Visual plasticity and exercise revisited: No evidence for a “cycling lane”

Abigail E. Finn
McGill Vision Research, Department of Ophthalmology & Visual Sciences, McGill University, Montreal, Quebec, Canada

Alex S. Baldwin
McGill Vision Research, Department of Ophthalmology & Visual Sciences, McGill University, Montreal, Quebec, Canada

Alexandre Reynaud
McGill Vision Research, Department of Ophthalmology & Visual Sciences, McGill University, Montreal, Quebec, Canada

Robert F. Hess
McGill Vision Research, Department of Ophthalmology & Visual Sciences, McGill University, Montreal, Quebec, Canada

Experiments using enriched environments have shown that physical exercise modulates visual plasticity in rodents. A recent study (Lunghi & Sale, 2015) investigated whether exercise also affects visual plasticity in adult humans. The plastic effect they measured was the shift in ocular dominance caused by 2 hr of monocular deprivation (e.g., by an eye patch). They used a binocular rivalry task to measure this shift. They found that the magnitude of the shift was increased by exercise during the deprivation period. This effect of exercise was later disputed by a study that used a different behavioral task (Zhou, Reynaud, & Hess, 2017). Our goal was to determine whether the difference in task was responsible for that study’s failure to find an exercise effect. We set out to replicate Lunghi and Sale (2015). We measured ocular dominance with a rivalry task before and after 2 hr of deprivation. We measured data from two conditions in 30 subjects. On two separate days, they either performed exercise or rested during the deprivation period. Contrary to the previous study, we find no significant effect of exercise. We hypothesize that exercise may affect rivalry dynamics in a way that interacts with the measurement of the deprivation effect.

Introduction

Brain plasticity is usually thought of in terms of critical periods in young animals. It is now recognized, however, that a degree of plasticity remains into adulthood. Studies using rodents have shown experience-dependent plasticity as a result of enriched environments. These can have both structural and functional benefits for the adult brain (Sale, Berardi, & Maffei, 2016). One key component of these enriched environments is physical exercise. Previous studies have also found that exercise increases neural responsivity in the visual cortex of adult rodents (Niell & Stryker, 2010).

Although exercise effects have been found in the visual cortex of rodents (Sale et al., 2007), their extrapolation to primates is not conclusive. In humans, evidence for an exercise effect on plasticity has previously been limited to prefrontal and hippocampal regions (Erickson, Gildengers, & Butters, 2013). This was the case until the recent study by Lunghi and Sale (2015). They argued that exercise could modulate the plasticity of the early visual cortex of adults. For their measure of plasticity, Lunghi and Sale used the shift of ocular dominance caused by short-term monocular deprivation. If one eye is deprived of visual input for a period of around 2 hr, there is a shift in dominance in favor of the patched eye. This effect has been demonstrated with deprivation from eye patches (Lunghi, Burr, & Morrone, 2011), dichoptically presented movies (Zhou, Reynaud, & Hess, 2014), processed video input from “altered reality” systems (Bai, Dong, He, & Bao, 2014).
2017), and continuous flash suppression (Kim, Kim, & Blake, 2017). The shift in ocular dominance has been confirmed with EEG (Zhou, Baker, Simard, Saint-Armour, & Hess, 2015), fMRI (Binda et al., 2018), MEG (Chadnova, Reynaud, Clavagnier, & Hess, 2017), and various psychophysical tasks (Baldwin & Hess, 2018; Spiegel, Baldwin, & Hess, 2017; Zhou, Clavagnier, & Hess, 2013). For their study on the effect of exercise, Lunghi and Sale measured the ocular dominance shift with a binocular rivalry paradigm. The perceptual dominance of each eye was monitored before and after deprivation. They found that exercise during the deprivation period enhanced the shift in ocular dominance. However, a later study by Zhou et al. (2017) failed to find an effect of exercise. Their methods were similar to Lunghi and Sale although, rather than binocular rivalry, they used a task that measured the relative strength of the input from each eye to a fused binocular percept.

