ORIGINAL RESEARCH ARTICLES

Agar ultrasound phantoms for low-cost training without refrigeration

Fantômes d'échographie utilisant l'agar-agar pour une formation à faible coût, sans réfrigération

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Introduction: Ultrasound is a technique that can be utilised to augment procedures to increase their safety and efficacy, but requires that health professionals be trained to use this imaging modality before it can be implemented. With the extremely high cost of manufactured phantoms, homemade alternatives are popular substitutes. Gelatine is commonly used as a matrix to suspend analogues and mimic tissue, but other substrates like ground meat can also be used. Both of these substrates require refrigeration and are subject to spoiling. Our research was designed to evaluate whether agar models would be superior to traditional Gelatine models in their sustainability and whether they would produce ultrasound images adequate for training.
Methods: Agar models of varying formulas (percent agar by weight with certain additives varied) were tested for acceptable fidelity to real tissue, ultrasound image quality, and durability compared to gelatine models and human tissue.
Results: A five percent by-mass agar model augmented with small amounts of suspended wheat flour presented as a model that could generate an ultrasound image that remarkably resembled that of real tissue. This agar model does not require refrigeration, is resistant to spoiling and desiccation, mimics tissue texture well, is durable enough to withstand high-volume training, and can be recycled to make new models.
Conclusion: Agar phantoms are easy to make, do not require refrigeration, and have multiple distinct advantages over gelatine models for ultrasound training in austere conditions.

Introduction: L'échographie est une technique qui peut être utilisée pour renforcer les procédures et viser à accroître leur sécurité et leur efficacité, mais elle exige que les professionnels de la santé soient formés à l'utilisation de cette modalité d'imagerie avant sa mise en œuvre. Le coût des fantômes industriels étant extrêmement élevé, les alternatives artisanales représentent des substituts répandus. La gélatine est couramment utilisée comme matrice pour mettre en suspension des produits analogues et imiter des tissus, mais d'autres substrats tels que de la viande hachée peut également être utilisés. Ces deux substrats doivent être réfrigérés et sont sujets à une détérioration. Notre étude visait à évaluer si les modèles utilisant l'agar-agar pourraient être supérieurs aux modèles traditionnels en gélatine en termes de durabilité et s'ils pouvaient produire des images échographiques adéquates pour la formation.
Méthodes: Des modèles utilisant l'agar-agar de formules différentes (variation du pourcentage d'agar-agar par rapport au poids avec certains additifs) ont été testés pour déterminer le degré de fidélité acceptable par rapport au tissu réel, la qualité d'image échographique, et la durabilité par rapport aux modèles de gélatine et aux tissus humains.
Résultats: Un modèle utilisant l'agar-agar à cinq pour cent par masse, renforcé de petites quantités de farine de blé en suspension, s'est avéré capable de générer une image échographique ressemblant de façon remarquable à un tissu réel. Ce modèle utilisant l'agar-agar ne nécessite pas de réfrigération, est résistant à la détérioration et à la dessiccation, imite bien la texture des tissus, est assez durable pour résister à un grand nombre de formations, et peut être recyclé pour fabriquer de nouveaux modèles.
Conclusion: Les fantômes utilisant l'agar-agar sont faciles à faire, ne nécessitent pas de réfrigération et présentent plusieurs avantages distincts par rapport aux modèles en gélatine pour la formation à l'échographie dans des conditions austères.

African relevance

- Agar is a promising material for constructing ultrasound training models, as it does not require refrigeration, resists melting and spoilage, and may be reused.
- Agar models provide images similar to real tissue and commonly used gelatine models while also withstanding more punctures than gelatine models of similar density for venepuncture.

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Introduction

Ultrasound is a technique that can be employed to great effect in the assessment of and treatment of patients, especially in emergent situations. As an example, protocols like the Focused Assessment with Sonography in Trauma (FAST) exam are widely used to screen patients for injuries that would otherwise not be evident, and possibly missed. With increased portability and decreasing costs, ultrasound machines provide quality images in many places where they are the only immediate imaging modality available.[1,2] Ultrasound can also augment

procedures like intravenous (IV) catheter placement, paracentesis or thoracentesis and central line placement to increase the procedure's efficacy and safety.[3] This considered, training providers to appropriately utilise ultrasound before introducing it as a part of the standard of care is necessary.

