ORIGINAL RESEARCH

Association Between EEG Microarousal During Nocturnal Sleep and Next-Day Selective Attention in Mild Sleep-Restricted Healthy Undergraduates

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Purpose: To explore whether sleep electroencephalogram (EEG) microarousals of different standard durations predict daytime mood and attention performance in healthy individuals after mild sleep restriction.

Participants and Methods: Sixteen (nine female) healthy college students were recruited to examine the correlations between nocturnal EEG microarousals of different standard durations ($\geq 3 \text{ s}$, $\geq 5 \text{ s}$, $\geq 7 \text{ s}$, $\geq 9 \text{ s}$) under mild sleep restriction (1.5 h) and the following morning's subjective alertness, mood, sustained attention, and selective attention task performance.

Results: Results revealed that mild sleep restriction significantly reduced subjective alertness and positive mood, while having no significant effect on negative mood, sustained attention and selective attention performance. The number of microarousals (\geq 5 s) was negatively associated with positive mood at 6:30. The number of microarousals was significantly and positively correlated with the response time difference value of disengagement component of the selective attention task at around 7:30 (\geq 5 s and \geq 7 s) and 9:00 (\geq 5 s). The number of microarousals (\geq 7 s) was significantly and positively correlated with the inaccuracy difference value of orientation component of the selective attention task at around 9:00.

Conclusion: The number of EEG microarousals during sleep in healthy adults with mild sleep restriction was significantly and negatively related to their daytime positive affect while positively associated with the deterioration of disengagement and orientation of selective attention performance, but this link is dependent on the standard duration of microarousals, test time and the type of task. **Keywords:** microarousal, alertness, sustained attention, selective attention, sleep restriction

Introduction

Microarousals refer to the events of electroencephalogram (EEG) frequency change lasting more than 3 s during sleep, including theta waves and brain waves with a frequency >16 Hz (excluding spindle waves).¹ As a transient arousal event that occurs during sleep, a microarousal is not strong enough to awaken people from sleep.² Microarousals can be generated directly by sensory input disturbances during sleep, such as blood pressure/heart rate changes and environmental noise.³ Excessive microarousals would cause sleep fragmentation, which would lead to the similar undesirable effects caused by sleep deprivation/sleep restriction, such as emotion deregulation,⁴ cognitive dysfunction⁵ and social withdrawal.⁶ Moreover, excessive microarousals is one of the core characteristics of some neurological disorders (such as depression)^{7,8} and breath-related problems (ie, obstructive sleep apnoea (OSA)).^{9,10} Thus, systematic investigations about microarousals are of vital role both theoretically and practically.

The minimum duration criteria for a microarousal has been under debated until now. Although the 3 s minimum duration criteria for a microarousal was acknowledged by the American Sleep Disorders Association in 1992,¹ this short-duration criteria was criticized by its poor inter-scorer reliability.^{11,12} Some scholars thus proposed that longer duration

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instead of the standard 3 s duration criteria might be better to characterize a microarousal and have improved clinical utility in assessing cognitive impairment in OSA patients. For instance, Schwartz and Moxley¹³ found that "long microarousals" (15 s to 60 s in duration) were better correlated with subjective sleepiness than "short microarousals" (3 s to 15 s in duration) did in OSA patients. A more recent study also confirmed this finding and revealed that compared to 3 s duration, microarousal with a duration of 5 s or longer had the better combination of sensitivity and specificity.¹⁴ Moreover, compared to the OSA group without neurocognitive impairment, the OSA group with neurocognitive impairment had a significantly greater number of microarousals longer than 5 s, 7 s, and 15 s in duration.

The current evidence supporting a longer duration criteria than standard duration (3 s) for microarousal is mainly retrieved from OSA patients.^{13,14} Whether this case could be found in the healthy population, however, still remains to be determined. Moreover, previous studies only focused on sleepiness^{13,14} and sustained attention,¹⁴ whether other domains of performance (ie, mood and selective attention) would be associated with microarousal remains unknown. The neurocognitive performance was only measured once in previous studies,^{13,14} the trajectory of the associations between microarousal and neurocognitive performance with increasing waking time needs to be revealed. Thus, by mildly restricting sleep (6.5 h), the current study recruited healthy undergraduates and recorded the EEG during nocturnal sleep and measured the next-day neurocognitive performance (sleepiness, mood, sustained attention and selective attention) multiple times to track the dynamics of the associations between microarousal and neurocognitive performance.

