enhanced when information of different items shows strongly separated neural representations (see, e.g., Heusser et al., 2016; Figure 1A). On the other hand, single-trial perceptual outcomes might depend on the timing of the sensory input as the representational mechanism activates item representations sequentially (i.e., phase 1, item A; phase 2, item B). Specifically, when neuronal input coincidentally arrives at phase 1, neuronal activation will spread through to item A representations (see, e.g., Ten Oever and Sack, 2015). As a consequence, stimuli presented at phase 1 should bias information processing to item A, both neuronally and perceptually (Figure 1B).

The current study aimed to test this hypothesis by systematically evaluating whether the magnitude of oscillatory phase separation of item representations relates to perceptual changes as predicted for a single trial as well as for overall perceptual performance. To this end, participants were presented with ripple sounds that they learned to categorize in one of two classes (determined by the pitch of the sound). Neuronal oscillations during task performance were measured with electroencephalography (EEG). Our results show that (1) oscillatory phase in the theta and alpha ranges at stimulus onset biased participants' perception to one of the two categories; (2) dependent on the phase at stimulus onset, evoked neuronal responses fitted the sound envelopes of either one of the two pitch categories more strongly; and (3) participants with stronger perceptual theta phase biases, i.e., stronger phase separation, demonstrated overall better discrimination performance. These results show how phase-coded representations interact with perceptual processes, with phase not only biasing perception but also improving item separation, which enhances overall perceptual discrimination.

RESULTS

We analyzed 32-channel EEG data in which participants (total of 21 participants) were required to perform an auditory categorization task. Sounds were ripple sounds varying in velocity, density, and pitch. The pitch variation determined the category. We presented ripple sounds based on a total of 12 different frequencies,
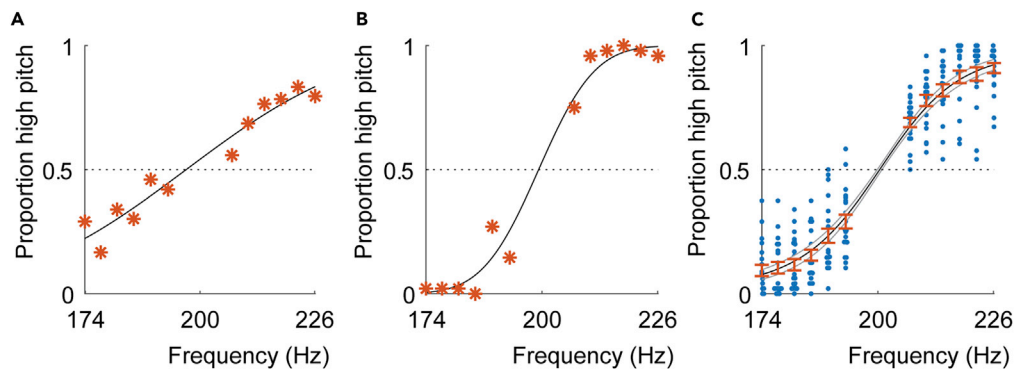
Figure 2. Behavioral Psychometric Functions
(A and B) Example of a participant with low (A) and high performance (B).
(C) All individual data points as well as the average psychometric function. Orange error bars indicate the mean and standard error of the mean for the individual sound categories. The black solid line and shaded areas indicate the mean of the fitted psychometric functions and their respective standard error of the mean.

straddling an arbitrary boundary of 200 Hz. Before the start of the experiment, the participants heard four random exemplars of the category extremes (without explicit instruction as to what feature determined the categorization). Then participants were required to categorize randomly presented sounds, learning the category dimension and boundary implicitly during the experiment based on feedback after each response. The interval after the participant's response and the next sound was jittered between 1.5 and 2.5 s. In total there were four blocks in which 576 sounds were presented, lasting approximately 35 min. All participants were able to classify the sound according to a typical psychometric function (Figure 2; fitted with a probit function with a guessing and lapsing rate at 0 using a maximum likelihood estimate). Our interest for this study was to investigate if single-trial perceptual and neuronal outcomes are influenced by oscillatory phase, as well as to investigate if phase separation strength influences overall perceptual discrimination.

Theta and Alpha Phase Bias Sound Categorization

If the two sound categories are represented by the brain on different phases, it is expected that single-trial behavioral responses are biased to the sound associated with the oscillatory phase at which (at random) an ambiguous sound stimulus happened to be presented on any given trial (Figure 1B). To investigate this hypothesis, for every trial we extracted for frequencies ranging from 2 to 15 Hz the pre-stimulus phase by calculating the fast Fourier transform over three cycles of data up to sound onset. No additional offline filters were applied to avoid smearing any post-stimulus effect to the pre-window interval. Then we investigated whether the category responses (sound category A or B) for ambiguous sounds (sounds with a categorization accuracy lower than 80%) were correlated with the pre-stimulus phases. We did this by first regressing out any effect that could be related to stimulus type by fitting a general linear model (GLM) with a binomial distribution on the response choice data with stimulus type as factor. The residuals of this analysis were used in a circular-linear correlation with pre-stimulus phase correction for multiple comparisons using cluster statistics (Maris and Oostenveld, 2007) (we repeated the analysis using the phase opposition index, VanRullen, 2016; this resulted in the same outcome; Figure S1). The correlation resulted in two significant clusters. One cluster encompassed the frequencies 4.6–5.6 Hz with a frontocentral topography (Figure 3A, clusterstat = 191.4, p = .034). The second cluster included the frequencies 8.5–10.9 Hz and had a right centroparietal topography (Figure 3B, clusterstat = 259.97, p = .020). Finally, a trend-significant cluster was found (6.5–7.5 Hz, clusterstat = 94.0, p = .090). This indicates that spontaneous oscillatory phase at the time of stimulus presentation biases participants' percept to either sound category A or B.

To investigate whether not only the category choice but also single-trial performance was influenced by pre-stimulus phase, we repeated the analysis, but correlating pre-stimulus phase with the single-trial GLM residuals of the categorization accuracy of participants (i.e., correct or incorrect). For this analysis, no significant or trend-significant clusters were found.

