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Article type : Letter to the Editor

Title

Potential treatment effect of the SQ tree SLIT-tablet on pollen food syndrome caused by apple

Capsule summary

Pollen food syndrome (PFS) is common and interferes with daily living. The results of this apple challenge suggest a potential treatment effect of birch AIT on PFS related to tree pollen.

Keywords

Pollen food syndrome, food challenge, apple, sublingual immunotherapy, tree SLIT-tablet

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Main text

To the editor

In adolescents and adults up to 60% of all food allergies are associated with a respiratory allergy⁽¹⁾. Around 70 percent of birch allergic individuals develop allergic symptoms against certain foods such as nuts and apples containing homologous cross-reactive allergens to the major birch allergen Bet v 1^(2,3,4). The symptoms are manifested as a condition called pollen food syndrome (PFS), and its occurrence typically involves pre-sensitisation to pollen allergens from the *Fagales* order. PFS is IgE-mediated and is caused by cross-reactivity between Bet v 1 and other protein family 10 (PR-10) proteins in a number of common food items such as apple, hazelnut, carrot and peach^(2,3). PFS interferes with activities of daily living, as patients will experience local oral symptoms such as itching, tingling, swelling in the mouth, or oral angioedema after intake of a number of common food items^(2,5). Some patients may also experience systemic reactions^(2,3), but PR-10 triggered PFS is not a severe condition in most cases⁽³⁾.

Birch allergen immunotherapy treatment (AIT) could be beneficial in the treatment of PFS due to the cross reactivity between Bet v 1 and common PR-10 allergens⁽⁶⁾. In a recently published position paper from EAACI on PFS, the authors concluded that trials were needed to assess the effect of birch AIT on PFS⁽¹⁾.

The original purpose of the study presented here investigates the potential treatment effect on PFS of a sublingual immunotherapy tablet for the treatment of tree pollen allergy, the SQ tree SLIT-tablet (12 SQ-Bet, ALK, Hørsholm, Denmark).

The treatment effect was measured in an open-label apple challenge conducted at the end of a double-blind, placebo-controlled phase III trial in a subgroup of the trial participants after 6.5-9.5 months of once-daily treatment with the SQ tree SLIT-tablet or placebo. The primary phase III trial results were reported previously⁽⁷⁾. In total, 124 participants (63 placebo, 61 SQ tree SLIT-tablet) from 12 centres in Germany and 12 centres in Poland with tree pollen allergy accepted to participate. They were challenged with apple slices (cut by standardised apple slicer) in 5 steps with increasing amounts corresponding to 4, 8, 16, 32 and 64 g given in intervals of 15 minutes. Objective and subjective PFS symptoms in response to the challenge were recorded. Participants reported a global evaluation of efficacy on PFS symptoms after intake of apple by comparing subjective PFS symptoms before and after treatment (see also online addendum). Specific IgE and IgG₄ responses to the apple PR-10 allergen Mal d 1 were measured throughout the trial.

The majority of participants in both groups (75% for SQ tree SLIT-tablet and 68% for placebo) managed to consume the highest dose of apple (64 g) without relevant objective symptoms. The subject-reported PFS symptoms by VAS during the apple challenge were generally low and many participants reported no symptoms (**Figure 1**). However, the mean levels of PFS symptoms after apple intake were lower for participants in the active group compared with placebo. Especially the increase in symptoms from before the apple challenge to after the first dose of apple (4 g) was more pronounced for participants in the placebo group, suggesting improved tolerance in the active group. This tendency was similar for individual symptoms and the overall PFS VAS score. A global evaluation of efficacy on PFS symptoms showed that more participants in the active group reported improved PFS symptoms after treatment compared with placebo (87% vs. 64%, OR=0.27, p=.0028).

Treatment with the SQ tree SLIT-tablet increased serum levels of apple specific IgE and IgG₄ compared with placebo at all measured time points (changes from baseline, **Figure 2**). Apple specific IgE increased in the active group until the off-season visit, after which it began to decrease. However, at end of trial serum levels of apple specific IgE (log-transformed) were still significantly higher in the active group than for placebo (p<.001 for difference in change from baseline).

