PEER REVIEW HISTORY

ARTICLE DETAILS

TITLE (PROVISIONAL)	Neurophysiological correlates of motor planning and movement initiation in ACL-reconstructed individuals: A case-control study
AUTHORS	Giesche, Florian; Engeroff, Tobias; Wilke, Jan; Niederer, Daniel; Vogt, Lutz; Banzer, Winfried

VERSION 1 – REVIEW

REVIEWER	Samuele Passigli University of Rome "Tor Vergata"
REVIEW RETURNED	17-Apr-2018

GENERAL COMMENTS	The study is very interesting and well written, congratulations. Neuroplasticity after ACL reconstruction is certainly a current topic to optimize the outcome of rehabilitation, encourage return to sport/play and reduce the risk of a new injury. Only some clarifications: line 58-59: The statement "ACL tears represent the most frequent injury of the knee" is very strong. Do you have any recent references to support this? Or would it be better to change "the most frequent injury" with "one of the most common sports-related injuries to the knee"? line 128: Why were only males included? Female athletes are at greater risk than male athletes for noncontact ACL injury. For example, see: https://www.ncbi.nlm.nih.gov/pubmed/27315457 and https://www.ncbi.nlm.nih.gov/pubmed/15722283. line 146-147: Hop tests are frequently used to determine return to sports after ACL reconstruction. Given that bilateral deficits are present after ACL reconstruction, this may result in a falsely high limb symmetry index (LSI), since LSI is calculated as a ratio between the values of the limbs. See https://www.ncbi.nlm.nih.gov/pubmed/28428033. line 175: Why did you choose a total of 70 successful jumps? Was it an arbitrary choice or did you use a protocol used previously? line 241: Add “:” after “Three parameters will be investigated”. line 242: Add “.” after “of pressure (COP) path length”. line 266: Delete “;”. line 268: Delete “;”.

	line 270: Reference?

REVIEWER	Nathan Schilaty, DC, PhD Assistant Professor Mayo Clinic, USA
REVIEW RETURNED	17-May-2018

GENERAL COMMENTS	In this protocol article, the authors have outlined their methodologies for investigation of neurophysiological correlates of motor planning with controls and ACL reconstructed individuals. They propose the use of force plates, EEG, and complex statistical methods. Major concerns: Methodologically, if unsuccessful trials are categorized as landing errors, will this bias the data? Not successfully landing could be due to an overbearance on the cortical processing and could be indicative of the injury mechanisms. Exclusion could lead the researchers away from the actual 'signal' they desire. It was not made clear why only male sex has been recruited. This should be addressed as sex is an important biological variable to consider, especially as females have higher risk for ACL injury. If this clinical trial is ongoing, will it involve both sexes? As cognitive function was not directly addressed in the manuscript, Table 1 is not necessary for inclusion in this manuscript unless it is further discussed as to how these variables will be utilized to parse the data. After Independent Component Analysis, it is mentioned that the trails with remaining artefacts will be rejected and only artefact-free trials will be used for analysis. How do the authors successfully determine what is artefact and what is not? Interpretation may describe it as artefact when it is actually signal. Minor revisions: Line 34 – Remove 'In' Line 77 – 'The authors' is not clear that it is reference to the previous study as it may be considered that it is the authors of the current study. This should be clarified. Line 93 – This statement is not clear and should be re-worded. How does this refer to single-leg landings, specifically. Line 103 – 'unimpaired individuals' is inaccurate as even controls could be impaired in some sense. Best to stick with 'control individuals' here. Lines 102-104 – the entire hypothesis is worded weird. Please re-write for improved clarity.

REVIEWER	Nyland, John Spalding University Louisville
REVIEW RETURNED	31-May-2018

GENERAL COMMENTS	Introduction, lines 58-59. I do not believe that this is the most frequent knee injury. Please confirm or revise. Introduction, lines 102-104. This sentence is confusing. Please rephrase for clarity. Methods, line 117. Confounders such as.....

