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# ASSESSMENT OF THE IMPACTS OF TROUT FARMING ON WATER QUALITY USING MACRO INVERTEBRATES AS BIO- INDICATORS

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## ABSTRACT

The study was conducted to assess the impacts of trout farming on water quality using macro invertebrates as bio-indicators. Two trout farms were selected for the study, viz., Gandaki Trout Farm (GTF) in Kaski district and Fall & Trout Fish Farm (FTF) in Nuwakot district. Reference and impacted sites were selected in each trout farms from where macro invertebrates were collected and physico-chemical parameters were measured. Sorensen's Index and Multiple Site Similarity Index were calculated to compare the macro-invertebrate assemblages between the impact and the reference sites. Water quality classes were also calculated using macro invertebrate-based tool, NEPBIOS/ASPT scores. Altogether 24 families of macro invertebrates were observed at GTF and 12 families at FTF. The Sorensen's Similarity Index was greater than 0.5 between reference and impacted sites at GTF; whereas it was only 0.28 at FTF indicating comparatively low level of similarity. In addition, Multiple Site Similarity Index (0.64) at GTF also indicated high similarity between the macro invertebrate assemblages. Reference sites showed higher scores with higher diversity of macro invertebrates. Both farms had suitable water quality for trout (dissolved oxygen and temperature) and most of the physico-chemical parameters did not show significant differences except pH and turbidity at GTF probably due to small scale operation and production.

**Keywords:** Trout farms, Water quality, Macro invertebrates, Bio-indicators, Sorensen's index.

## INTRODUCTION

Fish farms and aquaculture are one of the fastest growing food industries in the world as they have proven to be a reliable source of fish for consumption (Mainstone *et al.*, 1989; FAO, 2014). Rainbow trout (*Oncorhynchus mykiss*) are considered as one of the most popular cultured fish species (FAO, 2014) which are unique among carnivorous fishes and are natives of rivers draining into the Pacific Ocean (Boughton *et al.*, 2006). They survive only in cold running waters requiring high protein feeds and well oxygenated water (MOAD, 2013). Trout culture has become an established tourism industry throughout the world in terms of consumption as well as recreational fishing (Barrington, 1983). Rainbow trout was introduced in Nepal only in 1969 from India and in 1971 from the United Kingdom and had to be re-introduced again from

Japan in 1988 (Swar, 2008). Rainbow trout farming has become popular in the country and is practised over several hilly districts of Nuwakot, Rasuwa, Kathmandu, Lalitpur, Dolakha, Sindhupalchowk, Kavrepalanchowk, Makwanpur, Dhading, Kaski and Mustang (MOAD, 2013).

Although fish farming has socio-economic benefits, it does have some environmental impacts as well (Namin *et al.*, 2013). The impacts are from pollutants such as the uneaten feed, faecal matter, soluble metabolites and excretion of the fish farms (Kendra, 1991; Guilpart *et al.*, 2012). These pollutants in turn alter water quality parameters such as Dissolved Oxygen (DO), Turbidity, Conductivity, Total Dissolved Solids (TDS), pH of the fish farms as well as other receiving water bodies (Noroozrajabi *et al.*, 2013). Apart from these, consumption of fishes treated with drugs and residual chemicals may

cause health impacts on humans too (Authman *et al.*, 2015). Therefore, sustainable management of fish farming requires water quality assessment and monitoring of the fish farms and the receiving water bodies.

Routine water quality impact assessments of trout farming and aquaculture involve physico-chemical as well as biological assessments (Noroozrajab *et al.*, 2013; Namin *et al.*, 2013). The most common physico-chemical parameters used for aquaculture impact assessment in water bodies include pH, temperature, dissolved oxygen (DO), electrical conductivity (EC), nitrate, and phosphate. A number of studies have revealed increase in the nitrate and phosphate concentrations and electrical conductivity at the inlets (Namin *et al.*, 2013) and decrease in DO at the outlets (Kirkagaç *et al.*, 2004). Additionally, macro-invertebrate assemblages have been used successfully for biological assessments in a number of studies (Namin *et al.*, 2013; Kirkagaç *et al.*, 2004). These studies have shown that the inlet and outlet assemblages differ with increase in the pollution tolerant taxa in the outlets. In Nepal, few studies have been conducted to assess the impacts of trout farming (Bhagat & Barat, 2016) but specific studies using macro invertebrates as indicators to assess trout farming has not been conducted yet. Therefore, this study aims to assess the impacts on the water quality of the trout farms due to trout farming primarily focusing on macro invertebrates as bio indicators.