Materials and Methods

Participants and Screening

Sixteen undergraduates (nine females and seven males, 21.19 ± 2.23 years old) with non-extreme chronotypes were recruited in the formal study due to the fact that individuals with extreme chronotypes would exhibit alternations in both subjective sleep quality¹⁵⁻¹⁹ and objective sleep parameters,²⁰ which would potentially affect the number of microarousals. The exclusion criteria were based on previous studies^{21,22} and include: (1) habitual sleep duration >9 hours or <7 hours; (2) poor sleep quality (Pittsburgh Sleep Quality Index (PSQI) score >5);²³ (3) extreme chronotype (Chinese version of Morningness-Eveningness Questionnaire (MEQ) score <47 or >69),^{24,25} (4) shift-work or travel to a different time zone during the last three months; (5) presence of sleep problems/disorders; (6) emotional disturbance (Beck Depression Inventory-II (BDI-II) score >10);²⁶ (7) physically and mentally unhealthy (General Health Questionnaire-20 (GHQ-20) score ≥ 8);²⁷ (8) overweight/obesity (body mass index (BMI) \geq 24), or underweight (BMI <18.5); (9) addiction to drugs, caffeine, tobacco and alcohol; (10) having visual impairments other than myopia or hyperopia. The inclusion criteria were (1) 7 hours \leq habitual sleep duration \leq 9 hours; (2) good sleep quality (PSQI score \leq 5);²³ (3) non-extreme chronotype $(47 \le MEO \text{ score} \le 69)$;^{24,25} (4) no shift-work or travel to a different time zone during the last three months; (5) absence of sleep problems/disorders; (6) absence of emotional disturbance (BDI-II score ≤ 10),²⁶ (7) physically and mentally healthy (GHQ-20 score <8);²⁷ (8) normal weight (18.5 \leq BMI < 24); (9) no addiction to drugs, caffeine, tobacco or alcohol; (10) no visual impairments other than myopia or hyperopia. The study was conducted according to the guidelines of the Declaration of Helsinki and approved by the Ethics Committee of South China Normal University (protocol code: 138). Informed consent was obtained from all participants involved in the study.

Design

The current study used the one-factor (test time) within-participant design.

Measurements

Subjective Sleepiness

The Karolinska Sleepiness Scale (KSS)²⁸ was used to assess subjective sleepiness, which contains one 9-point Likert item ranging from "1" (extremely alert) to "9" (extremely sleepy-fighting sleep).

Subjective Mood

Subjective mood was evaluated by the Chinese version of the Positive Affect and Negative Affect Schedule (PANAS),^{29,30} which includes eighteen 5-point Likert items ("1" - "very slightly or not at all"; "5" - "extremely") with nine items assessing positive mood and the remaining nine items evaluating negative mood (see Tables A1 and A2).

Sustained Attention

Sustained attention was measured by an adapted version of the Psychomotor Vigilance Test (PVT).³¹ Following the rationale of the original PVT,³² in the adapted PVT, a red rectangular border³³ was always presented in the center of a computer screen with a black background, and a yellow triangle (which is similar to the red dot used in previous studies)^{34,35} appeared from time to time within the border, and the participants were asked to press the "space" key as soon as they saw the yellow triangle. The maximum duration of the yellow triangle is 30s and it will disappear after detecting the response. The interval between yellow triangles was randomized between 2s to 10s. The total duration of PVT lasted for 10 minutes.

Selective Attention

The Spatial Cueing Test (SCT)³⁶ was utilized to assess selective attention. In the SCT, a cue ("+" /" \leftarrow "/" \leftarrow "/" \rightarrow ") was presented at the center of the screen for 100 ms, which was followed by a black screen lasting for 150 ms. Then, a target ("X") was displayed either at the left or at the right side of the screen and the participants were asked to identify the position of the target accurately as soon as possible. The maximum duration of the target was set to 1000 ms and the target would disappear after detecting a response. There were three kinds of trials, which were neural trials (cue was "+", 50 trials), valid trials (the cue pointed to the position of the target, 40 trials) and invalid trials (the cue pointed to the opposite position of the target, 10 trials), which lead to 100 trials in total.

