



DEATH IN THE MOUTH: THE CURRENT STATUS OF PESTICIDE RESIDUES IN CURED FISH FROM SELECTED MARKETS IN RIVERS STATE NIGERIA

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ABSTRACT

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Pesticide residues in cured fish from selected markets in Rivers State, Nigeria were determined to compare their residues content with FAO/WHO MRLs. The research was carried out at Ignatius Ajuru University of Education and Austino Research Laboratories, Rivers State, Nigeria. Samples of cured fish were collected from different major markets in Rivers State, Nigeria. Each product was purchased from three different traders and kept in an air tight Ziploc envelope containing general information. Some of the samples were kept in 250 g plastic containers and kept on a laboratory bench for a period of 45 days to observe insect emergence while the other lots were taken to Austino Research Laboratory for pesticide residue analysis. The result shows varying degrees of damage ranging from slightly to undamaged and mean number of *D. maculatus* emergence was higher in smoked fish samples obtained at Rumuokwuta. The result also shows that in all the cured fish sampled, DDT, Captan, Hexazinone, and Tecnazene were found above the FAO/WHO MRLs and few compounds were below the MRLs. Conclusively, relevant authorities and agencies needs to increase surveillance and monitoring to check and control the illegal importation, sale and use of such compounds especially in controlling stored pests of cured fish. This could reduce cases of pesticide toxicity and food poisoning and other health implications due to its ingestion in such contaminated cured fish.

Contribution/Originality: The objective of this research therefore was to determine the levels of pesticide residues in cured fish sold in selected markets in Rivers State Nigeria and to compare their residues content with FAO/WHO MRLs.

1. INTRODUCTION

The sharp increase in global human population combined with increasing standards of living with much awareness on healthy diet has led the demand for animal-derived protein such as fish [1]. While human population in sub-Saharan Africa is growing at 3% per annum its food production is growing 1.5% pa [2] and currently none of the African countries is included in the list of the high Human Development Index- which is a measure of the quality of life; rather they were all ranked among the low Human Development Index United Nations Environment Programme [3]. To meet up with this food deficit especially daily protein intake, many people in Nigeria and some parts of Africa now engage in fish farming which is fast becoming a lucrative agro-business with high promises [4] and may seem to be an alternative to meeting future protein source of man in order to enhance global hunger index Boland, et al. [5]; Cashion, et al. [6]. Ajayi, et al. [7] stated that fish protein compares favourably with other animal

protein sources such as egg, meat and milk in terms of essential amino acids, lipids, vitamins, lysine methionine and other medicinal values Guizani, *et al.* [8]. Regular fish intake have been reported by various scientist to be both curative and preventive of several human diseases such as cardiovascular diseases, cancers, rheumatoid arthritis, inflammation [9, 10] and also regulate body water balance, bones formation and other metabolic reactions [11].

Fish once captured starts to deteriorate immediately; therefore, it is either consumed immediately or processed in order to extend its shelf life. Captured fish is known to be highly susceptible to insect pest infestation which commences from processing to storage thereby leading to losses attributed to net reductions in the amount of nutrients available to the consumer (nutritive quality) to declining consumer acceptability and market prices (economic losses) or both quantitative and qualitative losses [12]. *Dermestes maculatus* and *Necrobia rufipes* are key postharvest pest of smoked fish and both adults and larvae are capable causing damage [7] with infestation put in the range of 75 - 93% [13, 14]. Both processors and marketers of cured fish products in Nigeria are thereby faced with major challenges such as poor handling, packaging, marketing outlets and transportation [15] short storage duration and lack or expensive storage and processing facilities. In an attempt to reduce this high risk, fish merchants use several means within their reach in protecting the fish from pest infestation, disease infection and other deteriorating agents by irradiating [12] use of degradable and less toxic biopesticides with high repellency, synthetic pesticides [16] sufficiently toxic to the targeted insect pests and not leaving residues harmful to the consumer [17]. Only few insecticides are able meet up these criteria [16] therefore, the poor resources farmers and fish handlers in the tropics resolve to sun drying, salting and smoking where 45% of total fish catch in Nigeria is utilized as smoked fish [18]. Any food stuff to be acceptable for consumption must be free from insect exuvia, off-smell, unusual colour and mycotoxins and above all must not contain active substances of pesticides above the maximum residue limits (MRLs) [19]. Consuming pesticides in any form could lead to acute or chronic discomfort to carcinogenic, endocrine, disruption and organic disorder. Therefore, it makes it important to checked for permissible concentration of residues in food such as fish and grain [19, 20] since its accumulation/concentration down the food chain starts from the point of application both during processing or storage and up to the point when food is first offered for consumption Akunyili and Ivbijaro [21]. FAO/WHO [22] is of the view that pesticide toxicity does not differentiate between targeted and non-targeted species and its use especially in storage of food stuff should be handled with care. The highly stable compounds can last for several decades in the environment and are easily magnified through bioaccumulation and magnification with no global limitation. Pesticides released in one part of the world can be transported and deposited in other regions far away from the original source [23]. The objective of this research therefore was to determine the levels of pesticide residues in cured fish sold in selected markets in Rivers State Nigeria and to compare their residues content with FAO/WHO MRLs.