It is possible that the use of a fusion rather than a rivalry task is responsible for the failure to replicate the previous effect. Recent studies have demonstrated that different effects of monocular deprivation may be found depending on the psychophysical task used to measure them (Bai et al., 2017; Baldwin & Hess, 2018). The binocular fusion task used by Zhou et al. (2017) involves two grating stimuli of the same orientation and spatial frequency but slightly different spatial phase. These are fused into a single binocular percept with the phase of that perceived grating shifted to favor the dominant eye. This combination can be attributed to excitatory interactions. The rivalry task used by Lunghi and Sale (2015) is the perceptual alternation of stimuli of orthogonal orientation that stimulate different neuronal populations. Their perceptual competition is subserved by inhibitory interactions (Klink, Brascamp, Blake, & Van Wezel, 2010). Zhou et al. (2017) left open the possibility that differences between the results from the two studies may be due to the different methods used. One task measures inhibitory interactions between different neural populations tagging left and right eye responses. The other measures excitatory combination of left/right eye signals within a single neuron population.

To determine whether the difference in task was responsible for the previous failure to replicate, we set out to reassess the effect of exercise on visual plasticity. As before, we used the short-term monocular deprivation paradigm but with the binocular rivalry measure rather than the binocular fusion measure. This amounts to a replication of the study of Lunghi and Sale (2015). We have endeavored to use as similar a protocol to theirs as their described methods allow.

### Methods

#### Subjects

We recruited 30 subjects, including one author (AF), two experienced psychophysicists, and 27 naive subjects. The average age was 22 years (range 18–26). Of the subjects recruited, 21 were female. All procedures were approved by the research ethics board of the McGill University Health Centre and carried out in accordance with the Declaration of Helsinki. Informed written consent was obtained from the subjects.

#### Procedures

In the binocular rivalry experiment, oblique Gabors at +45° and −45° were presented separately to the two eyes using shutter glasses. The Gabors had a spatial frequency of 1.5 c/° and a spatial sigma of 1.3° of visual angle and a contrast of 50%. The experiment program randomly assigned the two grating orientations to the two eyes for each block to mitigate any orientation bias. However, due to a failure to consistently reset the random number seed in the experiment program, the two possibilities (left oblique for left eye and right oblique for right eye or vice versa) were not equally likely. Instead, there was a two-thirds chance that the left eye would receive the left oblique grating. This convergence was most extreme for the initial baseline measurement, in which only 3% of blocks had the opposite stimulus arrangement. Although unfortunate, we do not see any way that this oversight could be responsible for the pattern of results we find. For a 3-min testing period, subjects then used a keypad to continuously indicate their current percept. Subjects had three buttons to press to signal three perceptual states: “left oblique grating,” “right oblique grating,” and “mixed percept.” All three states were illustrated for the subject before they began testing, so they knew how to respond. Data were processed by assigning the two Gabors orientations to the role of the eye in which they were presented (patched or non-patched eye).

We measured the shift in ocular dominance after patching in two conditions. In the control condition, subjects rested during the patching period by sitting in a chair. In the cycling condition, subjects performed physical exercise by cycling on a stationary standing bike during the patching period. Subjects performed the two conditions on different days (separated by at least 24 hr). For both conditions, a baseline measurement was made using the binocular rivalry task. After this measurement, a translucent patch was fitted over the dominant eye of the subjects. We determined the
dominant eye using the Miles test for sighting dominance. This was different from Lunghi and Sale (2015), in which the dominant eye was determined using the rivalry task. This deviation is addressed in the appendix. Once the patch was fitted, the subject then viewed movies (cinematic films) on the projector screen for 2 hr. In the control condition, the subject sat in a chair throughout this period. In the cycling condition, subjects used a standing bike to alternate 10-min periods of exercise (target heart rate of 120 beats per minute) and rest. After 2 hr, the patch was removed, and subjects were tested at 0, 4, 8, 12, 30, 45, 60, 90, and 120 min following patch removal.

**Apparatus**

Subjects sat at 2.3 m from a projector screen. Both the stimuli for the binocular rivalry task and the movies viewed during the monocular patching period were presented on the screen by an Optoma HD26 DLP projector. The resolution of the projector gave 75 pixels per degree of visual angle. The mean luminance of the screen was 95 cd/m². The experiment was programmed in MATLAB (MathWorks, Natick, MA) using Psychtoolbox (Kleiner et al., 2007). During the binocular rivalry task, the grating stimuli were presented dichoptically using frame interleaving with a pair of Optoma ZD302 DLP Link Active Shutter 3-D glasses. During the binocular rivalry task, the control movie viewing, and the rest periods in the cycling movie viewing, subjects sat on a chair. During the active periods in the cycling condition, subjects pedaled a stationary bike at such a pace as to sustain a heart rate of 120 beats per minute (monitored by a Polar H7 heart rate sensor).

