**What happens to the brain when we don’t sleep?**

Everyone has experienced it at some point, staying up all night to finish an assignment, play video games or just not being able to sleep. The consequences of sleep deprivation are easily felt, reduced attention span, lack of energy, irritability, etc. How do such changes to cognition and mental states represent themselves, in the brain and are there ways to mitigate some of these changes?  
In this essay I will focus on one neurophysiological change that happens as sleep deprivation starts to mount, namely the lack of adenosine triphosphate and the build-up of adenosine in the brain.

Adenosine triphosphate (ATP) is the energy currency of the human body and most living systems. This molecule is built such that the three phosphate groups in the molecule can be released one by one and in the process release energy that can be utilized. The adenosine molecule acts as a carrier of the phosphate molecules, and these can be utilized to produce energy in either muscle cells for contraction or to neurons and cause ion channels to transport ions across the membrane potential hereby either polarizing or hyperpolarizing the neuron (Owen & Sunram-Lea, 2011). When phosphate molecules are utilized for energy in the cells from ATP the remaining molecule will, with help from the mitochondria, be made back into ATP, hereby restoring the homeostatic balance of energy in the overall system.   
The simplistic view of adenosine and its phosphatic relatives as the energy currency in the brain and body, with ATP being the most valuably and adenosine being the least valuable has proven remarkably useful for understanding how these molecules influence physiology and into the investigation of why we sleep.  
One tenable hypothesis is that as we are awake, we use up the phosphate groups in ATP to produce the energy needed for daily activity, and when we go to sleep, we resynthesize this ATP (Lazarus et al., 2019, Bjorness & Greene, 2009). A key assumption here is that the mitochondria cannot synthesize as much ATP from its phosphate relatives as is being used up when being awake and therefore needs a period (sleep) to resynthesize the ATP from adenosine (Chikahisa & Séi, 2011).

One of the effects of adenosine in the brain is that it works as an inhibitory neuromodulator. Meaning that increases in adenosine concentrations in the brain leads to decreased neural excitability especially in acetylcholine and norepinephrine diffuse modulatory systems, which appear to promote wakefulness (Dulla et al., 2005, bear et al p. 672-673). Several other converging lines of evidence support this notion that increased adenosine concentration is involved in the sleep-wake cycle (Basheer et al., 2008). In this essay I will be focusing on neuropharmacological studies and especially on the molecules creatine and caffeine. These will be the focus as they are easily supplemented in humans and have been shown to cross the blood brain barrier which is a necessity for the molecules to have a direct effect in the brain (McCall et al., 1982, Ohtsuki et al., 2002, Christie, 2007).

Caffeine is one of the most studied stimulants in the world, and its effects range from increased alertness, to decrease reaction time (McLellan et al., 2016). Caffeine’s effect is thought to be mediated by being an adenosine antagonist, meaning it binds to adenosine receptors and inhibits the decreased neural excitability caused by the accumulation of adenosine (McLellan et al., 2016). Some of the most convincing mechanistic animal evidence of this antagonistic effect of caffeine comes from knock-out mice where one of the 4 known adenosine receptors have been inactivated. The results from these studies suggest a dose-dependent relationship between the percent of knocked out receptors and the wakefulness effects of induced caffeine (Lazarus et al., 2011). It could therefore be that antagonists to adenosine receptors would reduce feelings of sleepiness and reverse some of the negative cognitive effects seen in prolonged sleep deprivation. This hypothesis would therefore be able to explain the increased alertness and reaction time findings mentioned earlier and supports the notion that the effects are more notable when individuals are sleep deprived (Souissi et al., 2014).

Creatine does not directly influence adenosine receptors as caffeine does, however creatine is thought to work as a buffer of phosphate, the energy storing component of ATP, essentially working as an energy reserve. Creatine is thought to help maintain a homeostatic ATP level by binding to a phosphate group and thereby becoming creatine phosphate which enters the cells and can donate the phosphate group to adenosine diphosphate or adenosine monophosphate.

Creatine can also cross the blood brain barrier and it has been hypothesized that it therefore might influence cognition and especially when lots of energy has been consumed, as with prolonged sleep deprivation. Magnetic resonance spectroscopy has been used to determine that oral supplementation of creatine can increase brain phosphor creatine levels as well as inorganic creatine levels in the brain. (Lyoo et al., 2003)

One of the most convincing lines of evidence indicating that creatine supplementation has cognitive implications is studies on patients with cerebral creatine deficiency syndrome, making them unable to produce and synthesize creatine. Symptoms of these conditions include autism like behaviors and intellectual disability of variable severity. These symptoms are treated with creatine supplementation, which strongly improves many of the symptoms of the patients (Stockler et al., 2007).  
Other studies have shown that creatine supplementation in healthy individuals can reduce the negative effects on executive function from sleep deprivation in healthy individuals (McMorris et al., 2007, McMorris et al., 2006), and that these effects are more pronounced in vegetarians and vegans that do not get creatine from their diets hereby further decreasing their creatine availability (Cooper et al., 2012).

Creatine and caffeine are widely studied supplements and the side effects of them are therefore also well established. Caffeine for one seems to have very few adverse effects, when consumed in normal ranges up till 500mg aside from a little shakiness, if the consumer is not used to it (Wikoff et al., 2017). However, it is to be noted that caffeine consumption at later stages of the day is normally advised against, to not disturb sleep. Whether this is true, is still an ongoing debate in the scientific community (Weibel et al., 2021, Kerpershoek et al., 2018).   
Creatine has a few reported side effects for a minority of subjects, including gastrointestinal disturbances, however these adverse effects are rare and not harmful (Poortmans & Francaux, 2000).

One of the reasons that we sleep seems to be to replenish the molecules that the body uses for energy. So, when we do not sleep the remains built up and interfere with normal functioning. There are several ways to combat this built up of remains, some temporary, caffeine supplementation and some more long lastly, creatine supplementation.

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**The Effects of Concreteness and Valence on Word Processing - an fMRI Study**

**Disclaimer**

The two papers below “The Effects of Concreteness and Valence on Word Processing - an fMRI Study” and “The Effects of Concreteness and Valence on Word Processing - an MEG Study” are written by the members of study group 2. These include Julie (201904671), Gaba (202202799), Jiaqi(202202798), Jesper(201910213), and Liv (201905665). The names of the authors of the various sections are noted with names throughout both papers.

**Abstract (Together)**

How meaning is extracted from words and represented in the brain remains one of the conundrums of cognitive neuroscience. Theoretical frameworks such as the dual code theory and the context availability theory provide possible explanations for how the issue might be understood, focusing on the separation between processing of abstract and concrete words. The study at hand applies functional magnetic resonance imaging (fMRI) to investigate the neural underpinnings involved in processing of concrete and abstract words. Also, the interplay between words’ concreteness and valence is investigated. This was done utilising an experimental paradigm where participants saw a word that was predictive of an upcoming face. Participants (n = 22) were then instructed to correctly identify if the face was positive or negative. Mixed effects modelling of the participants’ response times showed that participants learned the experimental paradigm, which was reflected in shorter response times for valenced words than neutral ones. Using a second level fMRI analysis, a main effect of the contrast between concrete and abstract words revealed that voxels in the left superior temporal gyrus were more active when viewing abstract words compared to concrete.  The results of this study are in line with theories suggesting a left lateralisation of processing of abstract words compared to that of concrete words. No significant evidence for increased activity in the precuneus for processing of concrete words was found. However, higher activity in precuneus was found in valenced words compared to neutral words suggesting a link between emotional valence and this area. Discussion of the results and limitations of the study are presented.

