# **Poster Presentation**

# Comparison of EPT (Ephemeroptera, Plecoptera, Trichoptera) adult assemblage between old-growth natural forest and planted coniferous forest basins in Japanese temperate region

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**Abstract:** The purpose of this study is to clarify the differences of EPT (Ephemeroptera, Plecoptera, Trichoptera) adult assemblage between the two basins that have the most common types of Japanese headstream forests, i.e. an old-growth natural forest (ONF) and a planted coniferous forest (PCF) in the Kuroson stream in the Shimanto River basin. In the previous study, we found that aquatic invertebrate assemblages (larvae) were different between an old-growth natural forest (ONF) and a planted coniferous forest (PCF) basins. However, it was better to confirm the result in other way. Principal components analysis showed the differences also in EPT adult assemblage between the two basins in this study. Leaf litter, stream water quality, and light availability can differ depending on the forest vegetation. Because the difference of assemblage between the two basins was found both in larvae and adults in the same stream, it is supposed that the differences of forest vegetation would strongly relate with the difference of aquatic invertebrate assemblage.

Key Words: forest type, forest management, benthic invertebrates, EPT, old-growth forest, planted coniferous forest, adults, larvae

#### Introduction

Aquatic invertebrate assemblage is different depending on the surrounding land use such as forest, agriculture and pasture (Quinn et al. 1997; Townsend et al. 1997; Hall et al. 2001). Biotic integrity and habitat quality in the stream are increased when forest is prominent and decrease when agriculture are prominent as the surrounding land use (Roth, Allan and Erickson 1996).

Composition of riparian forest affect on the larval aquatic invertebrate assemblage. Leaf litter fallen into the stream could play several role for aquatic invertebrates assemblage depend on the litter species (Cummins et al. 1989). More plecopteran shredders are in the maple litters than in the pine litters (Whiles and Wallace 1997). Though riparian willow forest can provide few number of aquatic invertebrate (Lester et al. 1994), riparian red alder forest can provide nutrient-rich stream ecosystem (Volk et al. 2003) and support more aquatic invertebrate biomass than riparian conifer forest (Piccolo and Wipfli 2001).

Different type of forest provides different water quality for the stream (Friberg 1997; Friberg et al. 1997). So, composition of basin forest would be one of the most important variables affecting on the aquatic invertebrate assemblage. Recently I revealed the difference of larval aquatic invertebrate assemblage depend on the basin forest types (Yoshimura and Maeto 2006; Yoshimura 2007). These studies are focused on the larval assemblage. Adult assemblage of aquatic invertebrate might be different from that of larval one because they could fly. In this study, adult EPT (Ephemeroptera, Plecoptera, Trichoptera) assemblage were compared between the two basins that have the most common types of Japanese headstream forests, i.e. an old-growth natural forest and a planted coniferous forest.

#### **Materials and Methods**

#### Study site

The study area is located on the Shimanto River basin in Shikoku Island, Japan. In the upper reaches of Kuroson stream, a tributary of the Shimanto River, one old-growth natural forest (ONF) basin (33°10′N, 132°38′E) and one planted coniferous forest (PCF) basin (33°09′N, 132°39′E) were selected (Fig. 1). There is no residential area and cutting area in both basins.

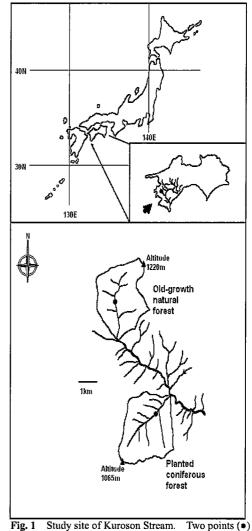
#### Basin and riparian forest

The vegetation of the basins forest was investigated using a 1/20,000 vegetation map (Map of National Forest for Management Plan in 1996) and field survey. Each basin was divided into  $100 \times 100$  m grids, and the number of grids covered with broad-leaved trees, coniferous trees and bamboo grasses was counted. When there were two categories in one grid, it was counted as 0.5 grids for each category.

The vegetation of the riparian forests was surveyed at the middle site in both basins (Fig. 1, •). Woody plant species observed in two 20 × 20 m plots along the streamside were recorded, and the diameter at breast height (DBH) of the woody plants (DBH > 5 cm) was measured. Observed species of herbs and ferns were also recorded.

