NeuroImage 195 (2019) 444–453

Contents lists available at ScienceDirect

NeuroImage

journal homepage: www.elsevier.com/locate/neuroimage

Eye movements explain decodability during perception and cued attention in MEG

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ARTICLE INFO

Keywords:
MEG
Decoding
Eye movements

ABSTRACT

Eye movements are an integral part of human perception, but can induce artifacts in many magneto-encephalography (MEG) and electroencephalography (EEG) studies. For this reason, investigators try to minimize eye movements and remove these artifacts from their data using different techniques. When these artifacts are not purely random, but consistent regarding certain stimuli or conditions, the possibility arises that eye movements are actually inducing effects in the MEG signal. It remains unclear how much of an influence eye movements can have on observed effects in MEG, since most MEG studies lack a control analysis to verify whether an effect found in the MEG signal is induced by eye movements. Here, we find that we can decode stimulus location from eye movements in two different stages of a working memory match-to-sample task that encompass different areas of research typically done with MEG. This means that the observed MEG effect might be (partly) due to eye movements instead of any true neural correlate. We suggest how to check for eye movement effects in the data and make suggestions on how to minimize eye movement artifacts from occurring in the first place.

1. Introduction

While eye movements are often treated as artifacts during neuroimaging studies, understanding their important role in visual perception makes it clear they cannot just be treated as random artifacts, that could not be influenced in a consistent way by the visual stimuli we present to our participants. Humans have a limited capacity to observe their outside world and the direction of our eyes determines which part of our world can be observed. This is the reason eye movements are constantly made. A long history of research into eye movements has identified several different types of eye movements. Large eye movements or 'saccades' serve to aim the most sensitive part of the retina, the fovea, at an area of interest in our visual environment (Kowler et al., 1995). Saccades can be triggered through different mechanisms, most notably by stimuli in the environment (exogenous) or by internal expectations (endogenous) (Godijn and Theeuwes, 2002). The role of small fixational eye movements or microsaccades is more debated, but seems also relevant for enhancing fine spatial detail during the recognition of gratings (Rucci et al., 2007).

In object recognition tasks, human participants tend to make consistent eye movements related to a particular object (Yarbus, 1967; Peterson and Eckstein, 2012). Eye movements may be used to efficiently extract task relevant information from the environment (Yang et al., 2016).

However, in many neuroimaging studies it is attempted to minimize the occurrence of eye movements, since the muscle contractions lead to an electrical current that causes large deviations in the magnetic field that are picked up by MEG, EEG and fMRI techniques. Besides these artifacts caused by muscle contractions, eye movements also induce neural effects, like motor planning and retinal shifts in the visual cortex. While the artifacts induced by muscle contractions depend on the imaging technique used (generally worse in MEG and EEG than in fMRI), there is no dependence of the neural effects on the imaging technique used.

Here we specifically focus on the effect of eye movements on the MEG signal. In MEG, electrooculography (EOG) signals are often recorded to detect any artifacts that are being caused by eye movements or blinks. These EOG signals can inform the analysis for artifact removal in the MEG signal. There are many different techniques for removing artifacts, but generally two strategies are adopted: either remove an entire trial or part of a trial that is contaminated with artifacts, or reduce these artifacts, often by linear transformations or regression techniques (Woestenburg et al., 1983; Vigário et al., 1998).

While it is attempted to reduce the influence of artifacts on the MEG

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https://doi.org/10.1016/j.neuroimage.2019.03.069
Received 25 October 2018; Received in revised form 27 March 2019; Accepted 28 March 2019
Available online 3 April 2019

signal as much as possible, it is not clear to what extent this actually succeeds and how much of a problem that is. It has been shown that even small fixational eye movements, that are difficult to exclude from the MEG signal, influence electrophysiological responses (Yuval-Greenberg et al., 2008; Dimigen et al., 2009).

If eye movements just lead to random artifacts that reduce the signal-to-noise ratio in our MEG signal, this is a nuisance that increases our chances of finding false negatives (type II errors). In contrast, systematic and condition-specific eye movements pose a more serious problem, since they could lead to false positives (type I errors) from the MEG signal that are not caused by any brain-related responses, but purely by the artifacts induced by the eye movements.

We investigated the potential confounding influence of eye movements on MEG effects here by analyzing an MEG dataset previously recorded in the context of a working memory match-to-sample task. We extracted two periods of interest from the trials that we further analyzed for potential eye movement related effects. The first period considered purely perceptual responses to presented orientation stimuli at one of four retinotopic quadrants. During the second period no stimuli were shown, but participants received a cue telling them from which quadrant they has to retrieve the stimulus from working memory. This condition closely matches cued-based attention paradigms and any results found here are expected to extend to similar setups. During both conditions an decoding analysis on the spatial location in which stimuli are presented (or cued) is performed to assess the decodability of retinotopic representations in MEG.

During recording and preprocessing of this data we follow any good practices for MEG experiments given in (Gross et al., 2013), to ensure solid methodology. We followed the standard procedure of excluding trials showing excessive blinks or eye movements and compare two commonly used methods to correct the remaining artifacts to quantify how efficiently they solve eye movement-related confounds. The first method uses an independent component analysis (ICA), where the individual components are correlated with the electrooculography (EOG) signal to remove any components that have a Pearson correlation higher than 0.3 with the vertical or horizontal EOG signal. The second method regresses the eye movement signal directly onto the MEG data to remove any activity explained by the eye movements. The remaining residuals are then used for further processing.

We find problematic artifacts from eye movements both in the perception condition and the cued attention condition that are systematic and lead to significant effects. Neither of the two commonly used artifact removal methods solves this problem. We propose that any study concerning MEG data should control their findings for effects caused by eye movements, especially in studies that use stimuli that are not foveally presented. We also show that controlling for eye movement artifacts by using the EOG signal is not sufficient to eliminate these effects. We strongly recommend using a high-quality eye tracker to record eye movements in MEG studies. An eye tracker is more sensitive to small eye movements and can pick up on artifacts that may otherwise be missed.

2. Methods

2.1. Participants

We recorded MEG and eye tracking data from 19 participants. The first two participants were removed from further analysis because of technical difficulties during acquisition. One other participant showed considerable movement (> 5 cm) throughout the experiment, and completed less than half of the trials. We removed this participant due to large artifacts. All participants gave written informed consent and were between 18 and 29 years old (11 female, 5 male). The study was approved by the local ethics committee and conducted according to the corresponding ethical guidelines (CMO Arnhem-Nijmegen).