**Introduction (Jesper)**

One of the greater questions of the brain concerns how words are meaningfully represented in the mind. One of the ways words can be classified is the distinction between abstract or concrete. Here, concrete refers to words describing actual things, which are faster to respond to compared to abstract words that are more conceptual (Kumar, 2012). This is also known as the concreteness effect (Yui et al., 2017)

These observations have been explained in Paivio’s (1991) theory called dual code theory where he proposes two systems associated with the processing of word meaning. One is composed of a word-based verbal system while the other one is composed of an image-based, nonverbal system. The systems are presumably interconnected, but they also function independently (Paivio & Lambert, 1981). The theory proposes that verbal and nonverbal representations are retrieved differently in memory. Thus, concrete words such as “thundering waterfall”, which  are encoded in a verbal and nonverbal format, would be faster to recall compared to abstract words such as “optimal interface”, which have a lower nonverbal representation (Sadoski et al., 1995).

Another approach to explain the concreteness effect is proposed by Kieras (1978) in his context availability model. This approach does not separate verbal and nonverbal codes, but assumes that language, no matter if it is concrete or abstract, is understood and remembered by integrating it into the person’s propositional network based on prior knowledge. Thus, the model proposes that all information in memory is encoded in an abstract, single, amodal representation system. This means that concrete language is easier to understand and remember simply because it is faster at being linked to prior knowledge integrated in the propositional network. The context availability model was also supported by the work of Schwanenflugel & Shoben (1983) who based the speed of processing abstract and concrete sentences on comprehension. Here, concrete words are processed faster than abstract words as they activate higher associative information. This is further supported by the findings of Schwanenflugel & Stowe (1989), which showed that abstract words were processed just as fast as concrete words when put in a meaningful context that increased association strength.

**Neural activation and the concreteness effect (Julie)**

The dual code theory suggests that concrete words activate both the verbal and the nonverbal system as they are related to mental imagery. As abstract words are suggested to be processed solely in the verbal system, it could be argued that the words would elicit higher activation in areas related to language processing as it relies more on linguistic features.

In the context availability theory, abstract words take longer time to process than concrete words because there are higher associations in concrete words. Thus, this theory suggests that the language area processing both types of words will be more active when processing abstract words. Therefore, both theories argue that the language system would elicit a higher activity when processing abstract words, but the dual code theory also suggests that concrete words would elicit activation in areas related to mental imagery. In a meta-analysis from Wang et al (2010), 19 neuroimaging studies of participants processing abstract words compared to concrete words were assessed and showed higher activity in areas related to language processing such as left middle temporal gyrus, left inferior frontal gyrus, and left superior temporal gyrus(LSTG) (Hoffman et al., 2015). Furthermore, results showed higher activity in the left parahippocampal gyrus, the left precuneus, and the left posterior cingulate when participants processed concrete words compared to abstract words. Earlier work has shown that these areas have been related to mental imagery in fMRI studies (Kosslyn et al., 2001).

**Emotional valence, the concreteness effect, and neural activation (Liv)**

As with the division into concreteness and abstractness, words can be classified according to their emotional significance. Several studies have identified effects of emotional valence on processing, such as facilitated processing of emotionally valenced words when compared to neutral words [(as referred to in Kauschke et al., 2019)](https://www.zotero.org/google-docs/?UcBpC1). For example [Zeelenberg et al. (2006)](https://www.zotero.org/google-docs/?cTkjOf) used a binary forced-choice perceptual identification task and found that emotionally valenced words were better identified than neutral ones. Also, differential neural processing of valenced and neutral words have been found, with greater activation in areas in the left orbitofrontal gyrus and bilateral inferior frontal gyrus for emotional words than for neutral words [(Kuchinke et al., 2005)](https://www.zotero.org/google-docs/?U3OjjY). These are brain regions associated with semantic and emotional information processing. Also, different emotion related effects have been found for event related potentials (ERPs) [(Palazova et al., 2013)](https://www.zotero.org/google-docs/?mGSSPW). These include the early posterior negativity (EPN), and the late positive complex (LPC). As reported in the paper by Palazova et al. (2013), the EPN consists of an increased occipito-temporal negativity to emotional as compared to neutral stimuli. The LPC consists of increased centro-parietal positivity to emotionally valenced stimuli, and has been set in relation to more elaborate processing of emotional words. Thus, there seems to be strong indications of differential processing of emotional and neutral words. However, this matter is not as simple as it may seem. Both positivity and negativity biases in word processing have been identified, each suggesting a processing advantage over the other [(Kauschke et al., 2019)](https://www.zotero.org/google-docs/?FkPrJY). Positivity advantages include faster processing of positive information, broader associations connected to positive stimuli and stronger congruency effects [(Unkelbach et al., 2020)](https://www.zotero.org/google-docs/?nUvErW). Negativity advantages include enhanced attention, higher recall, and larger placed emphasis on negative stimuli. These discrepancies are usually referred to in the literature as valence asymmetries. Various attempts of explaining these differences exist, with some pointing to a possibility that there are separate systems for processing positive and negative information. It is, however, difficult to pinpoint whether the asymmetries are due to differences in brain areas involved or due to inherent differences in the properties of positive and negative words. Additionally, relating to the previous paragraph concerning the concreteness effect, findings suggest that emotional valence as reflected in the EPN is anchored in words’ meaning [(Palazova et al., 2013)](https://www.zotero.org/google-docs/?3bPBpA). It seems that emotional meaning in concrete words is accessed earlier on than in abstract words. As such, when investigating neural underpinnings of the concreteness effects, it is relevant to control for the impact of emotional valence. Several of the above mentioned effects could not have been identified without sophisticated research tools. One of these tools is functional resonance imaging, which will be described in the section below.

**fMRI as a method (Liv)**

Since its emergence over 30 years ago, functional magnetic resonance imaging (fMRI) has become one of the most popular tools for investigating the human brain. By making use of the link between hemodynamics in the brain and neuronal activity for localisation and measurement of brain activity, fMRI revolutionised cognitive neuroscience [(Heeger & Ress, 2002)](https://www.zotero.org/google-docs/?cEY90b). With the method, researchers were suddenly able to produce dynamic brain activation maps in a fast, non-invasive way, with relatively high resolution [(Bandettini, 2012)](https://www.zotero.org/google-docs/?xYDq6H). This stood in contrast to magnetic resonance imaging (MRI) which solely provided researchers with anatomic and basic physiologic information. fMRI opened the door for investigating topics such as human memory, reward circuitry, and brain plasticity [(Rosen & Savoy, 2012)](https://www.zotero.org/google-docs/?3dBlar).

fMRI combines structural and functional imaging of the brain. It is based on the same technology as MRI, which uses information about how hydrogen atoms in the brain respond to perturbations of a strong magnetic field to produce detailed structural images of the brain [(Bear et al., 2016)](https://www.zotero.org/google-docs/?eO0qDg). When exposed to a magnetic field, the nuclei of hydrogen atoms (protons) can exist in either a high- or a low-energy state. The basis of MRI imaging is built on forcing protons to alter between these two energy states. This is done by adding energy to protons in the form of an electromagnetic wave sent through a person’s head positioned between the poles of a large magnet. Low-energy protons absorb energy from the electromagnetic wave, and are transferred to a high-energy state. When the magnetic field is turned off, some of the protons return to the low-energy state, thereby emitting a radio signal at a particular frequency. This signal can subsequently be detected by a radio receiver. A stronger signal is a sign of higher hydrogen atom concentration in a certain location. The reason why these principles allow for distinction between tissue types is that, for one, hydrogen density varies across different regions of the brain. Also, the time it takes for protons to realign to the magnetic field after being pushed to a high-energy state varies with tissue type [(Sprawls Jr., 2000)](https://www.zotero.org/google-docs/?M4ac0D).