To further explore the phase-categorization correlation, we (post hoc) extracted for each participant all phases for sound A categorization and sound B categorization at the channel and frequency of the
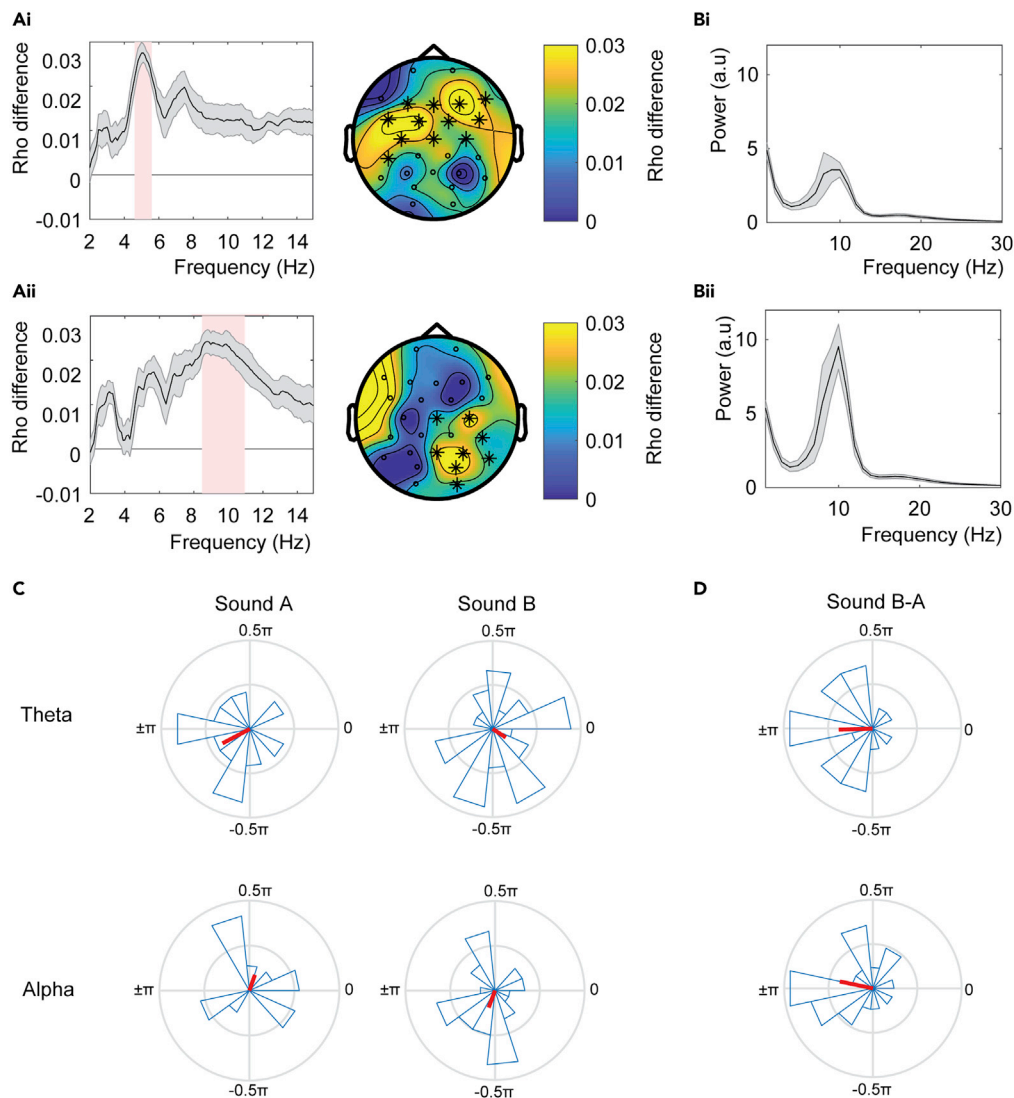
Figure 3. EEG Phase-Categorization Correlation

(A and B) The theta (A) and alpha (B) clusters are displayed. The left panels display the correlation difference relative to the permuted correlation for the channels included in the cluster. Shaded gray areas indicate the standard error of the mean. Pink areas include the frequencies belonging to the cluster. The right panels display the topographies for the respective significant frequency bins. Asterisks indicate the channels included in the cluster (p < 0.05).

(C) Circular histograms of a representative subject for sound A and sound B classification trials (at 50%–80% accuracy) for the peak significance frequency at the theta and alpha clusters.

(D) Circular histogram of phase difference of mean sound B versus sound A categorization over subjects for the peak significant frequency at the theta and alpha clusters.

maximum t-value of the cluster (see Figure 3C for an example participant). First, it was investigated if the mean phase over participants was consistent by performing a Rayleigh test over the mean phases over participants. We did not find a clear pattern of consistency of phases over participants. Only for the alpha cluster for sound A a significant effect was found (Z = 3.88, p = 0.019; Figure S2). However, the phase difference between phase B and phase A had a significant phase opposition for both the theta and alpha clusters (theta: p = 0.003; alpha: p = 0.015; Figure 3D). This indicates that whereas individual participants have a bias of responding sound A or sound B dependent on the phase, this exact phase of which participants say sound A or B is not consistent over participants (see Figure S2 for a split of the phase consistency into the different sound types).
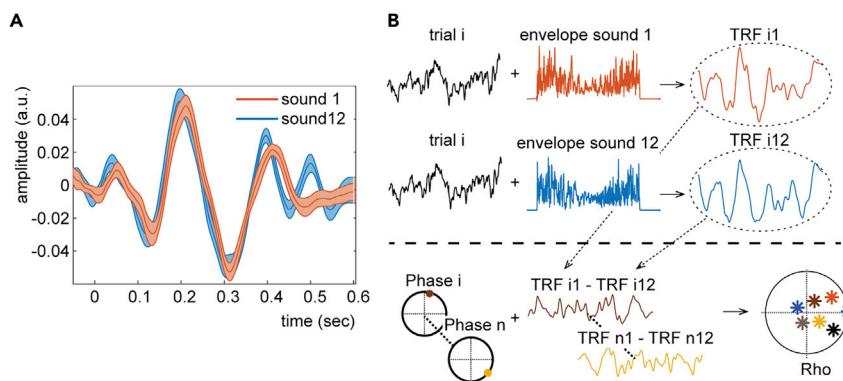
Figure 4. TRF Estimation

(A) Average TRF for the trials with sound 1 or 12 with the corresponding sound envelope.