**Results**

**Our replication attempt**

Measurements taken over the 120 min of recovery from 2 hr of patching are presented in Figure 1A. The response duration histograms were heavily skewed (Figure 2A and B). To give a measure that is independent of the shape of the histogram, we used the total time that the subject responded that the subject was seeing the stimulus for the patched or nonpatched eye. We used this to calculate the ocular dominance index (ODI) after Dieter, Sy, and Blake (2017), equation 1).

\[ \text{ODI} = \frac{d_p - d_n}{d_p + d_n}, \quad (1) \]

where \(d_p\) and \(d_n\) are the total response durations for the patched and nonpatched eyes, respectively.

The time courses of the recovery from 2 hr of patching with and without exercise are presented in Figure 1A for the ODI calculation. Five of our 30 subjects each had a single data point at which the data were not collected. When a subject was missing a data point for one condition, we also excluded data from the same time point in the other condition. These five subjects were also excluded from the two-way, repeated-measures ANOVA analysis we performed in RStudio (RStudio Team, 2016). The results from the ANOVA show a significant effect of time point, \(F(8, 192) = 11.09, p < 0.001\), but no effect of exercise, \(F(1, 24) = 0.82, p = 0.37\), and no interaction effect, \(F(8, 192) = 1.19, p = 0.31\).

We performed an additional analysis taking the area under the ratio versus linear time curve (Figure 1B). The area gives the overall deviation from baseline, representing the magnitude of the patching effect. We calculated the area for each subject (normalized for their individual baseline measurements). As removal of a single time point did not prevent an area being calculated, all subjects’ data were used in this analysis. We found no significant difference between the cycling and control conditions, \(t(29) = -0.84, p = 0.41\). In fact, in our data the (nonsignificant) trend is in the opposite
direction with a reduced patching effect in the cycling condition.

**Comparison with Lunghi and Sale (2015)**

In addition to the ODI, we considered two other ways to analyze the binocular rivalry data. In Lunghi and Sale (2015), the mean durations of the individual “patched eye” and “nonpatched eye” percepts were used to calculate the ocular dominance ratios. The duration distributions were skewed however (Figure 2A and B), so the mean would be affected by the relatively few responses that have much longer durations than the others. This departure from normality was highly significant for each of the distributions in Figure 2A and B, when tested with the normaltest function in SciPy (D’Agostino & Pearson, 1973; Jones et al., 2001). To account for this, we also analyzed the median response duration. To calculate the ocular dominance, we divided the average (mean or median) response
duration for the patched eye by the average duration for the nonpatched eye. We took the log₂ of that ratio so that a 2:1 ratio in favor of the patched eye would be a log₂ ratio of 1, whereas a 1:2 ratio in favor of the nonpatched eye would give a log₂ ratio of −1. For the mean and median ratio measures, the recovery from patching was similar to Figure 1A and gave the same pattern of results on the ANOVA analysis.

In Figure 2C–E, we present the mean-across-subjects values for the median duration, mean duration, and ODI measures. For each study, we performed a paired t test in SciPy (Jones et al., 2001). In all cases, the distribution of the differences was normal according to the SciPy normaltest function. In our data, we find no significant difference between the cycling and control conditions. In the data from Lunghi and Sale (2015), we find highly significant effects when the mean and ODI are used as the measures of eye dominance but no significant effect with the median response duration. It is worth noting that we find patching effects of comparable magnitude to Lunghi and Sale. For example, the mean ODI in the first measurement was 0.27 in their data, and in our study, it was 0.28 (taking the mean over the same time period). We do not, however, find the differences in patching effect magnitude between the cycling and control conditions that they report. We performed a post hoc power analysis using G*Power (Faul, Erdfelder, Lang, & Buchner, 2007) on the results from Lunghi and Sale. When comparing the area-under-the-curve patching effects based on the ODI calculation, the effect size was 0.73. To achieve a power of 95% based on a 5% significance level in our replication study, at least 27 subjects would have to be tested. Our 30 subjects gave a power of 97%. For the analysis based on the mean phase duration, the effect size in Lunghi and Sale’s data was larger (0.89). To achieve a power of 95%, only 19 subjects needed to be tested. With our 30 subjects, the power was more than 99%. Our study, therefore, should have been sufficiently powerful to replicate the previous finding.