When it comes to the functional aspect of fMRI, the method works by taking advantage of the fact that areas of increased neural activation in the brain require increased supplies of oxygen, and on the differences in magnetic resonance between oxygenated and deoxygenated haemoglobin [(Bear et al., 2016)](https://www.zotero.org/google-docs/?V5tCYS). More specifically, oxygen is provided to active brain regions through increased blood flow and deoxygenation of oxyhaemoglobin. Oxyhaemoglobin is diamagnetic, and deoxyhaemoglobin is paramagnetic. As such, the MR signal of the blood in the brain varies according to level of oxygenation, and it is possible to distinguish more active areas from less active ones by measuring the ratio of oxygenated to deoxygenated haemoglobin in the brain. This makes fMRI a form of blood oxygen level dependent (BOLD) imaging, which is the method we will use to investigate our hypotheses.

***Hypotheses (Together)***

As described in the introduction of this paper, the study at hand applies fMRI to investigate the neural underpinnings of the processing of concrete and abstract words. Based on the theories described above, we hypothesise the following:

*H1: We hypothesise that reaction times on the faces following a valenced word will be faster compared to a neutral word.*

*H2: We expect higher activity in the left superior temporal gyrus (LSTG) when participants view abstract words compared to concrete words.*

*H3. We expect higher activity in the precuneus when participants view concrete words compared to abstract words*

**Methods**

*Experiment* (Gaba)

The experiment had two conditions, the positive condition and the negative condition, and each session had 60 trials total. A fixation cross was shown at the top of the screen to signal the beginning of a trial. A word was displayed after the fixation cross, then another fixation cross, and finally a face that was either positive or negative. The experiment required participants to press one of two buttons indicating which face they saw. In total, all participants stayed in the scanner for approximately an hour as they had to run through 6 sessions each.

The words for this experiment were all taken from the created by Binder et al. (2016) corpus. In this copus each word was graded according to several criteria (e.g. colour, large, small). A sentiment score was derived using a principal component analysis and words were categorised into three categories, neutral, negative or positive. After having categorised the words that preceded the face the experiment was set up in such a way that a negative face could only be followed by a negative or neutral word, and a positive face could only be followed by a neutral or positive word. Emotionally valenced words seemed to be generally more abstract compared to concrete, which made us binarize the neutral, positive and negative words into two categories of either neutral or valence where valence refers to both positive and negative (Kousta et al., 2011).

*Participants (Gaba)*

Data were collected from 22 participants (17 females) aged 20-29. Participants were introduced to the process of the study and written consent was obtained for all of them. The participants were also informed about the option of leaving the trial at any time. The participants were prepared for the study under the supervision of the students and their supervisor and then placed in a magnet. All observers had normal or corrected-to-normal vision. The experiment was performed using Siemens 3T Magnetom Prisma fit MR scanner which were set to TR value at 1.8. The study tookplace in the Center of Functionally Integrative Neuroscience at Aarhus University Hospital.

Categorization of words (Gaba)

As we hypothesised activity differences between abstract and concrete words, the words shown to the participants were also categorised into a concrete or abstract category. This categorisation was done with the package “doc2concrete” (M. Yeomans, 2022) in Rstudio (*RStudio Team, 2022*), which gave each word a concreteness score from 0 to 5. To make the analysis easier these words were then categorised into either concrete or abstract based on a median split to ensure equal number of words in either condition.

**Analysis**

*Behavioural analysis (Julie)*

First we investigated the distribution of words in our defined binarization of concrete vs abstract in the neutral vs valenced categorization. This was done in order to determine if a confound in the valence of the words were present. This analysis showed a clear difference between how many words were labelled valenced in the concreteness binarization with many more words being concrete and neutral compared to abstract and concrete and vice versa (see Table 1).

|  |  |  |
| --- | --- | --- |
|  | **Concrete** | **Abstract** |
| Neutral | 1736 | 920 |
| Valenced | 2223 | 3076 |

*Table 1: Number of words in the defined categorisations of concrete and valence for all subjects for all runs.*

In order to investigate the participants’ response times to the different faces, we fitted a mixed effects model. The model predicted response time as a function of whether the word was concrete or abstract and whether the word was neutral or valenced. We included a random intercept for each participant. The model utilised a Gamma function with a log-link function to account for the fact that reaction times are notoriously known to be non-normally distributed and non-negative (Boeck & Jeon, 2019). See appendix A for assumption checks for both the Gamma and Guassian model.

*Preprocessing of fMRI data (Jesper)*

All fMRI data was stored in Brain Imaging Data Structure (BIDS) format and could therefore be preprocessed using the fMRIPrep pipeline (Esteban et al., 2018). See appendix B for the full boilerplate description of the preprocessing of the anatomical and functional images of the fMRI. We used fristons 24 motion regressions as well as the global signals to account for motion.

*fMRI analysis (Jesper)*

To investigate the hypotheses, we used the Nilearn module in Python to fit first and second level models on our data, which was in the BIDS dataset structure. This includes fitting the General Linear Model (GLM) to the data. More specifically, first level models can be understood as linear regression models that are run on a single subject or for a single session [(*5.2. First Level Models, Nilearn*)](https://www.zotero.org/google-docs/?7uNT8H). The model can be applied on the entire brain, or simply on a region of interest, and works on a voxel at the time.  A second level analysis is performed on all participants, making it a group level analysis. This is done in order to make more general inferences about the observed fMRI activity[*(5.3. Second Level Models, Nilearn.)*](https://www.zotero.org/google-docs/?FeWu67). Extra smoothing of full width a half a maximum of 8 mm was added for this second level analysis. The main analysis investigated the constrast in activation between concrete and abstract words. A follow-up control analysis was also performed on the contrast between neutral and valenced words.

Following the first and second level analyses, a searchlight analysis in the left hemisphere of a single participant was performed. The intention of this analysis was to assess whether we could classify whether the participant had observed a concrete or an abstract word.

Searchlight analysis is a method used for identifying locally informative areas in the brain (Etzel et al., 2013). It is a Multi-Voxel Pattern analysis (MVPA), which is also known as a classification analysis. The method works by creating maps of the brain through measuring information in globular subsets of voxels in the brain. These globular subsets are also referred to as searchlights, which illustrates the reasoning behind the name of the method. The final map value extracted from each voxel thus represents information available in the area of the searchlight, not the individual voxels. The activity of group voxels are expected to vary with experimental conditions, which is why the method is suitable for classification analyses. In practice, very informative voxel clusters are more suitable for identifying an experimental condition than a less informative one. The subject of our searchlight analysis was the one that looked the most promising on the basis of the first level analysis (e.g. subject 80.) For the sake of searchlight efficiency, a mask only covering the left part of the brain was conducted (see figure 6.) Lastly, a permutation test to examine the classification results was conducted.

**Results**

*Behavioural results (Jiaqi)*

First we investigated the distribution of words which can be seen in table 1. A chi-squared test showed that the distribution of words on the neutral-valence scale was not independent from the concrete-abstract scale x^2 = 387 df = 1, p < .0001. Afterwards we investigated the participants’ response times using a mixed effects gamma regression.

