
Research Article

Secondary production of the net-spinning caddisfly, *Cheumatopsyche* spp. (Trichoptera: Hydropsychidae) in mercuric contaminated riverGunawan Pratama Yoga^{1,2*}, Djamin T. Lumbanbatu³, Ety Riani³, Yusli Wardiatno³

1 PhD Program in Aquatic Resources Management, Graduate School of Bogor Agricultural University, Kampus IPB Darmaga, Bogor 16680, West Java, Indonesia

2 Research Centre for Limnology, Indonesian Institute for Sciences

3 Department of Aquatic Resources Management, Faculty of Fisheries and Marine Science, Bogor Agriculture University, Jl. Agathis, Kampus IPB Darmaga, Bogor 16680, West Java, Indonesia

*Corresponding author: yoga@limnologi.lipi.go.id

Abstract

Cikaniki River is located in Bogor regency, West Java, Indonesia. The river has been contaminated with mercury because of small-scale gold processing. In this paper, secondary production of *Cheumatopsyche* spp. in Cikaniki River was studied. The aim of this study was to determine the impact of mercury contamination in the river to the production and turnover of *Cheumatopsyche* spp. Three locations - Cisarua, Curug Bitung and Lukut, were chosen to represent gold amalgamation activities position in the river. Production of *Cheumatopsyche* spp. was determined using the size-frequency method. The dissolved mercury concentration at the three locations ranged from undetected at all locations to 4.01 $\mu\text{g dm}^{-3}$ at Cisarua. Higher mercury concentrations were observed on particulate form, which from upstream to downstream were 73.26 $\mu\text{g dm}^{-3}$, 114.44 $\mu\text{g dm}^{-3}$, and 52.37 $\mu\text{g dm}^{-3}$, respectively. Particulate Hg were related with the TSS concentration in the river. Particulate Hg were about 30 % to almost half (48 %) of the TSS. The highest annual production of *Cheumatopsyche* spp. in Cikaniki River occurred at Lukut with 7127.96 $\text{mg m}^{-2} \text{y}^{-1}$, followed by Cisarua at 793.94 $\text{mg m}^{-2} \text{y}^{-1}$, and the lowest was at Curug Bitung at 408.31 $\text{mg m}^{-2} \text{y}^{-1}$. The highest annual P/B (turn over) of the caddisfly in Cikaniki River was observed at the downstream of the river, Lukut 11.95, followed by Cisarua 7.32, and the last was Curug Bitung 6.00. Cohort P/Bs of *Cheumatopsyche* spp. were about half of their annual P/B. We conclude that *Cheumatopsyche* secondary production in Cikaniki River was affected by particulate Hg and TSS concentration in the river water. Our result shows that gold amalgamation at the river bank should be regulated to prevent mercuric pollution in Cikaniki River.

Keywords: Cikaniki River, Mercury, Gold amalgamation, Secondary production, *Cheumatopsyche* spp.

Introduction

Mercury (Hg) concentrations in the aquatic environment have increased substantially during the industrial age and are now present at levels which adversely affect aquatic organisms. Larva is apparently the most sensitive stage of the invertebrate's life cycle. Mercuric concentration 1-10 μgdm^{-3} usually causes acute toxicity for the most sensitive developmental stage of many different species of aquatic invertebrates (Boening, 2000). According to WHO (1989), toxicity of inorganic mercury (mercuric chloride) to freshwater invertebrates ranged from <2 to 7390 $\mu\text{g dm}^{-3}$.

In the gold mining process, mercury is used as the chemical agent to perform amalgam, which facilitates the separation of gold from unwanted materials. The mercury pollution from gold mining and processing plants causes contamination of the aquatic environment. Mercury is used in the amalgamation of gold and contamination from the amalgamation process waste is usually discharged directly into the environment. Once released, Hg will continue to cycle through soil, air and water.

Cikaniki River is located in Bogor Regency, West Java, Indonesia. The river has long been contaminated by mercury due to small-scale gold processing activities. The contamination level of mercury in Cikaniki River has exceeded to the maximum tolerable concentration (0.002 ppm) in relation to the Indonesian Government Regulation for river water (Yustiwati et al., 2003).