Procedure

During the week before the formal experiment, the participants were required to maintain their daily sleep routine, and the bedtime was determined by their sleep habits. During this period, the participants wore actiwatches and filled in a sleep diary every day to ensure that their sleep habits were in line with the requirements of the experiment.

The formal experimental period was scheduled during the luteal phase for female participants to ensure they were tested in the same phase. During the formal experimental period, each participant was instructed to rest in the sleep laboratory for two nights, separated by at least one week. The first night was an adaption night to avoid first-night effect³⁷ and allow the participants to adapt to the environment of the sleep laboratory and familiarize themselves with the formal experimental procedures and test tasks. On the second night, the participants arrived at the sleep laboratory four hours before habitual bedtime. First, they filled out the Groningen Sleep Quality Scale (GSQS)³⁸ to assess the sleep quality of their previous night. If the sleep quality was bad (GSOS score \geq 5), then the formal experimental date would be rescheduled. If the sleep quality was good, then the participants were asked to complete the baseline tests (KSS, PANAS, PVT and SCT) at about three hours before habitual bedtime. Subsequently, after completing wash-up, brushing teeth and taking a shower in the laboratory bathroom, the participants were ready for the preparatory work of polysomnography (PSG) which was recorded by Compumedics Grael system (Compumedics, Abbotsford, Victoria, Australia). After finishing the preparatory work of PSG, the time each participant went to bed was determined based on measurements taken one week prior to the beginning of the experiment. The sleep duration was set to 6.5 h and the participants were woken up by an alarm clock. Immediately upon awakening, they sat down before a computer and completed a series of scales and cognitive tasks. Cognitive tasks were tested half-hourly and six times in total; subjective alertness was measured once after participants woke up, and every half hour thereafter, for a total of seven times; subjective mood was measured once after participants woke up, and every hour thereafter, for a total of four times. During the experiment, it was forbidden to use self-luminous devices (such as cell phones and iPads) so as not to interfere with sleep.³⁹ At the end of the experiment, the experimenters removed the electrodes worn on the scalps of the participants, and the participants could leave the laboratory after washing and having breakfast. The procedure was displayed in Figure 1. All the data were collected during autumn (from 25th September to 5th December in Guangzhou, China).



Figure I The procedure of formal experiment.

Abbreviations: PANAS, positive affect and negative affect schedule; KSS, Karolinska Sleepiness Scale.

Data Analysis

For SCT, two components of selective attention were computed: orientation and disengagement. For orientation, the differences on response times and inaccuracy between valid trials and neutral trials were computed. For disengagement, the differences on response times and inaccuracy between invalid trials and neutral trials were computed. A one-factor (test time) repeated-measure ANOVA was conducted on all subjective sleepiness/mood/ sustained attention/selective attention indicators. Bonferroni corrections were applied to multiple comparisons. Spearman correlation analysis was conducted between number of microarousals of various duration criteria and subjective sleepiness/mood/sustained attention/selective attention performance to determine the potential associations between microarousal and neurocognitive performance. JASP 0.17.2 software⁴⁰ was used for all the analysis.

Results

The data from five participants were excluded from formal analysis (three due to the incomplete EEG data and two due to exceeding ± 3 SD from the mean reaction time of PVT response), which led to 11 participants in total in the formal analysis (eight females and three males, 20.73 ± 2.28 years old).

Sleep and Microarousal

The mean sleep efficacy was 98.45%. The total number of microarousals (≥ 3 s) was 46.91±19.18 on average, while total number of microarousals (≥ 5 s) was 26.00±12.18, total number of microarousals (≥ 7 s) was 18.91±9.89, and total number of microarousals (≥ 9 s) was 13.45±8.74.

Subjective Sleepiness and Mood

The main effect of test time was significant for subjective sleepiness (see Table 1). Further analysis revealed that the participants felt more sleepy at test time 1 (6.45 ± 0.46 , p = 0.003) than at baseline (3.18 ± 0.38). In addition, significantly more sleepiness was reported at test time 2 (5.09 ± 0.48 , p = 0.045) and test time 3 (5.18 ± 0.60 , p = 0.016) than at test time 7 (3.64 ± 0.47) (see Figure 2).