(B) Description of the analysis. For each trial we calculated the TRF for the envelope of sound 1 and sound 12 (category extremes). The difference between these TRFs was correlated with the pre-stimulus phase.

Theta and Alpha Phase Modulate Temporal Response Function Fit

If behavioral category responses are modulated by phase, the same should be expected for stimulus-evoked neuronal responses. Specifically, it is expected that the evoked responses should be more similar to either one of the two sound categories dependent on the pre-stimulus phase. The timing of the neuronal modulations could differentiate whether the phase-dependent behavioral bias is related to perception (early modulation) or decision making (later modulation). To test this, we extracted the temporal response functions for each trial over an epoch of -0.1 to 0.6 s (Crosse et al., 2016). This temporal response function (TRF) is the output of a model that assumes that the EEG response is a consequence of a linear convolution from the neuronal data and a weight vector (i.e., the TRF). The stronger the amplitude of the TRF, the better the fit (under normalized amplitudes of the stimulus output and EEG). As such, we use the amplitude of the TRF as a measure of the fit of the predicted stimulus type in the model.

First, we validated that a TRF can reliably be determined. For all extreme sounds (pitch 1 and 12) we extracted the TRF with the corresponding sound envelope. During this analysis, we also estimated the lambda parameter needed for an accurate TRF estimation used in the subsequent analysis (see Transparent Methods). Indeed, we were able to extract TRFs with an expected amplitude modulation between 0.1 and 0.4 s (Figures 4A and S3 for event-related potentials [ERPs]).

To investigate if the TRF was modulated by phase, we estimated two TRFs per trial corresponding to the envelope of the sound belonging to pitch 1 and 12 (Figure 4B; matching in velocity and density with those of the presented sound of that trial). Again, only trials with ambiguous sounds (below 80% accuracy) were used. This calculation provided us with two TRF time-series reflecting how well the EEG data (of a specific channel) fits the two extreme sounds. The difference of these two TRFs provides a time-series estimating for each trial to which sound envelope the trial response fits better. This TRF difference was then correlated (separately for each time point) with the corresponding pre-stimulus phase. If the phase of presentation influences the neuronal response, this difference (representing whether the response function fitted either one of the sounds more) should depend on the phase of presentation. The phase dependency of the TRF difference was estimated for the two separate frequency bins identified in the *EEG phase-categorization correlation* analysis. We repeated the analysis using a linear instead of ridge regression (i.e., the temporal response function), resulting in a similar pattern of results (data not shown). We choose to show the temporal response function as it corrects better for the autocorrelation in the signal.

For the theta bin we found four significant clusters (Figure 5; 0.11–0.15 s: clusterstat = 69.8, p = 0.013; 0.31–0.35 s: clusterstat = 79.2, p = 0.004; 0.38–0.43 s: clusterstat = 81.2, p = 0.004; 0.50–0.53 s: clusterstat = 76.1, p = 0.008). For the alpha bin two significant clusters were found (Figure 6; 0.03–0.45 s: clusterstat = 3,812.7, p < 0.001; 0.55–0.60 s: clusterstat = 356.7, p = 0.031). This indicates that the response function was modulated by the phase at the time of stimulation, fitting more closely one or the other sound category depending on the pre-stimulus phase. In other words, if an ambiguous sound is presented at the phase associated with sound category A, the neuronal response to that ambiguous sound will fit extreme exemplar of sound category A better than on another phase.
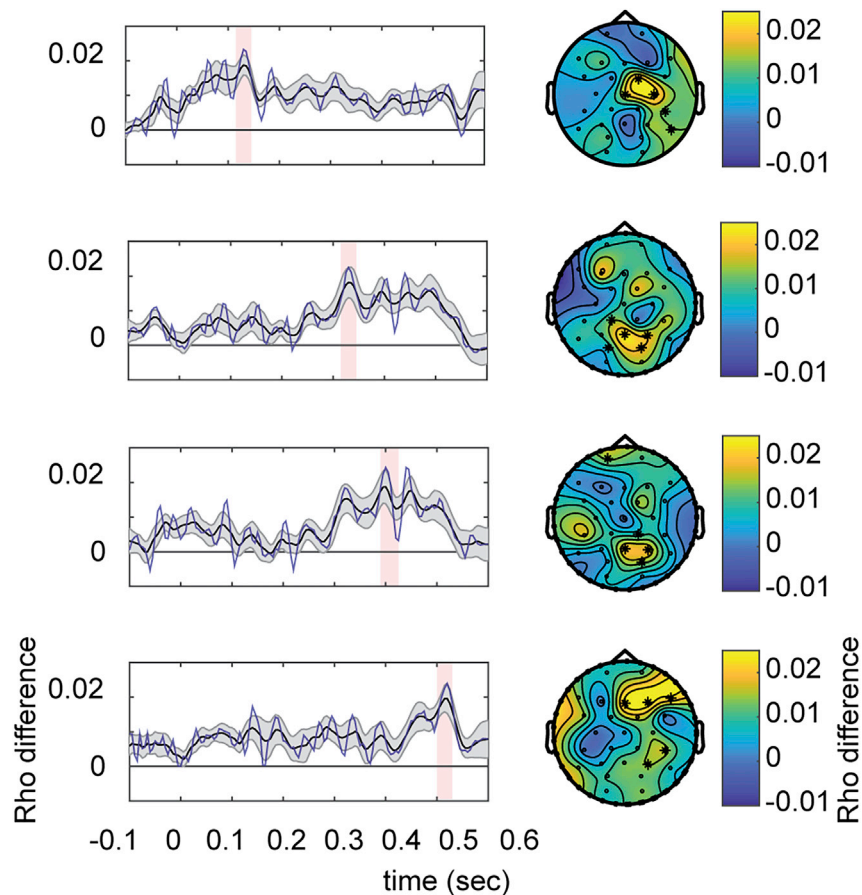
Figure 5. Phase-Dependent TRF, Theta Bin

Left panels represent the time course of the correlation difference between the permuted phase TRF difference correlation and actual phase TRF difference correlation. Right panels indicate the topographies of the clusters (p < 0.05). Shaded gray areas indicate the standard error of the mean. Pink areas indicate the significance window (and the window for the topography on the right panel). Channels that are displayed are the five channels with the highest summed T within the clusters and are highlighted on the right panels with asterisks. Time courses are smoothed with a nine-point moving average; purple lines show the non-smoothed data.