Yet, one of the main barriers noted amongst practitioners in low- and middle-income countries has been access to training.[4] Utilisation of realistic models has been shown to increase the competency of providers using ultrasound to augment standard of care treatment,[5] however, realistic models, also called phantoms, can be cost prohibitive.[6] Many programs create homemade phantoms to save on the cost of procuring commercially made models. However, most homemade phantoms lack durability and many are made of perishable materials that can only be kept for a short time before spoiling.[7–12]

Many homemade phantoms are constructed from a gelatine formula with particulate suspension (e.g. flour or soluble fibre medications) to increase echogenicity. However, gelatine requires refrigeration, both to set the model when poured and to minimise spoilage. The use of Agar-Agar (henceforth called agar) in place of gelatine (henceforth called gel) may provide advantages in durability, ability to hot-set without refrigeration, reusability, and mimicry of human tissue. This research sought to demonstrate feasibility of hot-set, spoilage resistant agar models in constructing low-cost ultrasonographic training models as an alternative to commonly used gelatine models.

Methods

Model formulas were created to produce models with a percent-agar by weight formula (e.g. 10% agar by-weight). The appropriate amount of dry agar powder (i.e. 900 g/cm^2 strength, 38 g for a 5% model) was mixed into 750 mL of cold distilled water until the powder homogenised in suspension (Table 1).

This mixture was brought to a light boil for about five minutes or until the agar solubilised under periodic stirring. Additives (e.g. wheat flour, dyes, ethanol) were mixed in at this stage if required. Model pouring was done in two stages: the first pour set a base for the vessel analogues to be placed upon, and the second pour created a cap to overlie the vessels and act as the tissue analogue. About 300 mL of hot mix was poured into a standard container (8.5″Lx 6″Wx 2″D) to set the base, which set at room temperature (82 °F/28 °C) in about 15–18 min. During this time, the remaining hot mix was kept hot to prevent solidification.

Vessel analogues were created using a variety of water-filled Penrose drains and long latex balloons. These analogues were placed onto the set agar base before the cap was poured. The remaining hot mix, approximately 450 mL, was poured as the cap over the analogues and allowed to set for another 15–20 min. The cap is intended to place about 1–2 cm of agar between the surface of the vessel analogue and the surface of the cap. A nitrile glove with fingers cut off and split down the thumb-side to create a single nitrile sheet was used to cover the surface of the model to simulate skin and to provide a completely opaque layer to obscure the placement of objects in the model. Models were stored in a refrigerator to increase their longevity.

Agar models could be made using a microwave in addition to a stovetop. The same agar slurry was made to the desired percent by-mass and portioned into two equal components. The fractions were microwaved separately and used to pour the base and cap. Complete solubilisation of the agar took about four minutes in a 1.58 kW microwave, with stirring every 45–60 s to prevent settling out of the agar suspension. Volume loss from heating was minimal and estimated to be 5–10 mLs. No physical property discrepancies were noted between the microwave and stovetop agar models.

Agar matrix can be melted down and reused to create new models using either microwave or stovetop methods. Careful heating re-liquefies the matrix and allows the model to be re-poured, with its vessel analogues recycled or replaced as needed. Volume loss during recycling can be compensated for adding the volume of water that was lost during heating.

One-quarter teaspoon of red dye and one teaspoon of wheat flour were tested as possible additives to increase the opacity in separate 5% agar models. Ethanol was tested as a de-gassing agent to remove the bubbles observed in the

Table 1 The optimal recipe and steps needed to make one 5% agar phantom.

Materials	Water* (750 mL)
	Agar 900 g/cm^2 (38 g)
	Latex tube/Analogues
	Flour (1 teaspoon)
	Stove/Microwave
Steps	Mix 750 mL of cold water with 38 g of 900 g/cm^2 agar gel
	Stir until agar is suspended in water without clumps
	Briefly bring mixture to boil, stirring periodically
	Sprinkle 1 teaspoon of flour into mix, trying to avoid clumping
	Stir flour into mix until homogenised
	Using about half of the prepared mix, pour the base layer into mold
	Let set at room temperature for 20 min
	Prepare vessel analogues (tie ends and fill with water)
	Place analogues on base and pour remaining mix over them as the cap layer
	Let set at room temperature for 20 min (add glove layer if desired)

* Water cannot be boiled before addition of agar or the powder will irreversibly clump.

models. Liquid latex was tested as a possible skin analogue alternative to latex gloves.

