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A review of the methodological challenges of combined eye tracking & EEG during natural vision begins on page “R”.

Co-Registration of Eye Movements and EEG in Natural Reading: Analyses & Review

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Brain-electric correlates of reading have traditionally been studied with word-by-word presentation, a condition that eliminates important aspects of the normal reading process and precludes direct comparisons between neural activity and oculomotor behavior. In the present study, we investigated effects of word predictability on eye movements (EM) and fixation-related brain potentials (FRPs) during natural sentence reading. Electroencephalogram (EEG) and EM (via video-based eye tracking) were recorded simultaneously while subjects read heterogeneous German sentences, moving their eyes freely over the text. FRPs were time-locked to first-pass reading fixations and analyzed according to the cloze probability of the currently fixated word. We replicated robust effects of word predictability on EMs and the N400 component in FRPs. The data were then used to model the relation among fixation duration, gaze duration, and N400 amplitude, and to trace the time course of EEG effects relative to effects in EM behavior. In an extended *Methodological Discussion* section, we review four technical and data-analytical problems that need to be addressed when FRPs are recorded in free-viewing situations (such as reading, visual search, or scene perception) and propose solutions. Results suggest that EEG recordings during normal vision are feasible and useful to consolidate findings from EEG and eye-tracking studies.

Keywords: EEG, eye tracking, fixation-related potentials, artifact correction, natural viewing

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Reading is a complex cognitive task, unfolding at the same time at visual, attentional, lexicosemantic, and oculomotor levels. Comprehension requires the processing of visual input across a complex series of brief fixation pauses and saccadic eye movements as well as retrieving, updating, and integrating contents of memory. Current research on reading makes heavy use of two methods: recording eye movement (EMs) and event-related brain potentials (ERPs). Traditionally, these research methods have used different experimental protocols: In EM studies, subjects read sentences or paragraphs of text while their fixation position is monitored with an eye tracker. The durations, positions, and sequences of fixations are then used to make inferences about the underlying cognitive processes (Rayner, 1998). Procedures in these studies often resemble everyday reading without unusual task demands. In contrast, in ERP studies of reading, serial visual presentation (SVP) has typically been used to avoid saccade-related measurement artifacts in the electroencephalogram (EEG). In SVP, readers fixate the center of the screen while sentences are presented word by word at a predefined pace. ERPs are then time-locked to stimulus presentations.

In the present study, we demonstrate effects of a critical variable — the predictability of a word from the prior sentence context — in EMs and ERPs that were recorded simultaneously during left-to-right sentence reading. The predictability effect has figured prominently in both the EM- and the ERP-research traditions. Coregistration of EM and EEG may grant new perspectives on the relation between fixation time and single-trial EEG amplitude, as well as on the time course of predictability effects in both measures. Their simultaneous recording also raises several methodological problems to which we propose solutions. We argue that methodological advances in coregistration, as exemplified for reading in the present article, will also apply to other free viewing situations. In the following, we summarize (a) the rationale for focusing on word predictability effects, (b) the potential benefits of simultaneous recordings, (c) previous EEG studies in which some form of EM coregistration has been used, and (d) the methodological challenges that have limited the use of this technique.

Word Predictability in Reading

A word's predictability in the context of a given sentence is known to modulate both oculomotor behavior (e.g., Balota, Pollatsek, & Rayner, 1985; Kliegl, Grabner, Rolfs, & Engbert, 2004; Rayner, Ashby, Pollatsek, & Reichle, 2004) and N400 amplitude (e.g., Dambacher, Kliegl, Hofmann, & Jacobs, 2006; Kutas & Hillyard, 1984). The well-established N400 component describes a negative-going ERP deflection, which is most pronounced around 400 ms after stimulus onset at centroparietal recording sites (Kutas & Hillyard, 1980). N400 amplitude is largest when a word violates the semantic context of a preceding sentence fragment, but is also larger for semantically correct words that are less predictable from

the context. Because of its context sensitivity, N400 amplitude is thought to reflect the difficulty in retrieving conceptual knowledge associated with a word from memory, or in integrating it into the context of the sentence or discourse (Kutas, Van Petten, & Kluender, 2006). However, it remains controversial whether N400 effects reflect facilitated access to lexicosemantic features (Lau, Phillips, & Poeppel, 2008), a late post-lexical process of semantic context integration (Holcomb, 1993; Brown & Hagoort, 1993), or semantic inhibition (Debrulle, 2007) and it is possible that multiple mechanisms contribute to the N400. Regardless of the theoretical viewpoint, the N400 provides information about the time course of semantic processing and its onset can be interpreted as an upper time limit for the initial access to word meaning. One aim of the present study was therefore to test for the existence of an N400 in a normal reading situation and describe its properties.

Predictability also figures prominently in current conceptualizations of reading from the perspective of eye movement control: Highly predictable words are skipped more frequently (e.g., Balota et al., 1985; Vitu, 1991; Drieghe, Rayner, & Pollatsek, 2005); fixations on them are shorter (e.g., Altarriba, Kroll, Sholl, & Rayner, 1996; Balota et al., 1985; Rayner et al., 2004); and high predictability of an upcoming word is associated with a longer fixation on the previous word (Kliegl, Nuthmann, & Engbert, 2006). Understanding the role of predictability in reading is also part of the broader question whether lexical processing is spatially distributed over several adjacent words, and whether lexicosemantic information – in addition to low-level visual and orthographic properties – is extracted from not-yet-fixated words in the parafovea (Kennedy, Pynte, & Ducrot, 2002; Kliegl et al., 2006; Kliegl, 2007; Rayner, Pollatsek, Drieghe, Slattery, & Reichle, 2007).

Potential Benefits of Simultaneous Recordings

From the perspective of ERP research, there is no doubt that SVP has proven itself to be extremely successful in studying the electrophysiological correlates of word recognition (Kutas et al., 2006). At the same time, it presents a strong simplification of the normal reading process, which differs in several ways from SVP: In normal reading, readers determine how long each word is fixated and which word to fixate next. Words are therefore not inspected in a strictly serial fashion, but frequently skipped or fixated several times, and regressive saccades towards earlier words are common. Unlike SVP, normal reading allows for the preprocessing of upcoming words in parafoveal vision. At the same time, words are not always fixated at their center (as in SVP), but are often processed from non-optimal viewing positions near the word boundaries (Nuthmann, Engbert, & Kliegl, 2005). Another major difference concerns speed: While most ERP studies present words at stimulus-onset asynchronies of 400 to 1000 ms (i.e., 60-150 words per minute), average reading fixations last only 200-250 ms and reading speeds of 250

words per minute are common. Accordingly, new visual input is obtained at much higher rates than in most SVP experiments. Finally, SVP imposes secondary-task demands – to maintain fixation and refrain from blinking – absent in normal reading.

As a result of these differences, it is largely unknown to what extent SVP results apply to normal reading. There have been several approaches to improve the ecological validity of SVP. One is to present words at speeds that are very fast (Kutas, 1987), reading-like (Dambacher, Rolfs, Göllner, Kliegl, & Jacobs, 2009), or under the control of the reader by pushing a button (Ditman, Holcomb, & Kuperberg, 2007). Another proposal is to grant a parafoveal preview on the upcoming word during SVP without eye movements (Barber, Donamayor, & Kutas, 2010). Finally, several studies used the same sentences in separate EM and SVP experiments with different participants (Dambacher & Kliegl, 2007; Raney & Rayner, 1993; Sereno, Rayner, & Posner, 1998).

All these techniques preclude direct comparisons between EEG measures and oculomotor behavior. As a consequence, the basic relationship and temporal contingency between the dependent variables in EM research (e.g., fixation duration) and ERP research (e.g., component amplitude) is unresolved (Sereno & Rayner, 2003). On the topic of predictability, one interesting question concerns the apparent discrepancy in the timing of effects in EMs and ERPs (Sereno & Rayner, 2003; Rayner & Clifton, 2009): In normal reading, predictability acts early enough to influence the initial fixation on a word (Rayner, Binder, Ashby, & Pollatsek, 2001; Rayner et al., 2004), which lasts less than 250 ms on average. In contrast, N400 effects from SVP studies only begin to arise at 200-250 ms and reach their maximum at 400 ms or later. This raises the question whether predictability effects in both methods reflect a common underlying process, or not.

A potential alternative is to record EMs and ERPs simultaneously from the same reader. Because little or no useful information is acquired during the saccade (Matin, 1974; Ross, Morrone, Goldberg, & Burr, 2001), fixation onsets provide natural EEG time-locking points to study information processing in normal vision. Averaged potentials aligned to fixation onsets are called fixation-related potentials (FRPs), while those aligned to saccade onsets are called saccade-related potentials (SRPs)¹.

¹ With regard to the family of visually-evoked components that follow saccade on- and offset, SRPs have also been referred to as *lambda waves* in the literature. Likewise FRPs have also been called *eye-fixation-related potentials* (EFRPs). We use SRP and FRP for averaged saccade- and fixation-aligned data, because these abbreviations are short, symmetric, and their meaning is not confined to early visual processing.

Existing Research Integrating EM and EEG

Recording SRPs and FRPs is not a new technique. Large and single saccades, measured via electro-oculogram (EOG) electrodes near the eyes, have frequently been used in basic EEG research on post-saccadic visual processing, oculomotor preparation, and decision making (e.g., Everling, Krappmann, & Flohr, 1996; see *Methodological Discussion* for additional references). In contrast, only a handful of studies on visual word recognition have allowed for saccades. Several early studies have recorded SRPs following a single saccade towards a word presented in the periphery (e.g., Marton, Szirtes, & Breuer, 1985). Two recent studies with eye tracking (Baccino & Manunta, 2005; Simola, Holmqvist, & Lindgren, 2009) have presented pairs of words in order to investigate whether the lexicality of a parafoveal word and its semantic relation to the foveal word influence the ERP while participants still fixate the foveal word. Baccino and Manunta have reported an effect of semantic relatedness before the saccade to the parafoveal word and as early as 110 ms after stimulus onset. Simola and colleagues found a lexicality effect for parafoveal words in the right hemifield, but no evidence for parafoveal semantic access. To avoid saccade-related measuring artifacts, both studies restricted data analysis to a short segment of EEG before the first saccade.

In a study by Hutzler et al. (2007), participants read an array of five unrelated words and had to judge whether the final word had been presented as part of the array or not. The old/new effect – a late positivity for correctly recognized old words – was observed in FRPs, and also during SVP of the same words. As part of several pioneering studies on SRPs (Marton, 1991), Marton and colleagues even allowed their participants to read a full sentence from left to right (Marton & Szirtes, 1988a; 1988b). However, due to various technical constraints, the sentence-final word was displaced 20° to the left or right. After time-locking the SRP to the saccade onset, the authors observed an N400-like effect when the final word violated the sentence context. As in the study of Hutzler et al., the critical saccade was last in the sequence, so it was not possible to compare neural activity with fixation durations.

Finally, several studies have recorded SRPs during largely unconstrained scanning behavior such as reading (Barlow, 1971; Kurtzberg & Vaughan, 1979; Takeda, Sugai, & Yagi, 2001, see also Burdette, Walrath, Gross, & Stern, 1986), REM sleep (Abe, Ogawa, Nittono, & Hori, 2004), or picture viewing (Kurtzberg & Vaughan, 1979). However, without concurrent eye tracking, SRPs could not be related to fixation durations or the fixated item, but were instead aggregated across all saccades or compared globally for different stimuli or saccade types (e.g. reading vs. picture scanning, Kurtzberg & Vaughan, 1979). A study by Graupner and colleagues (Graupner, Velichkovsky, Pannasch, & Marx, 2007) allowed for free EM behavior during picture viewing. Participants scanned a scene, and distracter stimuli were occasionally flashed

near current fixation. Different distracter conditions were then compared in terms of their effect on fixation duration and the visual potential evoked by distracter onset.

Methodological Challenges

To our knowledge, no study has co-registered EM and EEG in an unconstrained viewing situation in order to directly compare oculomotor behavior to brain activity as a function of the properties of the currently fixated item. This is likely due to at least four major methodological problems associated with such recordings: (1) the need to co-register precise gaze position without technical interference, (2) corneoretinal and myogenic eye movement artifacts, (3) varying degrees of overlap between brain responses elicited by successive fixations, and (4) low-level, visuomotor influences on cortical activity before and after fixation onset. In an extended *Methodological Discussion*, we review the relevant background information on each of these problems and propose solutions.

The Present Study

Given the important role of predictability in reading research, we deemed it a suitable proving ground for an attempt to co-register EMs and ERPs in saccadic vision with in- and outgoing saccades. Participants read sentences at their own pace, moving their eyes freely, with no other task than comprehension. We assumed that well-known effects could be recovered and that we may reap benefits from co-registration that go beyond what usually can be inferred from separate recordings. Data analyses are structured as follows: First, we describe basic properties of the artifact-corrected FRP and SRP in multi-saccadic vision. Second, we demonstrate that standard word predictability effects are recovered under co-registration. Third, we establish the basic relationship between EM behavior and N400 in the same set of fixations. We will model this relation at the level of individual fixations and trace the time course of semantic processing relative to fixation onset.