The model showed a main effect of concreteness X2 (df = 1) =18, p < .001, and a main effect of valence X2 (df = 2) = 250, p < .001. Pairwise comparison showed a difference between concrete and abstract words β = 0.02 se = 0.005 z = 4.3  p < .001. A significant difference was also found in response time between valenced and neutral words β = -0.08 se = 0.005 z = -16  p< 0.001 indicating that people were generally 9.5% faster when the word was valenced compared to neutral.

Chart

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*Figure 2; displays the raw response times in the fMRI experiment for n = 22 subjects stratified by valence and concreteness of the word. The response time was to the image presented after the word and measured the time to determine whether the face was positive or negative.*

*fMRI results (Jiaqi)*

First-level models were fitted to all 22 participants, and contrast between concrete vs abstract words were calculated. The results of the first level analysis are displayed in *Figure 3*.

A picture containing text

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*Figure 3: Shows the results of the first level analysis on the participants (n = 22) included in the current study for the contrast concrete-abstract words. Results are depicted at p<.001 uncorrected for multiple comparisons.*

Following the first level analyses on each subject, a second level group analysis was performed by fitting a second level model on the fitted first level models. This second level analysis revealed 3 clusters that were significant after correcting for multiple comparisons (Bonferroni correction)(*Table 2*). A visualisation of the second level results can be found in *Figure 4.*

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| Cluster id | Peak\_x | Peak-y | Peak-z | Peak-value | Volume mm | Harvard oxford atlas |
| 1 | -51.03 | -11.30 | -12.50 | -5.50 | 95.70 | Left\_superior\_temporal\_gyrus\_posterior |
| 2 | -48.51 | -13.80 | -9.50 | -5.00 | 19.10 | Left\_superior\_temporal\_gyrus\_posterior |
| 3 | -51.03 | 9.00 | -18.50 | -5.10 | 19.10 | Left\_temporal\_pole |

*Table 2: Shows the identified clusters that reached statistical significance after correcting for multiple comparison (bonferroni) at p < .05 for the contrast concrete vs abstract words.*

Diagram

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*Figure 4: Results from the second level group analysis, showing the contrast between concrete and abstract words uncorrected for multiple comparison for visualisation purposes.*

A control analysis was performed, because of the interaction between concrete and abstract words and valenced words. This analysis investigated the activity difference between neutral and valenced words. The results can be found in *Table 3.* A visualisation of the results can be found in *Figure 5.*

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| Cluster id | Peak\_x | Peak-y | Peak-z | Peak-value | Volume mm | Harvard oxford atlas |
| 1 | 9.60 | -49.10 | 44.50 | 5.20 | 76.60 | Right\_ precuneus |
| 2 | 29.80 | 14.00 | 50.50 | 5.10 | 38.30 | Right\_middle\_frontal\_Gyrus |
| 3 | 39.90 | -54.20 | -24.50 | -5.30 | 19.10 | Right\_temporal\_occipital\_fusiform |
| 4 | -38.40 | -18.80 | 65.50 | -5.10 | 19.10 | Left\_precentral\_gyrus |

*Table 3: shows the identified clusters that reached statistical significance after correcting for multiple comparison (Bonferroni) at p < .05 for the contrast neutral vs valenced (positive or negative) words.*

Diagram

Description automatically generated

*Figure 5: Results from the second level group control analysis, showing the contrast between neutral and valenced (positive or negative) words. Results are depicted uncorrected for multiple comparisons for visualisation purposes.*

The results of the general searchlight analysis can be seen in *Figure 6. Figure 7* represents the searchlight outcome for the visualisation of the 500 most predictive voxels.  No significant  prediction accuracies of whether the subject was seeing a concrete or an abstract word were found in either of the two analyses.

Shape

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Diagram

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*Figure 6: shows the outcome from the Searchlight analysis, showing the contrast between concrete and abstract words, unthresholded (top), with thresholded for an accuracy of  > 60% (bottom)*

*A picture containing linedrawing, porcelain

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*Figure 7: shows the visualisation of the 500 most predictive voxels plots with glass brain effects (top) and with anatomical background (bottom). Searchlight scores size = 8980290, percentile that makes the cutoff for the 100 best voxels = 99.99, cutoff = 0.60.*

Chart, histogram

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*Figure 8: Results from permutation test. Mean prediction score = 0.48, p-value = 0.57.*

**Discussion (Julie)**

The present study investigated the effect of concreteness and valence of words in the face-word task. In our first hypothesis we predicted that the response time to the faces after a valenced word would be faster compared to a neutral word. This hypothesis was confirmed as participants were much faster at responding to the faces after a valenced word, indicating that participants learned the experimental paradigm as valenced words were always associated with the face that matched the valence (e.g. positive word positive face). Furthermore we found that even after controlling for the valence of the word participants were still faster at responding to the faces after an abstract word compared to concrete words. This effect can be explained by the fact that participants didn’t know what category each word was in. Therefore if participants received a neutral abstract word then they had a higher probability of thinking it as a valenced word and therefore responded faster because of the inherent association between the valenced words and images see table 1 and Kousta et al (2011).

In our second hypothesis, we predicted that there would be higher activity in the LSTG when participants viewed abstract words compared to concrete words, as abstract words are suggested to be processed more in areas related to the verbal system. Based on the results from Table 2, there was a significant effect in the LSTG when participants viewed abstract words compared to concrete words. This also aligns with expectations from dual-code theory as well as context availability theory as they both assume higher activation for abstract words in this area. Meanwhile, there was no significant evidence showing higher activity in the precuneus when participants viewed concrete words compared to abstract words, which may lead to the failure of the third hypothesis, which predicted higher activity in the precuneus when participants looked at concrete words. According to dual code theory, the precuneus should be more active as this area is related to mental imagery (Kousta et al., 2011). However as our behavioural analysis showed an independent effect of concreteness based on reaction times, we ran a control fMRI analysis looking at how valence affected brain areas. We therefore conducted a new analysis looking at the contrast between “neutral” and “valence” (both negative and positive) words. This control analysis showed a different pattern of activity from the main analysis, indicating that the activity difference from the second level analysis cannot necessarily be explained away by the valence of the word. Table 3 shows that activation in the right precuneus was significantly more active when participants saw neutral words compared to valenced words. This is of particular interest as higher activation in this area seems to be more related to the emotional valence of a word and not its concreteness as suggested in the dual code theory.

A possible explanation could be that these two theories do not fully account for the representation and processing of the meaning behind words being concrete or abstract. According to Kousta et al. (2011), there seems to be a residual advantage for abstract words when both context availability and imageability have been accounted for. They argue that this difference can be explained by emotional valence as abstract words are statistically more emotionally valenced than concrete words. Thus, the activation in the precuneus is not related to the concreteness of a word, but may be more related to the valence of the word. Thus, previous literature not accounting for valence of the words may have interpreted the role of the precuneus differently. However, it must be noted that the stimuli of this study also included almost twice as many concrete neutral words compared to abstract neutral words (Table 1). Furthermore, there were also more valenced abstract words compared to valenced concrete words, which may have skewed the results.

Lastly our explorative searchlight analysis on the most promising subject did not reveal any results of particular interest. This might be caused by several factors, however one of the most obvious being that the analysis was only run on a single subject and that we restricted the searchlight to the left hemisphere.