Four aliquots of 100mL of the selected model mixes (10% agar, 7.5% agar, 5% agar and 10% gel) were mixed and poured into two separate trays. One tray with two replicates of each model mix was left open on a counter at room temperature (approximately 28 °C) for a week; the second was left for the same period uncovered in a refrigerator. A 5% gel was made in a covered container to monitor desiccation differences and also left on a counter at room temperature for a week.

All models were evaluated within five days of pouring and refrigeration for storage. Evaluation was in real-time using a Sonosite M-Turbo portable ultrasound with a high-frequency linear probe (HFL-38/13-6 MHz) by an attending emergency physician formally trained in emergency ultrasonography and a medical student, who were not blinded. An additional attending emergency physician reviewed images obtained from various models. The models were evaluated based on their echogenic properties, the presence of artefacts in the matrix, the ability to discern the lumen of the analogue vessels, and the ability to visualise venepuncture needle tips.

Figure 1 A completed 5% agar model with red dye in the matrix and two sets of vessel analogues.

Echogenicity, defined as the ability of the model matrix to generate a signal that accurately mimics the sonographic image generated by real tissue, was evaluated by visual comparison. Similarly, images generated by the models were visually examined to ensure that there were no significant artefacts affecting the quality of the generated image, and that the lumen of the analogue vessel was visually discernable from the surrounding matrix. These characteristics were subjectively evaluated in comparison to gel phantoms and real tissue.

The durability of the models was evaluated by a medical student placing repeated needle sticks within a 1 cm radius until the gel degenerated or significantly lost resistance to further sticks as determined by manual palpation. Degeneration was defined as a significant, palpable loss of resistance to further needle-sticks or fragmentation of the media to a point at which there was no ability to continue (i.e. complete fragmentation of media).
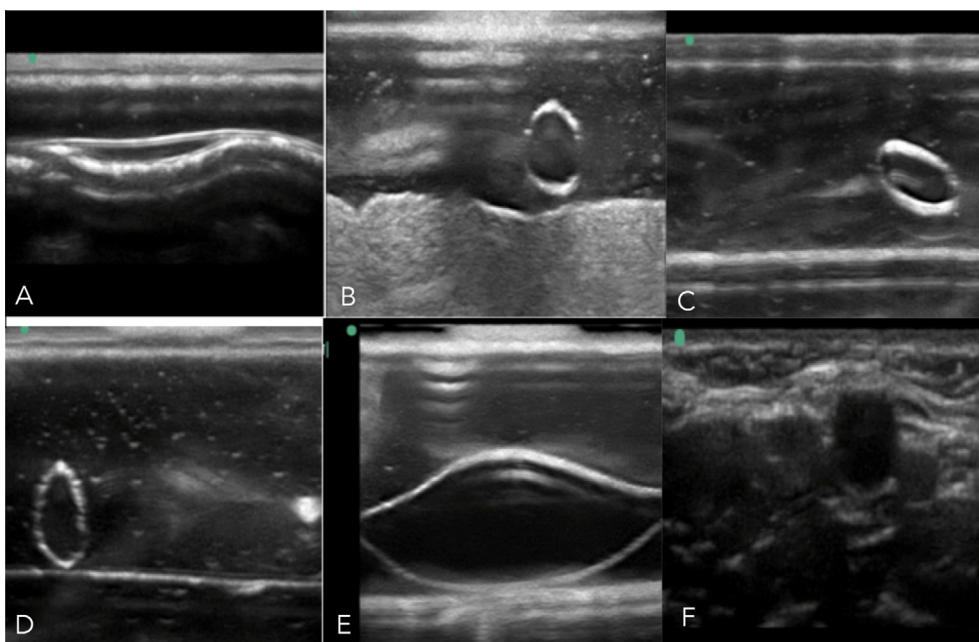
Results

Models were made with 10%, 7.5%, 5%, and 2.5% by-mass agar compositions and compared to 5% and 10% by-mass gelatine models (Fig. 1 and Table 2).