Method

Participants

Thirty students (22 women, 17-37 years, mean age 23.0 years) participated in the 2.5-hour session. All were native speakers of German with a mean of 15 years of education and no history of reading difficulties or neurological/psychiatric disorders. They were paid 25 € or received course credit. All participants had normal or corrected-to-normal visual acuity (Bach, 1996). Twenty-five participants were right-handed, one left-handed, and four ambidextrous (Oldfield, 1971). Data from four additional participants was recorded but not analyzed because of EEG voltage drifts.

Apparatus

Participants were seated in a dimly lit, sound-attenuated room at a distance of 85 cm from a 17" monitor (Samsung SyncMaster 171T TFT, resolution 800 × 600 pixel, 60 Hz

vertical refresh). The screen of the monitor was framed with a light grey cardboard mask that subtended 60° × 75°. The mask served to homogenize the characteristics of the visual field across different on-screen fixation locations and to reduce any resulting influences on the morphology of post-saccadic visually-evoked lambda waves (see *Methodological Discussion*).

Materials

Subjects read the *Potsdam Sentence Corpus* (Kliegl, Grabner, Rolfs, & Engbert, 2004) which contains 144 unrelated German sentences (1138 words) with a large variety of grammatical structures and semantic contents. All sentences are semantically and syntactically legal. Sentence length ranges from 5 to 11 words with a mean of 7.9 words. The corpus has previously been used to study predictability effects on EMs (Kliegl et al., 2006) and stimulus-locked ERPs (Dambacher et al., 2006). Thirty-two samples of these sentences are provided in Kliegl et al. (2004). For the present analyses, we considered only open-class words of the corpus (nouns, adjectives, verbs, adverbs; $n = 813$ words) and excluded words at the beginning (word position one and two) or end (final word) of the sentence. Sentence-initial words were excluded to avoid influences of the trial-initial fixation check on fixation behavior. Sentence-final words were excluded because they tend to be fixated longer (Rayner, Kambe, & Duffy, 2000; Just & Carpenter, 1980) and elicited more positive-going ERPs (Friedman, Simson, Ritter, & Rapin, 1975; Hagoort, 2003) than words at intermediate positions ("sentence wrap-up" effects).

To study predictability effects, we used only the normal range of cloze probabilities in the sentences of the Potsdam Corpus. In order to do so, the remaining 499 words (henceforth called *target words*) were categorized according to cloze probability. The cloze probability of a word in a given sentence context is defined as the probability of correctly guessing it as the upcoming word after knowing all preceding words of the sentence. Cloze probabilities for every word in the corpus were collected in a norming study with $N=282$ German native speakers (for details see Kliegl et al., 2004). Each participant generated predictions for a subset of the sentences, yielding 83 complete protocols for the entire corpus.

While predictability is always defined as cloze probability in the present study, it is important to note that cloze probability is typically correlated to, but not identical with, the amount of contextual constraint imposed by the preceding sentence. For example, both a weakly and a strongly constraining sentence frame can be completed by an equally unpredictable final word (e.g., Federmeier, 2007; Federmeier, Wlotko, De Ochoa-Dewald, & Kutas, 2007). We therefore also computed sentence constraint at the position of the target word, which was operationalized as the number of different predictions generated during the norming study². The theoretical range of this variable was therefore from 1 (perfectly constraining sentence frame; allows only one completion) to 83 (uninformative sentence frame; every rater guesses a different upcoming word). As it is typical for a corpus of normal sentences, cloze probability correlated not only with constraint ($r = -.43$), but also with word length ($r = -.23$), word position ($r = .18$), and CELEX-based word frequency ($r = .33$; Baayen, Piepenbrock, & van Rijn, 1995). To control the influences of these covariates, they were included as predictors in a linear mixed model of N400 amplitude.

² The same results pattern was obtained when we defined contextual constraint not as the number of different words expected in the cloze procedure, but as the cloze probability of the most expected word.

Fixation-Related Potentials in Reading

For most SRP/FRP analyses, we used a three-level categorization of *low predictable* words (cloze $p \leq .01$, $n = 187$ words), *medium predictable* words ($.01 < \text{cloze } p \leq .25$, $n = 235$) and *high predictable* words (cloze $p > .25$, $n = 83$). Mean cloze probabilities for these categories were .00, .07, and .55, respectively (Table 1). To compare effects on EMs and FRPs, we also used a finer categorization into five bins, where unpredictable words (cloze $p = 0$) were assigned to the first bin and the remaining words were assigned to four additional bins each containing the same number of words.

Procedure

The experimental procedure was designed to approximate a natural reading flow, including leftwards return saccades at the end of each trial. At the beginning of the trial, a fixation point appeared on the left side of the center line of the screen (Figure 1A). Five-hundred milliseconds after fixation point onset, the eye tracker started to poll the participants' eye position. Once it registered a stable (> 150 ms) fixation, a full sentence was presented as one line of text on the center line of the monitor, thereby replacing the fixation point. Text was displayed in black on a white background in a monospaced font (Courier 9) at a size of 0.26° per character. The horizontal position of the sentence was set so that the initial fixation was always located slightly left of the center of the first word (the optimal viewing position, O'Regan & Lévy-Schoen, 1987). Subjects then read the sentence at their individual pace, moving the eyes freely over the words. After they finished reading, participants looked for 500 ms at a second small point near the right margin of the screen. This fixation initiated a new trial: Sentence and right fixation point disappeared and were replaced by the left fixation point, the fixation of which triggered the next sentence presentation.

The participants' task was to read the sentences and to answer simple three-alternative multiple-choice questions presented after 20% of the sentences. Questions pertained to the content of the preceding sentence and were answered by a mouse click (mean accuracy: 96%). There was no instruction to suppress eye blinks. Subjects read ten warm-up sentences before the experiment.

EM Recording

EM were recorded from the right eye with a table-mounted IView-X Hi-Speed eye tracker (SensoMotoric Instruments, Germany) at a sampling rate of 240 Hz. Viewing was binocular. The infrared video-based system has an instrument spatial resolution of $< 0.025^\circ$ and an absolute gaze position accuracy of up to 0.2° . Thus, calibrated eye position was recorded accurately at the level of letters. Head movements were minimized by the eye tracker's built-in chin and forehead rests. Proper calibration of the eye tracker was automatically assessed at the onset of every trial: If gaze was not detected within an invisible $0.5^\circ \times 0.5^\circ$ box around the left fixation point within 2 s, the system was recalibrated with a 13-point grid.

EEG Recording

The EEG and EOG were recorded from 32 Ag/AgCl electrodes on the scalp and around the eyes. Twenty-eight electrodes were mounted in an elastic electrode cap (Easycap GmbH, Germany) at positions FP1, FP2, Fz, F3, F4, F7, F8, FC5, FC6, FT9, FT10, Cz, C3, C4, T7, T8, A2, CP5, CP6, Pz, P3, P4, P7, P8, PO9, PO10, O1, and O2 of the International 10/10 system. Four EOG electrodes were affixed to the outer canthi and infraorbital ridges of both eyes. Foam-cushions were fitted to the participants' forehead to preclude pressure artifacts from contact

between frontal electrodes and the eye tracker's forehead rest. Seating position and head position in the eye tracker were carefully adapted to avoid myogenic interspersions from neck and temple muscles. Impedances were kept below 5 k Ω . An additional electrode at FPz served as ground. Signals were amplified with a Brainamp AC amplifier (Brain Products GmbH, Germany) at a band-pass of 0.01 - 70 Hz and digitized at a rate of 250 Hz. All electrodes were initially referenced to left mastoid (A1), but converted to average reference offline, thereby recovering A1 as a recording electrode. Thus, the data of 33 electrodes entered the analyses. For use in artifact correction, 3D electrode locations were determined with a Zebris CMS20 digitizer (Zebris Medical GmbH, Germany). To synchronize EM and EEG records, a common TTL trigger was sent at the beginning and end of each trial from the stimulus presentation PC (running *Presentation* Software, Neurobehavioral Systems Inc, Albany, CA) and looped through to two additional PCs recording EMs and EEG.

EM Analysis

The EM record was screened for loss of measurement and eye blinks. If a sentence contained only a single blink very early (< 200 ms) or late (> 2 s) after sentence onset (12.8 % of trials), the remaining data was used for fixation detection. Otherwise, or if multiple blinks occurred, the trial was discarded (5.1 % of trials). Saccades were detected as outliers in two-dimensional-velocity space with the monocular variant of the algorithm detailed in Engbert and Mergenthaler (2006). Saccade detection led to a total pool of 38,538 reading fixations. In a first level of screening, we discarded 2,775 fixations that occurred during intervals in which the EEG contained nonocular artifacts. In a second step, the pool was constrained to 22,321 fixations that occurred more than 700 ms after sentence onset. Earlier fixations were excluded to avoid temporal overlap between FRPs and the ERP evoked by the screen onset of the sentence. In line with previous experiments with the Potsdam Corpus (Kliegl et al., 2006), we eliminated extremely short (< 50 ms, $n = 1,157$) or long (> 750 ms, $n = 64$) fixations. In a final step, the pool was restricted to *first* fixations on *target words* in *first-pass reading*: 12,607 of the remaining fixations were on targets, 9,237 fixations of these were first fixations rather than refixations, and 7,113 occurred in first-pass reading. All EM and EEG analyses were based on this final pool of 7,113 fixations. Because fixations with a bad concurrent EEG record were removed, EM and FRP analyses were always conducted on the exact same set of fixations.

Dependent variables for behavioral analyses were first-fixation duration (FFD) and gaze duration (GD). Gaze duration is defined as FFD plus the duration of all immediate refixations. FFD and GD were submitted to repeated-measures analyses of variance (ANOVAs) on the factor predictability. Results are reported with p values corrected for violations of sphericity according to Huynh & Feldt (1976), the original degrees of freedom, and the epsilon (ϵ) value.

EEG Ocular Artifact Correction

To correct for corneoretinal eye movement artifacts (see *Methodological Discussion*), we applied Surrogate Multiple Source Eye Correction (MSEC; Berg & Scherg, 1994; Ille, Berg, & Scherg, 2002) as implemented in BESA (v. 5.1; MEGIS Software GmbH, Germany). The method combines the recording of calibration eye movements, Principal Component Analysis (PCA), and dipole modeling to separate artifact and brain activity. In surrogate MSEC, characteristic scalp topographies for

different types of ocular artifacts are derived for each participant by averaging calibration eye movements. In addition to these empirically derived artifact topographies, a set of brain signal topographies is defined by a generic dipole model of the brain, which is identical for all participants. Importantly, this “surrogate” brain model is not used to directly model the artifact-free EEG, but its purpose is to preclude the subtraction of genuine brain activity that is spatially correlated to the artifact. Based on these spatial definitions for artifact and brain activity, a linear inverse operator is computed that decomposes the experimental data into linear combinations of brain and artifact activity, that is, the activation time courses for the artifact topographies are determined in the presence of the brain model. In a final step, this estimated artifact activity is subtracted from the raw EEG.

Technical details were as follows: In a 15-minute session before the experiment, participants performed 120 calibration saccades (15° amplitude) in the four cardinal directions. Saccades were aimed at targets on the mask surrounding the monitor. Saccade direction was indicated by an arrow, which appeared in the screen center every three seconds. In addition, 40 spontaneous eye blinks were recorded during fixation. Short EEG segments following each of the three movement types (vertical, horizontal, blink) were then averaged and subjected to three separate PCAs. The first PCA factor (typically explaining > 97% variance) was used to define the topography for each type of artifact. Note that PCA was used here as an optional preprocessing step (see Berg & Scherg, 1994) to extract the most characteristic artifact topographies from calibration data; PCA was not applied to the experimental data. Brain signal topographies were defined by BESA model *RS4.par*. This model contains 12 dipoles with fixed location and orientation, placed at spatially distributed, strategic positions of the brain. After defining artifact and brain topographies, the activity time course for each topography was determined in the experimental data using the spatial filter operator detailed in Ille et al. (2002, p. 123). For correction, the activity assigned to the artifact topographies was subtracted. After MSEC, the corrected continuous data were high pass-filtered at 0.25 Hz (48 dB/octave) and EOG channels were treated as regular EEG channels. Application of surrogate MSEC is detailed in Scherg (2003). Recommendations for recording clean calibration movements are given by Ruchkin (in Berg, 2002, p. 7-2). A comparison with other correction methods is provided by Ille et al. (2002).