For subjective mood, only the main effect of test time on the positive affect was revealed (see Table 1). Less positive affect was reported at test time 1 (9.46 \pm 0.28, *p* = 0.004) than at baseline (16.82 \pm 1.46) after mild sleep restriction (see Figure 3).

Sustained Attention and Selective Attention

The descriptive results of PVT and SCT performance are shown in Table 2. For PVT performance, only the main effects of mean RT of the fastest 10% of trials and lapse probability were significant (see Table 1). However, further analysis found that the differences among conditions did not survive after Bonferroni corrections. For SCT performance, the main effects of test time on four indicators did not reach significance (see Table 1).

	F	Þ	Partial η^2
KSS	5.86	0.005	0.37
PANAS _{PA}	5.35	0.018	0.349
PANAS _{NA}	1.26	0.29	0.112
PVT _{Fastest10%RT}	3.92	0.023	0.28
PVT _{Slowest10%RT}	1.69	0.221	0.145
PVT_{MeanRT}	2.02	0.182	0.168
PVT _{LapseProbability}	3.74	0.025	0.272
SCT _{Orientation}	0.81	0.567	0.075
SCT _{Orientation∆Inaccuracy}	0.82	0.493	0.075
SCT _{Disengagement}	0.40	0.726	0.038
SCT _{Disengagement}	0.96	0.459	0.088

 Table I The Statistical Results

Note: Significant differences are indicated in bold.

Abbreviations: KSS, Karolinska Sleepiness Scale; PANAS, positive affect and negative affect schedule; PVT, Psychomotor Vigilance Test; SCT, Spatial Cueing Test; PA, positive affect; NA, negative affect; RT, reaction time.

Correlational Results

The correlational results are presented in Table 3. Correlational analysis revealed that the number of microarousals (≥ 5 s) was negatively correlated with positive affect at test time 1 (r = -0.616, *p* = 0.043). For the attention performance, the differences between baseline performance and performance at each test time were obtained and then the associations between these differences and number of microarousals of various standard durations were computed. The results found



Figure 2 The trajectory of subjective sleepiness. Error bars indicate 1 standard error of the mean. *p < 0.05, **p < 0.01. Abbreviation: KSS, Karolinska Sleepiness Scale.



Figure 3 The trajectory of subjective mood. (A) Positive affect; (B) Negative affect. Error bars indicate 1 standard error of the mean. **p < 0.01. Abbreviation: PANAS, positive affect and negative affect schedule.

	PVT Fastest 10% RT (ms)	PVT Slowest 10% RT (ms)	PVT MeanRT (ms)	PVT Lapse Probability (%)	SCT Orientation ART (ms)	SCT Orientation ∆Inaccuracy (%)	SCT Disengagement ∆RT (ms)	SCT Disengagement ∆Inaccuracy (%)
Baseline	297.13 (9.63)	508.81 (42.72)	372.24 (19.89)	8.18 (3.92)	40.91 (4.60)	-0.68 (0.40)	40.64 (5.77)	-17.27 (4.38)
Test I	301.14 (8.69)	691.49 (71.19)	407.46 (17.66)	11.45 (2.52)	41.48 (4.82)	-1.14 (0.87)	34.69 (10.67)	-14.55 (4.55)
Test2	310.40 (9.97)	1064.39 (382.37)	494.57 (87.50)	16.73 (5.72)	46.91 (6.68)	-1.59 (0.53)	42.09 (4.86)	-16.36 (5.09)
Test3	314.51 (9.27)	1184.11 (342.52)	490.63 (55.59)	19.73 (5.10)	41.92 (4.40)	-0.91 (0.87)	30.90 (6.01)	-23.64 (4.79)
Test4	323.30 (13.46)	1146.97 (401.56)	511.01 (74.79)	22.91 (6.82)	49.62 (7.92)	-0.41 (1.19)	46.42 (18.55)	-21.64 (4.54)
Test5	319.66 (12.13)	928.71 (133.75)	452.41 (24.47)	19.18 (4.85)	48.52 (4.51)	-2.77 (1.22)	33.13 (11.20)	-20.18 (4.38)
Test6	312.13 (12.87)	1038.52 (367.96)	478.40 (55.42)	20.55 (6.24)	51.40 (5.75)	-1.77 (1.28)	40.33 (6.12)	-20.73 (6.32)

Table 2 The Descriptive Results of PVT and SCT Performance [M(SE)]

Abbreviations: RT, reaction time; PVT, Psychomotor Vigilance Test; SCT, Spatial Cueing Test.