The change from baseline in apple specific IgG₄ (log-transformed) increased throughout the duration of the trial in the active group (p<.001 for difference between groups at end of trial). No increases in apple specific IgE or IgG₄ were seen for the placebo group.

The apple specific immunological responses resembled that seen for birch. The ratio of IgG₄/IgE showed that the immunoglobulin production was in favour of Mal d 1 reactive IgG₄ over IgE from 4 weeks of treatment (**Figure 2**).

The apple challenge did not lead to any serious adverse events, severe local swellings and/or systemic allergic reactions. A summary of adverse events reported during the entire trial period showed that a slightly higher proportion of participants with PFS reported adverse events regardless of treatment group compared with participants without PFS (placebo: 59% vs. 50%, 12 SQ-Bet: 84% vs. 77%). This was primarily driven by more events of oral pruritus. The main safety phase III trial results have been published previously ⁽⁷⁾.

It is a limitation of this study that no baseline challenge was conducted. A study is warranted where participants are pre-selected specifically for PFS (e.g. by challenge) to confirm the findings of the present study. This would ensure distinct PFS symptoms and allow for a better detection of differences in desensitisation between the groups. However, despite the majority of participants reported no PFS symptoms in the present study, the results point at a potential treatment effect: we saw a significant difference between the groups in the global evaluation and a trend towards improved PFS symptoms in the active treatment group for each dose step (Figure 1).

It has been suggested that higher allergen doses are needed to reduce food-induced reactions than to reduce pollen-induced reactions ⁽⁸⁾. The present study suggests, that the SQ tree SLIT-tablet is of sufficiently high-dose to elicit an effect on PFS symptoms, and to modulate the antibody responses towards apple allergens (i.e. apple-specific IgE and IgG₄).

The results of the apple challenge suggest a potential treatment effect of the SQ tree SLIT-tablet on PFS related to tree pollen. Participants in the active group on average experienced less symptoms during the apple challenge, were more likely to reach the highest dose of apple and rated their global evaluation of PFS symptoms better than participants in the placebo group. In conclusion, this exploratory study indicated a potential improvement of PFS and increased tolerance to apple in participants with a history of apple induced symptoms treated with the SQ tree SLIT-tablet, compared with placebo. Treatment resulted in changes in serum levels of apple specific IgE and IgG₄ with the similar kinetics as seen for birch specific IgE and IgG₄. These results should be confirmed in a study with participants pre-selected specifically for PFS.

Figure legends

Figure 1 Reported overall PFS VAS score

The symptoms were measured on a VAS scale ranging from no symptoms to severe symptoms. No symptoms corresponded to 0 cm and severe symptoms corresponded to 10 cm. —: mean, —: median

Figure 2 Change from baseline of apple specific IgE and IgG₄ (log transformed)

Approximate time points: Randomisation: 0 months; Off-season: 1 month of treatment; Pre-tree pollen season: 4 months of treatment; Pre-birch pollen season: 7 months of treatment; End of trial: 9 months of treatment