2. MATERIALS AND METHOD

2.1. Sample Collection

The research was carried out at Ignatius Ajuru University of Education and Austino Research Laboratories, Rivers State, Nigeria. Cured fish samples were collected from (Creek Road market, Mile 1 market, Mile III market) in Port-Harcourt Local Government Area (PHALGA) and (Rumuokwuta market, Choba market, Rumuokoro market and Rumuokoro slaughter market) in Obio-Akpor Local Government Area (OBALGA). In each market, products were purchased from three different traders and such cured fish were kept in an air tight Ziploc envelope containing information such as market place, produce, date of purchase and conveyed to laboratory. Some of the samples were kept in 250 g plastic containers arranged in Completely Randomized Design (CRD) and replicated three times and kept on a laboratory bench for a period of about 45 days to observe insect emergence, while the remaining lots were taken to Austino Research Laboratory for pesticide residue analysis before and after four months of storage in the laboratory.

2.2. Chemicals and Reagents

The chemicals, reagents and solvents used in the analysis included; Air-Zero grade, Nitrogen gas -UHP grade, ethylene chloride, anhydrous sodium sulfate, n-hexane and the standard of the pesticide used was internal standard.

2.3. Preparation of Samples

A 20g aliquot of sample was homogenized and a 10 g aliquot was spiked. The sample was mixed with anhydrous sodium sulfate and was allowed to dry for about 30 minutes minimum. The samples were extracted within 18-24 hours using methylene chloride in a soxhlet extractor. The extract was evaporated to dryness and the lipid content was determined. Duplicate portions of 10 g of the samples were stored as cured fish in a well labeled glass bottles and stored in a refrigerator as backup samples.

2.4. Extraction and Clean-Up of Samples

Pesticide residues in sample were extracted using established methods though with slight modifications. In brief, extracts for pesticide analysis were subjected to a sequential methylene chloride-n-hexane (1:1) clean-up specifically for the analyses. 40 microL of the each sample was injected into a gas chromatograph, equipped with a wide-bore fused -silica capillary column in an Electron Capture Detector (ECD). The lipid content extracted using a 2.5 mL air tight syringe at a volume of 1 microL was injected through the injection port into the injector. The samples in the injector were held at a temperature ramp of 250°C and detector at 280°C. At initial oven temperature of 200°C held for 1 min but subsequently increased to 230°C at 1.5°C min⁻¹ and then held for 10 mins.

3. ANALYSIS OF PESTICIDE RESIDUE CONTENT

3.1. GC-MS Conditions

All compounds were determined and quantified using an Agilent 6890 gas chromatograph with a 5973 Mass Selective detector (GC-MS) 30 m × 0.25mm, i.d. fused silica capillary column chemically bonded with SE-54 (DB-5), 1 microm film thickness. The carrier gas was nitrogen at a low rate of 1 mL min⁻¹ using manual injection at injection volume of 1 microL. The split ratio was 50:1 and the sample size was 1 microL. Oven temperatures was maintained initially at 200°C held for 1 min and heated to 230°C (1.5°C min⁻¹) then held for 10 mins.

Table-1. Classification of damage in sampled stock and smoked catfish from selected major markets in Rivers State caused by *Dermestes maculatus* after 30 days of storage.