## Sampling

Light trap surveys were conducted at the middle reach of each basin (Fig. 1,  $\bullet$ ) on April 26, May 16, June 7, June 25, July 26, August 28 and October 2 in 2001. A piece of white cloth (1.5  $\times$  3.0 m) was positioned vertically and parallel to the stream 5 m



**Fig. 1** Study site of Kuroson Stream. Two points (•) are the collection sites in both basins.

from the bank. One 20-watt black and one 20-watt white fluorescent lamp were hung on the cloth 1-m apart. At 18:00, lights were turned on. Collection was started at 19:50 and finished at 23:05. At hourly intervals, adult EPT (Ephemeroptera, Plecoptera, Trichoptera) were collected from one side of the cloth for 15 minutes. It was very dark after sunset in the planted coniferous forest, even during the full moon. It was brighter and more open in the natural broad-leaved forest. Light intensity 0.1 m from the black lamp was 400 lux and from the white lamp was 2,000 lux. Air temperature and humidity were measured by Thermo Recorder (TR-72S, T&D) every 5 min during the collection. Samples of EPT were preserved in 80% ethanol, identified to family level and counted.

### Statistics

Abundance and the number of families were tested between two basins and among seven dates by ANOVA. The EPT adult assemblages sampled in each date were gathered and were analyzed using principal components analysis (PCA) of the family abundances. The values of the PCA axes were compared among seven dates and between basins using ANOVA. In order to select the families that tended to inhabit either basin, repeated-ANOVA was used between the two basins. These statistical analyses were done using SISTAT 10 (SPSS Inc., 2000).

### Results

#### Basin and riparian forest

ONF (old-growth natural forest) basin is 301.4 ha. Broad-leaved trees mixed with some native conifers (more than 128 years old) cover 82 %, planted conifers (*Cryptomeria japonica*) (64 years old) cover 13.1 % and bamboo grasses cover 4.9 % of this basin. PCF (planted coniferous forest) basin is 343.8 ha. Broad-leaved trees (36 - 43 years old, or more than 128 years old) cover 19.5 %, planted conifers (*Cryptomeria japonica*) (36 - 49 years old) cover 80.5 % of this basin.

The riparian forest in ONF basin was dominated with diverse deciduous trees (*Lindera erythrocarpa, Mallotus japonicus, Cornus controversa*) and evergreen broad-leaved trees (*Quercus salicina, Machilus japonica, Cleyera japonica*). Total basal area of conifers, deciduous trees and evergreen broad-leaved trees whose DBH are more than 5 cm (thirty species) was 0.015 m<sup>2</sup>, 1.215 m<sup>2</sup> and 0.706 m<sup>2</sup>, respectively. Totally, sixty-one species of woody plants including the woody plants whose DBH was less than 5 cm, twelve species of herbs and eight species of ferns could be found. The riparian forest in PCF basin was dominated with conifers (*Cryptomeria japonica*), deciduous trees (*Carpinus tschonoskii, Mallotus japonicus, Hovenia tomentella, Idesia polycarpa*) and evergreen broad-leaved trees (*Machilus japonica, Distylium raremosum*). Total basal area of these conifers, deciduous trees and evergreen broad-leaved trees and evergreen broad-leaved trees of these trees whose DBH are more than 5 cm (fifteen species) was 1.572 m<sup>2</sup>, 0.512 m<sup>2</sup> and 0.578 m<sup>2</sup>, respectively. Totally, forty-three species of woody plants including the woody plants whose DBH was less than 5 cm, fourteen species of these and evergreen broad-leaved trees of these trees whose DBH are more than 5 cm (fifteen species) was 1.572 m<sup>2</sup>, 0.512 m<sup>2</sup> and 0.578 m<sup>2</sup>, respectively. Totally, forty-three species of woody plants including the woody plants whose DBH was less than 5 cm, fourteen species of herbs and

#### eighteen species of ferns could be found.

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		April 26	May 16	June 7	June 25	July 26	August 28	October 2
ONF basin	Air temperature (°C) *	10.2	15.3	18.1	20.6	21.4	20.0	13.6
ONF basin	Humidity (%) *	75.8	64.9	94.4	98.4	98.5	90.0	96.1
DOD 1	Air temperature (°C) *	9.6	14.8	17.3	20.6	22.4	19.9	14.3
PCF basin	Humidity (%) *	79.9	68.8	99.0	99.0	94.5 94.9	94.9	97.4

# Comparison of EPT families between ONF and PCF basins

Table 1 Air temperature and humidity on each collection date

ONF: old-growth natural forest, PCF: planted coniferous forest, \*: Averaged value through the investigation.

Twenty-nine families were collected from the two basins over the seven collection dates. Most individuals and families were collected in June and July. Humidity during the collection times was between 65% and 99%. Air temperature ranged from 9.6°C (April 26) to 22.4°C (July 26) (Table 1).

Twenty-eight families and 919 individuals of EPT (Ephemeroptera, Plecoptera, Trichoptera) adults were collected in ONF basin and twenty-eight families and 1641 individuals of EPT adults were collected in PCF basin. There was significant difference of abundance between the two basins and among seven dates (Table 2). There was significant difference of the number of families among seven dates (Table 2).