*Limitations and directions for further studies (Liv)*

Some of the crucial limitations in the study at hand are linked to fMRI as a method, the chosen experimental procedure, and the analysis applied for investigating the hypothesis. Firstly, fMRI is accompanied with certain inherent issues. This has shown itself through a replication crisis in cognitive and clinical neuroscience. In 2015, replication rates in fMRI studies were as low as 39% [(Open Science Collaboration, 2015; Specht, 2020)](https://www.zotero.org/google-docs/?bd6LrP). Commonly identified issues with the method include the BOLD-signal itself and current fMRI-praxis. The BOLD-signal is an indirect measure of neural activity, at best. As such, one cannot be sure of the accuracy of the activity measured in this paper. Also, since fMRI is a physiological response, it is also easily influenced by other physiological occurrences such as variation in blood pressure, blood oxygenation and other factors that might influence the vascular system. This means that any observed variability in the BOLD signal does not necessarily represent variability in neuronal activity that elicited the response. Furthermore, indications that the metabolic demands of the brain vary throughout the day have been found [(Vaisvilaite et al., 2020)](https://www.zotero.org/google-docs/?ZUDSbw). This suggests the need for altering the way fMRI studies are conducted, for example by conducting experiments at the same time of day for all participants. This was for example, not accounted for in the current study, which might have led to unwanted between subject variability.

When it comes to the experimental procedure applied in this study, problems include its lengthiness, the unnatural setting in which it was conducted, and the fact that the experiment was not designed to investigate our hypotheses. Lengthiness is a general issue in fMRI studies, as the time is crucial to obtain the correct measurements. This is a problem in the sense that it puts the participants under great pressure. In our study, all participants were in the scanner for approximately an hour, often in the early hours of the day. Some participants found it very difficult to focus on the task for this long, and also with staying awake due to the very repetitive aspect of the experiment. These factors are highly likely to have affected their responses, and thus also the overall results of the analyses included in this paper. Also, the setup of the experiment does not really facilitate a naturalistic setting of investigating cognition. As such, making conclusions about the cognitive processes underlying the results in this paper is difficult. Lastly, the experiment applied in the study was not specifically designed to investigate the hypotheses in our study, meaning that it is difficult to state whether the patterns identified in our analysis actually portray effects of concrete and abstract word processing, or a different mechanism.

The last limitation concerns the searchlight analysis in this paper. This analysis was performed on a single subject in the interest of time. As such, the findings from this analysis cannot be generalised to other individuals. At best, they can function as indications of patterns that one might expect to see if the analysis was to be performed on a larger participant population. Secondly, only the left part of the brain for that single subject was covered in the analysis for the sake of increased searchlight efficiency. This is problematic, as performing the analysis on half the brain yet again reduces the generalisability and statistical power of the analysis. Future studies should focus on improving the limitations presented in this section in order to obtain more precise results.

**Conclusion (Together)**

In this paper, we investigated the differential effects of concreteness and valence on brain activation in a word-image recognition task. The behavioral analysis showed that participants generally responded faster following valenced words than neutral words. We argue that this is likely to reflect that participants learned the experimental paradigm, where the intention was to test whether participants were able to use emotional valence for predicting positive or negative faces.  Lastly participants were found to respond faster to abstract words compared to concrete words. This seems to be in opposition to the presented theory, but is likely to be connected to the experimental paradigm (i.e. participants responded to faces preceded by the words).. On the basis of the fMRI analyses, higher activation in the LSTG was found for processing of abstract words compared to concrete words, which is in concordance with both the dual code theory and context availability theory meaning that concrete words faster to process We also hypothesized to observe increased precuneus activation when participants viewed concrete words compared to abstract words, which we found no support for. However, our findings seem to suggest that precuneus activation may be connected to emotional valence of words rather than concreteness as suggested in the dual code theory. These findings should however be interpreted in the light of the limitations of this study. Overall, this study has contributed to literature supporting theories related to the concreteness effect as well as supporting the precuneus’s role in emotional valence. However, this study is accompanied with a set of limitations that affect the validity and generalizability of our results. Future studies should take these points into consideration when building on our findings.

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**The Effects of Concreteness and Valence on Word Processing - an MEG Study**

**Abstract (Together)**

Significant activity in the left superior temporal gyrus was found from the group level analysis in the fMRI experiment, which seemed to not be due to the effect of valence. The same experiment was therefore used in this follow-up study investigating classification of concrete and abstract words using magnetoencephalography. The current study utilised the results of the fMRI study and looked specifically at the superior temporal gyrus label from freesurfer to see if classification was feasible using two classification algorithms, logistic regression and support vector machines. It was found that neither of the classification algorithms could classify over chance level from stimulus onset to 800ms after stimulus onset. Limitations and further directions are discussed.

**Introduction (Julie)**

Based on previous findings from the fMRI section, it remains clear that concrete and abstract words are processed differently in the brain. Several other methods have been used to assess the processing of abstract and concrete words in relation to the two theories explained in our fMRI section. Some studies have used event-related brain potentials(ERPs) to determine at what point the concreteness effect emerges in the cognitive processes. A study conducted by West & Holcomb (2000) showed that imagery and semantic tasks elicited more negative ERP for concrete words compared to abstract words between 300 and 550 ms. In a study by Palmer et al. (2013), it was shown that newly learnt concrete words elicited a more negative N400 response compared to newly learnt abstract words, which support the dual-code theory as described in the fMRI section. Furthermore, the ERPs in the West & Holcomb study were also more negative for concrete words compared to abstract words between 550 and 800 ms. For the imagery task, the activation was more frontally distributed and more evident. This is related to an ERP component called N700, which is more sensitive to mental imagery. Thus, the results support the dual-code theory as mental imagery tasks contributed to a faster processing of concrete words compared to the abstract words. This is further supported by the findings from Huang et al. (2009) which suggested that potentials from concrete words between 250 and 500 ms were more negative over the front of the head compared to abstract words. As the time course differed, neural sources are suggested to be non-identical, which also favour the dual code theory.

*Duel-code and context availability theory* (liv)

These findings show that there is a difference between abstract and concrete words mainly based on the dual-code theory. However, some studies suggest that this difference is better supported by the context availability theory. Findings from an EEG study by Adorni & Proverbio (2012) showed that abstract words have a larger area of activation compared to concrete words in areas within the left hemisphere. Furthermore, Fahimi et al. (2018) has also reported stronger activation for abstract concepts in the right temporal cortex as they argue that the right hemisphere may play a role in interpreting and processing abstract language.

The above mentioned literature suggests that equipment such as MEG and EEG are effective when looking into the concreteness effect. Therefore, it could be interesting to see if a classification analysis with MEG data would be able to support our findings from the fMRI study. To understand what this method can add to our analysis, the section below seeks to explain the advantages of MEG.

**MEG as a method  (Jiaqi)**

The study at hand applied Magnetoencephalography (MEG) in order to investigate if it would be possible to classify if a participant saw a concrete or an abstract word from the MEG signal. MEG is a neuroimaging technique to investigate neuronal activity in the living human brain without invasion  (Hämäläinen, 1993). By measuring the magnetic fields created at the scalp by neuronal currents, it enables human brain electrophysiology to be directly imaged.(Boto, 2018). As the brain is processing information, small electrical currents flow in the neural system generating a weak magnetic field which can be measured by a superconducting quantum interference device (SQUID) magnetometer (Hämäläinen, 1993), by which means, MEG records the signal.

In contrast to localization techniques such as fMRI, MEG has a higher temporal resolution, which is better than 1ms (Hämäläinen, 1993). With this temporal resolution advantage, MEG is primarily applied in the measurement of time courses of activity. Applications of MEG include basic research into perceptual and cognitive brain processes, as well as in a clinical setting to find locations of abnormalities (Carlson, 2013). Furthermore, facilitated by machine-learning techniques for multidimensional signal classification, MEG also has been significantly applied in identifying early components of visual object categorization (Cichy, 2014) and in tracking the temporal organisation of spatial patterns of brain activity (King, 2014).