	Methods, line 128. Why just men? Methods, line 131. What is meant by "plastic and surgical access"? Methods. line 134. What about MCL injury status or laxity? Methods, line 141. What about trunk or core region injury? Methods, line 147. What is the validated basis of 85%? Measurements, lines 199-200, What is the weight of the back pack? Measurements, lines 206. Why not just ask them to close their eyes at rest, or to wear a blindfold for standardization purposes? Measurements, line 240. Will footwear be standardized? What shoe type will be worn? Discussion, lines 51-52. Might participants have differing jump task performance histories? For example, Volleyball versus Bicyclists? Might this affect neurocognitive patterning? Discussion, lines 70-71. Will anterior knee laxity, pivot shift, and lower extremity strength/ROM be evaluated prior to study participation? References. Reference style is not consistent. Please revise to Journal standard.

VERSION 1 – AUTHOR RESPONSE

Reviewer: 1

Line 58-59: The statement "ACL tears represent the most frequent injury of the knee" is very strong. Do you have any recent references to support this? Or would it be better to change "the most frequent injury" with "one of the most common sports-related injuries to the knee"?

- We agree and have changed the wording accordingly (Line 59).

Line 128: Why were only males included? Female athletes are at greater risk than male athletes for noncontact ACL injury. For example, see: https://www.ncbi.nlm.nih.gov/pubmed/27315457 and https://www.ncbi.nlm.nih.gov/pubmed/15722283.

- We agree with the reviewer. We decided to include only participants of one sex in order to exclude influences due to this variable. We apply the jump landing experiment for the first time. Male participants were chosen because they were more likely to achieve the required jump height of about 30 cm corresponding to (480 – 520 ms). Future studies need to evaluate if an approach with an adapted jump height is feasible to investigate female participants. We refer to this limitation in the discussion (lines 361 to 364).

Line 146-147: Hop tests are frequently used to determine return to sports after ACL reconstruction. Given that bilateral deficits are present after ACL reconstruction, this may result in a falsely high limb symmetry index (LSI), since LSI is calculated as a ratio between the values of the limbs. See https://www.ncbi.nlm.nih.gov/pubmed/28428033.

- We agree with the reviewer as based on the LSI only, the level of neuromuscular restoration may be overestimated. Unfortunately, we have no access to the preinjury performance data. So, we cannot exclude that a potential decline of the affected side leads to a falsely high LSI. However, limb symmetry indices (LSI) are often used to evaluate athlete's neuromuscular performance during the return to play/sport process. LSI of ≥85% are recommended as a higher limb symmetry has been demonstrated to lower the risk of re-injury (Kyritsis 2016; Paterno 2010). We apply the single leg hop for distance, which is a valid and reliable performance-based outcome measure reflecting the combination of leg strength, neuromuscular control and self-confidence in the ACL reconstructed knee (Reid et al. 2007), and the ability to tolerate sports-specific loads, respectively (Logerstedt et al. 2012). We refer to this point under 2.4 (Line 148 to 149)

Line 175: Why did you choose a total of 70 successful jumps? Was it an arbitrary choice or did you use a protocol used previously?

- Thank you for raising these important questions. There is no work published yet, investigating the cortical activity during such a sport- and injury related movement task. Due to the novelty of this approach we could not refer to previous studies.
A typical challenge in the design of EEG studies (event or movement related cortical potentials) is to choose a sufficient number of trials to include in order to maximize statistical power on the one hand, and to minimize the duration of the recording session on the other hand. The general principle is "the more trials that are included in the average, the better the quality of the data" as the signal-to-noise ratio improves with an increased number of trials (Boudewyn et al. 2018; Luck et al. 2014). In the pioneering studies (i.e. Shibasaki et al. 1980) investigating the Bereitschaftspotential prior to voluntarily performed finger movements (sitting position) about 200-250 trials were included. However, these numbers cannot be transferred to our jump landing experiment as the participants would become fatigued, which may probably lead to an increased noise level in the data and to an impaired neuromuscular performance. Therefore, we performed a pilot. The results indicated that a number of 35 successful trials (which leads to 70 overall trials) per condition (5-minute breaks after each 12 trials) are sufficient in order to produce stable results (EEG outcomes) on the one hand, without evoking measurable exhaustion in any assessed parameter on the other hand. Furthermore, a literature research revealed that the number of trials chosen lies in the range of other studies (Verleger et al. 2016; Spring et al. 2016). Nann et al. 2018 found that the Bereitschaftspotential can be successfully detected by averaging EEG data from even less than 15 trials.
Referring to this, we have added a short note in the method section of the manuscript. Nevertheless, based on the calculated statistical power, we will discuss this topic in detail in the main manuscript (original article).
We refer to this issue in the method section of this manuscript (Line 158 to 161)