2.2. Procedure and experimental design

The experimental design is shown in Fig. 1. At the start of a trial, three oriented gratings were shown sequentially, each at one of four locations. This was followed by a delay period, during which participants were instructed to remember the stimulus location and orientation. After this, a retro-cue indicated one of the locations where a stimulus was shown. After another delay, participants reproduced the orientation of the cued stimulus. A central fixation circle was visible during the entire trial. Participants were instructed to maintain fixation during the entire trial and only make eye movements between trials.

A trial started with a cue that filled the fixation circle. After 500 ms from the start of the trial the first stimulus was shown for 200 ms as a grating in one of four locations (left bottom, left top, right top or right bottom). After an interstimulus interval (ISI) of 800–1100 ms, the second grating stimulus was shown for 200 ms in one of the remaining locations. After another interstimulus interval (ISI) of 800–1100 ms the third grating stimulus was shown for 200 ms in one of the locations where no stimulus was shown yet. A delay period of 1400–1800 ms followed the third stimulus, after which a cue (shown for 500 ms) indicated which stimulus to retrieve. After another delay of 1400–1800 ms, participants performed an orientation matching task in which they rotated a bar to match the orientation of the cued stimulus. Participants responded by pressing 2 buttons rotating the bar left and right. The orientation at the end of the response period was registered as final answer. They had 3000 ms to perform the orientation matching. At the end of the trial, participants received feedback indicating how well they matched the cued stimulus. Between trials there was an intertrial interval (ITI) of 1800–2000 ms.

2.3. MEG recording

Data were recorded at 1200 Hz using a 275-channel MEG system with axial gradiometers (VSM/CTF Systems, Coquitlam, BC, Canada). For technical reasons, data from five sensors (MRF66, MLC11, MLC32,

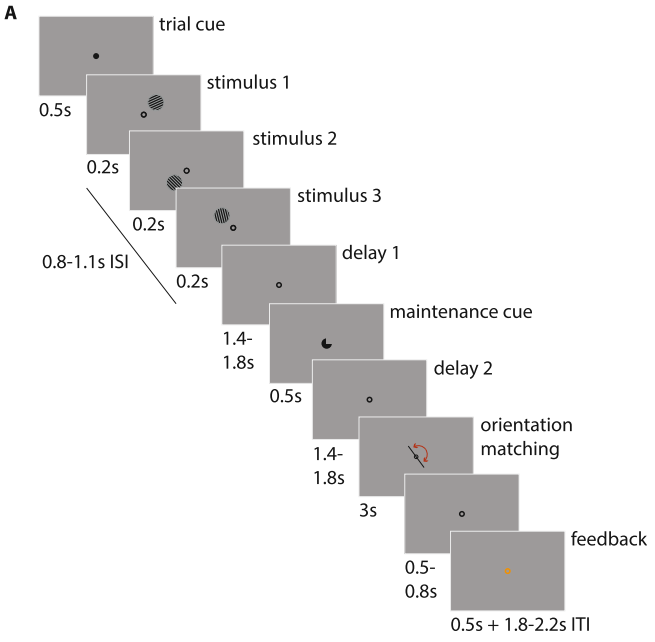

Fig. 1. Experimental design. Participants performed a working memory task in which three grating stimuli were shown sequentially for 200 ms each, at one of four different locations in the visual field. Between the stimuli there was a 800–1100 ms interval. After the final stimulus a delay of 1400–1800 ms followed. Next, one of the locations was cued for 500 ms and following another delay of 1400–1800 ms, participants had to match a rotating bar to the orientation of the cued stimulus.

MLF62, MLO33) were not recorded. Subjects were seated upright in a magnetically shielded room. Head position was measured using three coils: one in each ear and one on the nasion. Throughout the experiment head motion was monitored using a real-time head localizer (Stolk et al., 2013). If necessary, the experimenter instructed the participant back to the initial head position during the breaks. This way, head movement was kept below 8 mm in all participants included in the analysis. Furthermore, both horizontal and vertical electro-oculograms (EOGs), as well as an electrocardiogram (ECG) were recorded for subsequent offline removal of eye- and heart-related artifacts. For the recorded EOGs we used four electrodes. Two electrodes beside the left eye and right eye, in line with the pupil. And two underneath and above the left eye, in line with the pupil. Eye position and pupil size were also measured using an Eye Link 1000 Eye tracker (SR Research).

Data were analyzed with MATLAB version R2017b and FieldTrip (Oostenveld et al., 2011). Per trial, two stages were defined. The first perception stage was defined as 200 ms prior to onset of the first stimulus until 300 ms after the offset of the first stimulus. The second stage started at the moment that the maintenance cue appeared on the screen, until 1400 ms after offset of the maintenance cue. As a baseline correction, for each stage, the activity during 300 ms from the onset of the initial fixation of that trial was averaged per channel and subtracted from the corresponding signals. The data were down-sampled to 300 Hz to reduce memory and CPU load. Line noise at 50 Hz was removed from the data using a DFT notch filter. To identify artifacts, the variance of each trial was calculated. Trials with high variance were visually inspected and removed if they contained excessive artifacts. After artifact rejection, on average 255 trials per subject remained for analysis. To remove heart rate artifacts, independent components of the MEG data were calculated and correlated with the ECG signal. Components with high correlations were manually inspected before removal. The eye tracker data was cleaned separately by inspecting trials with high variance and removing them if they contained blinks or other excessive artifacts.

The main analysis was performed without correction of eye movement artifacts to determine the scale of the problem and ensure that the eye movement artifact correction methods themselves do not induce unwanted effects. Subsequently the effect of two commonly used eye movement artifact correction techniques was compared to determine to what extent they are able to solve the effects of eye movements in the MEG data. The first method uses an independent component analysis (ICA), where the individual components are correlated with the electrooculography (EOG) signal to remove any components that have a Pearson correlation higher than 0.3 with the vertical or horizontal EOG signal. The second method regresses the eye movement signal directly onto the MEG data to remove any activity explained by the eye movements. The remaining residuals are then used for further processing. We tested this both with regressing the EOG signal and the eye-tracker signal onto the MEG data.

2.4. Decoding analysis

To track the neural representations within the perception and the cued attention stage, we decoded the location in which the stimulus was presented from the preprocessed MEG signals during the first stimulus and after the maintenance-cue for every time point. We used a multinomial logistic regression classifier with the activity from the 270 MEG sensors as features (see ref (Bishop, 2006). for more details). A 5-fold cross-validation procedure was implemented where for each fold the classifier was trained on 80% of the trials and tested on the other 20%. To prevent a potential bias in the classifier, the number of trials per class was balanced per fold by randomly removing trials from the class with the most trials until the trial numbers were equal between the classes. During the cued attention stage a cross-condition decoder was used which was trained on the perception trials and tested on the cued attention trials. This ensured that decoding was due to stimulus specific neural activation and ruled out any influence the cue could have.