Theta Behavioral and TRF Phase Are the Same

If the TRF response relates to the behavioral choice, then the phase at which a participant chooses sound A should be the same as the phase at which the TRF amplitude is higher for sound A compared with sound B. To investigate this, we extracted the average phase at which participants choose sound A and sound B at the frequency and phase point of the maximum t-value of the clusters (same as phases in Figures 3C and S2). For the TRF we extracted the average phase for trials in which the TRF amplitude was larger for A and the average phase for which the TRF was larger for B (at the time point of the maximum t-value either of the significant clusters at the channel of the behavioral cluster). Then, we calculated the differences between the TRF and behavioral phases and calculated the v-statistics (Zar, 1998). The v-statistics tests for non-uniformity of a circular distribution assuming a known mean value (in this case zero). This test showed that for the theta cluster the behavioral and TRF phases were the same (Figure 7A; V = 13.442, p = 0.0017 at 4.89 Hz, F4, 0.515 s), but for the alpha no non-uniformity could be confirmed (V = 2.131, p = 0.350 at 10.27 Hz, PO4, 0.350 s).

Improved Performance for Stronger Behavioral Theta Phase Modulations

Last, we investigated whether the strength of the determined behavioral phase modulation has an influence on the overall categorization sensitivity. The rationale was that if participants are better at separating the categorical representations (i.e., higher phase modulation), they would be better at the task (Figure 1A).
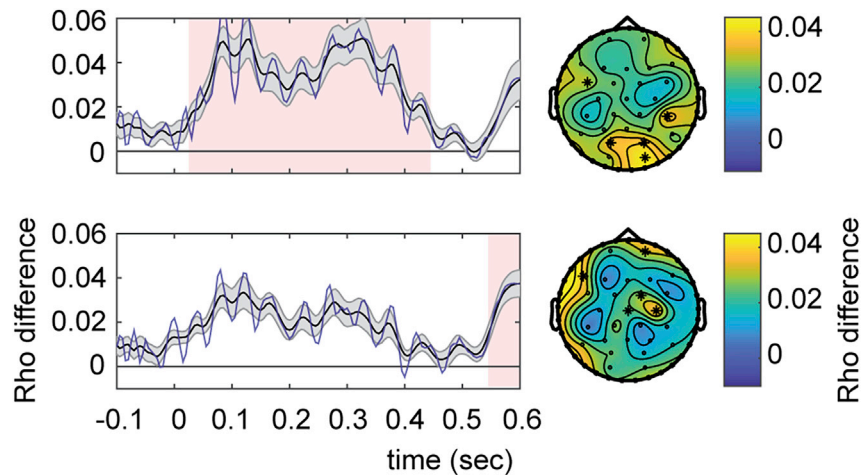
Figure 6. Phase-Dependent TRF, Alpha Bin
Conventions are the same as in Figure 5.

Therefore, we extracted the phase modulation index for both significant behavioral frequency clusters: the difference between the observed phase-categorization correlation from the chance correlation (the median of a created null distribution per participant) divided by the chance correlation. Note that we calculated the phase modulation index instead of the raw circular-linear correlation as the correlation will be higher for participants in whom fewer trials would be included. The phase modulation index controls for the trial amount effect. The phase modulation index was correlated with the slope of the psychometric curve (A versus B categorization across the 12 pitch levels; Figure 2). We found a significant correlation for the phase categorization modulation in the theta range (Figure 7B; r = 0.39, p = 0.037), but not for the phase categorization modulation in the alpha range (r = −0.03, p = 0.450). Thus, simplified, participants with brains in which theta phase strongly coded for sound categories A and B were better at performing the sound categorization task.

DISCUSSION

In this study we investigated whether neuronal and behavioral responses to stimuli are influenced by the phase of EEG-measured neuronal oscillations at the time of stimulus presentation. We presented ripple sounds of varying pitches with a category boundary at a fundamental frequency of 200 Hz. We found that oscillatory phase in the theta and alpha ranges, at the time of ambiguous sound presentations, influenced the sound categorization responses of participants. Moreover, ongoing oscillatory phase modulated the amplitude of the fit of the evoked neuronal response to one of the other sound category exemplars. Interestingly, the strength of the theta, but not alpha, phase modulation correlated with the overall discrimination abilities of the participants. These results show that categorical information is ordered on oscillatory phase and that this oscillatory ordering scheme promotes the separation of closely matched representations to optimize perceptual discrimination.