The MEG technology is advanced, however, when it comes to its application, one should also keep in mind its shortcomings. For instance, given the fact that MEG mainly measures activity from the fissures of the cortex, and only currents that have a component tangential to the surface of a spherically symmetric conductor produce a magnetic field outside, MEG frequently simplifies the interpretation of the data. (Hämäläinen, 1993). Another challenge, referred to as the inverse problem, is to determine the location of the activity within the brain, which requires advanced signal processing techniques that use the magnetic fields measured outside the head to estimate the location of the source (Müller & Kassubek, 2007). The spatial discrimination is, under favorable circumstances, 2-3 mm for sources in the cerebral cortex (Hämäläinen, 1993). In addition, the complexity and upkeep of MEG come at a significant financial and operational cost. The bright side is, there are increasing options for more affordable alternatives, which is good news for MEG's long-term viability and increased affordability as a research tool (Baillet, 2017).

As illustrated in the fMRI session, the fMRI method analyses the brain function based on the BOLD contrast and can provide excellent spatial sampling in the range of 1 mm (Kwong, 1992). Combining MEG and fMRI, therefore, can improve interpretation of functional neural organization (Müller & Kassubek, 2007). Taking use of the respective advantages of fMRI and MEG we discussed above and in the previous fMRI session, we combined those two methods together in our analysis aiming for optimal accuracy. Considering the localization of cortical function advantages of fMRI, we have applied it to detect which brain regions are more active while seeing concrete vs abstract words (see fMRI). We then utilised the temporal resolution advantages of MEG to examine the previous fMRI results.

**Methods** *(Gaba)*

In this study, we relied on the same paradigm as for the fMRI study. Please refer back to the methods in the fMRI study paper for details. A significant difference in the conduct of the experiment itself is the time taken, which was half as long for the MEG study and also, the number of participants changed to 6 (4 females) aged 21-27.  However, a different imaging modality also introduces other methods to prepare participants for the study, which we will describe below.

*Preparation of participants (Gaba)*

A number of electrodes were attached to the individual in preparation for the experiment. Six electrocardiogram (ECoG) electrodes were placed on the participant's face. The head's three cardinal points were measured using Polhemus FASTRAK.  To capture blinking one electrode were placed above and one below each eye and to measure vertical eye movement we attached one electrode on each temple. In addition to keeping track of head movement, four head position indicator (HPI) coils—one behind each ear and one on each side of the forehead—were fastened to the participants' heads. One electrode was placed on the left collarbone and the other on the right hip to assess heart rate. Right elbow and the right wrist were used as the locations for the reference and ground electrodes.

*Experiment setup (Gaba)*

Elektra Neuromag Triux MEG with 102 magnetometers and 204 planar gradiometers was used for the project. In order to get the participant's head as close to the helmet as feasible for better recordings, the participant was seated beneath the MEG detector with the seat lifted. The MEG recordings were performed in a space that was magnetically insulated. From a nearby room, the experiment stimuli were projected onto a screen. When the words and faces were displayed, a tiny white dot was also projected into the screen's bottom right corner, and the white dot was registered by a sensor to record stimulus onset time. This was done to ensure that the stimuli were time-locked with the MEG recordings. The study takes place in the Center of Functionally Integrative Neuroscience at Aarhus University Hospital.

*Preprocessing (Gaba)*

The raw sensor space data of the MEG was preprocessed using the mne package in python (Gramfort et al., 2013). Here, the upper pass band edge was set to 40Hz. Events were extracted from the event channel with a minimum duration of change being set to 2 ms. Hereafter epochs were extracted for abstract and concrete words from 200ms before stimulus onset to 500 ms after stimulus onset. The baseline correction applied in the current analysis was done by calculating the mean of the epoch in the interval between 200ms before stimulus onset to stimulus onset and subtracting this mean from the entire epoch of each channel and epoch individually.

*Behavioural analysis of MEG data (Liv)*

Figure 1; displays the raw response times in the MEG experiment for n = 6 subjects stratified by valence and concreteness of the word. The response time was to the image presented after the word and measured the time to determine whether the face was positive or negative.

As for the fMRI analysis we investigated the behavioural responses of the participants with the same model. Here we did again find that a gamma distribution with log link function fitted the reaction times the best (see appendix A).

*MEG analysis (Jesper)*

To see if we could predict if a participant saw an abstract or concrete word based on their activity of the MEG we ran a logistic regression classifier with a l2 penalty term and with an inverse of regularisation strength of 0.001, and a support vector machines algorithm. To further protect against overfitting our results, we cross validated our results for each subject 5-fold. We extracted a time-series for each participant from -200 ms before word-onset to 800 ms after stimulus onset. After running our classification algorithm through this time interval we extracted the proportion of rightly classified words and aggregated this for each participant. As we’ve seen from the literature and our fMRI results we expected there to be differences in activity in the left superior temporal lobe and we therefore limited our search for activity to this area with the freesurfer label “lh.superiortemporal.label”.

**Results**

*Behavioural results (Liv)*

The mixed effects Gamma model showed a main effect of valence X2 (df = 1) = 44, p < .001, but not for concreteness X2 (df = 1) = 0.05, p = .83. Post hoc comparisons showed that participants responded faster to valenced words compared to neutral words β = -0.80 se = 0.012 z = -6.60  p < 0.001 indicating that people were 8% faster at responding to the valenced words compared to the neutral words, with no difference between concrete and abstract words after correcting for valence.

Chart

Description automatically generated

Figure 1: This figure shows the participants’ response time of the different words. The words are divided into neutral or valenced words as well as concrete(blue) and abstract(red) words. The y-axis is the response time in milliseconds.

*MEG results (Jesper)*

The results of the classification analysis of the MEG signal can be seen in figure 2 and 3 where the percentage of correctly classified words are depicted on the y-axis and the time around stimulus onset 200ms before until 800 ms after onset is depicted for both algorithms.

Chart, line chart

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*Figure 2: Results of the classification analysis on the freesurfer label “lh.superiortemporal”, of the support vector machines algorithm. The red line depicts the mean % of correctly classified words for all participants at a given time point. The blue shaded area depicts the standard deviation of the % of correctly classified words for the subjects.*

Chart, line chart

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*Figure 3: Results of the classification analysis on the freesurfer label “lh.superiortemporal”, of the logistic regression algorithm. The red line depicts the mean % of correctly classified words for all participants at a given time point. The blue shaded area depicts the standard deviation of the % of correctly classified words for the subjects.*

**Discussion (Julie)**

The current study found, as with the fMRI study, that participants were faster at responding to faces after a valenced word compared to a neutral word. This means that the participants again learned the experimental paradigm. No main effect of concreteness was found which is counter to the findings of the fMRI study. There could be several reasons why the effect is not present in the current study, the first thing is that this task was twice as fast as the fMRI, meaning that participants had a shorter time to process the words. Therefore the confusion to neutral abstract words as alluded to in the fMRI discussion could be abolished. Another reason could be the low sample size in the current study, as only 6 participants completed this study and 22 participants went through the fMRI study.

As can be seen in figure 2 and figure 3, the results of the classification analysis of concrete and abstract words in the left hemisphere superior temporal gyrus seem to be no better than chance at every time point from 200ms before stimulus onset to 800 ms after stimulus onset at classifying whether the word was concrete or abstract. This was the case for both classification algorithms, indicating that the activity pattern from the left hemisphere superior temporal gyrus doesn’t seem to convey much information about whether the participant saw a concrete or abstract word.