The proportion of agar in the model correlated with its density, opacity and echogenicity. The models did not require the use of ultrasound gel to produce high-quality images in which the analogues were clearly discernible and venepuncture training could be readily accomplished. It was observed that the higher-density models (10% and 7.5%) tended to create reverberation artefacts under ultrasound, but that this effect could be reduced with the addition of ultrasound gel. A hyperechoic boundary that delineated the border of the base and cap layers was observed in all agar models as in previously described gel models. All models were slightly hypoechoic when compared to actual tissue. The more dense models had the greatest durability, but subjectively lacked the realistic feel of tissue during needle insertion. Their density also prevented compression of the analogue vessels during the ultrasound. The lower density models had a more realistic tissue texture, and were readily compressed with a more realistic feel during needle insertion in the opinion of the two live evaluators. The gelatine models were much more fragile and compressible than tissue and were prone to rapid degeneration from needle sticks and heat exposure. The 10% gel model had greater durability and was denser (better resembled tissue) than the 5% gel model. The 5% agar model withstood an average of 20 sticks within a 1 cm radius before degeneration ($n = 3$), while the gel degenerated after an average of 3 ($n = 3$).

Table 2 Physical properties of different agar model mixes compared to gelatin controls and tissue ideal.

Model	Density	Opacity	Echogenicity	Reverberation	Artefacts	Compressibility	Durability
10% Agar	+ + + +	+ + +	+ +	+ +	Base/Cap layer	+ +	+ + + +
7.5% Agar	+ + +	+ + +	+ +	+ +	Base/Cap layer	+ +	+ + + +
5% Agar	+ + +	+ +	+ +	+	Base/Cap layer	+ + +	+ + +
2.5% Agar	+	+	+	+	Bubbles in agar, base/Cap layer	+ + + +	+ +
5% Gelatine	+	+	+	+	Base/Cap layer	+ + + + +	+
10% Gelatine	+ +	+	+	+	Base/Cap layer	+ + + + +	+ +
Tissue	+ +	+ + + +	+ + +	–	–	+ + + +	+ + + +

Figure 2 The ultrasound images generated by the models. (A) 10% agar, (B) 7.5% agar, (C) 5% agar, (D) 2.5% agar, (E) 5% gel and (F) live tissues (brachial vessels).
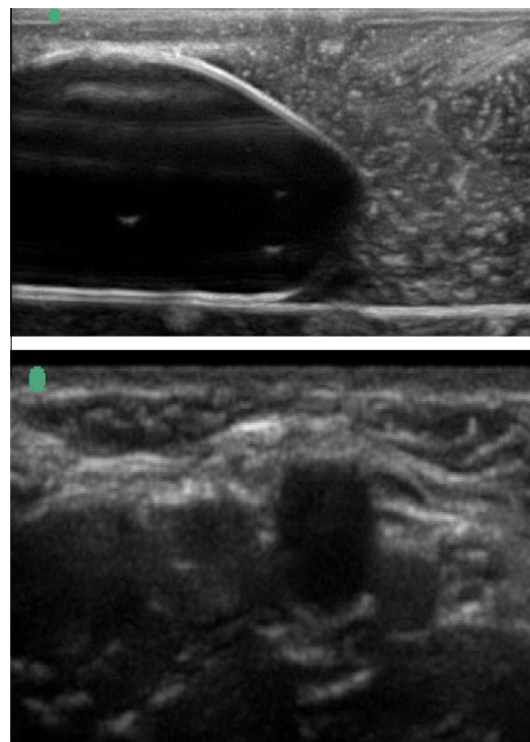
The agar models allowed for clear visualisation of introduced foreign bodies (metal or plastic objects, such as the tip of a needle) without ultrasound gel. The models did not introduce any artefacts to distort the image or complicate the viewed layout other than the previously mentioned reverberation artefact, which could be remedied by the addition of ultrasound gel and tended to be present only in the denser models (Fig. 2). Track artefacts (from the insertion of the needle) were evident in all models, and would likely be an additional limiting factor to the number of uses a model could withstand.