The role of oscillatory mechanisms for behavior has been investigated by extracting phase-dependent behavioral responses. Previous studies have shown that phase influences visual detection (Mathewson et al., 2009; de Graaf et al., 2013) and auditory detection (Ten Oever, Van Atteveldt and Sack, 2015; Ten Oever and Sack, 2015; Henry and Obleser, 2012; Henry et al., 2016) and modulates attentional processes (Fiebelkorn et al., 2013; Landau et al., 2015). The designs of these studies were inspired by experimental findings showing that neuronal activity clusters along one specific oscillatory phase (Buzsáki, 2004; Haegens et al., 2011). As such, it has been proposed that presenting any stimulus on an optimal phase should improve behavioral performance (Schroeder and Lakatos, 2009; Haegens and Golumbic, 2017). Here, we provide no evidence that phase improves performance (accuracy). The difference between our study and the ones described is that these previous studies have mostly focused on single item or detection tasks. In these tasks, it is impossible to investigate the role of oscillations for multi-item representations, as the behavioral output is only related to a single representation. However, some studies have reported accuracy effects for more complex auditory decision tasks (Strauß et al., 2015; Hansen et al., 2019) and
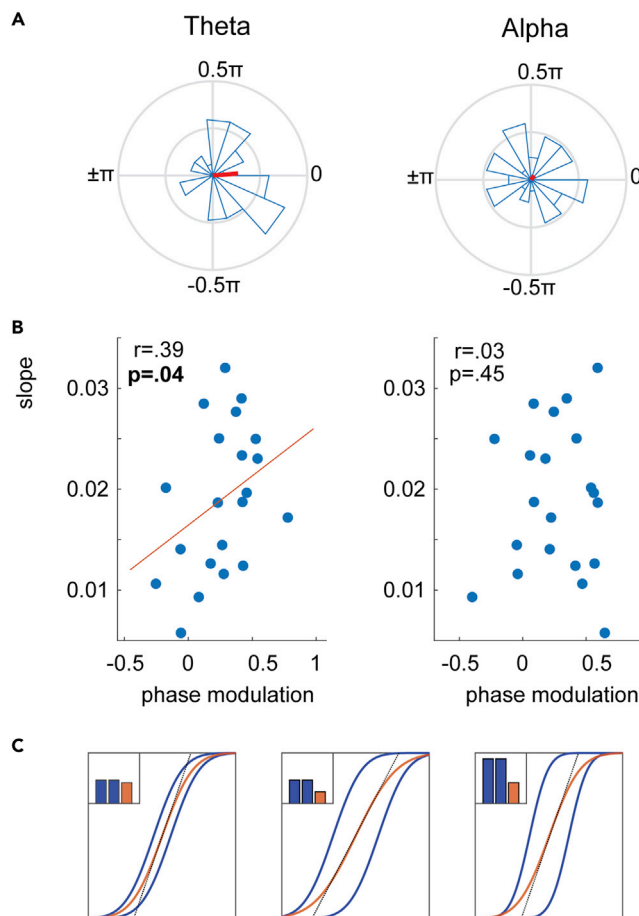
Figure 7. Behavioral-TRF Phase Comparison and Performance-Phase Modulation Correlations
(A) Phase difference over participants (separate for the two sound choices) for the behavior and the TRF.
(B) (Left) The phase modulation index for the theta cluster in the categorization modulation. (Right) The alpha cluster in the categorization modulation.
(C) Conceptual illustrations for low (left) and high (middle and right) phase modulations are shown with insets for slope estimations. Blue lines illustrate the mean phase at which bias is the strongest. The lower the bias, the more similar the overall psychometric curves (orange) and slope are to the bias extremes. To achieve a higher overall psychometric slope for higher phase modulations (stronger bias), the curves of the high bias must have a steeper slope than curves of bias extremes with low bias (compare middle with right). In other words, although the bias on its own must hurt performance, the greater categorical separation must sharpen the slope of the overall psychometric curve, yielding a net positive result.

effects of phase on the choice have also been reported (Kayser et al., 2016). Here, we investigated phase-dependent behavioral responses when two categorical representations had to be maintained simultaneously. We show that phase biases the percept to one of two categories. This finding suggests that phase modulates not only how well any input is processed but also that different neuronal representations are sorted on oscillatory phases (Lisman and Idiart, 1995; Lisman and Jensen, 2013). Indeed, also other studies have found that pre-stimulus power or phase might not solely improve stimulus detection, but merely the bias to respond (Weisz et al., 2014; Iemi et al., 2017; Woestmann et al., 2019).

We show improved categorical perception for participants with a stronger behavioral phase modulation. Although this positive correlation between phase modulation and psychometric slopes (i.e., perception) is in line with our expectations, i.e., enhanced behavioral separation with increased neuronal separation, overall slope increases are not possible when phase biases individual psychometric curves to opposing extremes. To elaborate, strong behavioral phase modulation suggests that the psychometric curves belonging to the two phases with the strongest modulation toward perceiving sound category A or B have the biggest bias differences (Figure 7C). If phase only has an influence

on the bias of this psychometric function, the slope of the overall psychometric curve in which all data are collapsed would be steeper for *low* phase modulation (i.e., low bias difference) compared with high phase modulation (i.e., high bias difference). This is the case because for high phase modulation two distant curves need to be combined, resulting in a lower overall steepness. The only way to increase this lowered overall psychometric slope is to have, in addition to the bias, steeper psychometric curves for the phase extremes (Figure 7C right). Indeed, the positive correlation found in our data suggests that phase modulation makes the steepness of the extremes stronger. This exemplifies that a strong phase modulation provides a better neuronal basis for sound categorization, which is translated to a high overall performance in this categorization task, showing that neuronal representation separation is critical for improving behavioral performance (see also Heusser et al., 2016; Bahramisharif et al., 2018; Lee et al., 2005).

With EEG it is not trivial to extract phase-dependent neuronal responses related to neuronal representations. Previously, phase-dependent neuronal responses have been found in ongoing spiking and gamma activity (Lee et al., 2005; Canolty et al., 2006; Axmacher et al., 2010). To evaluate similar mechanisms, we used stimulus-evoked EEG responses, which can reflect a readout of the state of the neuronal network (Wolff et al., 2017; Rose et al., 2016). Specifically, we found that phase modulated whether the evoked EEG responses were better fitted by the temporal envelope of one, versus the other, of the two sound categories. Sorting ERP based on phase is usually a problem as the phase at the moment of stimulus presentation directly influences the amplitude of the subsequent ERP. Taking the difference of two different TRFs avoids this problem as the absolute amplitude shift should be the same for both TRFs. Therefore, this method provides a solution to extract meaningful phase-dependent EEG-evoked responses.