Cured fish	Creek Road	Mile Iii	Mile I	Rumuokwuta	Rumuokoro	Rumuokoro Slaughter	Choba
Stockfish	Slight damage	Slight damage	Slight- moderate damage	Slight damage	Undamaged	Undamaged	Moderate damage
Catfish	Severe damage	Undamaged	Undamaged	Slight damage	Undamaged	Slight- moderate damage	Undamaged

Table-2. *D. maculatus* emergence in cured fish sampled from different markets in Rivers State and kept in a laboratory for 45 days

Selected major Markets	Stockfish	Smoked catfish	Total
<i>D. maculatus</i> emergence after 35 days of storage in the laboratory			
Creek Road	2	48	50
Mile 1	35	0	35
Mile 3	2	0	2
Rumuokwuta	20	150	170
Rumuokoro	0	0	0
Rumuokoro slaughter	1	8	9
Choba	71	0	71
Total	131	206	337
Mean ±SE	18.71±2.03	29.42±3.16	48.14±2.12

Table-3. Pesticide residues in stocked fish and smoked fish sampled from selected major market in River State, Nigeria.

Products		Captan	Chlordane Alpha (Cis)	Chrothalonil	DDT O P-	DDT P P-	Endosulphan-alpha	Endosulphan-Sulfate	Hexachlorobenzene	Methoxychor	Octachlorostyrene	Perthane	Quintozene	Tecnazene	Diazinon	Atrazine	Hexazinone	Malathion	DDE O,P	BHC1 Alpha	BHC1 beta	BHC delta	Simazine	Cyanazine
Choba Market																								
STOCKFISH	Before	0.10	0.13	0.02	0.08	0.09	0.15	0.07	0.09	0.01	0.06	0.03	0.19	0.23	0.07	0.11	0.25	0.15	0.02	0.19	0.04	0.02	0.10	0.18
	After M.R.L	0.02 0.07	0.06	- 0.81	0.06 0.05	0.06 0.05	0.03	0.02	0.04	-	0.06	0.09	0.11 0.64	0.13 1.15	0.05 0.22	0.08	0.16 0.01	0.10	- 0.05	0.16	0.02	0.05	0.03	0.11
CAT FISH	Before	0.03	0.08	0.02	0.16	0.12	0.07	0.15	0.08	0.06	0.09	0.03	0.13	0.10	0.02	0.09	0.07	0.03	0.14	0.06	0.17	0.10	0.21	0.05
	After M.R.L	- 0.07	0.06	0.03 0.81	0.06 0.05	0.05 0.05	0.05	0.03	0.04	0.01	0.05	-	0.05 0.64	0.03 1.15	- 0.22	0.07	0.03 0.01	0.05	0.03 0.05	0.05	0.11	0.04	0.15	-
Rumuokoro Market																								
STOCKFISH	Before	0.08	0.13	0.02	0.06	0.06	0.15	0.05	0.12	0.01	0.04	0.04	0.19	0.21	0.10	0.11	0.23	0.15	0.02	0.21	0.04	0.02	0.11	0.18
	After M.R.L	0.04 0.07	0.06	- 0.81	0.08 0.05	0.03 0.05	0.03	0.05	0.04	-	0.06	0.07	0.13 0.64	0.13 1.15	0.07 0.22	0.08	0.16 0.01	0.12	-	0.16	0.04	0.05	0.05	0.09
CAT FISH	Before	0.03	0.06	0.03	0.16	0.12	0.07	0.10	0.10	0.06	0.09	0.03	0.15	0.10	0.05	0.09	0.07	0.06	0.08	0.06	0.17	0.08	0.21	0.03