The t tests in Figure 2C–E were used as the basis for a replication Bayes factor calculation (Verhagen & Wagenmakers, 2014) in RStudio (Harms, 2018; RStudio Team, 2016). We used the t statistics for the ODI measure to quantify the evidence for the Lunghi and Sale (2015) effect in our data, relative to the null hypothesis in which exercise does not affect the patching effect. The ratio between the alternative and null hypotheses was 0.015, giving strong (Kass & Raftery, 1995) support to the null.

The use of these multiple measures allows us to draw a more nuanced conclusion about both the nature of the patching effect and its modulation by exercise. In both studies, patching increases the duration of the median patched eye response relative to the median nonpatched eye response. It also increases the total proportion of time during which patched eye responses are given. We were not able to find any significant effect of exercise in our data for any of the three measures we analyzed. Even the trend favoring a stronger effect in the cycling condition that we find for the median and mean analyses is reversed when we make our ODI analysis. In the Lunghi and Sale (2015) data, there is no significant effect on the median response duration from exercise. This means that there is no evidence that exercise increases the average response duration seen by the patched eye. Instead, it may be that exercise results in an increased duration of the longer responses. This would result in distributions that are more skewed after exercise. The results from Lunghi and Sale in Figure 2C and D support this with a highly significant increase in the mean phase duration but no significant change in the median.

### Analysis of the baseline behavior of the subjects

In our experiment, data were collected from the 30 subjects before analysis was conducted. We did not continuously monitor for any aberrant results or indications of poor data quality. It is possible that, had we done so, our subject pool would have been reduced to one in which the effect of exercise would be found. One possible metric that could have been considered for this purpose would be the proportion of “mixed” percepts reported by a subject. When more mixed percepts are reported, there will be less data regarding the dominance of the “left eye” or “right eye” percepts. These subjects may, therefore, show a weaker effect of patching and dilute the result. We performed a further analysis of our data to determine whether mixed percepts were responsible for our failure to replicate the results of Lunghi and Sale (2015). We calculated the mixed percepts index (MPI) as

\[
MPI = \frac{d_m - (d_p + d_n)}{d_m + d_p + d_n},
\]

where \(d_p\) and \(d_n\) are the total response durations for the patched and nonpatched eyes and \(d_m\) is the total response duration for the mixed percepts. The median MPI calculated across subjects is presented in Figure 3A. The horizontal black line shows the median MPI from the Lunghi and Sale data, whereas the rightmost gray circle shows the median MPI from our data (30 subjects). As the gray curve declines to the left of that point, it shows the effect of excluding subjects who showed the greater proportion of mixed percepts from our analysis. It is worth noting that only one of our subjects shows a proportion of mixed percepts lower than the median subject from Lunghi and Sale. The proportion of mixed percepts is largely dependent on the criterion that the subject uses. A previous study
instructed subjects to only identify the dominant grating when it was exclusively visible. They found that their subjects always reported mixed percepts for at least one third of the duration (O’Shea, Sims, & Govan, 1997). Plotting this on Figure 3A gives a horizontal line at −0.33. Our proportion of mixed percepts was lower (21% with 79% “exclusive” percepts) as we instructed our subjects to respond based on whichever grating was dominant. They were only to give the “mixed” response when the two appeared to be equally dominant. If similar instructions were given in Lunghi and Sale, then the differences between our studies are consistent with our subjects having different criterion levels for this “mixed” percept. Figure 3C shows how our conclusions regarding the effect of exercise on the ocular dominance shift from patching may have changed if we had excluded subjects who reported a greater proportion of mixed percepts. Only by excluding most of our subjects (more than two thirds) do we see a nonsignificant trend for exercise to increase the shift in ocular dominance. However, we would have no principled reason for setting the exclusion criterion at this point. It would perhaps have been more surprising if we could not find a way that our data could be “massaged” to show a trend in the same direction as the previous study.