Line 270: Reference?
- Reference has been added.

Reviewer: 2

Methodologically, if unsuccessful trials are categorized as landing errors, will this bias the data? Not successfully landing could be due to an overbearance on the cortical processing and could be indicative of the injury mechanisms. Exclusion could lead the researchers away from the actual 'signal' they desire.

- This is a very interesting point. We totally agree with the reviewer that inaccuracies of the cortical processing of the visual information (required landing leg during unanticipated trials) occuring in a

standardized experimental setting may put an athlete at high risk of injury in the real sports environment.
In terms of the assessed behavioural data, I am afraid that we will not have sufficient error trials to investigate properly, if the cortical activities differ between successful and unsuccessful landings as previously, most participants perform quite accurately. Pilot testing indicated that a number of 35 successful trials (which leads to 70 overall trials) per condition are sufficient in order to produce stable results (EEG outcomes) on the one hand without evoking measurable e(5-minute breaks after each 10 trials) xhaustion in any assessed parameter on the other hand. In order to produce a sufficient number of unsuccessful landings (about n = 35 as well) for analysis, the total number of trials would need to be considerable increased leading to a higher risk of fatigue effects which would negatively affect data quality. Alternatively, the measurements would be need to spread on several days which would increase the effort for the participants significantly. We apply the jump landing experiment for the first time. Therefore, we are aiming to focus on the successful trials first. If this novel approach described in this protocol will be proved feasible, we will adapt our jump landing paradigm to enable an analysis of unsuccessful trials in future studies. Nevertheless, we refer to this point in the discussion section of the manuscript (Line 362 to 369)

As cognitive function was not directly addressed in the manuscript, Table 1 is not necessary for inclusion in this manuscript unless it is further discussed as to how these variables will be utilized to parse the data.

- As indicated in the discussion of the manuscript the included cognitive functions are discussed as relevant confounders in earlier studies (i.e. Swanik et al. 2007). Nevertheless, we agree that table 1 is not necessary. We have therefore removed it.

After Independent Component Analysis, it is mentioned that the trials with remaining artefacts will be rejected and only artefact-free trials will be used for analysis. How do the authors successfully determine what is artefact and what is not? Interpretation may describe it as artefact when it is actually signal.

- According to other studies, besides ICA, artifact removal will be applied by removing segments with an absolute difference larger than 200 µV or a voltage step per sampling point larger than 50 µV by using automated artefact rejection (Saliasi et al. 2013). Additionally, semiautomatic artefact rejection methods will be applied afterwards to exclude trials with remaining artefacts. We have added these specific information in the corresponding part of the manuscript (Line 288 to 291)

Line 77 – ‘The authors’ is not clear that it is reference to the previous study as it may be considered that it is the authors of the current study. This should be clarified.
- We have clarified this (Line 78).

Line 93 – This statement is not clear and should be re-worded. How does this refer to single-leg landings, specifically.

- This refers to single leg landings insofar, that single leg landings have been demonstrated to represent one of the major causes for non-contact ACL injuries (Boden et al. 2000, Krupenevich et al. 2017) and most work investigating the effects of the anticipation status on injury risk using movement tasks including single leg landings and cuttings (Almonroeder et al. 2015). We have reworded this part in the introduction of the manuscript (Line 94 to 97)

Line 103 – ‘unimpaired individuals’ is inaccurate as even controls could be impaired in some sense. Best to stick with ‘control individuals’ here.
- Changed as requested (see line 104).