2.5. Statistical testing

Decoding accuracy was tested against chance using two-tailed cluster based permutation testing with 500 permutations that tested significance over participants (Maris and Oostenveld, 2007). Two dimensional clusters were formed over the training time and testing time dimensions. In the first step of each permutation, clusters were defined by adjacent points that crossed a threshold of $p < 0.05$. The t-values were summed within each cluster, but separately for positive and negative clusters, and the largest of these were included in the permutation distributions. A cluster in the true data was considered significant if its p-value was less than 0.05 based on the permutations.

2.6. Mutual information

Based on the decoding results on different trials, a measure of mutual information between the two sources of eye movement data and MEG data was calculated over all correctly classified MEG trials, to see how much information was shared between the MEG decoding results and eye movement decoding results. The mutual information gives in this case the amount of information that is obtained about the MEG decoding results by observing eye movement decoding results. Higher values indicate that the eye movement data can explain more of the decoding information in the MEG data. The mutual information between two sources of data, X and Y is given by

$$I(X;Y) = H(Y) - H(Y|X) \tag{1}$$

where $H(Y)$ and $H(Y|X)$ represent the entropy of Y and the conditional entropy of Y given X respectively. In terms of probability distributions this can be rewritten

$$I(X;Y) = \sum_{x\in X, y\in Y} p(x,y)\log\left(\frac{p(x,y)}{p(x)p(y)}\right) \tag{2}$$

where $p(x,y)$ in the joint probability distribution of X and Y and $p(x)$ and $p(y)$ are the marginal distributions of X and Y respectively.

2.7. Eye movements

To determine the size of the eye movements made during a trial the EYE-LINK signal was used. The size of the eye movements were calculated as the maximum eye position displacement during a trial relative to the eye position at onset of the trial. The size of eye movements is expressed in degrees of visual angle unless stated otherwise.

3. Results

3.1. Consistent eye movements decodable during perception and cued attention

To study whether eye movements are a confounding factor in typical MEG experiments, we used a working memory match-to-sample task that could be divided into two periods of interest. The first period was purely perceptual, and lasted from 200 ms before onset to 300 ms after onset of the first stimulus of every trial. The second period of interest started when the cue indicated which of the stimuli should be retrieved for the match-to-sample task at the end of the trial. This period lasted from cue onset to 1400 ms after cue offset. To assess the decodability of the location where a certain stimulus was presented, we trained and tested our classifier at different time points during the perception stage to create a generalization matrix (King and Dehaene, 2014).

First, we trained and tested this classifier on the MEG data without any of the artifact removal methods applied, to examine the scale of the problem and ensure that the artifact removal methods themselves do not induce unwanted effects. We could decode with high accuracy in which

of the four locations a stimulus was presented from the MEG signal (accuracy = 36.7%, $p < 0.002$) (Fig. 2A). The first peak of significant decoding was observed from 60 to 100 ms after stimulus onset, while the higher second peak was reached at around 100–150 ms. This second peak generalized well to later time points, indicating that it contained a stable signal representing the location of the presented class. At around 200 ms, we observed a longer period of prolonged decodability, which generalized well over time. This pattern of the generalization matrix is typically observed during perceptual tasks in MEG (Carlson et al., 2013; Cichy et al., 2014; Dijkstra et al., 2017).

Then, we attempted to decode stimulus location from the measured EOG signal, which is typically used for the removal of eye-movement artifacts. The EOG signal contained decodable information for the classifier (accuracy = 35.2%, $p < 0.002$) (Fig. 2B). We observed clearly generalizing decodability after around 200 ms from stimulus onset, coinciding with the block in decodability observed in the MEG signal (see Fig. 2A). This delay of 200 ms from stimulus onset matches typical delays for saccade onset in humans (Carpenter, 1988). We also measured eye movements using an EYE-LINK eye tracker, to see whether the typically used EOG signal is sufficient for removing eye movement confounds. From this eye-tracker signal we observed a similar block of decodability as in the EOG (accuracy = 41.8%, $p < 0.002$), but with higher accuracy than for the EOG signal ($p < 0.002$) (Fig. 2C).

Next, we investigated whether eye movements can also be a problem when there are no stimuli shown directly at one of the locations, but only a cue at the fixation dot is shown in the center of the screen. To this end, we tested our classifier on the second (cued attention) period. We used a cross-condition classifier that we trained on the perception period and tested on the cued attention period, to ensure that the trained classifiers were based on the actual location of the stimulus and not on some artifact induced by the cue.

The generalization matrix showed an decoding accuracy during the cued attention period that is a lower than during the attention period (Fig. 3A), but still shows significant decoding from the MEG signal. The same time points that showed large temporal generalization within perception also generalize to the cued attention period. The period of the second peak from 100 to 200 ms in the perception period generalized to the same 100–200 ms period after cue onset during the post-cue period. Subsequently, we observed a larger block that generalized over the entire post-cue period. Decodability only reached significant levels for the last part of this block (accuracy = 29.1%, $p < 0.05$). This second block was also clearly visible in the EOG and eye-tracker. While decoding was not significant from the EOG signal (Fig. 3B), it was very strong for the eye-tracker data (accuracy 41.0%, $p < 0.01$) (Fig. 3C).

These results show that both during perception as well as during cued attention, eye movements are induced that are consistent enough to significantly decode from. The data recorded via the eye-tracker eye tracker showed a much stronger effect than those from EOG.

3.2. Information from eye movements in MEG signal

The previous results indicate that there are indeed stimulus related eye movements during both tasks. These eye-movements could confound the MEG signal which, in the worst case scenario, would mean that MEG decoding can be fully explained by eye movements. It is also possible that the MEG and eye-movement related signals are independent and lead to good decoding accuracies on different trials.

To test this we checked whether the size of the eye movements correlated with the MEG decodability. The participants from whom we could decode best from the MEG signal after 200 ms, were also those that made the largest saccades on average ($c = 0.86$, $p < 0.0001$) (Fig. 4A).

To further test whether the decodability in the MEG signal is independent of eye movements or if they share the same information, we calculated the mutual information between correctly classified MEG trials and the corresponding classes decoded from the EOG and eye-tracker signals of each subject at every time point (Fig. 4B and C). This mutual information reveals how much information is shared between the eye-movement data and the MEG data. During the perception period, MEG decoding from 150 ms onward could be explained by eye-movements. The eye-tracker signal shares more information with the MEG signal than the EOG signal does, thus indicating that the eye-tracker is a much better predictor of eye-movement related effects in MEG. During the post-cue period, information from the eye movements was found in the MEG signal after 500 ms from cue onset, showing a much slower effect on eye movements due to cued attention. Here the EOG barely showed relevant information leaking into the MEG signal, while the eye-tracker signal had a very strong effect.