The agar spoilage samples lasted one week uncovered at room temperature without any change in texture or appearance. Slight desiccation was noted (the models shrunk by about 0.5 cm vertically) in the agar samples left at room temperature, however this effect was minimal in the refrigerated samples. No desiccation was noted in the 5% agar sample left covered at room temperature (approximately 82 °F, 28 °C). There was no notable change in the ultrasound signal generated by the gel models that were left at room temperature or refrigerated. The room temperature 10% gel samples began to degenerate notably on the fourth day, and were completely liquefied by the seventh. These samples also exhibited the growth of mould colonies (1 cm in diameter or smaller), which the agar samples lacked. The 10% gel samples that were refrigerated exhibited no notable changes in texture or appearance.

The 10% agar model that was recycled and re-poured demonstrated an ultrasound signal that was slightly hyperechoic compared to the signal it had generated before recycling. This is likely due to the trapping of gas within the matrix from renewed boiling and slight concentration from water loss. It is likely that this could be avoided by adding more water to compensate for losses.

Red dye greatly increased the opacity of the agar matrix, but the analogue vessels were still readily visible in the model. The dye did not alter the ultrasound signal. Wheat flour did

not significantly change the opacity of the matrix, but greatly increased the echogenicity of the matrix under ultrasound (Fig. 3), giving it greater fidelity to real tissue. The flour was prone to clumping, which created small imperfection artefacts

Figure 3 This compares the ultrasound image generated by the 5% agar model with added flour (top image) to actual tissue (bottom image). Actual tissue illustrates the brachial vessels.

under imaging. Ethanol proved an effective degassing agent, but the loss of the gas micro-bubbles resulted in a reduction of echogenicity of the matrix. The liquid latex provided an opaque skin analogue that was able to provide clear ultrasound images without the use of ultrasound gel. However, the latex took 4 days to set on the model surface at room temperature and tended to separate from the matrix upon needlestick.

The construction of models with agar on average cost more than using gelatine. With materials purchased through Amazon.com, the gelatine for a 5% model cost approximately 2.5 USD (33 ZAR) and agar for a single 5% model (50 g) cost approximately 5 USD (66 ZAR). Sourced through online suppliers in Cape Town, South Africa, gelatine for one model would cost 1.50 USD (20 ZAR)[13] and agar 3.60 USD (48 ZAR).[14] Though more expensive, agar delivers a higher quality model that is temperature stable and mimics tissue with fewer additives and at lower concentrations (thus less is needed to make a quality model) with the added benefit of durability. Our testing demonstrated that agar could be reused at least once, and likely more times if lost water were replaced. Thus, costs ultimately may favour an agar model as well.

Discussion

Currently, gelatine is used as the principle substrate for homemade models used in ultrasound training. This inherently requires that refrigeration is available and that gelatine is readily available in large quantities. These models are therefore less useful in environments that cannot provide refrigeration or readily supply new gelatine to replace the models as they wear out. Thus a substitute that is temperature stable, portable, resistant to spoiling and reusable over long periods could replace gelatine models in scenarios that do not suit the use of gelatine phantoms, such as in low to middle income countries. Agar fulfils these requirements and may be able to replace gelatine as a matrix in models. Agar, which is a vegan substitute for gelatine, can be found in specialty food stores and has been verified to be available from online sources or local markets in South Africa, India, Southeast Asia, Europe and the United States amongst many others.

When the agar models are visually compared between themselves, gelatine models and live tissue, the agar models represent excellent approximations of what would be seen on an ultrasound-guided procedure that involves placing a needle into a vessel lumen. The 10 and 7.5% agar models both exhibited significant reverberation artefacts, and although this could be mostly remedied with the addition of ultrasound gel, the artefacts still reduced the quality of the image. In addition, investigators felt the higher density models failed to resemble real tissue in terms of texture and compressibility. The 2.5% agar model and the 5% gel model both generated useful ultrasound images, but were much less durable and tended to be overly distensible (did not mimic tissue as well as possible). The 5% agar model provided an ideal texture and distensibility (mimicked tissue well) which minimised the amount of reverberation artefact generated. Additionally, the 5% models required less agar per model, and are thus more economical. The 10% gel model resembled real tissue much more than

the 5% gel model (in texture and distensibility), but generated a very hypoechoic signal.