Fixation-locked EEG

Around each fixation, a 1600 ms segment of EEG was cut (from 600 ms before to 1000 ms after fixation onset) and baseline-corrected by subtracting the mean voltage in the 100 ms interval prior to fixation onset. To reject muscle or drift artifacts, we discarded segments with absolute voltages in any channel > 100 μ V or with a peak-to-peak voltage difference > 150 μ V. Joint EM and EEG analyses were performed in MATLAB (The Mathworks Inc., USA) using selected functions of the EEGLAB toolbox (Delorme & Makeig, 2004).

Evaluation of Ocular Correction

The synchronized eye tracking data served as a new criterion to assess ocular correction quality. For this purpose, a corresponding set of fixation-locked segments was also cut from the original, uncorrected EEG. For each corrected and uncorrected segment, the correlation between each EEG channel (downsampled to 240 Hz) and the horizontal component of the eye track was computed in an interval from -100 to 1000 ms

around fixation onset. Correlation coefficients for individual segments were Fisher's z transformed, averaged within each participant and then across participants. Correlations before and after ocular correction were tested against zero for each channel with a paired t -test.

Analysis of Predictability Effects

Artifact-corrected segments were averaged according to the predictability of the fixated word. To test for the presence of an N400 effect, mean amplitude in the traditional N400 window (300-500 ms after fixation onset) was submitted to a repeated-measures ANOVA on the factors *predictability* and *electrode*. To estimate a discrete time point for the onset and peak of the N400 effect, we used the difference wave between the two extreme predictability conditions (low minus high predictable) at electrode Pz, low-pass filtered at 10 Hz (zero-phase). N400 onset was determined with consecutive, sample-by-sample t -tests on this difference wave between -300 and 600 ms around fixation onset. The t -max permutation test of Blair and Karniski (1993) was used to control for multiple testing.³

N400 peak latency was defined as the time of the maximum absolute voltage in the grand average difference wave between 0 - 800 ms. To test for a lateralization of N400 effects, effect amplitude (low minus high predictable words, 300-500 ms) was aggregated over all 15 left- and all 15 right-hemisphere electrodes and compared with a t -test, leaving out the three midline sites.

Analysis of EM-EEG Relationship

To explore the relative timing of EM and EEG measures, we analyzed on which word participants were fixating at the onset and peak of the N400 effect. For the same purpose, we also computed an additional average, aligned to the saccade that terminated the first fixation on the target. In analogy to response-locked averages in traditional ERPs, this SRP reveals whether or not, on average, N400 effects arise prior to the initiation of the next saccade, that is, during the initial fixation on the word. For this analysis, the baseline remained identical, that is, SRP segments were baseline-corrected with the baseline still placed before the onset of the preceding target fixation. An analogous t -max statistic was computed also for this average.

To test for a between-subject linear relation between predictability effects in EM and N400 amplitude, the size of the predictability effect on behavior (FFD and GD) was correlated with that on the FRP *across* participants. For this analysis, target words were categorized as low or high predictable via a split at the median cloze probability of 0.024.

Of special concern was the relation between fixation duration and N400 amplitude at the level of individual fixations. We specified linear mixed models, with the N400 amplitude following each individual fixation as dependent variable, and word and sentence characteristics (predictability, frequency,

³ During 10,000 data permutations, the sign of each single-subject difference wave was randomly assigned, t -values were again computed for every sampling point, and the t -value with the maximum absolute value was stored. This resulted in a distribution of 10,000 maximum t -values expected under the null hypothesis, i.e., with randomly shuffled conditions. N400 onset was defined as the first sample of the recorded waveform where the t -value was below (more negative than) the 5th percentile of the t -max distribution ($t = -3.33$ for the FRP). This tested the directed hypothesis of more negative voltages for low predictable words.

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length, constraint, word position) – as well as either the log of FFD or the log of GD as linear covariates (fixed effects). Predictability values were logit-transformed ($\text{logit } cloze\ p = 0.5 * \ln(cloze\ p / (1 - cloze\ p))$; see Kliegl et al., 2006) before they entered the model (and also for Figure 5A). Participants and words were included as crossed random factors. For parameter estimation we used the *lmer* program of the *lme4* package (Bates & Maechler, 2009) in the R system for statistical computing (R

Development Core Team, 2009). These regression analyses model the variance of the N400 differences between participants and between words on the assumption that they are normally distributed.

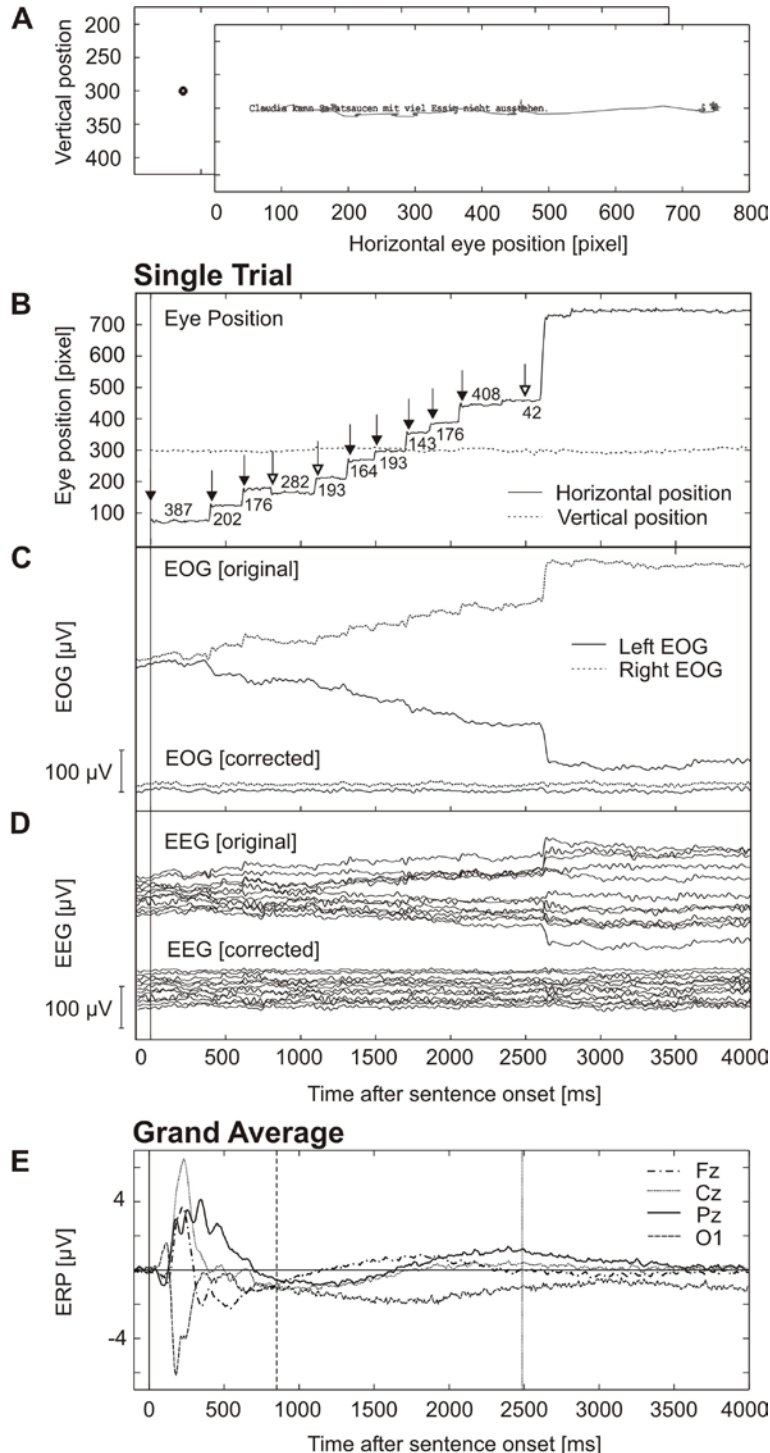


Figure 1.

A. Trial scheme and data for a typical sentence. Each trial began with a fixation point on the left. Once the eye tracker detected a precise fixation, a single sentence was displayed as one line of text. Participants read the sentence at their individual pace, moving the eyes freely over the text. Eye movements are plotted for one subject reading the sentence “*Claudia kann Salatsaucen mit viel Essig nicht ausstehen.*” (Claudia cannot stand salad dressings with lots of vinegar.). After reading the sentence, participants looked at a point on the right. Gaze-controlled presentation ensured a continuous reading flow including leftwards return saccades to read a new sentence.

B. Horizontal and vertical gaze position. The sentence appeared at time 0. Solid arrows indicate the onsets of first fixations, open arrows mark refixations. Fixation durations are given in milliseconds.

C. Signal at left and right horizontal EOG electrode before and after corneoretinal artifact correction.

D. Synchronized EEG record for a subset of channels before and after correction.

E. Grand average artifact-corrected ERP, time-locked to sentence onset. The dotted line indicates mean sentence-reading duration. To avoid overlap between potentials evoked by sentence onset and those evoked by individual reading fixations, only fixations were considered that occurred >700 ms after sentence onset (dashed line).

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Table 1

Target word properties and resulting effects on EM behavior and FRP amplitude

	Predictability				<i>F</i>	<i>p</i>
	All targets	Low	Medium	High		
A. Word and sentence properties						
Cloze probability	0.12 (0.21)	0.01 (0.00)	0.07 (0.07)	0.54 (0.21)	977.4	.000
Word length (char.)	5.8 (2.7)	6.6 (2.7)	5.4 (2.5)	4.9 (2.3)	17.2	.000
CELEX frequency (log, per million)	4.7 (1.3)	4.0 (1.2)	5.1 (1.2)	5.3 (1.2)	50.8	.000
Word position in sentence	5.0 (1.7)	4.6 (1.6)	5.1 (1.7)	5.8 (1.6)	14.8	.000
Sent. constraint (<i>N</i> guessed words)	25.1 (11.0)	27.8 (10.8)	26.3 (10.7)	15.4 (6.3)	45.6	.000
Sentence length (words)	8.2 (1.4)	8.2 (1.5)	8.2 (1.3)	8.4 (1.3)	0.6	<i>n.s.</i>
B. EM behavior						
FFD (ms)	224 (25)	235 (28)	219 (25)	213 (21)	39.2	.000
GD (ms)	278 (41)	304 (54)	268 (38)	247 (34)	57.5	.000
Refixation probability	0.24 (0.07)	0.28 (0.10)	0.22 (0.08)	0.18 (0.09)	24.9	.000
Duration previous fixation <i>n</i> -1 (ms)	213 (25)	214 (26)	210 (24)	217 (29)	3.5	.046
Duration next fixation <i>n</i> +1 (ms)	212 (32)	216 (29)	210 (34)	207 (38)	4.0	.025
Incoming saccade amplitude (°)	2.0 (0.5)	2.0 (0.5)	1.9 (0.5)	2.0 (0.5)	3.5	.037
Outgoing saccade amplitude (°)	1.4 (0.6)	1.4 (0.6)	1.4 (0.6)	1.5 (0.7)	1.2	<i>n.s.</i>
Fixated after sentence onset (ms)	1301 (198)	1244 (177)	1300 (208)	1431 (253)	63.9	.000
Sentence reading duration (ms)	2490 (713)	2566 (737)	2465 (704)	2400 (687)	29.1	.000
C. FRP						
Amplitude at Pz, 300-500 ms (μV)	-0.77 (0.62)	-1.27 (0.90)	-0.69 (0.74)	0.03 (1.31)	15.8	.000

Note. Given are means and standard deviations. Statistics are based on words in A. and on fixations in B. & C.

Results

Results are organized in six sections. First, we present standard EM effects that establish the ecological validity of the data. Second, we report measures for the quality of ocular artifact correction. Passing these checks was a precondition for the validity of FRP results. Third, we describe the FRP over the course of reading an entire sentence. In particular, sentence onset evoked a potential that spilled over to different degrees in the FRPs, forcing the exclusion of some fixations from the following analyses. Fourth, we introduce the artifact-corrected FRP as an EEG measure. Since FRPs have not been described in detail for natural viewing situations, we describe how pre-saccadic motor potentials, post-saccadic visual potentials, and overlapping potentials influence the results. Fifth, we present word predictability effects on FRPs. Finally, we compare EM and EEG effects and map them onto a common timeline.

Eye Movements

Eye movements for a typical trial are shown in Figure 1. On average, participants read the sentence for 2490 ms (*SE* = 130 ms) before they initiated the final saccade towards the right fixation point. Mean FFD on target words was 224 ms (*SE* = 5 ms). Targets received at least one refixation in 24% of the cases, resulting in a mean gaze duration of 278 ms (*SE* = 8 ms).