Together these results clearly reveal that the information coming from eye movements can be found in the MEG signal. Furthermore, the signal from the eye-tracker is a much better source to check whether our MEG data is contaminated than the EOG signal.

3.3. Effect of different eye movement artifact removal techniques

To see to what extent different techniques for the removal of eye movement artifacts reduced their confounding effect, we performed two

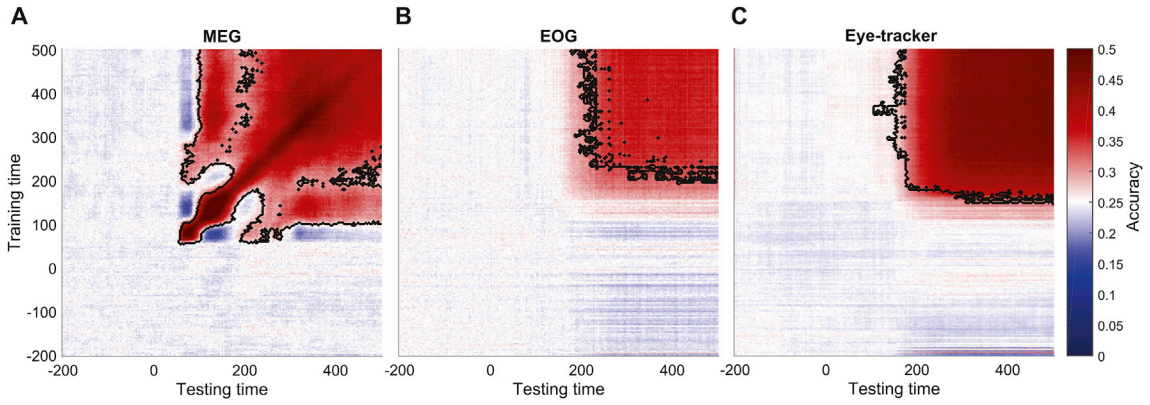

Fig. 2. Significant decoding from eye movements is observed during perception. The average generalization matrices over all participants during the perception stage for the MEG, EOG and eye-tracker signal are shown. **(A)** The MEG signal shows an early peak from 60 to 100 ms that does not generalize well over time. A second peak from 100 to 150 ms does generalize to later time points beyond 200 ms. From 200 ms onward the signal generalizes well across all time points. **(B)** In the EOG signal we do not observe the early peaks that were visible in the MEG signal. From 200 ms onward we see the same generalization as in the MEG signal though. **(C)** In the eye-tracker signal the same effect is visible, but decoding is much stronger than from the EOG signal.

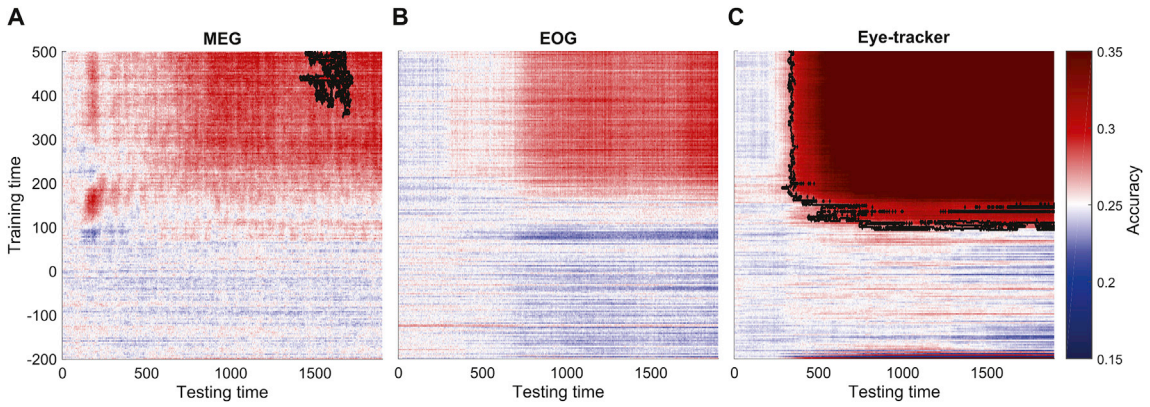

Fig. 3. Significant decoding from eye movements during cued attention. The average generalization matrices over all participants during the cued attention stage for the MEG, EOG and eye-tracker signal are shown. Classifiers are trained on the first perceptional stimulus of all trials. **(A)** The MEG signal shows an early non significant peak from 100 to 200 ms that does not generalize well over time. From 200 ms onward the signal starts to generalize across all time points, but significance is only reached after 1500 ms in the cued attention phase. **(B)** In the EOG signal the generalization beyond 200 ms is also visible, though weaker, not reaching significant values anywhere. **(C)** In the eye-tracker signal the decoding is much stronger than from the EOG signal, reaching significant values during the cued attention phase from 400 ms onward.

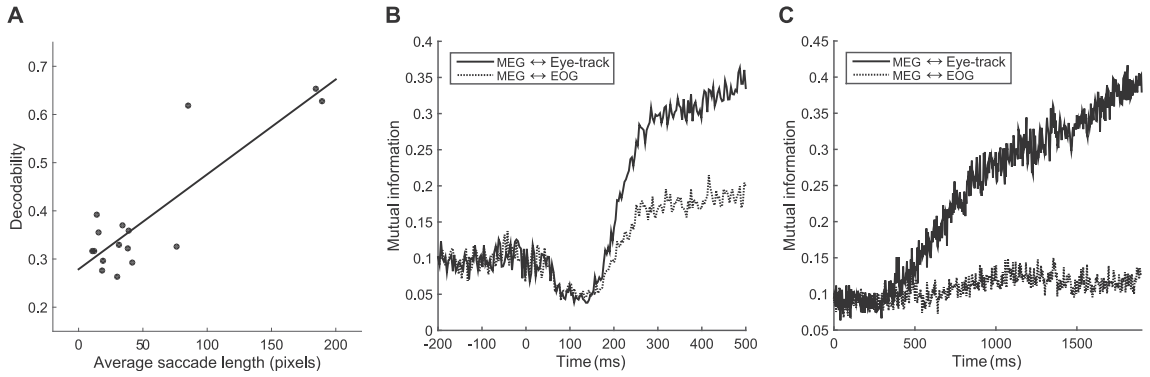

Fig. 4. Information from eye movements in MEG.(A) The participants making the largest eye movements were also those that showed the highest decodability from the MEG signal during the perception phase. **(B)** The mutual information between the MEG ↔ EOG and MEG ↔ eye-tracker data showed a strong increase after 200 ms during the perception stage. The eye-tracker data explained much more information from the MEG signal than the EOG data. **(C)** During the cued attention phase the difference is even more striking. Showing a strong increase in the information content of the eye-tracker data after 400 ms, while such an increase is absent in the EOG data.