Secondary production is defined as formation of heterotrophic biomass through time (Benke & Huryn, 2006). Secondary production is a measure of the relative importance of a species (population) to the aquatic ecosystem as a whole because production is the means by which energy is made available for transfer from one trophic level to another. Estimating production of an animal population is essential to understanding the role of a population in energy flow pathways (Benke & Wallace, 1980).

Larvae of filter feeding caddisflies of the family Hydropsychidae usually comprise an important portion of the invertebrate biomass in streams, especially in relation to abundance of fine organic particles which are retained by hydropsychid catch-nets (Sanchez & Hendricks, 1997). The larvae of hydropsychid caddisfly genus *Cheumatopsyche* are common filter-feeder in warmer streams with moderate to high loads of organic matter. They are distributed in all faunal regions except in South America (Kondratieff et al., 1997).

Application secondary production analyses in studies of anthropogenic stress are still uncommon (Benke, 1984). Several examples include the use of secondary production to assess the effects of a pesticide manipulation (Lugthart & Wallace, 1992; Whiles & Wallace, 1995), urbanization (Shieh et al., 2002), Zn contamination (Carlisle & Clements, 2003) and large wood addition (Entrekin et al., 2009) on stream communities.

The aim of our study was to determine whether secondary production of net-spinning caddisfly *Cheumatopsyche* spp. is influenced by mercuric contamination. Populations that persist in contaminated environments may reduce organism abundance either by increased mortality or reduced fecundity (Fleeger et al., 2003). In chronically mercuric contaminated environments aquatic biota may suffer from a decrease in foraging abilities, inability to avoid predators, decreased ability to reproduce, growth impairment and behavioral deviations (Desrosiers et al., 2006). Secondary production is the product of population biomass and individual growth rates; therefore in this paper we examined contaminant-induced variation in these processes as probable mechanisms for observed patterns in production.

Materials and Methods

Sampling sites

Cikaniki River, located in Western Bogor - West Java, is one of the headwater streams of Cisadane River which flows to Jakarta Bay. Mercuric-gold amalgamation is a common activity at villages along the river. Three sampling sites were chosen, from upstream to downstream, and the locations were Cisarua, Curug Bitung and Lukut. The land use activity in Cisarua and Curug Bitung is dominated by agriculture, especially paddy fields, while Lukut is dominated by domestic activities. The substrates were dominated by pebble and cobble with a few large boulders scattered within stream channels, with maximum water depth in Cisarua, Curug Bitung and Lukut measured at 50cm, 36.3 cm, and 32 cm, respectively. Average current speed in the three locations were 0.39 m sec^{-1} , 0.42 m sec^{-1} , 0.46 m sec^{-1} , respectively. Water temperature, dissolved oxygen, pH, and conductivity in the three locations ranged from 24.36 to 26.47 °C, 6.64 - 6.76 mg l^{-1} , 7.9 - 8.05, 163.47 - 237.83 mSsec $^{-1}$, respectively. The detailed locations of sampling sites are listed in Figure 1 and Table 1.