## MATERIALS AND METHODS

### Study Area

The study was carried out in two trout farms of Nepal- Gandaki Trout Farm (GTF) in Kaski and Fall & Trout Village Fish Farm (FTF) in Nuwakot (Figure 1). Gandaki Trout Farm lies in Kaski district of Western Nepal in Sardikhola Village Development Committee (VDC)(28°19'44.19"N and 83 ° 58'41.46" E). The farm is one of the most successful trout farms in Kaski and is located within ACAP (Annapurna Conservation Area Project) region. The main source of water for this farm is the Bhurjung River (commonly called Bhurjung "khola"). Fall & Trout Village Fish Farm is located in Kakani VDC within Nuwakot district (27°48'50.48"N and 85°13'13.93"E). The source of water for this fish farm comes from a spring

and the land used for trout culture is slightly sloped.

### Sampling Sites

Three sampling sites were selected for GTF. The reference site for the GTF (Figure 2) was selected at the upstream of the farm (1.5 km upstream) at Bhurjung Khola and the two impact sites (GTFI and GTFIM) were chosen downstream within the farm (Figure 2). GTFI was chosen to look at the impact on the artificial runway whereas GTFIM for the impact of the trout fishery on natural flow. In case of Fall & Trout Village Fish Farm, only two sampling points were selected i.e. the Reference site (FTFR) upstream of the farm and the impact site (FTFIM) downstream of the raceways of the farm (Figure 3).

At each site, two macro invertebrate samples were collected and a composite sample was made. Therefore, a total of three composite samples from GTF and two composite samples from FTF were collected. In GTF, around 50 m stretch was considered at each sampling site.