The use of nitrile gloves as a skin surrogate provided a very simple and accessible component that hid the placement of vessel analogues within the model while permitting ultrasound transmission without ultrasound gel. This conduction may be due to the trapping of a layer of moisture between the glove and surface of the agar. Both the Penrose drains and latex balloons proved to be proficient blood vessel analogues, though the latex balloons tended to be less durable and more likely to leak after multiple punctures. The larger diameter vessel analogues were found to be more compressible in all models, and would thus likely make better analogues of veins. The small diameter analogues were less distensible, better mimicking arterial vessels.

The addition of flour to the agar mix greatly increased the echogenicity of the model, such that it very closely resembled the ultrasound signal generated by real tissue layers (muscle, connective tissue, etc.). Though the flour was prone to clumping, vigorous mixing minimised the degree of precipitation. In fact, the small imperfections caused by the clumps helped to mimic the irregularity of real tissue.

Though the agar and gel models both produced similar ultrasound images, the agar models had distinct advantages in several areas. The agar models were much more durable than the gel, and were able to withstand more needle sticks. The agar was also more temperature stable and did not require refrigeration at any time, which allowed the models to hot set (at room temperature) rapidly. This rapid-setting time allowed models to be made faster, and caused fewer complications with analogue vessel placement (analogues tended to shift frequently during the settling of the gel models). The agar mix is more temperature stable, does not require refrigeration to be stored, and is more resistant to spoiling than gel, which allows it to be employed in more adverse conditions. The agar models can be stored at room temperature without desiccation when covered or sealed in a bag, which allows them to be transported very easily over long periods of time without deterioration. The agar mix was able to be melted down and reused once a model had deteriorated due to use (needle track artefacts were the limiting factor of durability). Overall, the agar model can provide a high-volume practice model that may be made, set and stored in adverse conditions, with limited resources and then reused to conserve materials.

Agar models have some disadvantages in comparison to gel models. Agar tends to be more expensive than gelatine per gram (about twice as expensive) in the United States and can be somewhat harder to procure, though it is readily available through online sources. However, this may be offset by the increased durability and reusability of the agar matrix. Both gel and agar models exhibited the ability to generate ultrasound signals without ultrasound gel and distinct layering artefacts at the base-cap boundary.

This feasibility study aimed to demonstrate that agar is an acceptable media, similar to currently used gelatine, with the additional benefits of the ability to hot-set and to resist spoilage. A small number of reviewers evaluated images produced using the phantom in an unblinded, qualitative manner. Quantitative data on durability and spoilage were limited to extremely small samples.

Future research could include blinded evaluations by trainees, EPs and radiologists to better evaluate acceptability of

images amongst a range of users. Assessment of trainees utilising these phantoms could be employed to determine the appropriateness of training provided for real-time venous access procedures after they have had the opportunity to complete several procedures on live patients. Further, future research to identify additional uses for agar-based ultrasound task trainers and to determine their longevity would be useful to those developing their own ultrasound training programs.

Overall, agar represents an exciting alternative to gel for the construction of low-cost, reusable ultrasound training phantoms. The ability of the models (ideally, the 5% agar with flour) to mimic the texture and ultrasound image generated by real tissue can allow for realistic, hands-on training with ultrasound guided venepuncture procedures. Obtaining intravenous access is pivotal in resuscitative efforts and patient care and is sometimes challenging for practitioners. We believe that the agar models allow for such practice. Other applications may include training on locating and removing small foreign bodies using ultrasound, or the construction of more complex models to simulate procedures like thoracentesis or suprapubic cystotomies. Agar models may be most applicable in scenarios where limited supplies are available, and models must be able to be constructed and stored without reliable refrigeration. The use of improvised materials in the models (nitrile gloves, Penrose drains, latex balloons) further increases the ability of the model to be improvised upon and used in non-traditional or austere environments.

Dissemination of results

This model will be presented at the University of Florida College of Medicine Medical Student Research Fair and has been submitted for presentation at International Emergency Medicine Congresses. Variations of the model have been informally utilised in several developing EM settings including Centre Hopspitalier Universitare, Kigali, in Kigali, Rwanda.

Author contribution

E.D. conceived the study, acquired materials and approved procedures. G.D. aided in acquisition of data and editing images. M.E. designed procedures, constructed models and acquired data and images. All authors contributed to drafting and revision of manuscript and approved the final version for accuracy and integrity.

Conflict of interest

The authors declare no conflict of interest.

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