We found that both theta and alpha phases modulated categorical choices. However, only theta, but not alpha, phase modulations influenced overall perceptual performance in our study, seemingly suggesting that specifically in the theta range this modulation is relevant for auditory categorization performance. Curiously, for the neuronal responses, the alpha phase modulation seemed to be stronger. This stronger alpha modulation might be a consequence of a more accurate phase estimation in the alpha range, as alpha power is higher compared with theta power. However, theta seems ultimately more relevant for behavior, which is in accordance with previous studies showing phase modulations for auditory categorization in the theta range (Köster et al., 2019; Wang et al., 2018) and theoretical accounts on phase-coded representations (O'Keefe and Recce, 1993; Watrous et al., 2015). Unfortunately, our results cannot address what the potential source of the theta modulation is in this study. It could relate to auditory responses or working memory processes. Both processes have been associated with theta activity (Roux and Uhlhaas, 2014; Poeppel, 2003; Lakatos et al., 2005) and are likely candidates to represent the types of stimuli presented here. However, future studies need to dissociate whether classical working memory areas, auditory areas, or both can code representations on phases.

Conclusions

Our brain has to keep track of multiple types of information in parallel. In this study we found that one way the brain may separate this information is by sorting multiple representations on different oscillatory phases. The current study demonstrated that oscillatory phase in the alpha and theta ranges biases behavioral and neuronal responses to either one of two sound categories. What is more, the better participants are able to separate information on different theta phases, the more sensitive they are in identifying sound categories, reflected by better perceptual discrimination performances. Future research should investigate which neuronal systems engage in this type of phase coding. Thereby, we can explore the full potential of the brain to represent many types of information in parallel.

Limitations of the Study

In the current study we show that phase modulates behavioral response choices. We did not find any effect of accuracy in contrast to previous articles (Hansen et al., 2019; Strauβ et al., 2015; Henry et al., 2016). It is still an open question what design choices lead to these differences. In addition, the contribution of the alpha modulation in the current study is unclear. Alpha showed a clear phase and TRF modulation, but the phases of the two effects were not identical and alpha did not contribute to the overall performance of the participants. The differentiation between this and theta effect needs to be investigated in the future.

METHODS

All methods can be found in the accompanying Transparent Methods supplemental file.

AUTHOR CONTRIBUTIONS

Conceptualization, S.T.O.; Methodology, S.T.O., F.D., and T.A.D.G.; Data collection, S.T.O. and T.M.; Formal analysis, S.T.O. and T.M.; Writing – Original draft: S.T.O.; Writing – Review and Editing: T.B., F.D., T.A.D.G., and A.T.S.; Supervision: A.T.S.; Funding Acquisition: A.T.S.

DECLARATION OF INTERESTS

The authors declare no competing interests.

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Supplemental Information

Phase-Coded Oscillatory Ordering Promotes

the Separation of Closely Matched Representations

to Optimize Perceptual Discrimination

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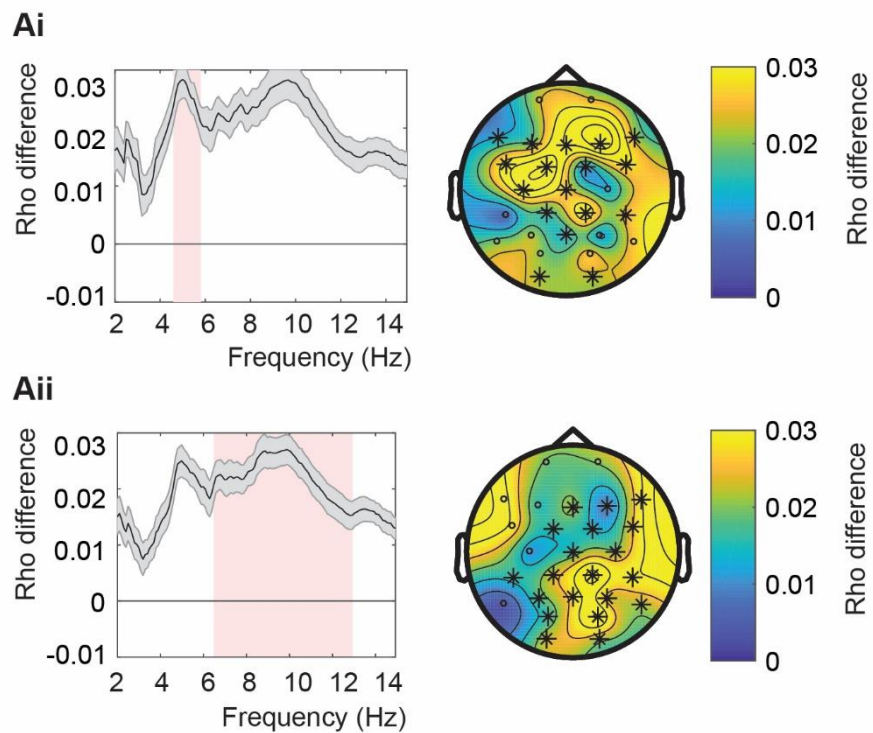
Supplemental Data Items