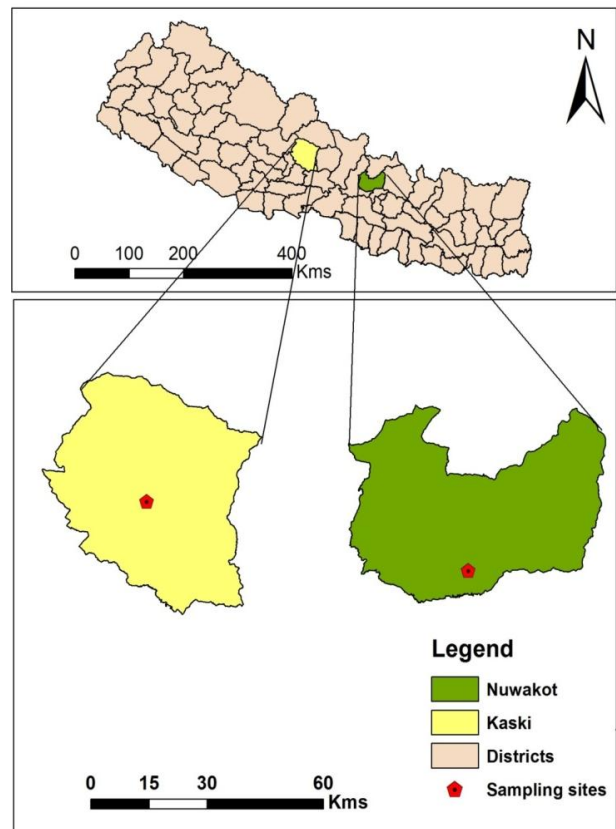
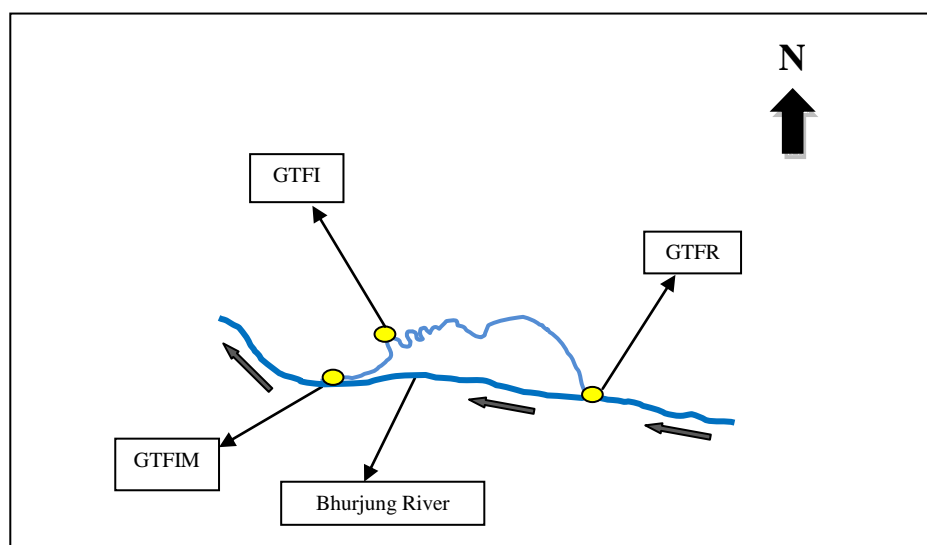
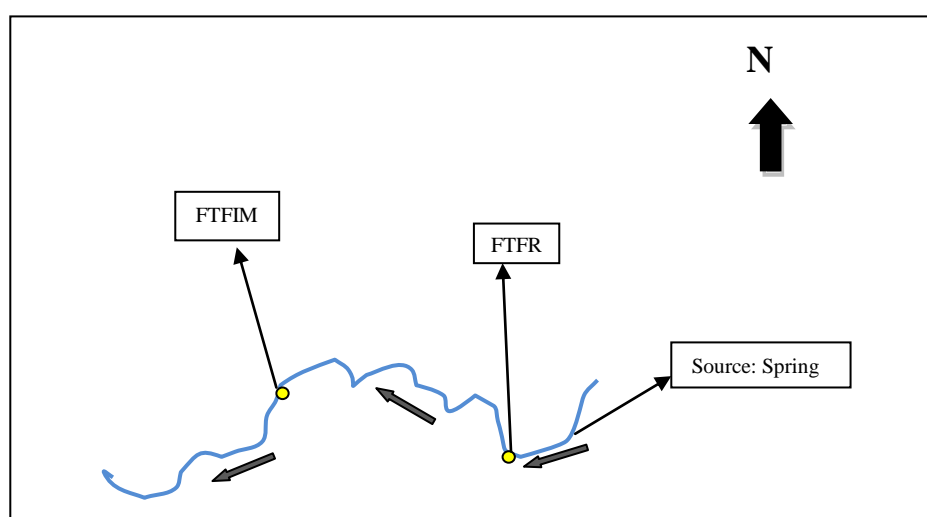


Fig. 1. Map showing the study sites.



*Fig. 2. Schematic diagram showing sampling sites of Gandaki Trout Farm.*



*Fig. 3. Schematic diagram showing sampling sites of Fall and Trout Village Fish Farm.*

### Sampling Method

Sampling was conducted in the month of June, 2016. Selected physico-chemical parameters such as pH, DO, conductivity, TDS, turbidity and temperature were measured on site using a portable kit (WagTech). Macro invertebrates were collected by Kick sampling method following Barbour (1999). The substrates were disturbed by kicking action using the heel to dislodge the macro invertebrates. Then the 250  $\mu\text{m}$  mesh sized nets were used to trap the macro invertebrates. Some benthic invertebrates were also picked from submerged stones and wooden debris with the help of forceps. Collected macro invertebrates were sorted in the field and were preserved in 70% Ethanol for further identification.

The mean values of the physico-chemical parameters were calculated using Windows Excel. Mann Whitney Rank Sum test was conducted to look at the variation between the reference and the impact sites using Sigma Plot version 12.3.

The macro invertebrates were sorted and identified up to Family level following relevant literatures and identification keys (Merritt & Cummins, 1996; Hartmann, 2007). The similarity between the macro-invertebrate assemblages of two sampling sites was assessed by calculating the Sorensen's Similarity Index which measures similarity in species composition for any two given sites (Magurran, 2004). This index can be calculated by using the following formula:

$$\text{Sorensen's Similarity Index} = \frac{2C}{A+B}$$

Where C = Number of Families that are common to the two sites undertaken

A = Number of Families in 1<sup>st</sup> site

B = Number of Families in 2<sup>nd</sup> site

The value of the Sorensen's Similarity Index ranges between 0 to 1. Values close to 1 indicate higher similarity and values close to 0 indicate lower similarity in assemblages from two sites (Magurran, 2004; Chao *et al.*, 2006) whereas 0 means no similarity and 1 means complete similarity between assemblages of two sites.