Word predictability clearly influenced EM behavior⁴. On average, low predictable words were fixated 22 ms

⁴ To ensure that predictability accounted for variance in EM measures under concurrent control of other word and sentence properties, we specified two control models with either FFD or GD as dependent variable, and the six variables from Model 2 (*pred, freq, pred x freq, length, constraint, and word position*, see Table 3) as predictors. Predictability was a significant predictor of both FFD (*t*-

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Table 2

Correlation between EEG & horiz. eye track

Hemisphere	Electr.	Original	Corrected
Midline	Fz	.07	.05
	Cz	-.02	.02
	Pz	.01	.03
Left	hEOG	-.97	-.04
	T7	-.63	-.05
	C3	-.32	.00
	P3	-.17	.00
	O1	-.06	.01
Right	hEOG	.97	-.02
	T8	.57	.00
	C4	.24	.01
	P4	.14	.03
	O2	.07	.04

Note. Shown are mean correlations for selected electrodes before and after MSEC. hEOG = horizontal EOG electrode. Correlations that differ significantly from zero are printed in bold.

longer than high predictable words upon first fixation (low: 235 ms, medium: 219 ms, high: 213 ms, $F(2,58) = 39.2$, $p < .001$, $\epsilon = .90$, $\eta_p^2 = .58$). Gaze duration was 57 ms longer for low predictable than high predictable words (low: 304, medium: 268, high: 247; $F(2,58) = 57.5$, $p < .001$, $\epsilon = .87$, $\eta_p^2 = .67$). Due to these effects, total sentence reading duration also differed between predictability levels, $F(2,58) = 29.1$, $p < .001$, $\epsilon = .96$, $\eta_p^2 = .50$ (Table 1).

Of special methodological importance were differences in the amplitude of the incoming and outgoing saccade. This is because saccade amplitude per se influences the morphology of saccade-related visuomotor brain potentials, independent of the distortion by eye movement artifacts (see *Methodological Discussion*). Importantly, outgoing saccade amplitude did not differ as a function of word predictability. Incoming saccade amplitude was slightly smaller for fixations on medium-predictable words (low: 2.0°, medium: 1.9°, high: 2.0°; $F(2,58) = 3.5$, $p < .05$, $\epsilon = .95$, $\eta^2 = .27$, see Table 1) but we will show later

value = -4.5) and GD ($t = -6.0$). Other significant predictors of FFD were *constraint* (-3.7), and the *pred x freq* (2.8) interaction. Other significant predictors for GD were *word length* (13.6), *word position* (3.2), and *pred x freq* (4.7).

on that this difference of less than 0.1° is unlikely to have a relevant impact on the FRP.

Quality of Artifact Correction

EEG correction quality was assessed with three criteria: (1) Visual impression of the continuous EEG, (2) voltage differences between left- and right-hemisphere electrodes in the averaged FRP, and (3) residual correlations between gaze position and EEG after MSEC correction. Figure 1 provides an example of the continuous EOG (Fig. 1C) and EEG (Fig. 1D) before and after correction. After correction, it was generally not possible to visually identify residual artifacts in the continuous data. Figure 2A shows the FRP, superimposed for all channels, before and after correction. Before correction, large distortions from the predominantly rightward-going saccades were evident, with positive distortions at right-hemisphere electrodes and negative distortions at left-hemisphere electrodes. As the incoming saccade was usually followed by more right-going saccades, artifacts from multiple saccades summed up towards the end of the segment. At +1000 ms, temporal electrodes T7 and T8 differed in voltage by about 100 μ V.

After correction, signals at all channels were in the typical ERP amplitude range (Figure 2A). Although artifacts were drastically reduced, some fronto-lateral channels still showed indications of reversed polarities on opposite sides of the head towards the very end of the segment. This suggests that correction across several saccades was not perfect for these electrodes. As a new quantitative measure, we computed linear correlations between each EEG channel and the horizontal component of the eye track. Before correction, 31 of 33 channels correlated significantly ($|t(29)| > 2.05$, $p < .05$) with gaze position, with maximum correlations at electrodes near the eyes (see Table 2). The horizontal EOG electrodes each showed a near-perfect correlation with gaze position ($r = \pm .97$), which increased to $r = .99$ in a bipolar EOG montage (right minus left). After MSEC, correlations at all channels were close to zero (max. $|r| = .07$; max. $R^2 = .005$), suggesting that the residual variance accounted for by horizontal saccades was small. Nevertheless, correlations remained significantly different from zero for about half (18 of 33) of the channels. Electrodes on the posterior sagittal midline were least affected by horizontal saccades, but Cz and Pz were the only two electrodes not significantly correlated with gaze, even in the uncorrected data. Because Pz contained little or no corneoretinal artifact in the first place, effects of correction were minimal at this electrode.

While MSEC removed most of the corneoretinal artifact, it only partially removed the brief muscle spike potential (Keren, Yuval-Greenberg, & Deouell, 2010) at saccade onset. Nevertheless, MSEC attenuated the spike potential because its topography overlaps with that of corneoretinal artifacts (see also *Methodological Discussion*).

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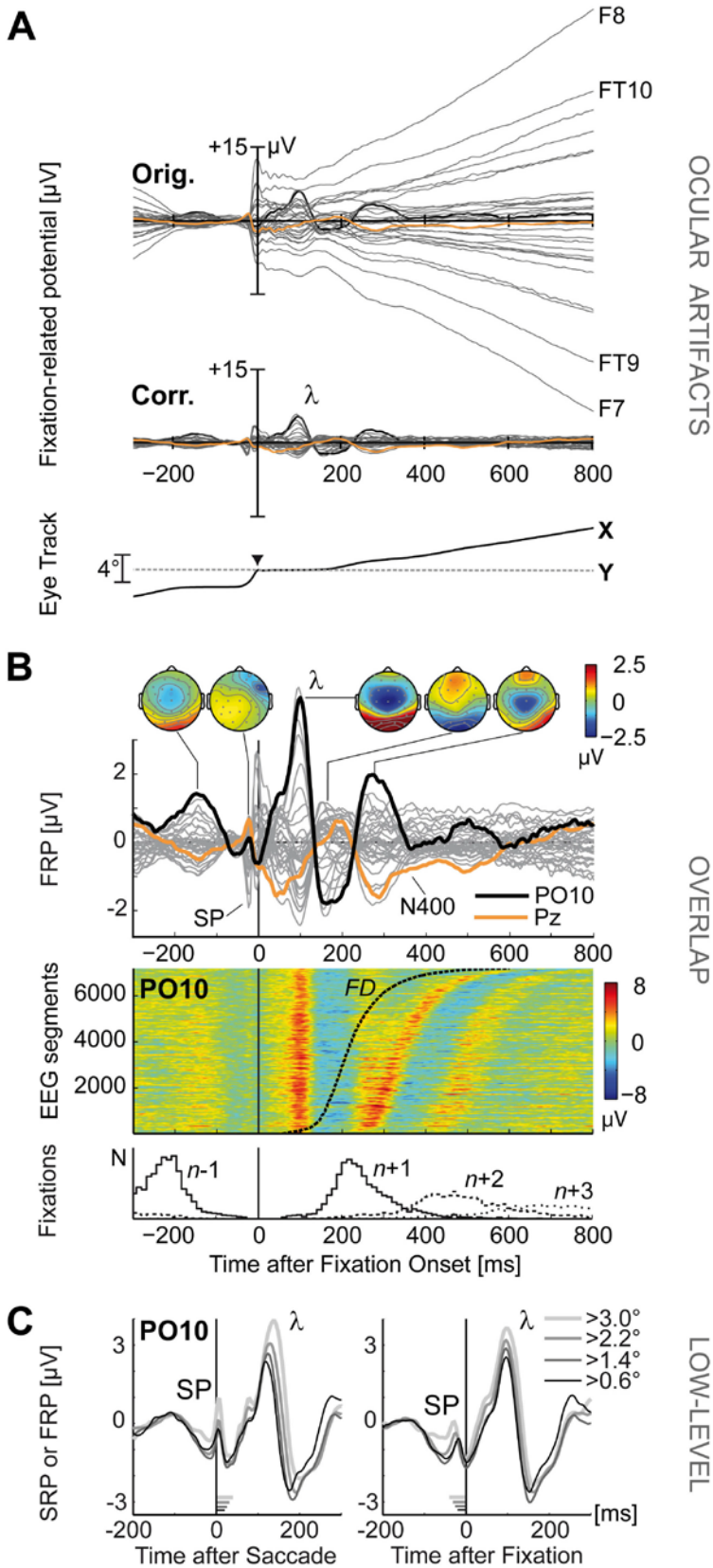


Figure 2. The fixation-related potential in multi-saccadic vision.

A. Grand average FRP for 7,113 target fixations before and after corneoretinal artifact correction. Time 0 marks fixation onset. Signals at all EEG electrodes are shown superimposed. Prior to MSEC (Orig.), frontal and temporal channels were strongly distorted. After MSEC (Corr.), artifacts are largely eliminated and genuine brain activity becomes visible. Midline electrode Pz (orange line) was unaffected by corneoretinal artifacts, even before correction.

B. Top. Properties of the grand average FRP. Maps depict scalp topographies at five component peaks: (1) the visually-evoked lambda response evoked by previous fixation $n-1$, (2) the myogenic spike potential (SP) at saccade onset, (3) the lambda response evoked by current fixation n , which peaked after 104 ms at parieto-occipital electrodes PO9 and PO10, (4) the equivalent of the N170 component, and finally (5) the N400 component, overlapped by the lambda response from fixation $n+1$. *Middle.* Each horizontal line in the *ERPimage* represents one of the 7,113 EEG segments that entered the average shown above. Amplitude is coded as color. Segments were sorted by first fixation duration and then smoothed vertically with a moving average across 50 adjacent segments. Sorting shows how the second positive peak at around 280 ms is partially explained by the lambda response from fixation $n+1$. Because contributions from successive fixations are increasingly time-jittered, the overall waveshape of the FRP resembles a damped oscillation. *Bottom.* Histogram of onset latencies of preceding and subsequent fixations.

C. Effect of saccade size on the morphology of SRPs and FRPs. The amplitude of the spike potential and the lambda response increased with saccade size.

Sentence-onset ERP

Figure 1E shows the artifact-corrected ERP time-locked to the screen onset of the sentence. Sentence onset elicited a large ERP, whose dominant component was a temporally extended P300 with a characteristic centroparietal-positive scalp distribution. To avoid a carry-over of this stimulus-evoked ERP into the fixation-locked segments, only fixations were analyzed that occurred > 700 ms after sentence onset, when this ERP had returned to baseline. Still, on average, the remaining low predictable words occurred at earlier sentence positions and were fixated sooner after sentence onset than high predictable words (see Table 1). To ensure that the EEG background activity was not different at the time the target was fixated, we calculated the mean amplitude in the 100 ms pre-fixation baseline relative to a 100 ms pre-stimulus baseline before sentence onset. An analysis of variance of these “absolute” pre-fixation baseline amplitudes as dependent variable, and predictability level and electrode as factors, yielded a non-significant result, $F(64,1856) < 1$. This suggests that FRPs occurred against a similar baseline in all predictability conditions.

Figure 1E shows that between 700 ms and the mean reading duration of 2490 ms, the sentence-locked ERP was characterized by a slow positive shift at frontal and central electrodes and a relative negativity at occipital sites. This sentence-level ERP during normal reading has not been described before and may be theoretically interesting as a phenomenon on its own. However, the present data does not allow us to distinguish whether these late ERP fluctuations reflect sentence-level processing demands (e.g., working memory load) or merely the superposition of many individual FRPs over the course of reading.

Fixation-Related Potentials in Natural Vision

Figure 2 summarizes the features of the FRP after artifact correction. The post-fixation waveshape was dominated by the visually-evoked lambda response (Kazai & Yagi, 2003), which peaked after 104 ms ($SE = 1.3$ ms). It was largest at lateral-occipital electrode PO10 over right visual cortex (amplitude: $M = 4.4 \mu V$, $SE = 0.3 \mu V$), but also influenced the waveshape at frontal electrodes with a reversed polarity (cf. also Figure 3A). The lambda response is considered primarily a visual response (Riemslog, van der Heijde, & van Dongen, 1987; Thickbroom, Knezevic, Carroll, & Mastaglia, 1991) which is most likely generated in striate or early extrastriate cortex (Kazai & Yagi, 2003; Dimigen, Valsecchi, Sommer, & Kliegl, 2009). Because the average interval between any two reading fixations was only 233 ms, the waveshape of the FRP was characterized by overlapping lambda responses from preceding and subsequent fixations. As shown in Figure 2B, a second occipital peak after 280 ms reflected the summation of a potential evoked by the current fixation n and the lambda response elicited by fixation $n+1$. Due to normal variance in fixation duration, overlapping contributions from adjacent fixations are time-jittered. The FRP in natural vision

therefore resembles a damped oscillation with an increasingly jittered occipital peak about every 250 ms.