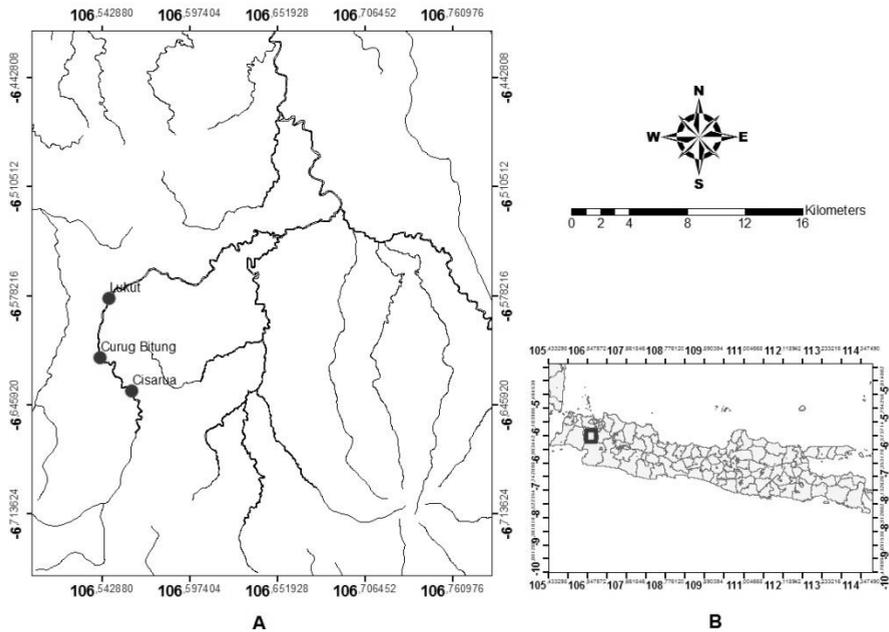


Figure 1. A Map of Cikaniki River showing sampling locations. B Map of Java Island showing Bogor Regency.

Sampling collection and processes

Quantitative samples for secondary production and mercuric concentration were taken at monthly intervals from January 2012 to October 2012. Samples for secondary production were collected with a surber sampler of 0.09 m^2 area and $200 \text{ }\mu\text{m}$ mesh size, preserved in 10 % formalin and sorted and identified under a dissecting microscope. *Cheumatopsyche* larvae were counted and their head widths measured with an ocular micrometer. Larvae from each sample were dried at 60°C for 24 hours and weighed in electrobalance to the nearest 0.1 mg. Average dry-mass for each instar was used to calculate mean biomass and biomass losses. One litre of water samples for mercury concentration analyses were taken using a Schott bottle that had been washed with 20 % nitric acid from each location. The samples were cooled at 4°C in a container until they were analysed at the laboratory.

Mercury Analyses

At the laboratory, the water samples were filtered using a $0.45 \text{ }\mu\text{m}$ membrane filter. The adsorbing mercury on suspended particle retained on the membrane filter was considered as particulate mercury, and the filtrates were analysed for dissolved mercury. The prepared samples were added by 1 cm^{-3} of H_2SO_4 (1:1), 1 cm^{-3} of HNO_3 concentrate, and 2 m^3 of KMnO_4 (50 gdm^{-3}), after

Table 1. Description of sampling locations.

Code	Name	Latitude/ longitude	Description
St. 1	Cisarua	6° 38' 15.16" S/ 106° 33' 39.7" E	Gold amalgamation activities were located on the river bank, paddy field area
St. 2	Curug Bitung	6° 37' 1.7" S/ 106° 32' 31.4" E	Gold amalgamation activities were located in the river as well as on river bank ,paddy field area
St. 3	Lukut	6° 34' 47.85" S/ 106° 32' 51.56" E	No gold amalgamation activities, village settlement

shaking for 15 minutes, then $1 \text{ cm}^{-3} \text{ K}_2\text{S}_2\text{O}_8$ was added. These sample solutions were heated at 95°C for two hours to complete the digestion process (Eaton et al., 1995). These were later cooled at the room temperature, and 1 cm^{-3} of hydroxylamine chloride was added to neutralize the excess of KMnO_4 , and the solution was diluted to 50 cm^{-3} with a volumetric flask and measured by Mercury Analyzer Hiranuma HG-300.

Total Suspended Solid analyses

Pre-ashed and pre-weighted $0.45 \mu\text{m}$ membrane filter were placed in a filtering apparatus and was moisturised with a small volume of pure water to seat it. Sample was stirred with a magnetic stirrer to obtain a more homogeneous particle size. While stirring, sample was pipetted $100 - 250 \text{ cm}^3$ onto the membrane filter. The membrane filter was washed three times with 10-mL volumes of aquadest before it was carefully removed from the filtration apparatus and transferred to an aluminum weighing dish for support. The membrane filter then was dried for at least 24 hours at 60°C in an oven, and was weighed (Eaton et al., 1995). To determine the TSS, the formula below was used:

$$\frac{A - B}{\text{sample volume}} \times 1000$$

where,

A = weight of filter + dried residue (mg) and B = weight of filter (mg).