Another metric would be the day-to-day variation in the measured baseline. A large difference in baseline between sessions may indicate that data measured from that subject are not reliable. This could add noise to the results, which would also reduce any measure of statistical significance. We calculated the day-to-day baseline variation as the difference in the ODI
measured on the two days of testing (one day for the cycling condition and one day for the control).
Performing this subtraction for each of our subjects gave us 30 values. The overall spread was characterized by the standard deviation of those differences. This is shown for our study in Figure 3B by the rightmost gray circle (30 subjects) in comparison to the solid black line from Lunghi and Sale’s (2015) data. Similar to Figure 3A, the data points to the left of that rightmost point indicate the effects of removing subjects showing the greater amount of day-to-day variation in their baselines. Our subjects were more variable than those from Lunghi and Sale. To reduce the standard deviation of our data to match theirs, we had to exclude two thirds of our subjects. Some of this variability comes from measurement error with the rest due to day-to-day variability in ocular dominance. We obtained an estimate of this day-to-day variability using data from another study in which 19 subjects were tested for six rivalry sessions (80 s each) across 2 days (Sheynin & Hess, personal communication, October 2018). The median ODI across six sessions has a much smaller measurement error than the ODI from a single session. We, therefore, used the median ODI from those six sessions to calculate the standard deviation of the day-to-day differences in ocular dominance. This is presented as the horizontal dashed line on Figure 3B and is similar to the standard deviation found with our full data set. The standard deviation found by Lunghi and Sale was much lower. This suggests that the subjects selected in that study had a particularly stable ocular dominance baseline. Figure 3D shows how our results would have changed if we excluded subjects who showed greater day-to-day baseline variation. There is no evidence that the more variable subjects were hiding any underlying enhancement of the ocular dominance shift. If anything, we find a significant effect in the opposite direction for certain combinations of subjects (indicated by asterisks).

**Discussion**

We find no effect of exercise on the transient neuroplastic changes caused by monocular deprivation. This result is surprising considering the size of the effect reported by Lunghi and Sale (2015). Their ANOVA found a large effect of exercise, $F(1, 19) = 9.58, p = 0.006$. It is, therefore, unlikely that the difference between our studies is due to chance. We calculated a replication Bayes factor (Verhagen & Wagenmakers, 2014) using the ReplicationBF package in RStudio (Harms, 2018; RStudio Team, 2016). We found a Bayes factor of 0.015, indicating strong support (Kass & Raftery, 1995) for the null hypothesis (in which exercise does not influence the patching effect). This evidence adds to the lack of an exercise effect found in a previous study using a binocular combination method (Zhou et al., 2017) and an as-yet unpublished study we have conducted with dichoptic surround suppression.

The procedures and subject population were similar between our study and Lunghi and Sale (2015). Our binocular rivalry statistics were within the normal range (O’Shea et al., 1997). The stimuli had small differences. The gratings we presented were around 30% larger. Although the contrast was the same (50%), the luminance of our display was 2.5 times higher. None of these differences seem likely to account for the absence of an exercise effect. We performed further analyses to see whether an effect exists in a subset of our cohort. We found that our failure to replicate the previous result was not due to including subjects with more day-to-day variability. Nor was it due to including subjects who gave more “mixed percept” responses. We did not find any post hoc “inclusion criteria” for our subjects that would have led us to replicate the finding of Lunghi and Sale.

We chose the ocular dominance index to present in Figure 1, but an analysis of the mean phase duration did not find a significant effect in our data either. The ODI analysis was significant for the data of Lunghi and Sale (2015) although a median phase duration analysis was not. The general effect of patching, however, is seen across all three measures, including median phase duration. Therefore, if exercise were to enhance that effect, we would expect it to have an equal impact on all three measures. It is possible that exercise may further skew the phase duration distributions in binocular rivalry. This would result in an effect that is driven by a small number of longer percepts whose durations are further extended.

A previous study found no effect of exercise on the patching effect measured using a binocular fusion task (Zhou et al., 2017). This result suggested that any modulation by exercise did not generalize to other measurement methods. The current study attempted to confirm the existence of the exercise effect with binocular rivalry as was used by Lunghi and Sale (2015). We were not able to replicate their finding. In the light of our results and the additional analysis of the data from Lunghi and Sale, we propose that, rather than exercise enhancing the patching effect per se (as they purport), exercise may be affecting the behavior of the subjects in a binocular rivalry task in a way that inflates some measures of the patching effect. Controlling for this would require a study that investigated the effect exercise has on binocular rivalry dynamics in the absence of patching.

**Keywords:** vision, psychophysics, binocular, rivalry, exercise, ocular dominance, plasticity, monocular deprivation
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Commercial relationships: none.
Corresponding author: Robert F. Hess.
Email: robert.hess@mcgill.ca.
Address: McGill Vision Research, Department of Ophthalmology, McGill University, Montreal, Quebec, Canada.