The pre-fixation waveshape was influenced by correlates of oculomotor preparation and execution, in particular the pre-saccadic spike potential. The spike potential is a sharp, biphasic spike at saccade onset that is believed to reflect summated electric activity of the oculomotor nerves or muscles. It is best seen in SRPs (cf. Figures 2C and 4A), and smeared in FRPs due to variance in saccade duration. The spike potential showed the typical topography, which is reversed relative to the corneoretinal artifact: a frontal negativity, shifted ipsilateral to saccade direction, and a parietal positivity, shifted contralateral to saccade direction. For large reading saccades ($> 3^\circ$), there was also some indication of an earlier posterior positivity that culminated into the spike potential (Figure 2C, right panel). This may be the pre-saccadic positivity (also called antecedent potential) a slow, ramp-like potential found prior to voluntary saccades and believed to reflect saccade preparation in cortical structures (Everling et al., 1996; Richards, 2003).

Figure 2C compares FRPs and SRPs for different saccade sizes. Both the spike potential and the visual lambda response increased with increasing saccade size (see also *Methodological Discussion*).

Predictability Effects in FRPs

Figure 3A shows that predictability clearly modulated the FRP evoked after the initial fixation on the word: Words with little contextual support elicited more negative voltages at centroparietal scalp sites. The presence of an N400 effect was confirmed by a *Predictability* \times *Electrode* interaction, $F(64, 1856) = 4.84$, $p < .001$, $\epsilon = .13$, $\eta_p^2 = .14$, in the 300-500 ms time window (effects across the whole scalp are only meaningful in interaction with electrode, because the average reference sets the mean of all electrodes to zero). Pairwise comparisons between predictability levels showed that the interaction with electrode was significant for the contrast *low* vs. *high*, $F(32,928)=4.82$, $p < .001$, marginally significant for *low* vs. *medium* ($p = .07$), and not significant ($p = .19$) for *medium* vs. *high* predictable words. All three comparisons were significant (at $p < 0.01$) when the main effect of predictability was tested only at electrode Pz.

Importantly, the centroparietal distribution of the N400 with a maximum over Pz (Figure 3C) resembled that observed in many SVP experiments (Kutas, Van Petten, & Kluender, 2006). However, while effects in SVP studies are often shifted slightly towards the right hemisphere, no evidence for lateralization was found in normal reading; effect amplitude did not differ between the left and the right hemisphere ($p = .49$).

For comparison with fixation durations, a discrete time point for the onset and peak of the N400 effect was determined in the difference wave at Pz (see Figure 4B). At Pz, a sustained N400 effect began 248 ms after fixation onset and peaked 384 ms after fixation onset with an effect amplitude of $1.53 \mu V$. Interestingly, much weaker N400-

like central negativities, qualitatively resembling the topography between 300-500 ms, could also be seen in earlier intervals (in particular between about 120-160 ms, see Fig. 3C), but did not survive correction for multiple comparisons. Nevertheless, they indicate that the semantic processing underlying the N400 may begin earlier - possibly only on a subset of fixations - than suggested by our strict onset criterion. No predictability effects were observed prior to the onset of the first fixation (i.e., there was no parafovea-on-fovea effect). This was also the case when the baseline interval was moved further away from fixation onset.

Finally, N400 onset was also determined relative to the *offset* of the first fixation by time-locking backward to the following saccade (SRP). In this analysis, a sustained N400 effect began 20 ms after the end of the first fixation. Additionally, an only temporary significant effect was observed in an early interval from -228 to -192 ms before fixation offset.

EM-EEG relationship

A unique feature of the dataset was the possibility of comparing FRP effects to corresponding modulations in EM behavior. Four analyses explored this relationship: First, we investigated at what time FRP effects arose relative to those on behavior. Figure 4 shows the predictability effect at Pz, relative to the beginning and end of the first fixation. When the N400 effect peaked in FRPs (384 ms), readers had already terminated the initial fixation on the target word (fixation n) in 96% of the cases. Instead, as Figure 4D shows, readers were typically already engaged in fixation $n+1$ (75%) or $n+2$ (19%; saccade intervals were assigned to the following fixation in this analysis). On the level of words, we found that in only 25% of the cases, readers were still looking at the target (word w) at the latency of the N400 peak. These were mostly cases where the word was refixated. Instead, readers had typically moved on to the next words $w+1$ (45%) or $w+2$ (20%). A somewhat different picture emerged when N400 onset latency was considered: In 30% of the cases, the statistical onset latency of the N400 effect (248 ms) fell into the first fixation on the word. In many more cases (67%), it fell only into the following fixation $n+1$. However, because fixation $n+1$ was sometimes a refixation (in 24% of the cases), readers were still looking at the target word in about half (47%) of the cases at the statistical N400 onset latency.

Second, we compared EM and FRP effects over five levels of predictability. As Figure 5A shows, both N400 amplitude and GD (as well as refixation probability, which is one aspect of GD) were monotonic functions of predictability. Only for FFD was there a discontinuity in the higher predictability range. While N400 amplitude was an approximately linear function of logit-scaled predictability, Figure 5A indicates that behavioral measures, in particular FFD, differentiated better in the low than in the high predictability range.

Third, we investigated whether participants with large predictability effects in FRPs also exhibit large behavioral effects. Figure 5b shows that of 30 participants, 25 showed a predictability effect in the expected direction in both measures, supporting the reliability of the co-registration data. However, when these difference scores were correlated across *participants*, N400 amplitude correlated neither with FFD ($r = -.07, p = .70$) nor GD ($r = .15, p = .42$).

Fourth, in separate linear mixed models, we regressed FRP amplitude after each individual fixation in the N400 time window on the two EM measures, FFD and GD, respectively. Gaze duration was found to be a strong and significant (i.e., $|t| > 2$) predictor of N400 amplitude ($b = -0.62, SE = 0.17, t = -3.71$) whereas FFD showed only a numerical trend in the expected direction ($b = -0.37, SE = 0.21, t = -1.73$; see Model 1 in Table 3). In a second step, we included lexical and sentential properties into the model. Both N400 amplitude and the EM measures are known to relate to the (logit of) word predictability and the (logarithm of) word frequency of the fixated words, as well as to the interaction between both variables (Dambacher et al., 2006). Predictability, frequency, and their multiplicative interaction were therefore included as predictors in the model. Additionally, we included the covariates word length, contextual constraint, and word position.

Results are shown in Table 3 (Model 2). Of the newly included predictors, predictability, frequency, word position, and the predictability \times frequency interaction were highly significant, while constraint and length had no effect on N400. Predictability was therefore a significant predictor of N400 amplitude under statistical control of other variables correlated with predictability in the sentence material. Importantly, gaze duration remained a significant predictor in Model 2. Thus, there is shared variance between GD and N400 amplitude that is not covered by the word or sentence properties included in our model.⁵

⁵ The same inferences resulted from likelihood-ratio tests of these models. Adding sentence- and word-properties as predictors significantly improved the model fit both with FFD, Chi-square(6) = 58.0, $p < .0001$, and with GD as first predictor in the model, Chi-square(6) = 101.1, $p < .0001$. Conversely, dropping FFD from the model did not significantly decrease the fit, Chi-square(1) = 2.1, $p = .143$, whereas dropping GD did decrease Chi-square(1) = 24.6, $p < 0.0001$.

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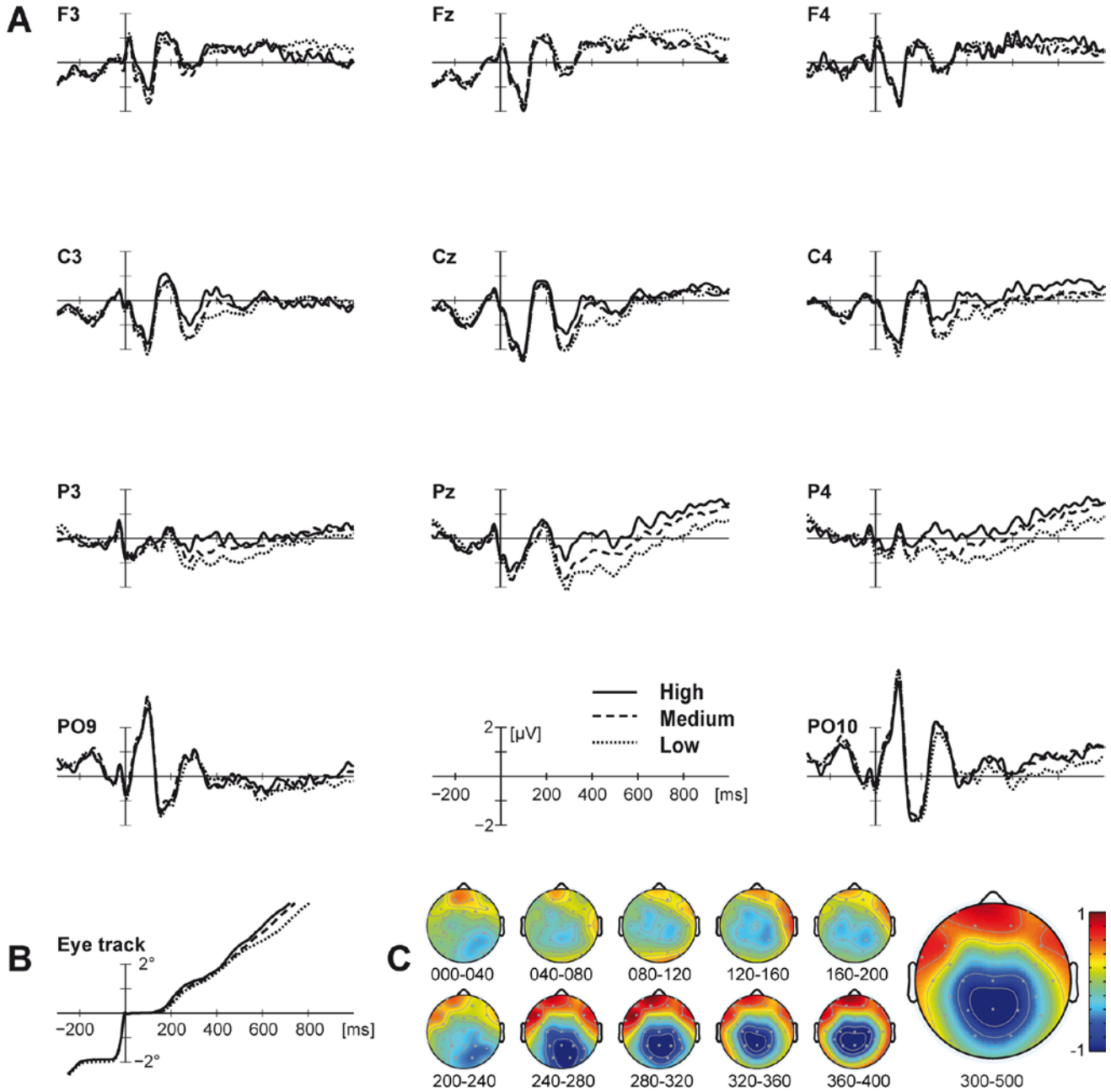


Figure 3. Predictability effect in FRPs. **A.** The grand average FRP, time-locked to first fixation on the target word (time 0) shows a graded effect of word predictability that is largest at centroparietal electrode Pz. **B.** Mean horizontal component of the eye track. **C.** Scalp distributions of the predictability effect (low minus high) are shown for successive 40 ms windows after fixation onset and for the traditional N400 window (300-500 ms).