Similarly, the similarity between the macro invertebrate assemblages of three sampling sites was assessed by calculating the Multiple Site Similarity Index which measures similarity in species composition for any given sites (Diserud & Ødegaard, 2007). The index can be calculated using the following formula:

$$C_S^T = \frac{T}{T-1} \left( \frac{\sum_{i<j} a_{ij} - \sum_{i<j<k} a_{ijk} + \sum_{i<j<k<l} a_{ijkl} \dots}{\sum_i a_i} \right)$$

Where  $a_i$  is the number of species in site  $A_i$ ,  $i=1, \dots, T$ ;  $a_{ij}$  is the number of species shared by sites  $A_i$  and  $A_j$ ; and  $a_{ijk}$  is the number of species shared by sites  $A_i$ ,  $A_j$  and  $A_k$  etc. and the value ranges from 0-1, 1 indicating complete similarity and 0 indicating no similarity (Diserud & Ødegaard, 2007).

Water Quality Class (WQC) was calculated using NEPBIOS/ASPT (Nepalese Biotic Score/Average Score Per Taxon) (Sharma, 1996). This is a macro invertebrate-based tool where scores ranging from 1-10 are given to around 80 macro-invertebrate Families. Higher scores are given to pollution sensitive taxa whereas lower scores are assigned to pollution tolerant taxa (Sharma, 1996). The NEPBIOS/ASPT value is calculated by dividing the total scores of the organisms by the total number of taxa present at a particular sampling site as shown below:

$$\text{NEPBIOS/ASPT} = \frac{\sum_i \text{NEPBIOS Score}_i}{\text{Number of taxa}}$$

From the obtained NEPBIOS/ASPT values, WQC is calculated using the transformation table (Table 1).

**Table 1: Transformation table and corresponding water quality class.**

Water Quality Class (WQC)	NEPBIOS/ASPT Original scale	NEPBIOS/ ASPT for midland	NEPBIOS/ ASPT for lowland	Description
I	8.00-10.00	7.50-10.00	6.50-10.00	Not Polluted
I-II	7.00-7.99	6.51-7.49	6.00-6.49	Slightly Polluted
II	5.50-6.99	5.51-6.50	5.00-5.99	Moderately Polluted
II-III	4.00-5.49	4.51-5.50	4.00-4.99	Critically Polluted
III	2.50-3.99	3.51-4.50	2.50-3.99	Heavily Polluted
III-IV	1.09-2.49	2.01-3.50	1.01-2.49	Very Heavily Polluted
IV	1.00	1.00-2.00	1.00	Extremely Polluted

(Source: Sharma, 1996)

## RESULTS AND DISCUSSION

### Physico-chemical parameters

The results of the on-site estimation of the selected physico-chemical parameters are given in Table 2.

DO, conductivity and TDS did not show significant variation ( $p < 0.05$ ) between the reference and the impact sites in both trout farms. Although DO was higher in the reference sites, conductivity and TDS

were higher in the impact sites (except at GTFIM where conductivity and TDS were lower at the impact site) that could be due to increased dissolved salts from the seeds (Ghimire *et al.*, 2017). pH was lower and turbidity was higher in the impact sites than the reference site in both farms. However, Mann Whitney Rank Sum Test showed a significant variation ( $p < 0.05$ ) in pH and turbidity only at GTF.