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Klink, P. C., Brascamp, J. W., Blake, R., & Van Wezel,


**Appendix A: Further analyses**

We considered the possibility that variations in rivalry dynamics over our 3-min testing period may obscure the presence of an exercise effect. We performed a further analysis in which our 3-min data blocks were subdivided into three 1-min mini-blocks (data from 0 to 60 s were labeled the *early* rivalry response, data from 60 to 120 s were the *middle* response, and data from 120 to 180 s were the *late* response). We performed a three-way ANOVA (factors were time point prepatching/postpatching, whether it was a cycling or control condition, and the three mini-block time bins: early, middle, and late). In some cases, subjects reported extended mixed percepts that lasted long enough that the entire 60-s period may contain only a single response. These would result in missing data in our analysis, so the six subjects for whom this occurred in any of our 20 measurements had to be excluded from the ANOVA. This left 19 subjects for the ANOVA analysis (as we also removed those subjects with missing data who were omitted from the original ANOVA analysis). Consistent with the original ANOVA analysis, we find a significant effect of time point, \( F(8, 144) = 8.05, p < 0.001 \), but no significant effect of exercise, \( F(1, 18) = 0.06, p = 0.803 \), nor any significant interaction between the two, \( F(8, 144) = 0.428, p = 0.902 \). There was a significant difference between the three 1-min mini-blocks, \( F(2, 36) = 4.531, p = 0.018 \); however, there was no significant interaction with time point, \( F(16, 288) = 0.843, p = 0.635 \), or exercise versus control, \( F(2, 36) = 0.446, p = 0.643 \). There was also no significant three-way interaction between exercise versus control, time point, and mini-block, \( F(16, 288) = 0.895, p = 0.575 \).

We also processed the data from the early, middle, and late mini-blocks in the same way as the combined data was processed in Figure 1 (Figure A1). There does not appear to be any indication that an enhancement from exercise occurred in any of the three mini-blocks. We performed a \( t \) test comparing the area under the curve from the cycling and control conditions for each bar plot. None of these showed a significant difference: early: \( t(29) = 1.18, p = 0.25 \); middle: \( t(29) = -0.13, p = 0.89 \); late: \( t(29) = 1.42, p = 0.17 \).
One difference between this study and that of Lunghi and Sale (2015) is that we determined the eye to be patched using sighting dominance (the Miles test). Lunghi and Sale used the eye dominance found with their binocular rivalry task. It has been demonstrated that there is a large degree of separation between the two types of dominance. One cannot simply predict the dominant eye in rivalry using sighting dominance (Coren & Kaplan, 1973; Ding, Naber, Gayet, Van der Stigchel, & Paffen, 2018; Mapp, Ono, & Barbeito, 2003). This raised the possibility that our failure to replicate an effect of exercise may be due to it only arising in cases in which the rivalry-dominant eye was patched. Our inclusion of subjects whose dominant eye in binocular rivalry was not the same as that for sighting dominance would, therefore, dilute this effect.

We analyzed the average baseline ocular dominance indices of our subjects. We found that 13 of the 30 subjects did not have the same dominant eye for sighting dominance and rivalry dominance. We then analyzed the data from the remaining 17 subjects (whose rivalry-dominant eyes were patched). The results of this analysis are shown in Figure A2. In agreement with the rest of our analyses, we find no effect of exercise. The difference between the control and cycling conditions shown in the bar plot is not statistically significant.

One difference between this study and that of Lunghi and Sale (2015) is that we determined the eye to be patched using sighting dominance (the Miles test). Lunghi and Sale used the eye dominance found with their binocular rivalry task. It has been demonstrated that there is a large degree of separation between the two types of dominance. One cannot simply predict the dominant eye in rivalry using sighting dominance (Coren & Kaplan, 1973; Ding, Naber, Gayet, Van der Stigchel, & Paffen, 2018; Mapp, Ono, & Barbeito, 2003). This raised the possibility that our failure to replicate an effect of exercise may be due to it only arising in cases in which the rivalry-dominant eye was patched. Our inclusion of subjects whose dominant eye in binocular rivalry was not the same as that for sighting dominance would, therefore, dilute this effect. We analyzed the average baseline ocular dominance indices of our subjects. We found that 13 of the 30 subjects did not have the same dominant eye for sighting dominance and rivalry dominance. We then analyzed the data from the remaining 17 subjects (whose rivalry-dominant eyes were patched). The results of this analysis are shown in Figure A2. In agreement with the rest of our analyses, we find no effect of exercise. The difference between the areas under the curve found in the cycling and control conditions was compared using a t test. It was not significant: \( t(16) = 0.33, p = 0.75 \). We recalculated the statistical power of our t test for this reduced sample size based on the result from Lunghi and Sale. Although we have excluded 13 subjects, we still achieve a power of 81%.