Fixation-Related Potentials in Reading

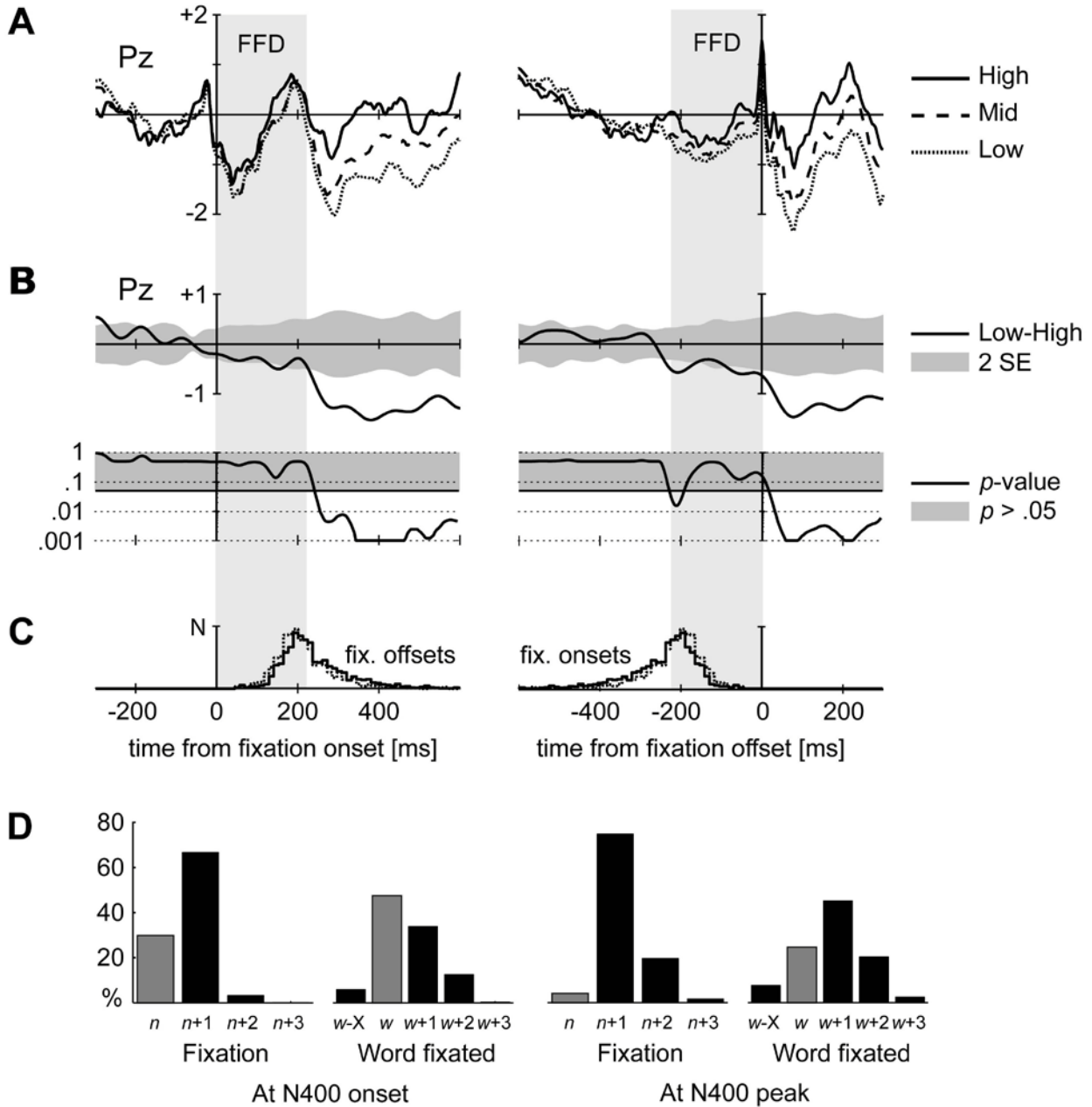


Figure 4. Relative timing of predictability effects. **A.** Grand mean FRP at Pz aligned to the onset (FRP, left side) and offset (SRP, right side) of the first fixation on the word. Grey boxes indicate mean FFD. **B.** *Upper panel:* Difference wave between low and high predictable words, which shows the effect of word predictability devoid of common overlapping activities. Shading indicates the 95% confidence interval without correction for multiple testing. *Lower panel:* corresponding *p*-values from the permutation test. Significant effects ($p < .05$) are indicated by points outside the grey shaded area. In the FRP, an effect of word predictability was observed starting 248 ms after fixation onset, which peaked at 384 ms in the grand average. **C.** Distribution of fixation offset and onset latencies relative to the time-locking point. **D.** Gaze position at onset (248 ms) and peak (384 ms) of the N400 predictability effect. *w-X:* The reader regressed to an earlier word in the sentence.

Fixation-Related Potentials in Reading

Table 3

Regressions of N400 amplitude on eye movement behavior and word/sentence properties

	FFD as predictor			GD as predictor				
		<i>b</i>	<i>SE</i>	<i>t</i>		<i>b</i>	<i>SE</i>	<i>t</i>
Model 1	Intercept	1.12	1.15	0.98	Intercept	2.63	0.94	2.81
	log(FFD)	-0.37	0.21	-1.73	log(GD)	-0.62	0.17	-3.71
Model 2	Intercept	3.03	1.30	2.34	Intercept	4.95	1.20	4.11
	log(FFD)	-0.31	0.21	-1.47	log(GD)	-0.67	0.18	-3.71
	pred	1.00	0.30	3.34	pred	0.91	0.33	2.73
	freq	-0.35	0.13	-2.77	freq	-0.34	0.14	-2.42
	length	0.02	0.03	0.69	length	0.05	0.04	1.38
	constraint	-0.01	0.01	-1.63	constraint	-0.01	0.01	-1.59
	pos	0.29	0.05	6.09	pos	0.30	0.05	5.72
pred×freq	-0.15	0.06	-2.48	pred×freq	-0.13	0.07	-2.05	

Note. Dependent variable is always N400 amplitude (mean amplitude 300-500 ms at Pz). Predictors: first fixation duration (FFD), gaze duration (GD), logit of predictability (pred), log of frequency (freq), word length (length), number of words predicted (constraint), word position (pos), interaction of pred and freq (pred×freq). *N* of observations: 7,113; *N* of subjects: 30, *N* of unique words: 499.

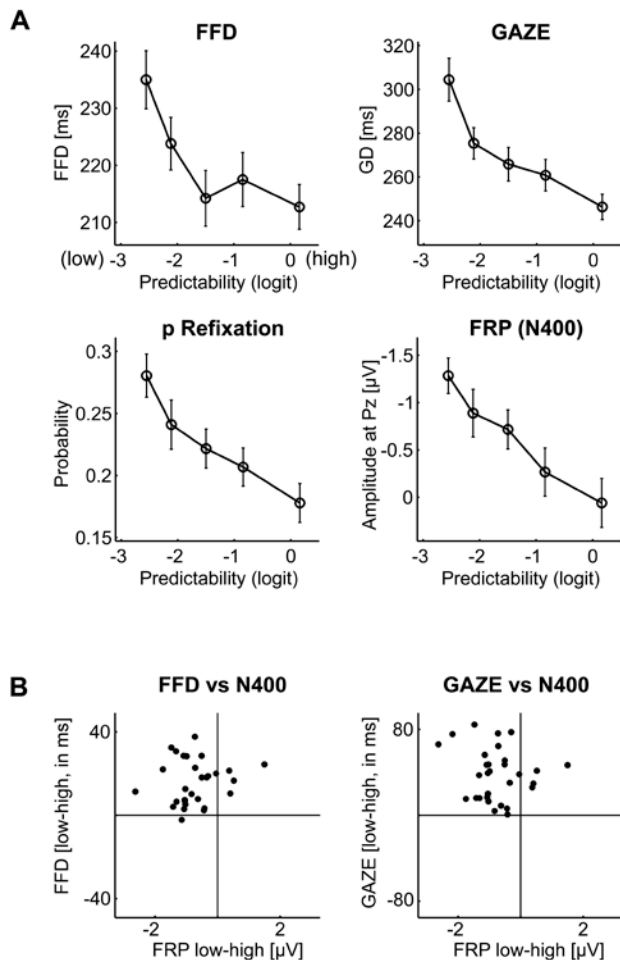


Figure 5. Comparison of EM and EEG effects.

A. Eye movement behavior (FFD, GD, and refixations probability) and FRP amplitude (at Pz between 300-500 ms) is plotted across five levels of word predictability (logit scaled). Mean cloze probabilities in the five bins were .00, .01, .05, .16, and .58. Note that negative voltages are plotted upwards in this panel only. Compared to N400 amplitude, FFDs showed stronger modulations in the low than in the high predictability range.

B. Size of the predictability effect (low minus high predictable words) in fixation times and FRPs. Each point indicates the data of one subject. Of 30 participants, 25 showed effects in the expected direction in both measures: FRPs were more negative, and fixation times were prolonged for low-predictable words. The size of EM and EEG effects did not correlate *across* subjects.

Discussion

Psycholinguistic Discussion

In the present experiment, participants read sentences from left-to-right, while eye movements and EEG were recorded. By time-locking the EEG to fixations on words that were expected to various degrees in the sentence context, we could replicate robust effects of word predictability on behavior and concurrent brain activity. The demonstration of predictability effects in an ordinary reading situation with heterogeneous sentence materials and in- and out-going saccades suggests that EEG recordings in natural vision are feasible in principal. In this first part of the Discussion, we comment on the psycholinguistic aspects of our results. A second, methodologically-oriented part of the Discussion reviews the technical challenges that emerged in the present experiment.

N400 effects

Decreasing word predictability increased a parietal negative-going component in the FRP, that reached a maximum at 384 ms. Importantly, this effect was observed despite the limited range of cloze probabilities in the Potsdam Corpus, which contains normal sentences and mostly words of low and moderate cloze probability. Because of its time course, polarity, scalp distribution, and sensitivity to word predictability, we take this component to reflect the N400. The fact that the topography of the effect was very similar to the N400 effect commonly observed during word-by-word presentation is reassuring evidence for the ecological validity of ERP data collected in traditional SVP paradigms.

Although the present study was mainly concerned with demonstrating the feasibility of this approach and did not primarily aim at covering new psycholinguistic ground – clearly a topic for future research – it also provided one indication that co-registration may yield somewhat different results than SVP. This concerns the N400 time course: The present N400 appeared to begin earlier than what is commonly reported from SVP. In visual word presentation, N400 effects typically arise at around 200-250 ms (Kutas et al., 2006). While our conservative onset criterion yielded a latency in this range (248 ms), we also observed much weaker N400-like effect topographies in earlier intervals after fixation onset, which did not survive correction for multiple comparisons. As we did not specify mixed models for these early intervals, we cannot exclude the possibility that this pattern was due to other variables correlated with predictability (e.g. constraint). Importantly, however, such early deviations were not observed by Dambacher et al. (2006) who presented the same sentences word-by-word and tested for early predictability effects in the P200 time window. In contrast, in an unpublished follow-up experiment that used an experimental manipulation of predictability (Dimigen, Sommer,

Dambacher, & Kliegl, 2008) we could replicate the observation of a comparatively early N400 onset in natural reading. An early onset of N400 effects under natural reading conditions was also reported in a recent study by Kretschmar, Bornkessel-Schlesewsky, & Schlesewsky (2009). These authors co-registered eye movements and EEG from centroparietal electrodes while participants read sentences that contained an entirely unpredictable target word. In a condition where the unpredictable target word was also semantically unrelated to the most expected word, an N400 effect arose soon after the first fixation on the target word (i.e., between 250-400 ms after the onset of the last *pre-target* fixation, which lasted 186 ms on average).

Although caution is necessary in the absence of a within-subject comparison to SVP, these observations indicate that the time line of word recognition in normal reading can differ from that commonly found in SVP experiments. An earlier N400 onset in normal reading is very plausible because of the parafoveal preview obtained during the previous fixation, a benefit absent in SVP. This could also explain the early N400-like deviations in the present study. But there are also other reasons why processing speed could differ in normal reading. For example, the fact that saccades are self-initiated should reduce temporal uncertainty about the arrival of new visual input. In the absence of parafoveal preview, Marton et al. (1985) still observed faster word-categorization after a 24° saccade, compared to foveal presentation. Similarly, Dimigen et al. (submitted) compared manual reaction times to small symbols presented either at fixation or at 10° eccentricity. Although parafoveal preview was unavailable in the 10° condition, post-saccadic RT (measured from saccade offset to reaction) was 30-70 ms shorter than RT to the same stimulus presented at fixation. Both results indicate that the time to prepare and execute a saccade can act as a foreperiod, which allows participants to optimize temporal preparation (e.g., Niemi & Näätänen, 1981) and thus enhances post-saccadic processing.

EM-EEG relationship

To our knowledge, the present study is the first to offer a detailed comparison of oculomotor and electrophysiological effects of a fixated item in free vision. Several exploratory analyses were carried out to investigate the EM-FRP relationship. As expected, both EM and FRP measures were sensitive to word predictability, suggesting that they are driven by common underlying processes. While gaze duration and especially refixation probability aligned well with N400 amplitude across five levels of logit predictability (Figure 5A), a different function was observed for first fixation durations. In an earlier study, Dambacher and Kliegl (2007) compared fixation times and ERPs for the same words, but measured in different groups of participants; EMs originated from natural reading whereas ERPs were collected in SVP. Dambacher and Kliegl reported remarkably similar functions for the duration of single

fixations and N400 amplitude across five levels of log frequency and four levels of logit predictability (their Figure 2). The profiles included even a disordinal trend with slightly longer single fixation durations and slightly larger N400 amplitude for words of medium log frequency. The study of Dambacher and Kliegl differs in several details from the present one (e.g., N400 from SVP rather than simultaneous recordings; aggregation over words, not over identical fixations) so it is difficult to speculate about the reasons for this difference in the exact relationship between fixation durations and N400 amplitude.