*Relation to the fMRI results* (Jesper)

This result is counter to our second level analysis from the Functional magnetic resonance imaging results, as we three found significant voxel activity difference between concrete and abstract words. These null-results do not seem to be related to the classification algorithm as both the logistic regression and the support vector machines failed to get a good accuracy score. As alluded to in the introduction, other studies have found (Fahimi et al. 2018, Palmer et al. 2013) using event related potential paradigms a processing difference between concrete and abstract words, it could therefore be argued that the current study should have utilised this statistical approach instead of a classification approach. It should however be noted that as alluded to in the introduction that most of the literature using high temporal imaging strategies such as EEG or MEG do not agree on specific time-courses where a processing difference can be found (Fahimi et al. 2018, Palmer et al. 2013).

*Further direction* (Liv)

There could be several reasons why we couldn’t find a better than chance level accuracy score with any of the classification algorithms, including sample size and the experimental paradigm. The experimental paradigm was not ideally suited to find a difference between concrete and abstract words. Further investigations into classification of concrete and abstract words using MEG, should make an experimental paradigm that makes sure that the participants are highly focused and perhaps also have to remember the word seen, making them pay more attention to the word and its meaning.

*Limitations* (Jiaqi)

There are certain limitations in the current study. Firstly, as it’s stated in the fMRI session, there are 22 participants in the fMRI study. However, the available data for MEG analysis only contains 6 participants. Considering that significant body movement in the MEG scanner can cause abnormality in the data, we can not rule out the possibility that the study didn’t cover enough applicable data. Thus, the inadequacy of experimental subjects may weaken the prediction accuracy.

Secondly, the prediction for seeing concrete vs abstract words are based on the analysis results from the fMRI participants who are different from those in MEG. Even though the second level fMRI analysis revealed significant activation contrast seeing concrete vs abstract words in the LSTG, it’s worth attention that in the first level analysis, the pattern of activation contrast in each participant varies in a considerably wide range. This indicates that the regions that are most promising for optimal prediction in MEG can vary from individual to individual. Given the fact that the 6 participants are excluded in the fMRI participants, e.g. they are entirely different subjects from those participating in fMRI, it is reasonable that the activation contrasts in their LSTG region are not as salient as those in fMRI, which explains a potential reason for the low prediction accuracy.

Last but not least, in both MEG and fMRI experiments, the required reaction behaviour for participants was to differentiate a positive vs negative face shown after the word, while each word and face only lasted on the screen for 350ms. Considering the short period of time for participants to complete the task, some of them therefore may stress more attention on differentiating faces instead of processing the words, which led to a less significant activation contrast seeing concrete vs abstract words than it was supposed to. In addition, all of the 6 participants are non-native English speakers, which may also affect the participants’ neural process and consequently affect the results.

**Conclusion (Together)**

The results from the current study found no evidence that it is possible to classify whether  a participant saw a concrete or abstract word using MEG. Several limitations to the experimental paradigm and the statistical methods are outlined, because previous work in event related potentials seem to find activity differences.

**Code availability**

All code used for the analysis can be found on the following github:

https://github.com/JesperFischer/MEG\_ADV\_cognitive\_neuro

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**Appendix**

**A)**

DHARMa residual plot for the Gamma log link function

fMRI:   MEG:

Chart, box and whisker chart

Description automatically generatedChart, box and whisker chart

Description automatically generated

DHARMa residual plot for the Guassian identity link function :

fMRI:   MEG:

Chart, box and whisker chart

Description automatically generatedChart, box and whisker chart

Description automatically generated

**B)**

Bolier-plate (fMRI preprocessing)

**Fmri preprocessing:**

**Results included in this manuscript come from preprocessing performed using \*fMRIPrep\* 22.0.2 (@fmriprep1; @fmriprep2; RRID:SCR\_016216), which is based on \*Nipype\* 1.8.5 (@nipype1; @nipype2; RRID:SCR\_002502).**

**Anatomical data preprocessing : A total of 1 T1-weighted (T1w) images were found within the input BIDS dataset.The T1-weighted (T1w) image was corrected for intensity non-uniformity (INU) with `N4BiasFieldCorrection` [@n4], distributed with ANTs 2.3.3 [@ants, RRID:SCR\_004757], and used as T1w-reference throughout the workflow. The T1w-reference was then skull-stripped with a \*Nipype\* implementation of the `antsBrainExtraction.sh` workflow (from ANTs), using OASIS30ANTs as target template. Brain tissue segmentation of cerebrospinal fluid (CSF), white-matter (WM) and gray-matter (GM) was performed on the brain-extracted T1w using `fast` [FSL 6.0.5.1:57b01774, RRID:SCR\_002823, @fsl\_fast]. Volume-based spatial normalization to one standard space (MNI152NLin2009cAsym) was performed through nonlinear registration with `antsRegistration` (ANTs 2.3.3), using brain-extracted versions of both T1w reference and the T1w template. The following template was selected for spatial normalization: \*ICBM 152 Nonlinear Asymmetrical template version 2009c\* [@mni152nlin2009casym, RRID:SCR\_008796; TemplateFlow ID: MNI152NLin2009cAsym].**

**Functional data preprocessing :**

**For each of the 6 BOLD runs found per subject (across all tasks and sessions), the following preprocessing was performed. First, a reference volume and its skull-stripped version were generated using a custom methodology of \*fMRIPrep\*. Head-motion parameters with respect to the BOLD reference (transformation matrices, and six corresponding rotation and translation parameters) are estimated before any spatiotemporal filtering using `mcflirt` [FSL 6.0.5.1:57b01774, @mcflirt]. The BOLD time-series (including slice-timing correction when applied) were resampled onto their original, native space by applying the transforms to correct for head-motion. These resampled BOLD time-series will be referred to as \*preprocessed BOLD in original space\*, or just \*preprocessed BOLD\*. The BOLD reference was then co-registered to the T1w reference using `mri\_coreg` (FreeSurfer) followed by `flirt` [FSL 6.0.5.1:57b01774, @flirt] with the boundary-based registration [@bbr] cost-function. Co-registration was configured with six degrees of freedom. Several confounding time-series were calculated based on the \*preprocessed BOLD\*: framewise displacement (FD), DVARS and three region-wise global signals. FD was computed using two formulations following Power (absolute sum of relative motions, @power\_fd\_dvars) and Jenkinson (relative root mean square displacement between affines, @mcflirt). FD and DVARS are calculated for each functional run, both using their implementations in \*Nipype\* [following the definitions by @power\_fd\_dvars]. The three global signals are extracted within the CSF, the WM, and the whole-brain masks. Additionally, a set of physiological regressors were extracted to allow for component-based noise correction [\*CompCor\*, @compcor]. Principal components are estimated after high-pass filtering the \*preprocessed BOLD\* time-series (using a discrete cosine filter with 128s cut-off) for the two \*CompCor\* variants: temporal (tCompCor) and anatomical (aCompCor). tCompCor components are then calculated from the top 2% variable voxels within the brain mask. For aCompCor, three probabilistic masks (CSF, WM and combined CSF+WM) are generated in anatomical space. The implementation differs from that of Behzadi et al. in that instead of eroding the masks by 2 pixels on BOLD space, a mask of pixels that likely contain a volume fraction of GM is subtracted from the aCompCor masks. This mask is obtained by thresholding the corresponding partial volume map at 0.05, and it ensures components are not extracted from voxels containing a minimal fraction of GM. Finally, these masks are resampled into BOLD space and binarized by thresholding at 0.99 (as in the original implementation). Components are also calculated separately within the WM and CSF masks. For each CompCor decomposition, the \*k\* components with the largest singular values are retained, such that the retained components' time series are sufficient to explain 50 percent of variance across the nuisance mask (CSF, WM, combined, or temporal). The remaining components are dropped from consideration. The head-motion estimates calculated in the correction step were also placed within the corresponding confounds file. The confound time series derived from head motion estimates and global signals were expanded with the inclusion of temporal derivatives and quadratic terms for each [@confounds\_satterthwaite\_2013]. Frames that exceeded a threshold of 0.5 mm FD or 1.5 standardized DVARS were annotated as motion outliers. Additional nuisance timeseries are calculated by means of principal components analysis of the signal found within a thin band (\*crown\*) of voxels around the edge of the brain, as proposed by [@patriat\_improved\_2017]. The BOLD time-series were resampled into standard space, generating a \*preprocessed BOLD run in MNI152NLin2009cAsym space\*. First, a reference volume and its skull-stripped version were generated using a custom methodology of \*fMRIPrep\*. All resamplings can be performed with \*a single interpolation step\* by composing all the pertinent transformations (i.e. head-motion transform matrices, susceptibility distortion correction when available, and co-registrations to anatomical and output spaces). Gridded (volumetric) resamplings were performed using `antsApplyTransforms` (ANTs), configured with Lanczos interpolation to minimize the smoothing effects of other kernels [@lanczos]. Non-gridded (surface) resamplings were performed using `mri\_vol2surf` (FreeSurfer). Many internal operations of \*fMRIPrep\* use \*Nilearn\* 0.9.1 [@nilearn, RRID:SCR\_001362], mostly within the functional processing workflow. For more details of the pipeline, see [the section corresponding to workflows in \*fMRIPrep\*'s documentation](https://fmriprep.readthedocs.io/en/latest/workflows.html "FMRIPrep's documentation").**