Table 3 The Correlations Between Microarousal and Subjective Mood and Selective Attention Performance [r(p)]

	MA3s	MA5s	MA7s	MA9s
PAI	-0.574 (0.065)	-0.616 (0.043)	-0.493 (0.124)	-0.369 (0.264)
PA2	-0.153 (0.653)	0.014 (0.966)	0.103 (0.762)	0.257 (0.446)
PA3	-0.120 (0.726)	-0.019 (0.955)	0.070 (0.838)	0.190 (0.576)
PA4	-0.507 (0.111)	-0.340 (0.306)	-0.215 (0.526)	-0.037 (0.913)
SCTDRTI	0.469 (0.145)	0.437 (0.179)	0.324 (0.331)	0.018 (0.958)
SCTDRT2	-0.100 (0.769)	-0.137 (0.689)	-0.050 (0.883)	-0.118 (0.729)
SCTDRT3	0.433 (0.184)	0.661 (0.027)	0.726 (0.011)	0.579 (0.062)
SCTDRT4	0.105 (0.759)	0.182 (0.592)	0.100 (0.769)	0.105 (0.759)
SCTDRT5	0.055 (0.873)	0.050 (0.884)	0.068 (0.841)	-0.223 (0.509)
SCTDRT6	0.465 (0.150)	0.620 (0.042)	0.525 (0.097)	0.310 (0.354)
SCTOINACCI	-0.023 (0.946)	0.187 (0.583)	0.390 (0.235)	0.336 (0.312)
SCTOINACC2	0.212 (0.531)	0.299 (0.371)	0.406 (0.215)	0.232 (0.493)
SCTOINACC3	0.316 (0.344)	0.281 (0.402)	0.247 (0.464)	0.138 (0.685)
SCTOINACC4	-0.555 (0.076)	-0.591 (0.055)	-0.442 (0.174)	-0.361 (0.276)
SCTOINACC5	0.124 (0.717)	0.349 (0.293)	0.593 (0.054)	0.555 (0.076)
SCTOINACC6	0.422 (0.196)	0.532 (0.293)	0.640 (0.034)	0.415 (0.205)

Note: Significant differences are indicated in bold.

Abbreviations: PA, positive affect; NA, negative affect; SCTDRT, The difference on reaction time of disengagement on Spatial Cueing Test; SCTOINACC, The difference on inaccuracy of orientation on Spatial Cueing Test; MA, microarousal.

that the number of microarousals (≥ 5 s and ≥ 7 s) was positively associated with the ΔRT on disengagement of SCT at test time 3 (r = 0.661, p = 0.027; r = 0.726, p = 0.011). In addition, the number of microarousals (≥ 5 s) was positively associated with the ΔRT on disengagement of SCT at test time 6 (r = 0.62, p = 0.042). The number of microarousals (≥ 7 s) was positively associated with the Δ Inaccuracy on orientation of SCT at test time 6 (r = 0.64, p = 0.034). No significant associations were revealed between PVT performance/subjective sleepiness and microarousal.

Discussion

Expanding previous studies in OSA patients,^{13,14} the current study transferred the sample from population with sleep disorder to healthy population to explore the potential associations between microarousal and neurocognitive performance. Except the generalization of population, the current investigation measured neurocognitive performance multiple times to track the dynamics of these potential associations. The results revealed that subjective sleepiness did not significantly correlate with the number of microarousals while the number of microarousals (≥ 5 s) was negatively related to positive affect immediately after waking up. Combined with the fact that mild sleep restriction impaired the positive affect at test

time 1, these findings suggested that microarousal might be closely related to affective experience and the number of microarousals (≥ 5 s) may be a promising candidate to represent emotional disturbances. These findings also supported the claim that the minimum duration criteria for a microarousal needs to extend from 3 s to 5 s or even longer.¹⁴