A model of N400 amplitude at the level of individual fixations (Table 3) provided no evidence for common variance between FFD and N400 amplitude that was not explained by the properties of the word or preceding sentence fragment. However, such covariance was observed between GD and N400 amplitude. Of course, this relationship could be mediated by other lexical variables not included in our model. The alternative explanation is that N400 amplitude and GD are directly related to each other, for example due to moment-to-moment fluctuations in the efficiency of word processing, which would affect both measures.

Since both EM and FRP measures are sensitive to word predictability, one might expect readers who show strong N400 effects to also show strong behavioral effects, and vice versa. For example, good readers should make better use of sentence context, and this may show up as larger predictability effects in EMs and FRPs. This was not the case: we found no evidence indicating that the size of the behavioral effects correlated with N400 effect amplitude across *participants* (Figure 5B). This result was surprising because Dambacher and Kliegl (2007) established such a correlation *across words* by using data from separate experiments. The lack of a correlation across *participants* is most likely caused by the notoriously unreliable difference-scores entering the correlations, which may represent too weak a signal to overcome individual differences in brain anatomy (e.g., cortical folding). Such anatomical differences between participants may influence the strength at which activity of an additional neural generator propagates to the scalp and could be a stronger source of N400 amplitude variation than differences in underlying brain activation, possibly concealing any existing relationship.

A final set of comparisons concerned the relative timing of predictability effects. Based on SVP data, it has been argued that there is a discrepancy between the latency of the N400 – the primary and so far the only robust index of semantic processing in psycholinguistic ERP research – and the fixation durations measured in eye tracking studies. The N400 typically peaks at around 400 ms in ERP studies, a time when the eyes have already left the critical word in natural reading (Sereno & Rayner, 2003). As Rayner and Clifton (2009) have pointed out, this “conundrum” is difficult to explain: How can the eyes react faster than the brain? We were able to address this

time lag question within the same dataset. Despite a relatively early N400 peak (386 ms) in normal reading, the pattern was still the same as in SVP: While predictability clearly influenced the duration of the first fixation on the target word, this fixation had almost always ended when the predictability effect peaked in the FRP (Figure 4D).

One common view on the N400 assumes that it reflects a late, post-lexical process of semantic context integration. Likewise, it is a commonly held view in EM research that these processes are reflected in gaze duration, which is seen as a measure of late processing. It is therefore interesting to note that the N400 peak did not fall into the mean gaze duration (278 ms) either. Of course, reading a low predictable word increases not only FFD and GD, but can also prolong later fixations on the following words (*spill-over or lag effects*; Rayner & Duffy, 1986; Kliegl et al., 2006). Nevertheless, the present data make it hard to conceive the measurable neural effects of predictability as being causal in some way for the behavioral effects, because the bulk of the predictability effects in ERPs only followed those in behavior. This raises questions about the functional interpretation of the N400 peak, whose latency does not seem to correspond to the maximum processing difficulty as reflected in the EM record.

Alternatively, one could consider the onset of the N400, rather than its peak, as the critical event. At the statistical N400 onset latency, readers were still looking at the target in more than half of the cases and were still in the first fixation in 38% of the cases. However, in order for lexicosemantic processing to influence FFD, it must do so before saccadic motor programming enters the non-labile stage, that is, at an estimated 80 ms before the end of the fixation (Becker, 1991; Findlay & Harris, 1984). The onset of the N400 effect therefore still seems to occur surprisingly late in comparison to the FFD effect. However, in contrast to the FRP analyses, the analyses of the SRP aligned to fixation *offset* provided some evidence that N400 effects may begin within the first fixation. The temporal contingencies observed here therefore do not rule out completely the possibility that the processes reflected in the N400 onset are also responsible for - or “driving” - the early effects in FFD. An answer to this question requires the design of dedicated experiments with strongly expected or unexpected words. Such experiments will allow very precise measurements of onset latency and possibly indicate that N400 onsets can occur early enough to influence behavior (see also Dimigen et al, 2008; Kretschmar et al., 2009). They will also clarify whether N400 effects can arise *before* the direct fixation of a target word due to parafoveal preview, a hypothesis that was not supported by the current results.

Possible applications

Apart from validating traditional ERP findings for more natural reading situations, co-registration can be used to investigate aspects of the reading process that are difficult or impossible to study with SVP. As described in the Introduction, one such aspect is the availability of

parafoveal information in natural reading. The timing and extent to which upcoming words are preprocessed is still controversial and can be studied in greater detail with FRPs. A promising approach in this context is the combination of simultaneous recordings with gaze-contingent changes of the computer display, as they are often used in eye tracking studies to manipulate preview (e.g., the boundary technique; Rayner, 1975). The question whether word meaning can be extracted from parafoveal words is one issue that could be further investigated with this technique. Co-registration is also the only viable approach to study EEG correlates of complex reading behavior. Interesting questions concern the EEG signature before a word is skipped rather than fixated, the FRPs that precede and follow the decision to trigger a regressive saccade (and their relationship to established syntactic ERP components), or the functional localization of individual differences in reading ability and reading speed. The final section reviews the relevant technical aspects for conducting such studies.

Methodological Discussion

Researchers who wish to record the EEG during reading or other free viewing tasks are faced with several technical and data-analytical problems, which are the likely reason why such recordings have rarely been attempted. The four main challenges we identified are (1) the need for precise co-registration of gaze position (2) the correction of corneoretinal and myogenic eye movement artifacts, (3) varying degrees of overlap between successive FRPs as well as between FRPs and background ERPs, and (4) variation of saccade-related cortical potentials according to low-level visuomotor factors. In the following, we will discuss each problem and possible solutions in some detail.

Co-registration of gaze position

A basic requirement for fixation-based averaging is accurate information about the latency and location of each fixation. Traditionally, ERP researchers have used electro-oculogram (EOG, Oster & Stern, 1980) electrodes near the eyes to control for a steady fixation. Basis of the EOG is an electrical gradient of 0.4 - 1 mV (Young & Sheena, 1988) between cornea and retina, which can be modeled by an equivalent electric dipole near the eye-ball (Berg & Scherg, 1991). Because changes in the orientation of the eye ball change the potential at peri-ocular electrodes, the EOG is well-suited to determine the onset latency of single, large saccades. However, with a spatial accuracy of $\pm 1.5 - 2^\circ$ (Malmivuo & Plonsey, 1995; Young & Sheena, 1988; see Joyce, Gorodnitsky, King, & Kutas, 2002, for a method to optimize EOG accuracy) it does not provide absolute gaze position with the single-letter accuracy required for reading analysis.

Current video-based eye trackers afford spatiotemporal resolutions up to $0.01^\circ / 2$ kHz, and both table-mounted

(Bodis-Wollner et al., 2002; Baccino & Manunta, 2005; Kennett, Van Velzen, Eimer, & Driver, 2007; Hutzler et al., 2007; Valsecchi, Dimigen, Sommer, Kliegl, & Turatto, 2009; Dimigen et al., 2009; Kretzschmar et al., 2009; for MEG see Herdman & Ryan, 2007) and head-mounted (Graupner, Velichkovsky, Pannasch, & Marx, 2007; Yuval-Greenberg, Tomer, Keren, Nelken, & Deouell, 2008) systems have been used for co-registration. Technical concerns about concurrent eye tracking are (1) pressure artifacts from contact between electrodes and eye tracker, (2) muscle artifacts resulting from head stabilization or unnatural sitting positions, (3) proper synchronization of the data records, and (4) electromagnetic artifacts from an electric device operating close to the EEG sensors. In the present study, these problems were minimized by (1) foam-cushioning forehead electrodes, (2) careful adaptation of the participant's sitting position, and (3) synchronization of EM and EEG records with a shared TTL pulse every few seconds. To double-check proper synchronization, we also use an A/D-converter in the eye tracker that feeds an analog copy of the gaze position as an additional channel into the EEG record. To test for (4) electromagnetic artifacts, we compared the EEG spectrum during steady fixation while the eye tracker was either recording or disconnected from power. We found that eye tracker operation introduced a weak 50 Hz line noise artifact at frontal electrodes near the eye tracker. However, this high-frequency artifact was irrelevant for the present FRP analyses and can be minimized by a notch filter (Yuval-Greenberg et al., 2008) or by using a remote eye tracker outside the shielded cabin.

In summary, advances in video-based eye tracking allow the routine recording of high-resolution EMs without obstructing EEG recordings. Moreover, two recent studies suggest that eye tracking can improve EEG data quality even in experiments that require steady fixation, as it allows to identify myogenic (Yuval-Greenberg et al., 2008) and visuocortical (Dimigen et al., 2009) potentials from involuntary microsaccades.

Eye movement artifacts

Eye movement artifacts in the EEG are generated by three mechanisms: rotation of the eye ball's corneoretinal dipole (Berg & Scherg, 1991), relative movements of the eye lid during blinks and upwards saccades (Picton et al., 2000), and electrical eye muscle activity at saccade onset, which propagates to the EEG as a spike potential (Thickbroom & Mastaglia, 1986). In normal vision, the strongest artifact source are corneoretinal artifacts. The changes in the corneoretinal potential, which provide the basis for the EOG, also propagate to the EEG electrodes, although they attenuate with increasing distance to the eyes (Picton et al., 2000). The horizontal saccades that are dominant in reading produce largest distortions at lateral-frontal channels and smallest distortions at posterior electrodes along the sagittal midline. While excluding of contaminated trials is not an option during natural vision,

there are simple ways to minimize EEG contamination despite saccades. Early studies with saccades recorded only from occipital midline sites (e.g., Gaarder, Krauskopf, Graf, Kropfl, & Armington, 1964) or used equal numbers of left- and right-going saccades, based on the assumption that both artifacts cancel out during averaging (e.g., Kurtzberg & Vaughan, 1981; Marton, Szirtes, & Breuer, 1985). Others have avoided the problem by limiting data analyses to the short interval of EEG before the first saccade (Baccino & Manunta, 2005; Simola et al., 2009) or after the terminal saccade in a sequence of saccades (e.g., Hutzler et al., 2007; Marton & Szirtes, 1988a; 1988b). Obviously, these approaches place severe limitations on the time segment, electrode site, and study design.

A large variety of algorithms have been proposed to correct mathematically for ocular artifacts (for reviews see Brunia et al., 1989; Gratton, 1998; Croft & Barry, 2000; Ille et al., 2002; Delorme, Sejnowski, & Makeig, 2007). Interestingly, the application of these algorithms has been largely restricted to correcting blink artifacts and accidental saccades in experiments that require fixation. Here, we applied surrogate MSEC (Berg & Scherg, 1994; Ille et al., 2002) to correct the heavily contaminated data. Although the collection of clean calibration EMs from each subject is time-consuming, the method was chosen for four reasons. First, MSEC can reduce the elimination of genuine brain activity compared to traditional regression-based methods (Berg & Scherg, 1994), because brain activity is modeled. Second, MSEC can be applied to continuous rather than averaged data, which supports a flexible re-segmentation of the corrected EEG to different time-locking points (e.g., the onset of fixation $n-1$). Third, the method does not make assumptions on the spatial or temporal orthogonality or independence of artifact and brain activities (the application of PCA during preprocessing is optional). Fourth, it does not require subjective choices from the experimenter apart from the one-time selection of a surrogate brain model.

Inspection of the continuous EEG, analysis of the averaged FRP, and analysis of residual correlations with the eye tracker converged to suggest that it was feasible to compensate for most of the artifact. While artifacts from the incoming saccade were completely abolished, residual artifact remained towards the very end of the fixation-locked segment, when several saccade artifacts had summated. This became apparent as small but significant correlations between about half of the corrected EEG channels and the eye track. Also, MSEC did not fully remove the spike potential (cf. Figure 4A, right panel), because its topography was not pre-defined as an artifact source. In summary, residual artifacts were small and correction quality was sufficient for the analyses that were being conducted. The use of more electrodes and a more realistic surrogate model may further improve MSEC correction.

Utility of eye tracking to improve ocular correction

Although correction worked well for the present purposes, other algorithms may have performed equally well or better. However, a fundamental problem with ocular correction methods is a lack of objective external criteria (Brunia et al., 1989) to compare and evaluate their performance on experimental data (for simulated data see Delorme et al., 2007; Klemm, Hauelsen, & Ivanova, 2009; Wallstrom, Kass, Miller, Cohn, & Fox, 2004). Choice of an appropriate correction method is particularly important for natural vision recordings. We therefore propose that concurrent eye tracking is useful to evaluate, compare, and improve correction methods:

First, unlike the EOG, the eye track provides a measure of eye position that is electrically independent of the EEG. Correlations between eye track and EEG after correction are therefore likely to result from residual corneoretinal or myogenic artifacts (or, less likely, from saccade-related brain activity occurring in synchrony with the saccade). The degree to which the EEG depends on eye position after correction (exemplified here by a linear correlation) can help to evaluate correction quality across studies and algorithms.