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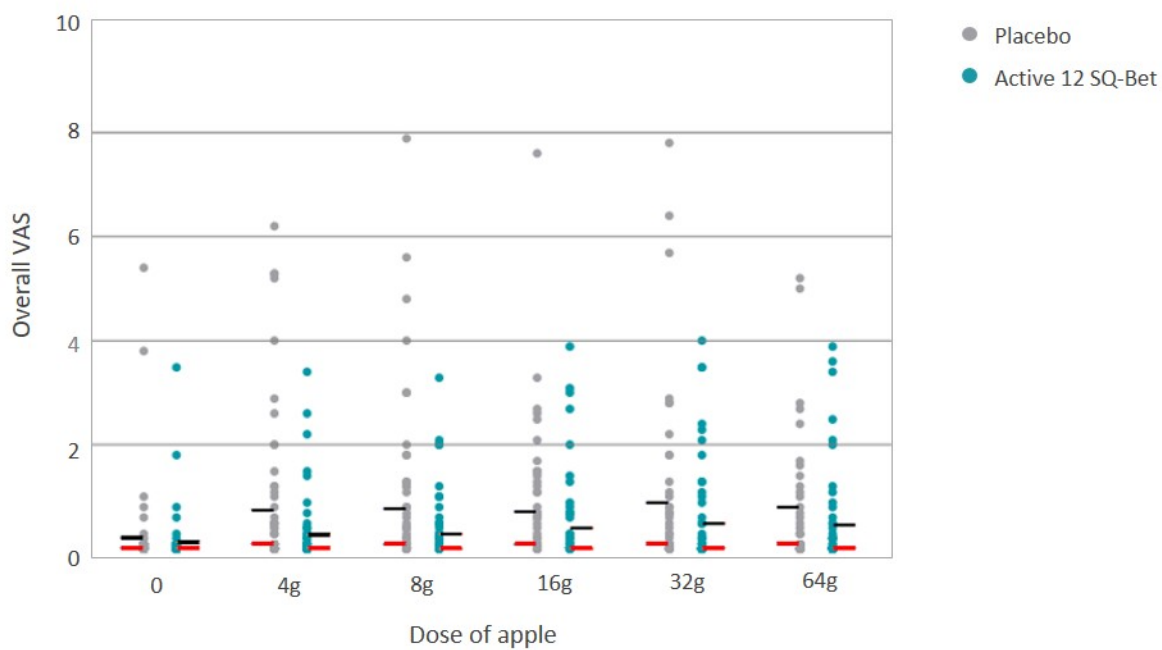
Author contributions

All authors made substantial contributions to conception and design of, or acquisition of data or analysis and interpretation of data. All authors revised the manuscript critically for important intellectual content. All authors gave final approval of the version to be published. TB were signatory investigator in the trial, SJT and IS were involved in designing the apple challenge and BSS was involved in trial design and responsible for reporting of trial results.

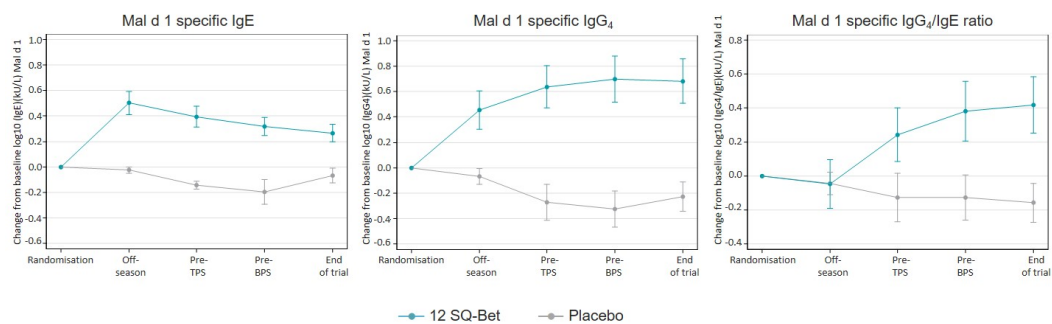
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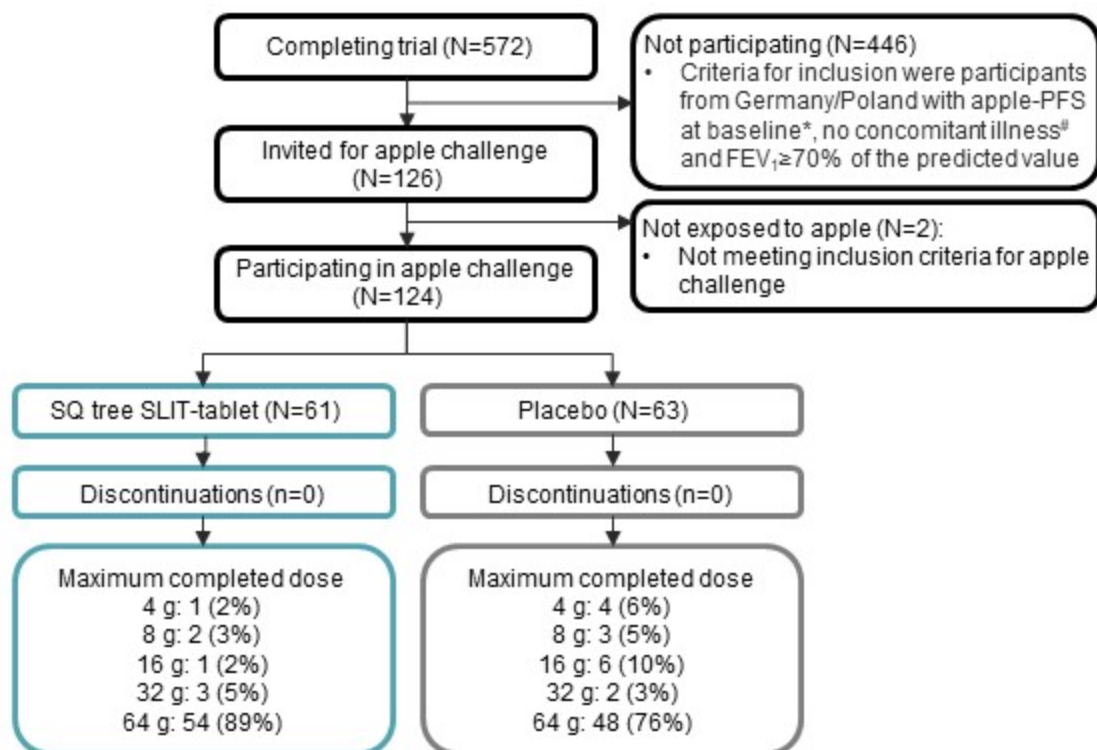
Dr. Till reports grants from ASIT Biotech and consultancy fees from Aimmune during the conduct of the study; Dr. Stage reports he was employed by ALK during the conduct of the study. Dr. Skypala has nothing to disclose; Dr. Biedermann reports he gave advice to or got an honorarium for talks or received a research grant from: ALK, Mylan, Novartis, Sanofi-Regeneron, Phadia Thermo-Fisher and Celgene during the conduct of the study.