	After M.R.L	- 0.07	0.08	0.05 0.81	0.06 0.05	0.07 0.05	0.05	0.03	0.04	0.02	0.05	-	0.07 0.64	0.05 1.15	- 0.22	0.07	0.03 0.01	0.07	0.02 0.05	0.05	0.11	0.06	0.15	-
Rumuokwuta Market																								
STOCKFISH	Before	0.08	0.15	0.02	0.06	0.03	0.15	0.05	0.09	0.01	0.08	0.03	0.19	0.21	0.07	0.11	0.23	0.15	0.04	0.19	0.04	0.06	0.10	0.18
	After M.R.L	0.02 0.07	0.06	ND 0.81	0.08 0.05	0.06 0.05	0.03	0.04	0.04	ND	0.06	0.07	0.09 0.64	0.13 1.15	0.05 0.22	0.08	0.19 0.01	0.12	ND 0.05	0.17	0.02	0.03	0.03	0.09
CAT FISH	Before	0.10	0.06	0.02	0.16	0.12	0.07	0.10	0.08	0.10	0.09	0.07	0.13	0.10	0.02	0.09	0.07	0.03	0.08	0.17	0.17	0.08	0.21	0.03
	After M.R.L	ND 0.07	0.08	0.03 0.81	0.06	0.07	0.05	0.03	0.04	0.01	0.05	ND	0.07 0.64	0.03 1.15	ND 0.05	0.07	0.03 0.01	0.05	0.02 0.05	0.05	0.11	0.04	0.15	ND
Mile III Market																								
STOCKFISH	Before	0.08	0.13	0.02	0.06	0.03	0.15	0.05	0.09	0.01	0.04	0.03	0.19	0.21	0.07	0.11	0.23	0.15	0.02	0.19	0.04	0.02	0.10	0.18
	After M.R.L	0.02 0.07	0.06	- 0.81	0.08 0.05	0.06 0.05	0.03	0.02	0.04	-	0.06	0.07	0.11 0.64	0.13 1.15	0.05 0.22	0.08	0.16 0.01	0.12	- 0.05	0.16	0.02	0.05	0.03	0.09
CAT FISH	Before	0.03	0.06	0.02	0.16	0.12	0.07	0.08	0.08	0.06	0.11	0.03	0.15	0.10	0.02	0.09	0.09	0.03	0.08	0.06	0.19	0.05	0.21	0.03
	After M.R.L	- 0.07	0.08	0.03 0.81	0.06 0.05	0.07 0.05	0.05	0.03	0.04	0.01	0.05	-	0.07 0.64	0.03 1.15	- 0.22	0.07	0.03 0.01	0.05	0.02 0.05	0.05	0.11	0.04	0.15	-
Mile I Market																								
STOCKFISH	Before	0.08	0.13	0.03	0.06	0.03	0.15	0.10	0.09	0.01	0.04	0.03	0.24	0.21	0.07	0.11	0.23	0.15	0.06	0.19	0.04	0.07	0.10	0.18
	After M.R.L	0.02 0.07	0.06	- 0.81	0.08 0.05	0.06 0.05	0.03	0.02	0.04	-	0.06	0.06	0.11 0.64	0.13 1.15	0.05 0.22	0.08	0.16 0.01	0.12	- 0.05	0.16	0.02	0.05	0.03	0.09
CAT FISH	Before	0.03	0.11	0.02	0.16	0.16	0.07	0.10	0.08	0.06	0.09	0.03	0.18	0.10	0.07	0.09	0.07	0.09	0.08	0.06	0.17	0.08	0.26	0.03
	After M.R.L	- 0.07	0.08	0.3 0.81	0.06 0.05	0.06 0.05	0.05	0.03	0.04	0.01	0.05	-	0.07 0.64	0.03 1.15	- 0.22	0.07	0.03 0.01	0.05	0.02 0.05	0.05	0.11	0.04	0.15	-
Rumuokoro Slaughter Market																								
STOCKFISH	Before	0.08	0.13	0.02	0.03	0.03	0.15	0.05	0.07	0.01	0.04	0.03	0.19	0.21	0.07	0.11	0.21	0.15	0.02	0.17	0.04	0.02	0.10	0.18
	After	0.03	0.06	-	0.08	0.06	0.03	0.04	0.04	-	0.08	0.07	0.13	0.13	0.05	0.08	0.16	0.12	-	0.16	0.03	0.05	0.05	0.09