Secondary Production Analyses

Larval instars were determined by plotting size frequency distributions of the head capsule width to obtain width ranges for instars I through V for each site and to generate regression lines of larval growth for three sites. Body mass relationship was determined by general power equation $DM = aL^b$, or after

logarithmic transformation as $\log DM = \log a + b \log L$ (where a , b = regression constants, DM = dry mass, and L = length parameter, which in this case was head width) (Benke & Huryn, 2006).

Production of *Cheumatopsyche* spp. was determined by using the size-frequency or Hynes method (Benke & Huryn, 2006), because keys for species identification of immature *Cheumatopsyche* larvae are unavailable, therefore the cohort was not distinguishable. Negative values in the “times number of size classes” column were excluded from calculation in the early life stage larva. These negative values most likely happened when larvae in smaller size classes were inadequately sampled (Benke & Wallace, 1980).

P/B ratio was determined as the ratio of production per unit time to mean biomass. According to this method, both annual production and annual P/B need a calibration based on cohort production interval (CPI), which is development time from larval instar I to larval instar V. CPI for *Cheumatopsyche* in tropic region is 182.5 days (Jacobsen et al., 2008). Cohort P/B = annual P/B x CPI (days)/365.

Results

The dissolved mercury concentration at the three locations varied from undetected at all locations to $4.01 \mu\text{g dm}^{-3}$ at Cisarua. However, the average concentration ranged from 1.35 to $1.57 \mu\text{g dm}^{-3}$. Much higher mercury concentrations were observed on particulate form. Average particulate mercury concentration from upstream to downstream were $73.26 \mu\text{g dm}^{-3}$, $114.44 \mu\text{g dm}^{-3}$, and $52.37 \mu\text{g dm}^{-3}$, respectively. Particulate Hg were related with the TSS concentration in the river. In Cisarua and Curug Bitung, particulate Hg were about 30 % of the TSS, while in Lukut the ratio was almost half (48 %). Table 2 shows the statistic summary of those parameters.

Secondary production calculation using size-frequency method (Hynes method) are illustrated for *Cheumatopsyche* spp. in Cisarua (Table 3). The size category was represented by the five larval instars. The sums of the second, fourth, and eighth columns of Table 3 are annual mean density (N), annual mean biomass (B) and annual production (P), respectively. Negative values of the total biomass losses for the first two small size categories were excluded and considered as zero in the production summation. Early instar larvae were usually under-represented in samples, and resulted in negative biomass loss values for that size class on secondary production calculation. It is

Table 2. Statistic summary of dissolved, particulate mercury, and FPOM concentration at sampling locations. Value in the parentheses were minimum and maximum values.

Parameters	Cisarua	Curug Bitung	Lukut
Dissolved Hg (μgdm^{-3})	1.37 (0.00* - 4.01)	1.53 (0.00* - 3.08)	1.35 (0.00* - 2.99)
Particulate Hg (μgdm^{-3})	73.26 (11.43 - 223.81)	114.44 (5.28 - 354.67)	52.37 (18.05 - 73.26)
TSS (mg dry mass dm^{-3})	214.56 (24.00 - 972.00)	374.68 (45.20 - 1582.00)	109.32 (55.20 - 236.40)

* Below detection limit

Table 3. Size-frequency calculation of *Cheumatopsyche* spp. in Cisarua.

Instar	Density (ind m^{-2})	Individual Mass (mg)	Biomass (mg m^{-2})	No. Lost (ind m^{-2})	Ind.	Total	Correction factors (x5) ($\text{mg m}^{-2}\text{y}^{-1}$)
					Biomass at loss (mg)	biomass loss (mg m^{-2})	
I	4.44	0.30	1.33	-6.67	0.93	-6.17	-30.87*
II	11.11	0.56	6.20	-15.56	1.38	-21.54	-107.68*
III	26.67	1.36	36.24	20.00	1.87	37.37	186.87
IV	6.67	2.13	14.22	-2.22	2.49	-5.53	-27.63
V	8.89	4.05	36.00	8.89	4.05	36.00	180.00
N= 57.78		B= 93.99		P= 688.10			

* Not included in secondary production calculation

theoretically impossible to have lower densities for the first to third instars than for later instars (Benke & Wallace, 1980).