standard procedures. The first, independent component analysis (ICA), is used to identify independent components of MEG activity that have a high correlation with the EOG signal, which are subsequently removed (see Methods section for more details). The second technique uses a linear regression to regress the eye movement signal directly on every MEG sensor. The remaining residuals of the MEG signal are then used for further analysis. We tested both regressing the EOG and regressing the eye-tracker signal from the MEG data. For all these three cases we performed the same analysis as in Figs. 2 and 3. The decoding accuracies were similar to the MEG data without eye movements removed both during perception (Fig. 5A–C) and cued attention (Fig. S1 A-C). The largest decrease in accuracy was observed when the eye-tracker signal was regressed out (Fig. 5, S1 D-F). However, for all these techniques there was only a small decrease in the mutual information between the decoding results from the MEG data and the decoding results from the EOG and eye-tracker data (Figs. 5 and S1 G-I), indicating that these techniques are not sufficient to completely remove the confounding effect of eye movements.

All these techniques use a linear relation between the MEG data and eye movement signals to identify artifacts, however, it is very likely that eye movements induce strongly non-linear effects in the neural activity of the brain, for example by retinal shifts or motor preparation. This would explain why the used methods are so poor at removing actual decoding effects due to eye movements from the MEG data.

3.4. Source of the eye movement artifacts

It is important to know how the eye movements exactly induce artifacts in the MEG signal, to understand what kind of techniques have to be developed to solve these confounds. There are different ways through which eye movements can cause artifacts in neuroimaging data. Generally we can distinguish neural effects (e.g. motor planning, or retinal shifts) from measurement effects (e.g. disturbances in the electrical current or magnetic field due to muscle contraction). How much both of these contribute is unclear.

Although it is hard to identify exactly how the eye movements induce artifacts, we expect measurement effects to be more apparent in frontal sensors, while neural effects would be more pronounced in occipital sensors. Identifying how eye movements contribute to the signal in these sensors could at least give is an idea about the source of these artifacts.

We used the linear regression of the previous section to identify the explained variance that the eye movements signal has on every MEG sensor (Fig. 6A). This revealed a much larger effect in frontal sensors compared to posterior sensors. A regression of the eye-tracker signal onto every sensor revealed a similar pattern (Fig. 6B, although the explained variance was much lower. These results would suggest that we are mainly dealing with measurement effects in our data. However, these regressions only explain linear effects and do not account for any non-linear effects in the data.