			Dates					ANOVA		
		April 26	May 16	June 7	June 25	July 26	August 28	October 2	Basin ( <i>d.f.</i> = 1,6) F - <i>ratio</i>	Dates (d.f. = 6, 6) F - ratio
• • • • • • • • •	ONF basin	22	186	115	195	273	112	11	8.43*	8.04*
Abundance	PCF basin	5	270	297	354	516	122	77		
The number	ONF basin	8	21	16	21	20	9	4	0.62	8.67**
of families	PCF basin	4	20	21	18	20	15	10		

Table 2. Result of ANOVA for abundance and the number of families.

ONF: old-growth natural forest, PCF: planted coniferous forest, \*: P < 0.05, \*\*: P < 0.01.

The first three axes of the PCA (principal components analysis) ordination of the EPT adult family compositions explained 56.9% of the variation (Fig. 2). In terms of EPT adult family

composition, plots of sampling dates in ONF basin had a small contained look and clustered at the left, whereas plots of sampling dates in PCF basin were extended and located at the right of the ordination diagram (Fig. 2). Axis 1 differed significantly between the two basins. Axis 2 and axis 3 differed significantly among the seven dates (Table 3).

**Table 3.** Result of ANOVA for axis 1, axis 2 and axis 3 ofPCA.

	ANG	ANOVA			
	Basin	Dates			
	(d.f. = 1, 6)	(d.f. = 6, 6)			
	F - ratio	F - ratio			
Axis 1	6.77*	4.01			
Axis 2	1.70	3.83*			
Axis 3	1.05	11.00**			

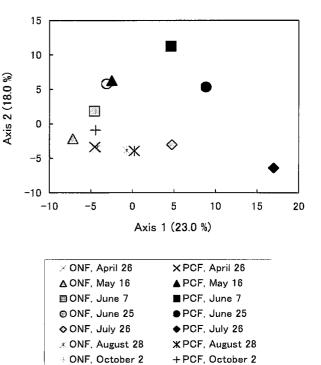


Fig. 2 Principal components analysis ordination diagram of EPT adult family compositions in old-growth natural forest (ONF) basin and planted coniferous forest (PCF) basin.

\*: *P* < 0.05, \*\*: *P* < 0.01.

Individuals of Nemouridae, Chloroperlidae, Leptophlebiidae and Philopotamidae were significantly more abundant in ONF basin. Individuals of Peltoperlidae, Ephemeridae, Goeridae, Stenopsychidae, Glossosomatidae, Hydropsychidae, Polycentropodidae and Lepidostomatidae were significantly more abundant in PCF basin.

### DISCUSSION

	Averaged	abundance	ANOVA			
	ONF basin (n / date)	PCF basin (n / date)	Basin (d.f. = 1) F - ratio	Dates (d.f. = 6) F - ratio	Basin × Dates (d.f. = 6) F - ratio	
Nemouridae	7.3	2.9	6.51*	7.69****	8.94***	
Chloroperlidae	9.0	1.4	19.60****	8.30****	6.34****	
Peltoperlidae	1.7	10.7	5.44*	3.88***	1.82	
Leptophlebiidae	4.3	0.0	6.99*	5.49****	5.49****	
Ephemeridae	4.9	12.6	10.41***	14.08****	3.32**	
Goeridae	1.0	6.6	20.28****	9.52****	4.85****	
Stenopsychidae	3.1	11.3	10.94***	4.91****	3.14*	
Philopotamidae	10.1	3.4	21.04****	7.82****	4.95****	
Glossosomatidae	6.1	30.9	23.19****	5.78****	2.56*	
Hydropsychidae	18.6	35.6	10.02***	16.60****	2.63*	
Polycentropodidae	2.7	10.0	7.89**	13.28****	4.78****	
Lepidostomatidae	4.9	13.9	14.40****	15.44****	6.13****	

Table 4. Result of repeated-ANOVA for twelve families that have significant difference in between basins.

ONF: old-growth natural forest, PCF: planted coniferous forest, \*: P < 0.05, \*\*: P < 0.01, \*\*: P < 0.005, \*\*\*: P < 0.001.

Abundance of EPT (Ephemeroptera, Plecoptera, Trichoptera) adult assemblage was different depends on the basin forest type, and it was more in ONF (old-growth natural forest) basin, though the number of families was not different between the two basins. It was brighter and more open in the old-growth natural forest. It was very dark after sunset in the planted coniferous forest, even during the full moon. This difference of brightness might effect on the number of individuals that attract to the light. Composition of EPT assemblage was also different between the two basins, and the difference was clear from June to July and unclear in April and October. Adults that emerge in early spring have difficulty in flying in the night due to lower air temperature. And the number of families that emerge in late summer are few. So, the individuals that gather toward light in these seasons would be few and the difference of adult assemblage between the basins by principal components analysis would be unclear in April and October.