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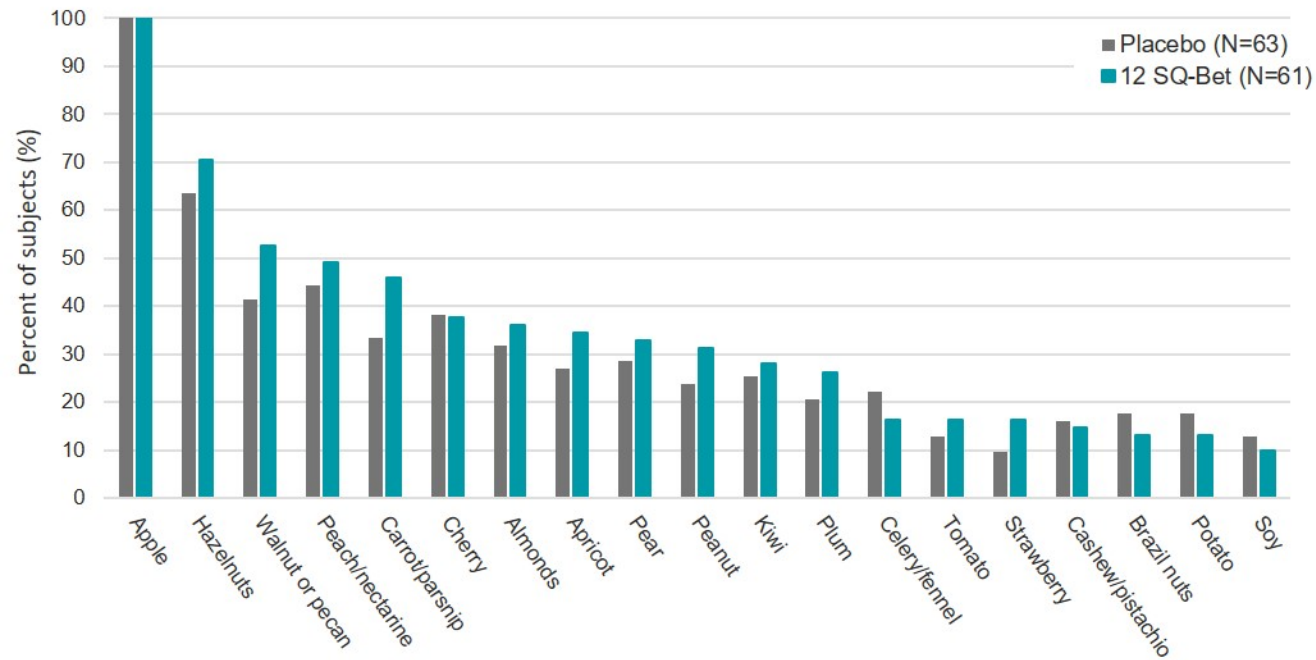