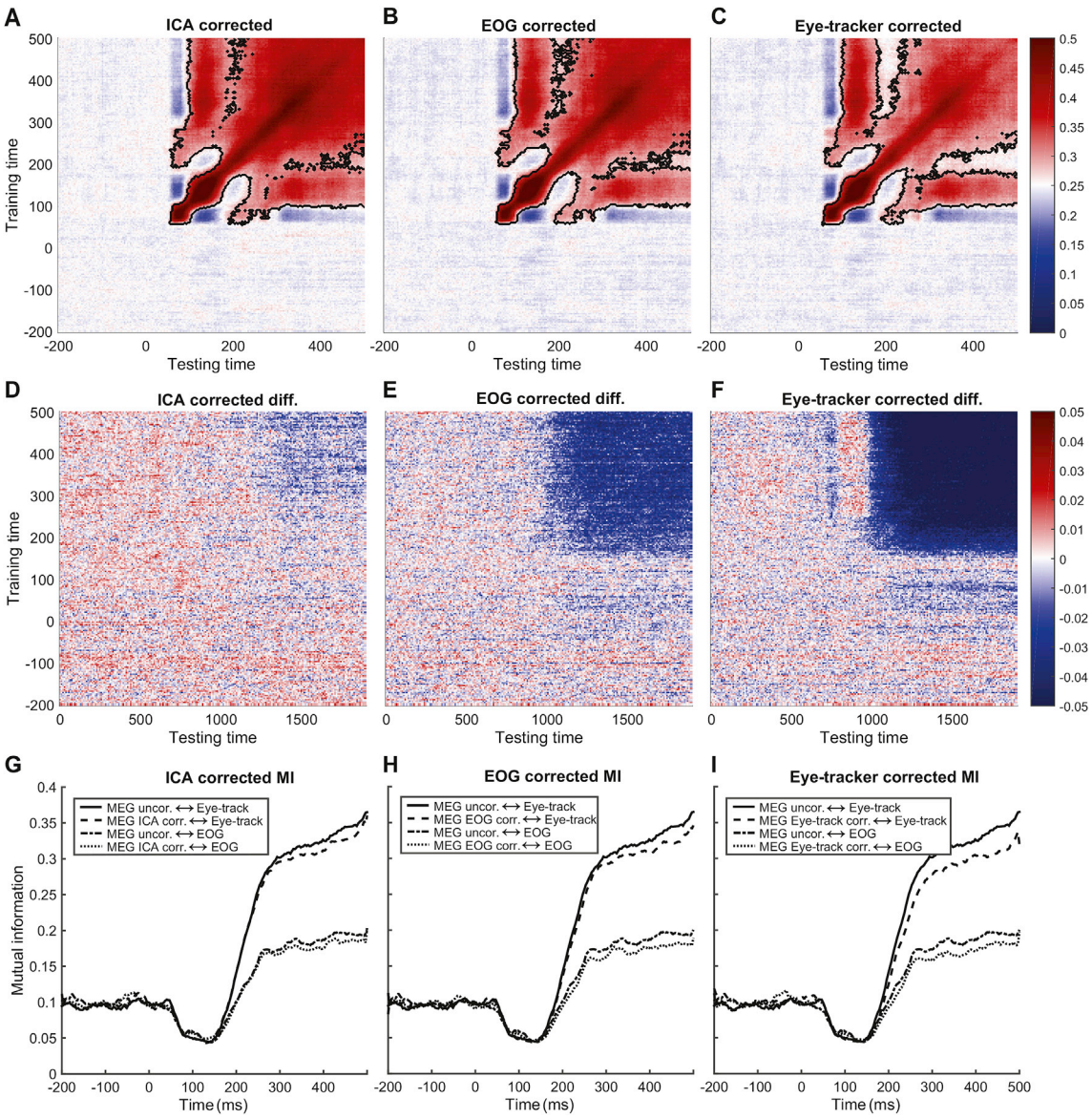

Fig. 5. Comparison of different techniques to remove eye movements during perception stage.(A,D,G) The first column shows results when the MEG data is corrected using ICA as described in the Methods section. The second and third column show the results when the MEG data is corrected using a linear regression of respectively the EOG **(B,E,H)** or eye-tracker **(C,F,I)** data onto every sensor separately. The top row shows the actual decoding results using the corrected MEG data. The middle row shows the difference in decoding between the uncorrected and corrected MEG data. The biggest decrease in decoding accuracy results from regressing out the eye-tracker data. The bottom row shows the mutual information between the uncorrected MEG data and both the EOG and eye-tracker, as well as the mutual information between the differently corrected MEG data and the EOG and eye-tracker signal. The data where the eye-tracker signal has been regressed also shows the largest decrease in mutual information, but there still remains a lot of shared information between the corrected MEG data and both the EOG and eye-tracker signal.

To get a better idea of these non-linear effects, we also performed our decoding analysis on a selection of the 40 most posterior sensors and a selection of the 40 most frontal sensors and used our mutual information measure to determine how these selections are influenced by eye movements (Fig. 6C and D). This revealed that the frontal sensors are indeed more strongly influenced by eye movements, but also the occipital sensors are influenced by these eye movements, most clearly revealed by the eye-tracker signal. These analyses reveal that both measurement and neural effects play a role in eye movement related confounds.

3.5. Eye-tracker, but not EOG picks up small fixational eye movements

To further reveal what drives how MEG decodability is influenced by eye-movements, we analyzed the eye-movements of the individual subjects more closely. For every subject, we averaged the eye movements over the trials of the different conditions, to see whether systematic eye-movements were made to the different locations where the stimuli were presented. This revealed subgroups of subjects that made strong saccades during the perception period (subjects 7, 10, 15, 16, average over participants: $3.3^{\circ} \pm 1.2^{\circ}$) and/or during the post-cue period (subjects 9, 10, 11, 16, average over participants: $3.2^{\circ} \pm 0.9^{\circ}$). These subjects were clearly not following the instruction to maintain fixation during the duration of respective part of the trial. The rest of the participants made much smaller eye movements, which can best be described as micro-saccades (average over participants: $0.6^{\circ} \pm 0.3^{\circ}$).

The average saccades for the large-saccade subgroups, corresponding to the different stimulus locations during both the perception period as well as the post-cue period, are shown in Fig. 7A,D. These subgroups made large saccades towards the respective location the stimulus was shown in, or where they were cued towards during the post-cue period.

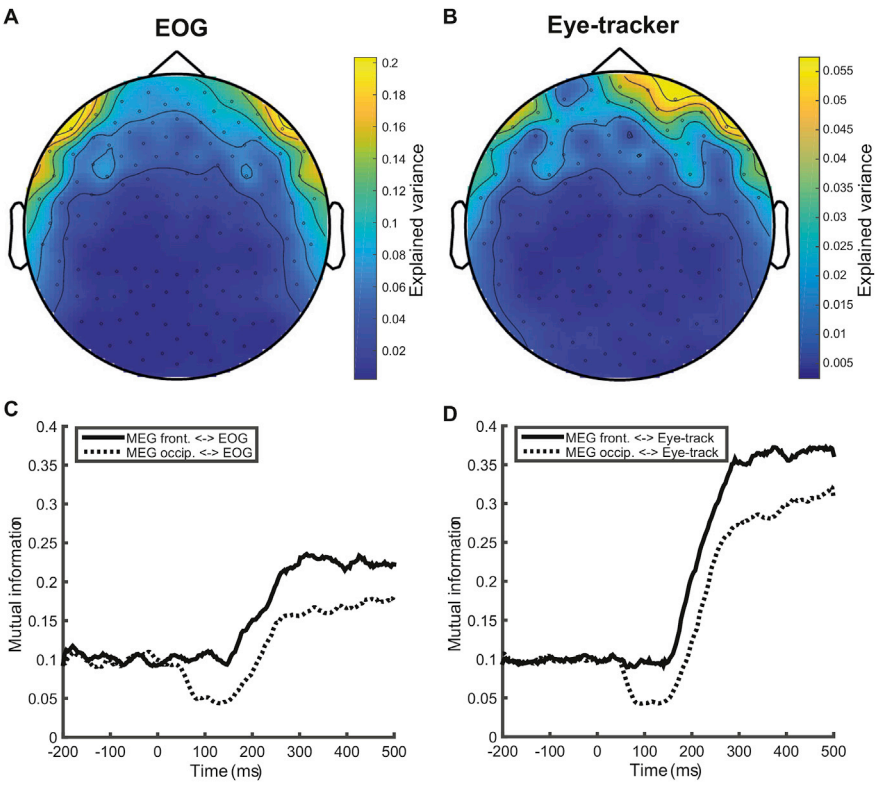

Fig. 6. Source of the eye movement confounds.(A,B) The explained variance of the regression of both the EOG and eye-tracker signal per sensor are shown. The explained variance was much stronger for the EOG signal, but both figures showed the strongest explained variance in frontal sensors. While this suggests that it are mostly frontal sensors that are influenced by eye movements and thus measurement effects are the main problem, it is possible that there are strong nonlinear effects that can not be revealed by such a linear regression. To check this possibility a decoding analysis was performed separately on 40 frontal and 40 occipital sensors. **(C,D)** The mutual information between decoding from frontal and occipital sensors and the EOG signal and EYE-LINK signal was determined, to identify the source of the confounding information in the MEG signal. While the mutual information between frontal sensors and the eye movement signals was largest, there is also a strong mutual information between occipital sensors and the eye movements signals. This indicates that also neural effects play a role in the eye movement confounds.