Besides the positive affect, the domain-specific correlations between microarousal and attentional performance were also identified. Specifically, selective attention but not sustained attention was significantly associated with microarousal, which was partially replicated with the findings of Duce et al.¹⁴ In their study, the significant association between microarousal and sustained attention assessed by PVT was reported. Using the same task, however,no such association was revealed in the current study. These inconsistencies might be due to the differences on sample characteristics. Duce et al¹⁴ employed OSA patients which were characterized by poor sleep quality and excessive number of microarousals, while we recruited healthy undergraduates showinggood sleep quality and a relatively few number of microarousals. All these results implied that the predictive role of microarousals on attentional performance might be dependent on group characteristics. More specifically, microarousals might better predict the simple attentional function (ie, sustained attention) in groups with sleep disorders, while microarousals showed some predictive power to more complex attentional components (ie, selective attention) in healthy groups. However, this possibility needs further empirical support. Moreover, the number of microarousals (\geq 5 s and \geq 7 s) but not the number of microarousals (\geq 3 s) was found to be associated with the selective attention performance, further supporting the extension of current microarousal minimum duration criteria.¹⁴

It should be noted that the significant associations between microarousal and affect/selective attention did not appear at all test times. On the contrary, these significant correlations only appeared at specific test times. For positive affect, it appeared at test time 1 (about 6:30), while it appeared at test time 3 (about 7:30) and test time 6 (about 9:00) for selective attention, which replicated previous studies about ultradian rhythms.^{41–45} These results also added novel insights into previous studies^{13,14} which only measured neurocognitive performance once, and emphasized the necessity of inclusion of test time as an important factor to be considered for future studies.

Previous studies on mild sleep restriction (less than 2 hours) are limited,^{4,46} the current study also provided novel empirical findings about the trajectory of neurocognitive consequence of mild sleep restriction in healthy populations. The results found that after 6.5 h nocturnal sleep, the participants felt most sleepy immediately after waking up, and the sleepiness fluctuated during the following 2.5 hours and did not get significantly less until the end of the experiment. This was the similar case for positive affect, which replicated a previous finding.⁴⁷ In addition, we found that the negative affect, sustained attention and selective attention did not change much during the three hours after waking up. It should be noted that although the main effects of test time on mean RT of the fastest 10% of trials and lapse probability were significant, the difference among different test times did not survive Bonferroni corrections. This might be due to the limited number of valid sample size in the current investigation. Thus, the interpretation of this null effect on sustained attention should be taken with caution. This null effect of mild sleep restriction seems to contradict Lo et al.⁴⁸ Besides the differences of sample characteristics (n = 11 vs n = 29; adults vs adolescents), the manipulation of sleep restriction might also explain the above-mentioned discrepancies. The participants were restricted for only one day in the current study while participants in the Lo et al study⁴⁸ were sleep-restricted for five days, which would amplify the undesirable effects of sleep restriction.

Some limitations need to be noted. First, only healthy young adults were recruited in the current study. Whether the current findings could be generalized to other populations (such as extreme chronotypes, individuals who are underweight or overweight, unhealthy sleepers, or depression inpatients) and other age groups (eg, the elderly and adolescents), needs to be determined. Secondly, previous studies found the effects of sleep restriction were task-specific^{49,50} and dependent on task difficulty.^{50,51} Thus, multiple tasks to assess the same cognition and various difficulty levels can be used in future studies to explore the potential task-specific association between microarousal and cognitive performance. Thirdly, although the formal experimental period was set at the same phase for female participants, which might be a strength of the methodology used, the potential menstrual effects on sleep⁵² still can not be excluded. Lastly, although the current sample size was comparable with previous studies, ^{53–55} the current sample size is still small, which needs to be enlarged in future studies to replicate and strengthen the current findings.

Conclusion

Mild sleep restriction increases subjective sleepiness, and decreases positive affects, while exerting a limited effect on negative affects, sustained attention and selective attention. The number of microarousals (≥ 5 s) might be negatively related to the decrease of positive affects caused by mild sleep restriction. And the numbers of microarousals (≥ 5 s and ≥ 7 s) are positively associated with both the orientation and disengagement component in selective attention. Duration criteria of longer than 3 s for microarousals is suggested to be used in the future studies.

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Disclosure

The authors report no conflicts of interest in this work.

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