Lines 102-104 – the entire hypothesis is worded weird. Please re-write for improved clarity.
- The hypothesis has been rewritten (see Line 103 to 106)

Reviewer: 3

Methods, Line 117. Confounders such as.....
- We have made a reference to the corresponding subchapter (Line 119)

Methods, Line 131. What is meant by "plastic and surgical access"?
- We have revised this accordingly (Line 132).

Methods. Line 134. What about MCL injury status or laxity?
- This comment is highly comprehensible as MCL injury is a very frequent concomitant injury. We are documenting all concomitant injuries from the doctor's letter, including MCL injury. To maintain sample size we did not consider to exclude those participants. Nevertheless, individuals with a history of serious concomitant injury, such as unhappy triad (incl. MCL injury) will be not included in the study as indicated in the method section of the manuscript. If possible we will conduct a subgroup analysis to compare those with vs. without (isolated ACL injury) concomitant injuries. Apart from that, we will definitely discuss this topic in detail in the main manuscript (original article).

Methods, Line 141. What about trunk or core region injury?
- We have clarified this (Line 138, 142)

Discussion, Lines 70-71. Will anterior knee laxity, pivot shift, and lower extremity strength/ROM be evaluated prior to study participation?

- Only participants who are cleared for return to sports by their treated physician (including check-ups of restored daily function, i.e. knee ROM) will be considered for inclusion. Additionally, we assessed the self-reported daily knee function (see 2.6.3). We refer to this point in chapter 2.3 (Line 132).

Measurements, Lines 199-200. What is the weight of the back pack?
- We have added this information accordingly (Line 206 to 208)

Measurements, Lines 206. Why not just ask them to close their eyes at rest, or to wear a blindfold for standardization purposes?

- We tested EEG at rest with both eyes open and eyes closed. We found that eyes closed conditions produced constant eye-movement artifacts. Surprisingly, the signal was much cleaner with eyes open (incl. fixation cross).

Measurements, Line 240. Will footwear be standardized? What shoe type will be worn?

- This is a good point. To avoid external distraction and the potential of discomfort by providing standardized footwear we ask the participants to bring along their own shoes. To ensure comparability only shoes used for indoor sports are permitted. During the jump landing tasks all participants are required to wear sports clothes (t-shirt and shorts). We have added this accordingly in the manuscript (Line 190 to 191).

Discussion, Lines 51-52. Might participants have differing jump task performance histories? For example, Volleyball versus Bicyclists? Might this affect neurocognitive patterning?

- Yes, that is possible. Open skill sports are defined as those in which players are required to react in a dynamically changing, unpredictable and externally-paced environment (i.e. team game sports, such as football; Wang et al. 2013). Therefore participants engaging in team game sports may be rather used to the unanticipated jump landing tasks compared to endurance athletes (closes skill sports) usually moving in a rather consistent, predictable and self-paced environment. To meet these potential influences, we ask all participants about their current and former physical/sports activities (including: primary sport, amount, expertise level, and years of experience). If possible, we will consider this by conducting a subgroup analysis. Irrespectively of this, we will match both groups (cases vs. controls) based on age, neurocognitive performance, sports (open and closed skill sports), and jump perfomance (counter movement jumps). We have added the assessment of physical and sportive activities as potential confounders (2.6.3) as well as our intended matching criteria, which can be found in the statistical section of the manuscript (see Line 274 to 275 and 322 to 324)

Formatting

Reference style is not consistent. Please revise to Journal standard.
- We have changed the reference style to the Journal standard accordingly.

Please provide another copy of your figures with better qualities and please ensure that Figures are of better quality or not pixelated when zoom in and make sure that they have a resolution of at least 300 dpi.
- The resolution of the figures have been adapted to 300 dpi.

You have cited Figures 3 and 4 rights after Figure 1 which makes your citations incorrect. Please review again your main document and ensure that all figures will be cited and will appear in numerical order.
- We agree and have changed the order of the figures accordingly.