	M.R.L	0.07		0.81	0.05	0.05							0.64	1.15	0.22		0.01		0.05					
CAT FISH	Before	0.14	0.23	0.08	0.05	0.01	0.22	0.14	0.03	0.14	0.09	0.17	0.02	0.01	0.11	0.06	0.16	0.27	0.34	0.18	0.10	0.04	0.02	0.11
	After	0.08	0.17	0.03	0.01	0.03	0.14	0.07	0.02	0.05	0.04	0.11	-	-	0.04	0.01	0.10	0.17	0.19	0.12	.	0.06	-	0.09
	M.R.L	0.07		0.81	0.05	0.05							0.64	1.15	0.22		0.01		0.05		0.04			
Creek Road Market																								
STOCKFISH	Before	0.08	0.13	0.02	0.06	0.03	0.15	0.05	0.12	0.01	0.04	0.04	0.19	0.21	0.10	0.11	0.13	0.15	0.02	0.21	0.04	0.02	0.11	0.18
	After	0.02	0.05	-	0.08	0.06	0.03	0.02	0.04	-	0.06	0.07	0.10	0.13	0.05	0.08	0.16	0.12	-	0.16	0.02	0.05	0.03	0.07
	M.R.L	0.07		0.81	0.05	0.05							0.64	1.15	0.22		0.01		0.05					
CAT FISH	Before	0.17	0.12	0.04	0.12	0.06	0.11	0.15	0.02	0.16	0.06	0.05	0.19	0.33	0.13	0.10	0.36	0.12	0.16	0.10	0.17	0.02	0.04-	0.03
	After	0.13	0.06	0.09	0.03	0.01	0.05	0.08	0.03	0.21	0.08	0.02	0.11	0.21	0.10	0.12	0.23	0.07	0.10	0.05	0.09	-		0.06
	M.R.L	0.07		0.81	0.05	0.05							0.64	1.15	0.22		0.01		0.05					

3.2. Characterization and Identification

The characterization and identification of pesticides from the samples was completed in the m/z range varying from 35 to 450. The composition of the pesticides from the sample was determined using an Agilent 6820 gas chromatograph.

4. RESULTS

Table 1 shows the degree of damage of cured fish sampled at various major markets and kept for 45 days in the laboratories. From the damage scale adopted samples of smoked catfish collected from Creek road markets were severely damaged while other samples obtained from other major markets in Port Harcourt were undamaged. From the same result, stocked fish sampled were mostly slightly damaged with only samples obtained at Rumuokoro and Rumuokoro slaughter were undamaged. Table 2 shows *D. maculatus* emergence in cured fish sampled from different markets in Rivers State and kept in a laboratory for 45 days. Mean number of *D. maculatus* emergence was higher in smoked fish with highest emergence from samples obtained at Rumuokwuta and none from samples obtained from Miles 1 and 3 and Choba markets. *D. maculatus* emergence in stoked fish was highest in samples collected from Choba markets followed by samples collected from Mile 1 with least emergence in Rumuokoro slaughter.

Table 3 shows the pesticide residues in stocked and smoked fish sampled from selected major market in River State, Nigeria. Stocked fish sampled at Choba market showed higher values of Captan and DDT above the MRL while sampled smoked fish had higher levels of Methoxychlor, Hexazinone, and DDE OP above the MRL. Stocked fish sampled at Rumuokoro market showed higher levels of Captan, DDT, Tecnazene and Hexazinone above MRL while smoked cat fish had Chlorothalonil and Tecnazene above MRL. Samples of stocked fish and smoked fish sampled at Rumuokwuta market had higher levels of Captan, DDT, Tecnozene, Endosulphane, Hexazinone and Captan and Hexazinone respectively. Similar results were obtained in the other fish samples analyzed from the other markets. From the same table the result shows that in all the fish sampled in all the markets, DDT, Captan, Hexazinone, and Tecnazene were found above the MRLs and few compounds were below the MRLs.