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*: assessed by a PFS diagnostic questionnaire; #: severe allergic rhinoconjunctivitis symptoms, or acute respiratory tract infection on the day of the challenge

Subject reported PFS food items in >10% of population



Treatment group	Placebo N=63	SQ tree SLIT-tablet N=61
Gender, n (%)		
Male	25 (40%)	27 (44%)
Female	38 (60%)	34 (56%)
Age, years		
Mean (SD)	35 (13)	39 (14)
Min – max	12 - 65	14 - 65
Years with birch allergy, mean (SD)	18.5 (11.8)	16.3 (11.1)
Concomitant asthma, n (%)	26 (41%)	29 (48%)
Years with asthma, mean (SD)	12.5 (11.5)	14.4 (12.4)
Years with PFS, mean (SD)	14.7 (12.2)	14.9 (11.6)
Mal d 1 specific IgE, kU/L (SD)	0.63 (0.74)	0.72 (0.76)
PFS questionnaire scores		
PFS status positive	63 (100%)	61 (100%)
Allergic reactions to apple	63 (100%)	61 (100%)
... to nuts	45 (71%)	48 (79%)
... to root vegetables	13 (21%)	9 (15%)
... to raw foods only	55 (87%)	50 (82%)
Allergic reactions within 5 minutes	50 (79%)	51 (84%)
PFS onset age, mean (SD)	21.8 (13.4)	24.9 (12.3)

N=number of participants, SD=standard deviation, PFS=pollen food syndrome

Online supplementary

Full methods description

Trial design

The trial was an open-label apple challenge conducted at the end of a phase III trial with the SQ tree SLIT-tablet (EudraCT 2015-004821-15) that is an oral lyophilisate for at-home use containing standardised allergen extract from birch pollen (*Betula verrucosa*).

In short, 634 participants with tree pollen allergy were randomised 1:1 to once-daily treatment with the SQ tree SLIT-tablet (12 SQ-Bet/12 DU/60 microgram Bet v 1) or placebo for at least 16 weeks before the anticipated start of the tree pollen season (including alder, hazel and birch) and until the end of the season (in total 6.5-9.5 months of treatment). The trial was designed and conducted in accordance with the Declaration of Helsinki ⁽¹⁾ and in compliance with Good Clinical Practice guidelines as set by the International Conference on Harmonisation (ICH) ⁽²⁾. Relevant national ethic committees and regulatory authorities approved the trial protocol and amendments.

From the trial population, participants from Germany and Poland with PFS associated with oral symptoms after intake of apples prior to randomisation (assessed by a PFS diagnostic questionnaire ⁽³⁾) were invited to take part in the apple challenge. In total, 124 participants from 12 centers in Germany and 12 centers in Poland accepted to participate. (**Figure E1**). The apple challenge was conducted on the same day as the end of trial visit between mid-May to mid-June 2017. No intake of antihistamine tablets or eye drops, or nasal steroids were allowed during the 5 days before the apple challenge.

For the apple challenge, Golden delicious apples from South Africa was used. The apples were kept cooled at 2-8°C from shipment and until use to maintain the allergenicity of the apples. Stability studies suggest that the majority of the allergenicity can be retained for 6 months ⁽⁴⁾. The

weight of a Golden delicious apple was estimated to 128 g without the core. Participants were challenged with apple slices that were not peeled (cut by standardised apple slicer) in 5 steps with increasing amounts corresponding to 4, 8, 16, 32 and 64 g apple in intervals of 15 minutes. Stopping criteria for the challenge included drop in lung function (>20% drop in peak flow), wheezing, swelling/oedema of the face/oral cavity, vocal changes, vomiting, or diarrhoea. Re-challenge with the same dose was performed if the subject experienced one or more of the following symptoms; cough, oedema lips, flushing/erythema, chest tightness, shortness of breath, nausea, or abdominal discomfort. Re-challenge with the same dose was performed only after symptoms with the previous dose had subsided (a minimum of 15 minutes after the previous dose). The apple challenge could be stopped at any time at the discretion of the investigator or the subject. After the apple challenge, participants were observed for a minimum of 120 minutes.