**Table 2: Physico-chemical parameters in two trout farms (GTF and FTF)**

Site	Site Code	DO (mg/L)	Turbidity (NTU)	pH	Conductivity ( $\mu\text{S/cm}$ )	TDS (ppm)	Temperature ( $^{\circ}\text{C}$ )
Kaski	<b>GTFR</b>	9.7 $\pm$ 1.32	0.7 $\pm$ 0.76	8 $\pm$ 0.11	163.5 $\pm$ 40.16	84.1 $\pm$ 23.54	20.7 $\pm$ 2.72
	<b>GTFI</b>	9.2 $\pm$ 0.26	1.7 $\pm$ 0.48	7.9 $\pm$ 0.06	202.4 $\pm$ 1.89	100.8 $\pm$ 0.60	18.6 $\pm$ 0.43
	<b>GTFIM</b>	10.4 $\pm$ 0.14	8.6 $\pm$ 2.84	7.8	148.4	72.7	19.7 $\pm$ 0.14
Nuwakot	<b>FTFR</b>	11.2 $\pm$ 0.84	0.5 $\pm$ 0.03	7.6 $\pm$ 0.12	38.7 $\pm$ 1.69	19.4 $\pm$ 0.77	18.3 $\pm$ 0.83
	<b>FTFIM</b>	8.8 $\pm$ 1.35	3.2 $\pm$ 1.66	7 $\pm$ 0.12	43.2 $\pm$ 1.34	21.7 $\pm$ 0.56	19.8 $\pm$ 3.02

\***p**< 0.05

DO, pH, and water temperature are of crucial importance for the abundance of rainbow trout (Swales, 2006). They require high DO concentrations (Matthews & Berg, 1997) and concentrations greater than  $7\text{mgL}^{-1}$  is considered optimum for their survival (Raleigh *et al.*, 1984). Temperatures below  $21^{\circ}\text{C}$  ( $18.3\pm 0.83$  to  $20.7\pm 2.72^{\circ}\text{C}$ ) and high DO values ( $8.8\pm 1.35$  to  $11.2\pm 0.84\text{mgL}^{-1}$ ) in the studied fish farms indicate optimal conditions for the growth and survival of rainbow trout. The water for farming was brought to the farms through pipes with high velocity thereby facilitating mechanical aeration which could have attributed to higher DO values (Pulatsu *et al.*, 2004, Boaventura *et al.*, 1997). The pH in the study area

was alkaline ( $7\pm 0.12$  to  $8\pm 0.11$ ) and similar findings have been reported in other trout farms as well (Kirkagaç *et al.*, 2004; Namin *et al.*, 2013). Similarly, increase in turbidity in water bodies due to trout farming has also been reported in Iran too (Namin *et al.*, 2013; Noroozrajabi *et al.*, 2013). Increase in TDS and total suspended solids (TSS) in fish farms are from uneaten fish feed and the faecal matters of the fish (Guilpart *et al.*, 2012).

#### Macro invertebrate assemblages

A total of 27 Families of macro invertebrates belonging to 10 Orders were observed from five sampling sites of the two investigated farms (Table 3a and 3b).

**Table 3a: Macroinvertebrate Families from Gandaki Trout Farm.**

Site Codes	Macroinvertebrate Families
<b>GTFR*</b>	Elmidae, Dryopidae, Chironomidae, Simuliidae, Limoniidae, Baetidae, Ephemerellidae, Heptageniidae, Leptoceridae, Hydropsychidae, Stenopsychidae, Uenoidae, Philopotamidae, Gomphidae, Perlidae, Capniidae.
<b>GTFI</b>	Elmidae, Dryopidae, Chironomidae, Simuliidae, Limoniidae, Ceratopogonidae, Baetidae, Ephemerellidae, Planorbidae, Hydropsychidae, Planariidae, Sphaeriidae.
<b>GTFIM</b>	Elmidae, Dryopidae, Chironomidae, Simuliidae, Limoniidae, Muscidae, Baetidae, Ephemerellidae, Caenidae, Planorbidae, Lymnaeidae, Mesovallidae, Hydropsychidae, Planariidae.

\*Bhurjung Khola

**Table 3b: Macroinvertebrate Families from Fall and Trout Village Fish Farm.**

Site Code	Macroinvertebrate Families
<b>FTFR**</b>	Elmidae, Gyrinidae, Chironomidae, Limoniidae, Baetidae, Ephemeridae, Leptophlebiidae, Hydropsychidae, Philopotamidae, Gomphidae, Perlidae.
<b>FTFIM</b>	Chironomidae, Hydropsychidae, Planariidae.