5. DISCUSSION

The presence of skin beetle in all the fish sampled across the major markets in Rivers State may be attributed what Alabi, et al. [24] stated that food for man is also a food for the pests and infestation starts at any point and/or stage of food production. Although Zakka, et al. [25] reported 15 species of insects as major pests on cured fish, hide beetles (Coleoptera: Dermestidae and Cleridae) remains the most abundant among them all, this explains why it was the only species found in the fish sampled. Eke, et al. [26] described *D. maculatus* also a highly destructive pest of protein based food which include smoked fish, dried meat and blood meal, bacon and cheese as well as any material containing steroids such as leather goods, poultry feeds and allied produce. Therefore its management is an essential component of any effort to increase food production and thus ensure food security. Haines and Rees [27] earlier reported *D. maculatus* to be a major pest of processed fish and both the adults and the larvae of the beetles feed thereby causing between 50-100 % quantitative and qualitative losses of the edible fish during storage [28, 29]. Fish traders in order to avert this colossal damage tend to adopt the use of insecticides of different types or class as reported by Alabi, et al. [24] dating pest control using synthetic insecticide to 19th century to provide means of reducing pest populations. The residues are most a time left on the fish and increase along food chain either due to biomagnifications or bioaccumulation process. Although various degree of damage which ranged from slightly to moderately damaged were recorded, agrees with the works of earlier scientists that application of insecticide does not preclude total insect infestation [30-32] and complete reliance on pesticides for insect pest management does not guarantee food security. The presence of different chemical residues found in smoked and stock fish sold in major markets in Rivers State in Nigeria may be due to the fact fish is prone to high insect pest infestation once captured. Therefore fish merchants tend to use heavy doses of chemicals to confer protection from

insect pest attack so as to reduce loss which when left unchecked could result to huge economic and nutritional loss [20]. Earlier Ogah, et al. [33] reported similar result on cowpea samples containing residues of one or more organochlorine pesticides exceeding the Maximum Residual Levels (MRLs) with some exceeding with up to 8%. The danger in consuming such was also highlighted by Akinneye, et al. [19] that when pesticide residue are found to exceed the MRL in any food, such commodity is said to be unsafe for human consumption and may usually suffers set back at international markets for lacking to meet international standards or standards of the receiving country. On the contrary Ogah, et al. [33] argued that the presence of pesticide residue in food below MRLs may not likely cause significant health hazards even on a long term basis, including the hardcore insecticides like DDT, endosulfan and endrin once they are below their maximum permissible intakes (MPIs). Egbecho, et al. [20] stated the danger of such assumption where other factors such as synergistic effects, bio accumulation and bio-magnification theories are not factored into the equation. And health challenges resulting from intake of such at whatever dose may include its ability to interface with other drugs and/or food which could be carcinogenic, neurotoxic and teratogenic and should not be compromised for whatever reason [33-35].

However, the presence of residual pesticide compounds in the processed fish agrees with similar findings by Anzene, et al. [36] where Lindane and Aldrin, Bakore, et al. [37] and Riaz, et al. [38] DDTs, and its metabolites and γ -HCH, Heptachlor, Aldrin, Endosulfan and Diazinon, Chlorpyrifos methyl and Pirimiphos methyl in stored grains were above the MRLs. The presence of these toxic substances found in cured fish in major markets that serves majority of the populations in the metropolis and environs calls for the attention of relevant monitoring authorities. Ogah, et al. [33] and Egbecho, et al. [20] made similar call on National Food and Drug Agency and Control (NAFDAC) to monitor the presence of such pesticide residues in cured fish and all other food stuff in the market. Akunyili and Ivbijaro [21] stated that these pesticides have been banned in Nigeria and most countries of the world and its mere presence in whatever form in food stuff calls for strict monitoring to ban its manufacture and/or importation into the country. Egbecho, et al. [20] in a similar report called on government to enforce the rule of prior informed consent (PIC) FAO of UN that countries that wish to import banned or severely restricted pesticides shall give the pesticide manufacturer or exporting country their formal consent before importation. They listed such pesticides to include carbon disulphide, hydrogen cyanide, ethylene dibromide, ethylene dichloride, ethylene oxide, methylbromide, trichloroacetonitrile, chloropicrin, aluminium phosphide and aluminium sulphide. Others include Aldrin, dieldrin, endrin, lindane, HCH, DDT, Chlordane, mercury compounds and 2, 4,5T [2, 4, 5] (trichloropheno-axyacetic acid) [21].

6. CONCLUSION

The result of this study shows that there is high incidence of pesticide residues in cured fish sold in the major markets in Rivers State, Nigeria. While few were below the MRLs majority of the chlorinated insecticides were above the MRLs. Relevant authorities needs to increase surveillance and monitoring to check and control the illegal importation, sale and use of such compounds especially in cured fish where some people sometimes eat raw before further processing such as cooking. This when enforced will help in reducing cases of pesticide toxicity and food poisoning and other health implications due to its ingestion and will ensure strict compliance with the Stockholm Convention 2001 which Nigeria is a signatory to.

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