Trial population

Participants in the apple challenge comprised adults (18-65 years) with persistent moderate-severe allergic rhinoconjunctivitis induced by birch (prior to randomisation), and PFS (assessed using a validated PFS diagnostic questionnaire ⁽³⁾) with previous oral symptoms after intake of apples, who had completed the treatment phase of the trial. All participants gave written informed consent prior to the apple challenge.

Participants with prior severe reactions, e.g. severe throat swelling, severe systemic reactions or significant respiratory symptoms, after intake of apples were not eligible. Participants with severe allergic rhinoconjunctivitis symptoms, acute respiratory tract infection, or FEV₁ <70% of the predicted value on the day of the challenge were also not included.

Endpoints and assessments

After each apple challenge step, the investigator assessed if the subject had any objective PFS symptoms and asked the subject about the occurrence of subjective symptoms (for full list of

1 symptoms refer to the PFS diagnostic questionnaire ⁽³⁾. The average amount of apple tolerated
2 without any objective symptoms predefined in the observer list was assessed. Peak flow was
3 measured if the subject experienced any respiratory symptoms e.g. wheeze, cough, chest
4 tightness, or shortness of breath. Vital signs were assessed if considered necessary by the
5 investigator.

6
7 Participants completed a PFS symptom assessment before and after each step of the apple
8 challenge. Individual PFS symptoms (tingling/numbness of lips or mouth; itching of lips, mouth or
9 palate; scratchy/sore throat; swelling of lips, tongue, mouth or throat) were scored on a 0-10 cm
10 VAS scale indicating no symptoms (0) to severe symptoms (10). Furthermore, participants
11 assessed the overall severity of the PFS symptoms by VAS.

12
13 After the apple challenge, the participants gave a global evaluation of efficacy on PFS symptoms
14 by answering the following question: "Compared to the allergic symptoms you experienced after
15 intake of apple before you started treatment, how are you feeling now?" Response options were:
16 much better, better, the same, worse, or much worse (for the analysis, much better and better were
17 considered 'improved' and the other categories were 'not improved').

18
19 For assessment of the immunological response to treatment, blood samples were collected at
20 randomisation and 4 times during the trial (approximate time points after randomisation; off-
21 season: 1 month; pre-tree pollen season: 4 months; pre-birch pollen season: 7 months; end of trial:
22 9 months. The samples were analysed by ALK using the ImmunoCAP assays (Thermo Fisher
23 Scientific). Immunological assessments included apple specific (Mal d 1) IgE and IgG₄ for
24 participants in the apple challenge.

Statistical methodology

This study was an addendum to a phase III trial with the SQ tree SLIT-tablet (EudraCT 2015-004821-15), and as such the endpoints, although pre-specified before the challenge, were exploratory. All statistical analyses were carried out by the sponsor. The principal statistical software used was SAS, version 9.4 (SAS Institute, Cary, North Carolina).

Analysis of the global evaluation of PFS (improved vs not improved) was done by a generalised logistic regression model with treatment as fixed effect. The analyses of average changes from baseline to end of treatment in specific IgE and IgG₄ were performed with a linear mixed effects model with change from baseline in log₁₀ IgE (or IgG₄) at end of treatment as dependent variable, treatment group and country as fixed effects and log₁₀ IgE (or IgG₄) at baseline as a covariate. The remaining exploratory endpoints related to the apple challenge were presented by descriptive statistics only.

Results

Population

124 participants (63 placebo, 61 SQ tree SLIT-tablet) were included in the apple challenge. The groups were balanced in terms of baseline characteristics (**Table E1**).

In addition to reactions to apple, the PFS questionnaire revealed that the most common foods causing PFS were hazelnut (reactions noted by 67% of the participants), walnut/pecan (47%), peach/nectarine (47%), and carrot/parsnip (40%) (**Figure E2**).

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