\*\*Spring upstream of the Fall and Trout Village Fish Farm

The total number of macro invertebrate Families observed at GTF was 24 and at FTF were 12. Insect fauna formed the bulk of the macro-invertebrate assemblages with 23 Families in contrast to the non-insect fauna which was represented by only four families of Mollusca (Planorbidae, Lymnaeidae, Planariidae and Sphaeriidae). At GTF, site GTFR had the highest diversity with 16 macro invertebrate Families followed by site GTFIM (14 Families) and GTFI (12 Families) (Table 3a). Similarly, at FTF, the reference site FTFR was found to have more number of macro invertebrate Families (11 Families) in contrast to only three Families in the impact site (Table 3b). In both the farms, the reference sites had the highest number of macro invertebrate Families (Table 3a and 3b).

The taxon Perlidae was found only in the reference sites of both the farms. This taxon is considered to be pollution sensitive and is typical of cold running waters with high DO concentrations (DeWalt *et al.*, 2005). Chironomidae, Planariidae and Simuliidae were observed in the impact sites which are considered as pollution tolerant taxa (Hatami *et al.*, 2011) known to survive in low DO concentrations (Kirkagaç *et al.*, 2004). In contrast, the

abundance of pollution sensitive taxa such as Hydropsychidae and Philopotamidae have decreased in the impact sites in both the farms. The abundance of pollution sensitive and pollution tolerant taxa in the reference and impact sites respectively from our study have also been reported elsewhere (Namin *et al.*, 2013). The Sorenson's Similarity Index value of 0.57 (between GTFR and GTFI) and 0.53 (GTFR and GTFIM) are almost equal indicating a comparatively similar composition of macro invertebrate assemblages in the impact sites and the reference sites. The highest Sorensen's similarity index value of 0.76 was observed between GTFI and GTFIM indicating a comparatively higher similarity in assemblages between the impact sites. Similarly, the Sorensen's similarity index between the macro invertebrate Families observed at two sites (FTFR and FTFIM) of FTF was found to be 0.28 suggesting a comparatively low level of similarity of macro invertebrate assemblages between these two sites (Krebs, 2014). The Multiple Site Similarity Index (Table 4) between the macro invertebrate families observed at three sites of GTF was found to be 0.64 suggesting a high similarity between the assemblages.

**Table 4: Table showing the similarity indices between Macro invertebrate Families observed at different sites.**

Site Combinations	Similarity Indexes
GTFR, GTFI & GTFIM	0.64 (Multiple Site Similarity Index)
GTFR & GTFI	0.57 (Sorenson's Similarity Index)
GTFR & GTFIM	0.53 (Sorenson's Similarity Index)
GTFI & GTFIM	0.76 (Sorenson's Similarity Index)
FTFR & FTFIM	0.28 (Sorenson's Similarity Index)

**Water quality**

The result of the NEPBIOS/ASPT scores and corresponding WQC and water quality of the sites is given in Table 5.

The overall Water Quality Class ranged from I-II to II-III (Table 5). The NEPBIOS/ASPT score of site GTFR is 6.42 which indicate that the Water Quality Class (WQC) is II. Similarly, the WQC of sites GTFI, GTFIM and FTFIM are found to be II-III

and site FTFR has the WQC of I-II. On the basis of NEPBIOS/ASPT scores, the WQC distribution shows that the water of site GTFR is moderately polluted; sites GTFI, GTFIM and FTFIM have similar water quality and are found to be critically polluted whereas the site FTFR is slightly polluted. The NEPBIOS/ASPT scores of the sampling sites showed that the water of the impact sites was more polluted than that of the reference sites.

**Table 5: Table showing the Water Quality and Water Quality Class of different sites using NEPBIOS/ASPT scores.**

Site Code	NEPBIOS/ASPT Score	Water Quality Class	Water Quality
GTFR	6.42	II	Moderately polluted
GTFI	5.45	II-III	Critically polluted
GTFIM	5.3	II-III	Critically polluted
FTFR	7.27	I-II	Slightly polluted
FTFIM	4	II-III	Critically polluted

## CONCLUSION

The impact of trout farming on water quality based on physico-chemical parameters was not significant although small differences were observed in the outlets. In contrast, macro invertebrate assemblages differed in the reference and the impact sites and the difference was more pronounced in Fall and Trout Farm. Nevertheless, impacts detected in this study were not severe probably due to small scale production of the fish farms.

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