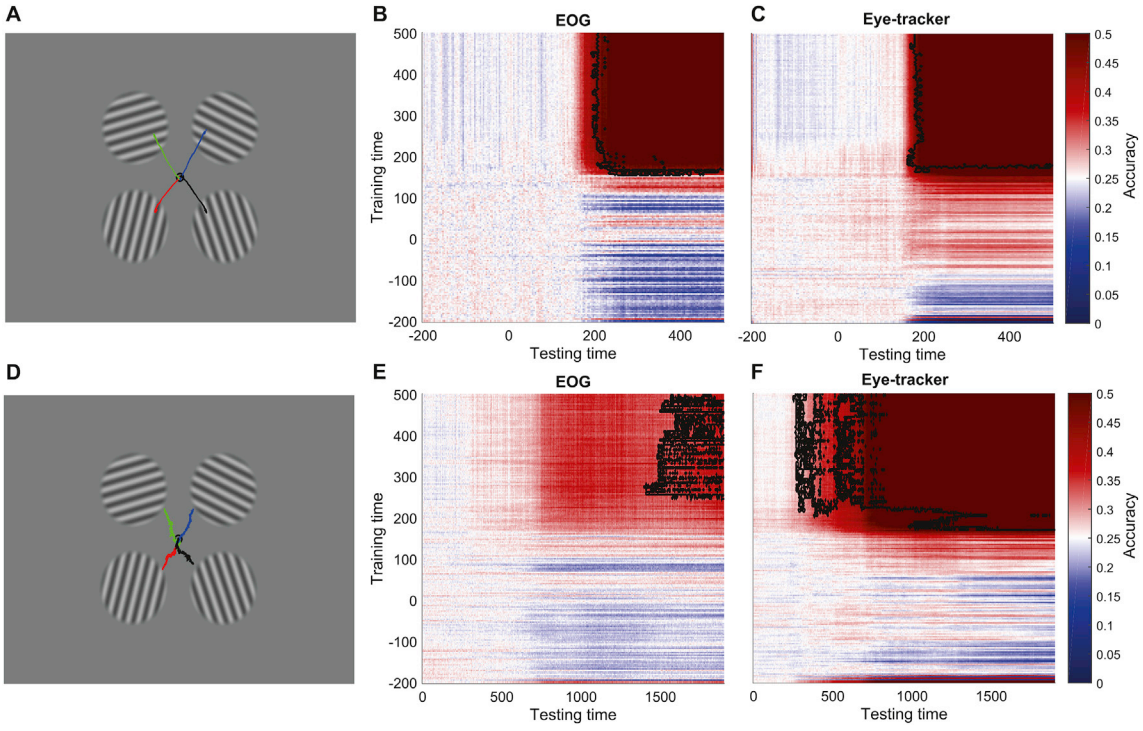

Fig. 7. Subgroup with large eye movements. A subgroup of the subjects made large eye movements against the instructions to fixate during the trial. **(A)** During perception subjects 7, 10, 15, 16 made large eye movements. **(B,C)** These were easily decodable both from the EOG data and the eye-tracker data. **(D)** During the cued attention stage large eye movements were made by subjects 9, 10, 11, 16. **(E,F)** These were also easily decoded from the eye-tracker data, but led to lower decoding accuracies in the EOG data.

From these subgroups with large saccades it was possible to decode stimulus location during the perception period both from the eye-tracker (accuracy = 78.3%, $p < 0.002$) as well as the EOG data (accuracy = 58.1%, $p < 0.01$) (Fig. 7B and C). Also during the post-cue period there was significant decoding of the cued location from both the eye-tracker (accuracy = 64.3%, $p < 0.002$) and EOG data (accuracy = 34.5%, $p < 0.002$), although the eye-tracker decodability was much stronger ($p < 0.002$) (Fig. 7E and F).

The average saccades for the subgroup making micro-saccades are shown in Fig. 8A,D). Although these eye-movements are much smaller, they are still consistently in the direction of the stimulated location. Interestingly there is still good decoding from the eye-tracker signal during both the perception period (accuracy = 31.1%, $p < 0.002$) and the post-cue period (accuracy = 36.5%, $p < 0.05$) from these micro-saccades (Fig. 8C,F), while there is very weak decoding from the EOG signals during the perception period (accuracy = 27.7%, $p < 0.01$) and no significant decoding during the post-cue period (Fig. 8B,E). This indicates that just using the EOG signal to check for eye movement induced effects is not enough, and makes a strong case for the recording of eye movements using an actual eye tracker.

4. Discussion

4.1. Summary results

We have shown that eye movements can be a major confounding factor in interpreting MEG signals. The effect of eye movements on results found in the MEG signal seems an underestimated problem, given that these types of control analyses are very rarely reported in MEG studies. Here we have shown two types of conditions (perception and cued attention) during which eye movements are a problem. The reported conditions are widely used in cognitive neuroscience study designs. The eye-movements lead to decodable information in the MEG even during the cued attention condition where no actual stimulus is shown. These effects are visible ~ 200 ms after stimulus onset during the perceptual condition or ~ 300 ms after cue onset during the cued attention condition. Given our approach we cannot say whether really is no eye movement effect during these time frames, or whether it is just a lack of sensitivity of our decoder. Experiments investigating early visual processing that happens within this time frame could use saccade onset times to determine from what time point eye movements could play a confounding role.

Yet more problematic is the fact that even participants that do tend to fixate properly, still make small eye movements that are consistent and decodable. This means that even throwing out 'bad' participants will not resolve our problem. We also showed that these eye-movements related effects cannot be fully picked up by EOG and that it is important to use actual eye-tracker data to check for impairing effects. This is especially the case for small eye-movements.

4.2. Eye movement effects in different cognitive tasks

Here we used a study design where stimuli were parafoveally presented. It is likely that this design makes it harder for the participants to suppress eye-movements than in a task where stimuli are presented foveally, as is also apparent from the fact that some of the participants did not strictly follow the instructions to fixate properly (Fig. 7). Many MEG studies use this type of stimulus presentation, though. Especially in attention research parafoveally presented stimuli are mostly unavoidable.

Recently, more studies have been reporting eye movement-related confounds in MEG studies that used foveally presented stimuli though. Consistent eye movement related differences were reported during the perception of house and face stimuli, though these did not seem to have a significant effect on the MEG signal (Dijkstra et al., 2018). During the perception of oriented gratings, consistent eye movements were reported that did confound the MEG signal (Mostert et al., 2017). Both studies used stimuli that were foveally presented and instructed their participants to keep fixation during relevant periods of the trials. This indicates that eye movements are not only a problem in studies using stimuli that