Figure S1. Phase opposition analysis, related to Figure 3. The theta (A) and alpha (Aii) clusters are displayed. The left panels display the correlation difference relative to the permuted correlation for the channels included in the cluster. Shaded grey areas indicate the standard error of the mean. Pink areas include the frequencies belonging to the cluster. The right panels display the topographies for the respective significant frequency bins. Asterisks indicate the channels included in the cluster.
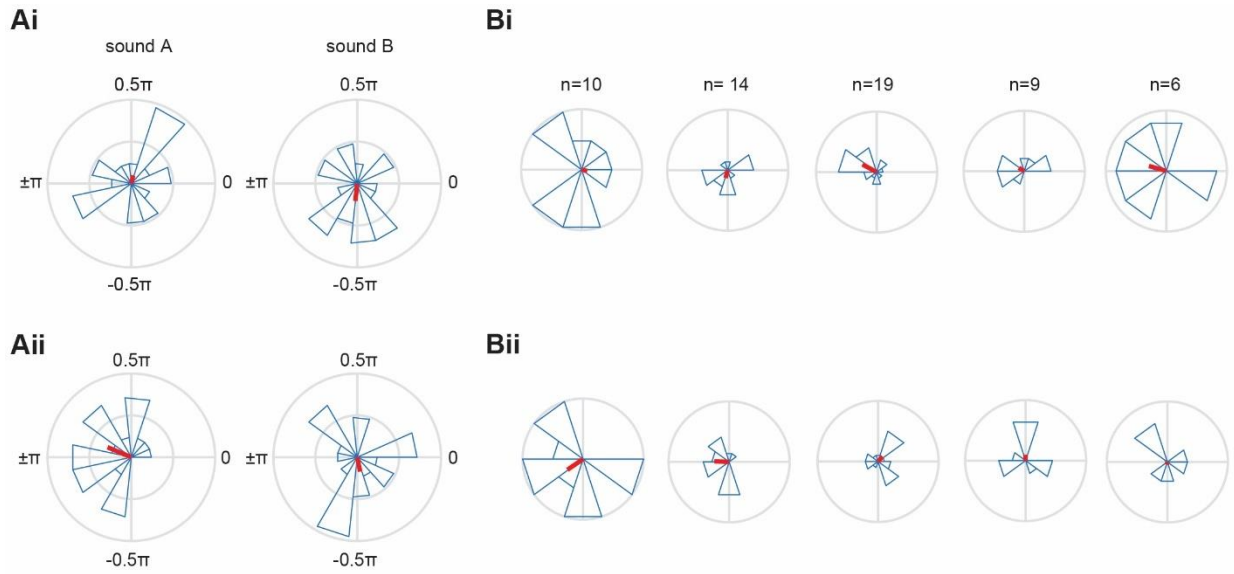
Figure S2. Phase specific effects, related to Figure 3. A) For the maximum t-value in the cluster the phase histograms of sound A and sound B are presented (over subjects) for the theta (Ai) and alpha (Aii) cluster. B) Phase difference between sound A and B split up for different sound types (sound 5-9) for the theta (Bi) and alpha (Bii) cluster.
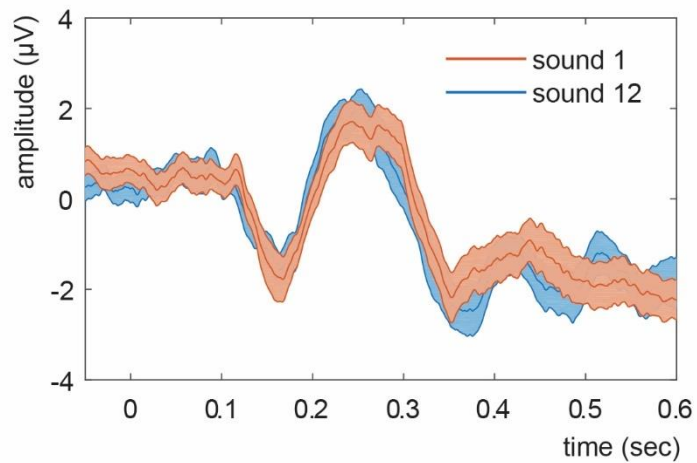

Figure S3. ERPs of the two extremes sounds, related to Figure 4. Shaded area indicates the standard error of the mean.

Transparent Methods

Experimental model and subject details

Participants

Twenty-one participants completed the experiment (participants' demographics were not recorded). All were informed about the experiment after given informed consent. The study was approved by the local ethical committee at the Faculty of Psychology and Neuroscience at Maastricht University (ethical approval number: ECP-127 14_04_2013). Participants received course credits or monetary compensation for their time.

Method details

Stimuli and procedure

Ripple sounds were presented to the participants consisting of 50 logarithmically spaced sinusoids spanning 5 octaves. Sounds had varying velocities (six velocities linearly spaced between 1 and 1.63 cycles/second) and densities (0.25 and 0.125 cycles/octave). The fundamental frequency determined the category boundary, which was arbitrarily set at 200 Hz. Six sounds were created in each category and were logarithmically spaced 7.2 until 26.1 Hz away from the category boundary. Modulation was set to 100 percent and sounds lasted for 500 ms.

First, participants were provided with some examples of extremes of both categories to familiarize them with the sounds. Subsequently, they performed one block of baseline categorization to which they did not get any feedback (not reported here). During the main experiment sounds were randomly presented and participants had to identify the sounds as either belonging to category A or B. Which sound was categorized as A or B was counterbalanced over participants. Participants were required to close their eyes during the whole sessions and received auditory feedback to their performance. The interval after the participant's response and the next sounds was jittered between 1.5 and 2.5 sec. In total there were four blocks in which 576 sounds were presented in total, lasting approximately 35 minutes. EventIDE was used for stimulus presentation (OkazoLab Ltd, The Netherlands) and sound were presented via ER-30 insert earphones (Etymotic Research) at a comfortable sound level.

EEG recordings and pre-processing

32 channels EEG data was recorded with Brain-Vision Recorder (Brain Products; BrainCap MR). Channels included: Fp1, Fp2, F2, F3, C4, C4, P3, P4, O1, O2, F7, F8, PO3, PO4, P7, P8, Fz, Cz, Pz, FC1, FC2, CP1, CP2, FC5, FC6, CP5, CP6, Oz, and A1. A2 was used as online reference, and Afz as ground. Three additional channels were included to measure eye movements (left and right from outer cantus and below the left eye). Data was recorded at a 5000 Hz sampling rate using hardware filters

with a bandpass of 0.01-1000 Hz and an additional 100 Hz low-pass software filter. A BrainAmp MR Plus EEG amplifier was used. Impedance was kept below 10 kiloOhm.

For the pre-processing we cut the data from -3 to 2 around sound onset. Data was re-referenced to the average of all channels, demeaned, and resampled to 1000 Hz. Bad trials were removed via visual inspection and bad channels were interpolated. ICA was performed to remove remaining eye movements and muscle artefacts.

Quantification and statistical analysis

Behavioural analysis: We fitted a psychometric curve to the data assessing the proportion that the participant identified the sounds as sound A (for the participants with reversed categorization, we recoded the sound identities). A psychometric function was fitted to this data using a probit function (guessing and lapsing rate at 0, using the frequency as independent variable, and proportion sound A as dependent variable; Modelfree fitting toolbox version 1.1. (Zchaluk and Foster, 2009)). For later analysis we extracted the 20 and 80 percentile for each participant.