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**### References Results included in this manuscript come from preprocessing performed using \*fMRIPrep\* 22.0.2 (@fmriprep1; @fmriprep2; RRID:SCR\_016216), which is based on \*Nipype\* 1.8.5 (@nipype1; @nipype2; RRID:SCR\_002502).**

**Homeostasis**

**Introduction**

Homeostasis is a self-regulating process that all humans have, it is a process that maintains internal processes at stable set-points. A concrete example of homeostasis is how the body maintains a stable internal temperature of around 37 degrees Celsius, if the temperature raises the body cools itself and vice versa.

investigations have also shown that one of such homeostatic processes are the circadian rhythm, which is a 24-hour cycle of many different processes. Most humans do for instance follow such a circadian rhythm when it comes to the sleep-wake cycle, research has however found that different hormones e.g. melatonin and cortisol also follow a similar pattern (Kim et al., 2015). It has also been hypothesized that other human experiences follow a circadian rhythm, indicating that the 24-hour cycle is important and worth investigating (McClung, 2013).

To statistically investigate the circadian rhythm a mathematical function that repeats itself after 24 hours is needed. The sinusoidal wave has this oscillating property see eq 1.

eq (1)

**hypothesis**

Tiredness, mood and hunger would follow an oscillating pattern best described by a sinusoidal function with an oscillation time of one day e.g. a frequency of 1/24 hours.

**Methods**

Using an experience sampling paradigm, student of the masters of cognitive science was promoted Between the 8th of September and the 21st  of November. Students were prompted by the teacher and answered the prompts to the best of their abilities at the given time, and were also encouraged to fill the questionnaires without being prompted.

**Prompts.**

When prompted students were instructed answer the questions which can be seen in appendix A. For the following paper the students’ tiredness/freshness and hunger scores were investigated which was given by a score of 0 to 100.

**Statistical analysis**

All code to reproduce the following results can be found in appendix C.   
To investigate the hypothesis two different models were fitted. First, a non-linear mixed effects model was fitted, where the amplitude the intercept and the phase of the sinusoidal wave could vary, and the frequency was set to 1/24 hours (eq. 1). Next a linear mixed effects model was fitted that was a reparameterization of eq 1. The random effects for each model were subject ids. All participants with less than 10 answered prompts were excluded leaving 13 participants in the analysis. The analysis on mood and hunger was only calculated by the non-linear model.

The linear mixed effects model was parameterized in the following way:

This parameterization implies that the amplitude of the model and the phase can be determined from the two beta estimates in the following way, for derivation see appendix B, furthermore the uncertainties on these can be determined using error propagation (Hughes & Hase, 2010).

Uncertainty on the amplitude:

**Results**

The linear mixed effects model investigating freshness showed a significant intercept ( = 45.6 se = 2.9 t = 15.8 p < .0001 and that both beta estimates were statically significant , se = 1.6 t = -11 p< .0001, , se = 2.3 t = -6.1 p< .0001. Recalculating this to the amplitude we get A = (se = 1.7) and 1.1

Results from the non-linear mixed effects model on freshness showed A = 20.75 se = 1.9 t = 11.2 p<.0001 se = 0.07 t = 61.4 p<.0001 and k = 42.3 se = 2.2 t = 18.7 p<.0001.

Chart

Description automatically generated with low confidence

Figure 1: Displays the results of the three nonlinear mixed effects models for freshness, hunger and mood for each participant.

Results from the non-linear mixed effects model on mood showed A = 4.8 se = 1.5 t = 3.3 p<.005 se = 0.25 t = 13.4 p<.0001 and k = 66 se = 2.8 t = 23.3 p<.0001.

Results from the non-linear mixed effects model on hunger showed A = 7.4 se = 1.9 t = 3.9 p<.0001 se = 0.36 t = 5.7 p<.0001 and k = 61 se = 2.6 t = 23.5 p<.0001.

**Discussion**

The results of the current study suggest that the freshness of students can be explained with a sinusoidal function where the oscillation time of the wave is set to 24 hours. These results therefore support the idea that freshness follows a circadian rhythm. The results also showed that hunger and mood followed a similar pattern, however not to the same degree as with freshness.

**Limitations**

Big limitations of the current study come from the low participation rate of students, but especially also the low number of answered prompts in the time interval from midnight to 6 in the morning. This is most likely because people slept at this time, however inference is especially difficult to make with one fourth of the time missing. Due to the low sample size a nonlinear mixed effects model where the frequency of the oscillation wasn’t feasible, which could have further strengthened the hypothesis that these experiences follow a 24-hour cycle.

**Appendix**

A:

To see the questions the students were prompted by follow the following link to try the questionnaire itself:

<https://chani.cogsciexperiment.au.dk/BodyFeelingExp.html?fbclid=IwAR175uXWlcbLCOH9zS-L0wrY51ca8B2UOkG8CNCAZESOXjLoWgH1nTUP_e8>

B:

deviation of phi.

Deviation of the amplitude

C:

All code can be found on the following GitHub:

<https://github.com/JesperFischer/MEG_ADV_cognitive_neuro/tree/master/Homeostasis>

**Literature:**

Hughes, I., & Hase, T. (2010). *Measurements and their Uncertainties: A practical guide to modern error analysis*. OUP Oxford.

Kim, T. W., Jeong, J.-H., & Hong, S.-C. (2015). The Impact of Sleep and Circadian Disturbance on Hormones and Metabolism. *International Journal of Endocrinology*, *2015*, 591729. <https://doi.org/10.1155/2015/591729>

McClung, C. A. (2013). How might circadian rhythms control mood? Let me count the ways….. *Biological Psychiatry*, *74*(4), 242–249. <https://doi.org/10.1016/j.biopsych.2013.02.019>