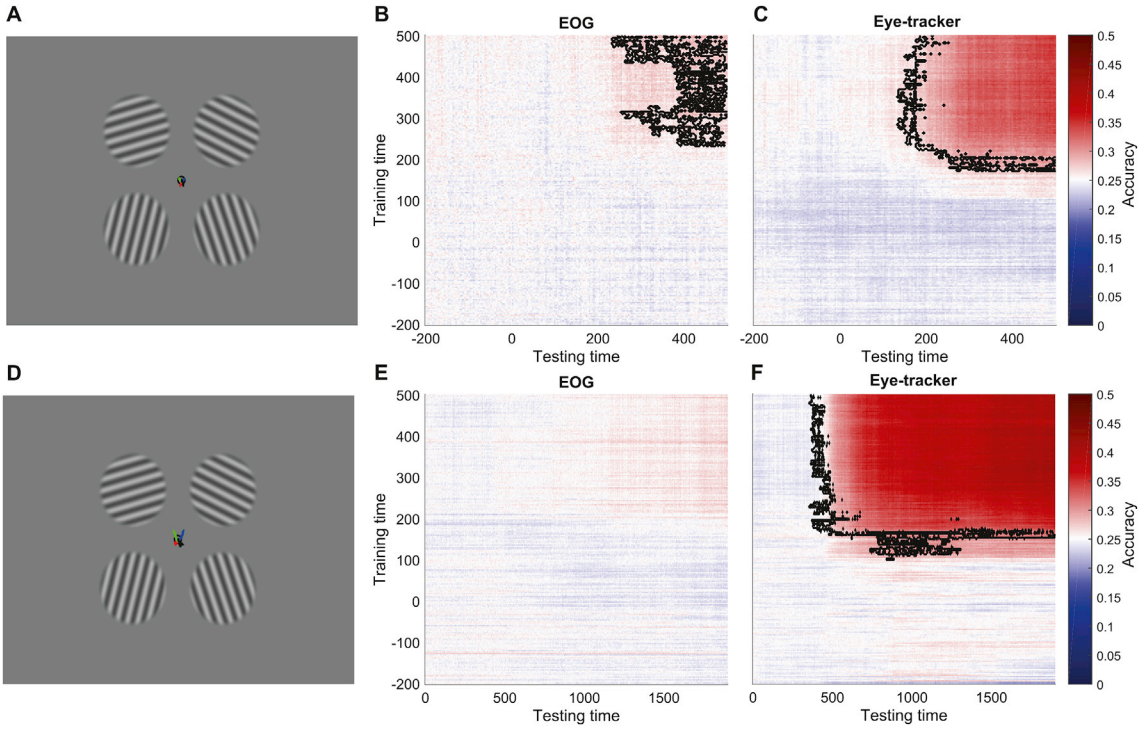

Fig. 8. Subgroup with small eye movements. The rest of the participants made small eye movements and followed the instruction to fixate correctly. **(A–C)** During perception eye movements were very small and very weakly decodable from the EOG data, but still easily decodable from the eye-tracker data. **(D–F)** During the cued attention stage the small eye movements could not be decoded from the EOG data, but led to strong decodability in the eye-tracker data.

are not presented foveally, but that it could be potential problem in any MEG study using visual stimuli. Further investigation to determine the scale of this problem is needed though. Reporting control analysis on eye movements in MEG studies will help us identify under which conditions these problems arise.

Whether consistent eye movements occur is also strongly task dependent. While an active task on grating stimuli induces consistent eye movement, there are no consistent eye movements during passive viewing of gratings (Mostert et al., 2017; Thielen et al., 2018).

4.3. Implications for other neuroimaging techniques

There are different ways through which eye movements can cause artifacts in neuroimaging data. Generally we can distinguish neural effects (e.g. motor planning, or retinal shifts) from measurement effects (e.g. disturbances in the electrical current or magnetic field due to muscle contraction). Both of these effects influence our MEG data. The measurement effects depend very much an the specific neuroimaging technique used. Where muscle contractions can cause large fluctuations in MEG and EEG sensors, it affects the fMRI signal much less. Neural effects do not depend on the neuroimaging technique used, and will affect any of them. If these are the major contributing factor to the effects found in our study, we expect that major confounding effects caused by eye-movements also affect fMRI studies. Future research should dissociate the different mechanisms through which eye movements contribute to experimental effects found with different neuroimaging techniques. Method development should aim to reduce the influence of measurement effects, while neural effects could be incorporated in better models that incorporate the important role eye-movements play in cognition.

4.4. Recommendations

The results of this study indicate that eye movements can explain effects in our MEG signal, that in turn can lead us to draw the wrong conclusions from our data. We therefore deem it essential that effects of eye movements are clearly reported in any MEG study. Performing the same effect size analysis as done on the MEG signal also on the EOG, or preferably actual eye tracker data, can inform to what extent eye movements can be the source of these effects. For studies using classification analyses, we recommend the use of mutual information measures between classifications from MEG and from a source of eye movement signal to check whether eye movements are actually a source of these effects, and whether the time window of interest is contaminated. For effect sizes based directly on the ERF signal, correlation between individual trial effect sizes might be used to check if effects are induced in the MEG signal by eye movements.

Given our comparison of the effects found by the EOG signal and those found by the eye tracker signal, we strongly recommend the use of an eye tracker. While the EOG might be suitable to detect strong effects on the MEG signal, there is a great risk of missing eye-movement-related confounds.

Any researcher conducting MEG experiments should be aware of the dangers of consistent eye movements, and should aim to reduce eye movements as much as possible (Tal and Yuval-Greenberg, 2018; Thaler et al., 2013). Experimental choices such as better fixation targets or online feedback to participants about their fixation can help reduce eye movements (Tal and Yuval-Greenberg, 2018; Thaler et al., 2013). Using decoders trained during passive viewing of the task stimuli can help to prevent eye movement confounds, since passive viewing seems to prevent consistent eye movements from occurring (Mostert et al., 2017; Thielen et al., 2018). To what extent such passive decoders work when neural activity is expected to differ greatly between passive viewing and an active task, such as when higher order areas of the brain are involved, has to be investigated further. Despite these different ways in which eye movement-related confounds can be prevented or reduced, we cannot ignore that eye movements are an important part of human cognition, and working towards models that explicitly incorporate eye movements will help elucidate their functional role in human cognition.

5. Conclusions

We conclude that when performing an MEG experiment it is essential to report a good analysis of eye movement related effects in your data. Especially studies with parafoveally presented stimuli are at risk and should be reported with great caution. More research should point out to what extend these effects occur under different types of tasks and to what extent they are detrimental to other neuroimaging techniques. Since eye-movements are inextricably linked to cognition there remains an important challenge for future research to find ways to move away from treating eye-movements as mere artifacts, but move towards more complete models which include eye-movements as interesting data points.

Data availability

All accompanying data to this article can be found online at http://hdl.handle.net/11633/di.dcc.DSC_2018.00111_468.

Acknowledgements

This research was supported by VIDI grant number 639.072.513 of The Netherlands Organisation for Scientific Research.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.neuroimage.2019.03.069.

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