There was clear difference of abundance between the two basins in twelve families. These results might show that individuals of these families would be strongly attracted to either ONF or PCF basin. Difference in the abundance of Chloroperlidae, Ephemeridae, Stenopsychidae and Hydropsychidae between ONF and PCF basin was the same with that of larval one (Yoshimura and Maeto 2006). Because larvae of Peltoperlidae, Goeridae and Polycentropodidae could not be enough collected for the statistical analysis by quadrat sampling (Yoshimura and Maeto 2006), the difference in larvae could not be detected. However, the difference between ONF and PCF basins could be detected in adults of these families. On the other hand, there was no difference of larval abundance (but had minor difference) in Nemouridae, Leptophlebiidae, Philopotamidae, Glossosomatidae and Lepidostomatidae (Yoshimura and Maeto 2006). But clear difference of abundance between the basins could be detected in adult of these families. Plecopteran families of Nemouridae are generally shredders-detritivores (Merritt and Cummins 1996). Early instar larvae eat the bacterial and fungal scum grown on the leaves that have soaked for many months in the stream. The amount of broad-leaves in the stream might be more in ONF basin than in PCF basin. Deciduous broad-leaved vegetation may provide a larger and higher quality food for shredders than the coniferous vegetation (Eggert and Burton 1994). So, the existence of Nemouridae larvae ought to depend on the forest types. Adults of Nemouridae prefer deciduous trees and shrubs (Harper 1973). Then, the difference of abundance in Nemouridae would be clear in adults. Leptophlebiidae (Paraleptophlebia) larvae feed on fine particles throughout their lives (Fuller and Desmond 1997), but could not obtain the clear difference in Larvae. Howevre, there was a difference in adults. Probably these families tended to inhabit ONF basin where detritus and broad-leaves are much more existing.

Some are found only as larvae, for example, Taeniopterygidae (Yoshimura and Maeto 2006), because they emerge early spring and adults could not be collected by the light trap in this study. Some are found only as adult, for example, Pertoperlidae, because their larval habitat is restricted to the special area such as near the bank or splash area, and can not be collected by quadrat sampling. In this study, the difference of larval assemblage between the two forest types (Yoshimura and Maeto 2006) was almost the same with that of adult assemblage. It could be concluded that the benthic invertebrate assemblage differ depend on the forest types and that some peculiar family inhabit either in the old-growth natural forest or planted coniferous forest.

Forest composition surrounding the stream and associated factors such as shade over the stream

might have an effect on the habitat of aquatic invertebrates (Nakamura and Dokai 1989; Wallance et al. 1997; Whiles and Wallace 1997, Flory and Milner 1999). The difference of basin and riparian vegetation between the two basins in this study might bring the difference of amount and types of leaf litter input, temperature and radiation related to canopy opening between the two basins. And they all would associate and alter the structure of stream habitat for aquatic invertebrates and reflect the composition of adult assemblage.

Altitude is a primary factor associated with aquatic invertebrate assemblage (Ward 1992). Altitude of ONF sites was higher than the sites of PCF in this study, but it was slightly and could consider to be ignored. Water and air temperature are also affect on the aquatic invertebrate assemblage (Ward 1992). However, there was no difference of air temperature between the two basins. Difference of adult assemblage and peculiarity of some families appears to be a reflection of the differences in forest vegetation. There still remains the possibility that other environmental factors affect on the adult assemblage. Though old-growth natural forest was located along a main stream, planted coniferous forest was located along a tributary stream. Besides, we compared only a single basin for each type of forest. At this stage, difference of aquatic invertebrate assemblage between the two basins is supposed to be strongly related with the differences of forest vegetation and it was also true in adult assemblage in this Kuroson stream.

Slight impact of human residence, even at a small scale, affects on the benthic invertebrate assemblage (Yoshimura 2008), perhaps accounting for the benthos differences between the forests that have been used by humans (e.g., forestry, camping and mountaineering) and those that never has. Human activity in the forested area surely affects the benthic invertebrates. There are few natural forests left in western Japan. Adults have little ability to fly long distances. What might happen when natural forest is converted to other forest types or other style of forest use? There is likely to be a negative effect on the families, which are most abundant in natural broad-leaved forest. The conversion these few remaining natural forests to other forest types or other style of forest use may cause extinction of species that inhabit only natural broad-leaved forest. For the sake of reducing the unnecessary disturbance and keeping the human impact to a minimum on benthic invertebrate, we may need some kind of instruction which leads the sustainable coexistence with nature.

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