EEG phase-categorization correlation: for frequencies ranging from 2 to 15 Hz (in steps of 0.1 Hz) we extracted the phase at stimulus onset. We did this by cutting the data 3 cycles prior to sound onset until sound onset and performing a fast Fourier analysis with Hanning tapers. Thus, for each frequency another window was chosen. All analyses were restricted to sounds that were identified below 80% correct to avoid ceiling effects. Still, to ensure that the effects were not due to physical differences in the stimuli, we performed a GLM with a binomial distribution on the response choice data (sound A or sound B) per participant with stimulus type as factor to remove any effects of stimulus type. The residuals of this analysis were used in a circular-linear correlation with pre-stimulus phase. The same correlation was repeated for 1000 times using permuted labels of the categorization, thereby creating a null distribution for the correlation. Dependent samples t-tests were performed between the actual correlation and the median of each individual's null distribution. Cluster statistics was used to correct for multiple comparisons ('nonparametric_individual' cluster threshold, with 'maxsum' clusterstatistics. We tested one-sided as circular-linear correlations are only positive;(Maris and Oostenveld, 2007)). The same analysis was repeated but using the phase opposition index as proposed in (VanRullen, 2016).

Positive clusters were further investigated by extracting for the maximum t-value within the cluster the phase angles per participant. We performed a Rayleigh test of the mean phases for sound A and sound B categorization over participants to test for phase consistency over participants. To test for any systematic phase clustering the phase opposition index (VanRullen, 2016) was calculated per participant. Group statistics was performed by inversing the p-values of the permutations of individual participants to z-values and performing a z-test.

EEG phase-accuracy correlation: Instead of modulating the response of the participants, phase could modulate the behavioural performance of the participant, as previously found for detection

studies(Mathewson, Gratton, Fabiani, Beck and Ro, 2009; Ten Oever, Van Atteveldt and Sack, 2015; Busch, Dubois and VanRullen, 2009; Hanslmayr, Volberg, Wimber, Dalal and Greenlee, 2013). To test this hypothesis, we repeated the same analysis was performed as described above in *"EEG phase-categorization correlation"*, but instead the correlation was based on phase with residuals of the accuracy instead of categorization.

Phase dependent TRF: For the significant *EEG phase-categorization correlation* frequency bins we investigated whether the evoked responses' similarity to either one of the two sound categories was also modulated by phase. To do so, we estimated the temporal response function (TRF) for each trial with the sound envelope of pitch 1 sounds and the envelope of pitch 12 sounds. The TRF is an encoding model and is calculated via the linear convolution of a specific input (here, the sound envelope) with a measured output (here, EEG), thereby providing an estimation over time how well the systems output can be estimated with a particular input property (Crosse, Di Liberto, Bednar and Lalor, 2016; Lalor, Power, Reilly and Foxe, 2009). The envelope was estimated by zero padding the sounds with 100 ms on either side, extracting the absolute of the Hilbert transform and resampling the sounds to 1000 Hz (matching the sampling rate of the EEG). EEG was epoched for -0.1 – 0.6 seconds around sound onset. Trials with sound pitches that were identified under 80% accuracy were extracted and for each trial we estimated the TRF with sound envelope of pitch 1 and pitch 12 sounds (using envelopes of sounds matching the velocity and density of the original sounds) using the mTRF toolbox (Crosse, Di Liberto, Bednar and Lalor, 2016). The lambda of the estimation was set to a 1000, based on fitting the TRF of the trials containing the extreme sounds (independent trials).

To estimate phase dependency of the estimated TRF we subtracted for each trial the TRF estimated with pitch 12 sound envelopes from the TRF estimated with pitch 1 sound envelopes (TRF1-TRF12). This TRF difference was used in a circular correlation with pre-stimulus phase (at frequencies determined by the *EEG phase-categorization correlation* analysis, using the same pre-stimulus estimates).

Estimating the circular correlation on the TRF difference allowed us to control for baseline TRF amplitude shifts caused by a different phase at stimulus onset: the baseline shift would be subtracted out from the TRF12-TRF1 calculation. Thereby, we could investigate whether the amplitude of the TRF1 vs TRF12 was modulated by the phase at stimulus onset, that is, whether the EEG response resembled sound 1 or sound 12 more dependent on the phase of the sound presentation. The TRF difference estimates were statistically compared to an estimated chance correlation calculated with permuted TRF difference – pre-stimulus phase comparisons (n = 500; using the median of the null distribution for each participant per time-frequency point). Dependent samples t-tests were performed between the actual correlation and the median of each individual's null distribution. Cluster statistics were used to correct for multiple comparisons ('nonparametric_individual' cluster threshold, with 'maxsum' clusterstatistics. One-sided alpha).

The same analysis was repeated but subtracting the TRF of the correct sound category of the TRF from the incorrect sound category at frequency ranges identified in the "*EEG phase-accuracy*

correlation". If phase modulates the accuracy of the participants, it is expected that the difference between TRF for correct and incorrect sound categories is bigger for specific phase ranges.

Behavioral and TRF phase comparison: The phase of the behavioral and TRF results were compared by calculating the phase difference per participants. The phase of the behavior was estimated from the frequency and channel of the maximum t-value within the cluster. The phase of the TRF was estimated at the time point of the maximum t-value within any cluster at the channel of the behavioral effect. The non-uniformity of the phase difference around zero was tested using the v-statistics.

Performance- phase-modulation correlation: In the final analysis we investigated whether the strength of this phase modulation had an influence on their overall discrimination performance. As such we extracted for each participant the phase modulation index: the difference between the observed correlation from the median of the null distribution divided by the median of the null distribution. This was extracted for all frequency ranges previously identified to influence behavioural responses, for the phase modulation of discrimination and accuracy. This phase modulation index was correlated with the slope of the psychometric curve, an index of how well participants could discriminate the sounds. A positive correlation would indicate that participants with stronger phase modulation had a higher discriminative performance.

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