Second, eye tracking may inform about whether an algorithm overcorrected the data and distorted genuine brain activity. Provided that an experiment contains at least some intervals with steady fixation, high-resolution eye tracking allows the researcher to select EEG intervals *objectively* free of any ocular artifact (eye blinks, saccades, and microsaccades). These intervals should not be altered by ocular correction and therefore provide a test-case to quantify the distortion of brain signals introduced by the method.

Third, eye tracking may directly improve correction. Correction methods based on PCA or independent component analysis (ICA) decompose the EEG into multiple uncorrelated (e.g., Lagerlund, Sharbrough, & Busacker, 1997) or statistically independent (Delorme et al., 2007) signal components and correction is then performed by removing components classified as artifact. This classification is typically performed manually and based on criteria such as the component's scalp distribution and spectrum (Rong & Contreras-Vidal, 2006; Okada, Jung, & Kobayashi, 2007; Li, Ma, Lu, & Li, 2006), or correlation with the EOG (Joyce, Gorodnitsky, & Kutas, 2004; Wallstrom et al., 2004). However, classification can be ambiguous, especially when many components are produced, and many studies do not report selection-criteria (Fatourech, Bashashati, Ward, & Birch, 2007). Considering the relationship between the components' time series and gaze position (i.e., eye tracker-informed ICA) should greatly improve the reliability of component selection.

Finally, we propose that ocular correction is entirely unnecessary for certain research questions. Reading studies with SVP have provided some evidence of early ERP correlates of lexicosemantic processing within 200 ms after stimulus onset (Dambacher, Rolfs, Göllner, Kliegl, & Jacobs, 2009; Penolazzi, Hauk, & Pulvermüller,

2007; Hauk & Pulvermüller, 2004; Sereno, Brewer, & O'Donnell, 2003; for review see Pulvermüller, Shtyrov, & Hauk, 2009). An interesting question will be whether similar effects, within the duration of the current fixation, can be reliably established in FRPs and whether they predict the current fixation duration or the upcoming saccade target. Each fixation, however, is by definition a short interval of EEG that is free of EMs and that can be analyzed without prior correction. This approach requires a sufficient number of fixations with a minimum duration (e.g., 200 ms) after shorter fixations have been excluded. Second, because the artifact of the incoming saccade precedes fixation onset, a pre-fixation baseline - as in the present study - is not feasible. Instead, the baseline could be placed in the first few milliseconds of the fixation itself (provided that effects are not already present in the baseline due to parafoveal preview). Finally, direct current (DC) amplifiers should be used to prevent a spill-over of the pre-fixation artifact into the post-fixation segment due to the amplifier's time constant (Yagi, Kazai, & Takeda, 2000).

In summary, results suggest that corneoretinal artifacts are not a principal - and not the most serious - obstacle for EEG recordings during natural vision: Correction was sufficient for the present analyses and can likely be improved further. Eye tracking provides new criteria to validate and optimize correction while certain analyses do not require artifact correction at all.

Differential overlap

Serial presentation allows full control over the stimulus that is presented to the visual system at any time. In normal vision, on the other hand, the experimenter has little control over the spatio-temporal pattern of fixations, in particular over the latency and duration at which a participant chooses to fixate a target item. This leads to two problems of EEG overlap:

The first problem is the varying degree of temporal overlap between the potentials evoked by a target fixation and existing EEG *background activity* that is related, for example, to the stimulus onset. In the present experiment, sentence onset evoked a P300, which returned to baseline level only after about 700 ms. If target items in two conditions are fixated at systematically different latencies after stimulus onset - for example salient versus non-salient parts of a picture during scene perception - fixation-related potentials occur against a different background. This will distort the FRP waveshape and topography between conditions⁶. It is therefore important

⁶ Consider this example: In normal sentences, low predictable words tend to occur at earlier word positions and will be fixated sooner after sentence onset. As a consequence, the pre-fixation baseline interval for these fixations will overlap more often with the sentence-evoked P300. Via the process of baseline correction, which involves the subtraction of the baseline voltage from each channel, the sentence-onset P300 (a centroparietal

to ensure that target fixations do not differ in terms of overlapping background activity. In the current case, this was done by excluding early fixations and by ensuring that the pre-fixation baseline activity did not differ between predictability levels.

The second problem is temporal overlap between the potentials elicited by *successive reading fixations*. With inter-fixation intervals of around 250 ms, reading proceeds considerably faster than typical SVP paradigms, leading to massive overlap between the potentials evoked by subsequent fixations. This means that late, endogenous components from the previous fixation *n-1* overlap early, exogenous components from fixation *n*. Likewise, late components from *n* will overlap early components from *n+1* (cf. Figure 2B). Temporal overlap and, hence, summation of successive fixation-related responses over the duration of the trial was also the likely reason for the highly significant effect of word position on EEG amplitude in the N400 window (see Table 3); an effect that did not reach significance when the same sentences were presented in SVP (Dambacher et al, 2006).

Due to variance in fixation duration, the overlapping potentials will be latency-jittered relative to the current fixation and therefore low-pass filtered (Woldorff, 1993). While overlap is also encountered in fast SVP experiments (which have presented words at up to 10 Hz; Kutas, 1987), overlap is problematic if it differs systematically between conditions. This is clearly the case in reading and many other viewing tasks, where readers modulate fixation time according to processing difficulty. Because any fixation duration effect translates into a change in the phase of the overlapping potentials, EEG effects that reflect stimulus processing can be easily confused with changes that merely result from different degrees of overlap. Because differential overlap occurs with any fast and self-paced stimulation, the problem also applies to self-paced SVP (Ditman, Holcomb, & Kuperberg, 2007).

The extent to which overlap is a problem depends on the exact paradigm and the size of the behavioral effect. For example, the last fixation in a sequence is only influenced by overlap from previous fixations but not from subsequent fixations. In addition, in the absence of behavioral effects on fixation *n-1* (e.g., a parafovea-on-fovea effect), different overlap will influence the FRP only after the outgoing saccade is executed (this is not entirely true, because correlates of saccade preparation like the pre-saccadic positivity may precede the outgoing saccade).

We are not aware of a simple solution to the problem of differential overlap in FRPs, and this problem has been ignored in previous SRP/FRP studies. Several deconvolution methods have been proposed to separate overlapping potentials in ERP experiments with fast stimulation and variable inter-stimulus intervals

positivity) will carry over into the fixation-locked segment with a reversed polarity (as a centroparietal negativity), thereby creating a bogus, N400-like effect for low predictable words.

(Woldorff, 1993; Jewett et al., 2004; Wang et al., 2006; Hansen, 1983; Delgado & Özdamar, 2004). For example, *ADJAR* (level 2 implementation, Woldorff, 1993) is a time-domain technique that iteratively deconvolves overlapping waveforms based on knowledge about the temporal distribution of the ERP-eliciting events. In natural vision, this information is provided by the eye tracker. However, deconvolution methods typically rely on high signal-to-noise ratios (Talsma & Woldorff, 2004), require knowledge about the non-overlapped template waveform, or make the assumption that each successive event evokes an identical response. As discussed in the next section, the last condition, in particular, is unlikely to be met during free vision and it needs to be tested whether deconvolution can be successfully applied to FRPs.

However, the influence of differential overlap can at least be approximated by convolving an estimate of the non-overlapped FRP with the latency distribution of fixation onsets in each condition (here: low, medium, and high predictable words)⁷. As an alternative, the large pool of fixations that is easily obtained during natural vision allows the selection of fixation subsamples from each condition that are matched in terms of fixation duration. Because experimental effects on fixation duration are often small in reading, only a small percentage of fixations must be excluded to equalize the distribution of fixation durations post-hoc. Obviously, the resulting fixation samples, matched for fixation duration, present a biased selection, which may exclude the theoretically most interesting fixations. However, this procedure provides a simple test whether FRP effects persist once overlap is controlled.

Low-level influences on saccade-related brain potentials

Saccade- and fixation-related brain potentials are not only modulated by higher cognitive processing demands, but their waveshape is also influenced by visuomotor low-level factors that cannot be controlled during natural scanning behavior. These influences must be carefully delineated from effects that reflect higher-level cognitive processing (e.g., semantic processing) of the fixated item. Among these low level influences are (1) the exact visual input at saccade offset, and (2) the kinematics of the incoming saccade.

Most ERP studies of higher cognitive processing invest great care to match their visual stimuli in terms of low-level properties such as contrast, luminance, and spatial frequency. In contrast, retinal inputs vary considerably across different fixation locations (e.g., the center vs. the edge of a bright screen). Both amplitude and latency of the visually-driven lambda response around 100 ms after fixation onset (cf. Figure 2) vary as a function of the

luminance and contrast (Gaarder et al., 1964; Kazai & Yagi, 1999; 2006; Marton & Szirtes, 1982) and spatial frequency (Kazai & Yagi, 2006; Armington & Bloom, 1974) of the background. The lambda response therefore resembles the P1 component in ERPs, which is generated in overlapping areas of visual cortex (Kazai & Yagi, 2003) and modulated by the luminance, size, contrast, and frequency content of the field of presentation (Tobimatsu & Celesia, 2006). Visual influences on the lambda response have been observed with stimuli that covered large parts of the visual field (Riemslog et al., 1987). To our knowledge, no study has investigated the question whether differences in the foveal input across different fixation locations – for example local changes in luminance and contrast when viewing different parts of a scene – cause relevant modulations of the fixation-related EEG. For the sentence stimuli presented in the current study, we assumed that visual field properties would be comparable for different fixation locations on the screen. In addition, we attempted to reduce purely visually-driven effects by covering the peripheral visual field with a mask.

A second important low-level factor that influences SRPs and FRPs is saccade size. Saccade amplitude modulates not only the size of the pre-saccadic muscle spike (Boylan & Ross Doig, 1989), but also the waveshape of the post-saccadic EEG. The visual lambda response, in particular, comprises both saccade onset- and saccade offset-related responses (Thickbroom et al., 1991; Kurtzberg & Vaughan, 1977). For long saccades (> 10°), it dissociates into two subcomponents, a first subcomponent, presumably related to visual changes at saccade onset and a larger second one, presumably evoked by the inflow of new visual input at saccade offset (Thickbroom et al., 1991). However, as Figure 2C shows, saccade amplitude modulates the lambda response even for the limited range of saccade amplitudes found in reading. Therefore one needs to be cautious when conditions are compared that differ in terms of incoming saccade amplitude. This is not necessarily a fundamental limitation because reading saccades have fairly constant amplitudes of around 7-9 characters (Rayner, 1998) and eye tracking allows the post-hoc exclusion of very short or long saccades. In the present case, condition differences in incoming or outgoing saccade amplitude were too small (< 0.1°) to cause relevant changes in FRP waveshape. However, when we specified a mixed model (not shown here) in which we added incoming saccade amplitude as an additional predictor, this predictor explained EEG variance at electrode Pz in all time windows between 40 and 280 ms after fixation onset. We therefore propose to include incoming saccade amplitude as a covariate in FRP analyses. Finally, we are not aware of any study that has tested the influence of saccade *direction* on FRPs, a question that seems important for applications of the technique to visual search or scene perception.

⁷ Such a simulation (not shown here), conducted for the present dataset with the grand-average FRP (averaged across conditions) as the waveform estimate, suggested that effects of differential overlap were small compared to the much larger effect of word predictability.

Summary: Feasibility of co-registration

Fixation-related potentials are influenced by corneoretinal artifacts, differential overlap, and visual- and motor-related brain potentials, all of which vary with oculomotor behavior. These indirect influences of EM behavior on the EEG can be easily mistaken for genuine condition differences in the brain's processing of the fixated item. Special care must be taken during the analysis and interpretation of multi-saccadic EEG experiments, and researchers should account for how these problems were addressed. In particular, details about oculomotor behavior (fixation durations and saccade amplitudes) on the fixations preceding and following a target fixation should be provided. The aforementioned problems are not exclusive to the analysis of FRPs in the time domain but also apply to EEG analyses in the frequency domain, that is, to saccade- or fixation-related oscillations (SROs or FROs). However, EEG recordings in natural vision also have the major advantage in that many fixations can be collected in a short time and with little strain on the subject. In future studies, these large fixation pools could be used to model and disentangle the influences of artifacts, low-level processing, and higher level processing on the fixation-related EEG.

Conclusions

Visual perception outside the ERP laboratory is fundamentally trans-saccadic and involves an active sampling of the environment several times per second. What types of information are obtained on each fixation and how is it integrated with the information from previous and subsequent fixations? Despite their advantages, EEG recordings have been largely precluded from natural viewing conditions. We demonstrated here that EEG indices of semantic processing can be obtained in natural reading and compared to EM behavior. With the appropriate consideration of technical and data-analytic issues, concurrent recordings may contribute new answers to long-standing questions.

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