Annual mean density (N), mean standing stock biomass (B), production (P) and P/B ratios are compared for each location (Table 4). *Cheumatopsyche* spp. secondary production in Lukut was the highest among the three locations, followed by Cisarua and Curug Bitung. The higher production in Lukut were due to annual mean density and mean standing stock biomass which were much higher compared to the other locations. Annual secondary production estimates for *Cheumatopsyche* spp. in the three locations ranged from 408.31 - 14458.96 $\text{mgm}^{-2}\text{y}^{-1}$ (dry mass).

Annual P/Bs of *Cheumatopsyche* spp. in the three locations vary from 6 to 11.95 per year. Lukut was higher at about 1.6 times to twice compared to Cisarua and Curug Bitung, respectively. Cohort P/Bs of *Cheumatopsyche* spp.

Table 4. Summary of mean values for secondary production parameters of *Cheumatopsyche* spp.

Location	N (Ind m ⁻²)	B (mg m ⁻²)	P (mg m ⁻² y ⁻¹)	Annual P/B	Cohort P/B
Cisarua	57.78	93.99	688.10	7.32	3.66
Curug Bitung	44.44	68.09	408.31	6.00	3.00
Lukut	900.00	1210.18	14458.96	11.95	5.97

were about half of their annual P/B. The lowest cohort P/B was found in Curug Bitung, i.e. 3.00, dan the highest was observed in Lukut, i.e. 5.97.

Discussion

Yustiawati et al. (2003) observed that the concentration of mercury as adsorbing species on suspended species is highest compared to other species of mercury in Cikaniki River water. Coquery et al. (1997) stated that mercury in river water mostly presents itself as adsorbing species on suspended particle. Total mercuric concentration in the river surface water in the three locations had exceeded the tolerable value (0.002 mg l⁻¹) of the Indonesian Government Regulation No. 82/2001.

Production of *Cheumatopsyche* spp. in Cikaniki River was higher than secondary production of *Cheumatopsyche spinosa*, 265.2 mgm⁻²y⁻¹, and *Cheumatopsyche quadrata*, 150 mg m⁻²y⁻¹, as calculated by Dudgeon (1997) in Tai Po Kau forest stream, Hong Kong. However, they were still lower than those calculated by Benke & Wallace (1997) and Bowles & Allen (1991) which reached 13692 mg m⁻² y⁻¹ and 17.02 gm⁻² y⁻¹, respectively for *Cheumatopsyche* spp. According to Sanchez & Hendriks (1997), higher annual production values of *Cheumatopsyche* spp. were affected by several factors such as higher larval density and different voltinism of the organism. The shorter CPI value resulted in a higher adjustment of the annual production, higher organic seston and variation of food types. Calculation of secondary production from tropical streams are usually not high because of the difficulty in determining growth and turnover ratios of populations that exhibit continuous and synchronous development (Jacobsen et al., 2008).

The contrasting roles of growth rate and biomass in determining secondary production are summarised by the P/B ratio (Huryn & Wallace, 2000). Annual P/B of *Cheumatopsyche* spp. that have been calculated by other researchers

vary broadly, which correspond to different frequency of bivoltinism of the organism at the sites observed (Sanchez & Hendriks, 1997). For the tropical region, the annual P/B ratio of Lukut was higher than those reported by Dudgeon (1997) for *Cheumatopsyche spinosa* and *Cheumatopsyche ventricosa* which were 8.4 and 9.7, respectively, while in Curug Bitung and Cisarua, the ratio was smaller. However, our annual P/B ratios were lower than the *Cheumatopsyche* spp annual P/B ratio reported by Benke & Wallace (1997).

Cohort P/B ratios (the P/B ratio over the life-span of a cohort) for stream macroinvertebrates is usually about five (Waters, 1987), but it can however range from two to eight (Huryn & Wallace, 2000). According to that range, our cohort P/B ratios were low in Cisarua and Curug Bitung, but in Lukut the ratio was about five. Our finding is consistent with that of Parker & Voshell (1983), and Benke et al. (1984) which found the cohort P/B ratio for Trichoptera ranged 3.5 - 6 and 3.2 - 5.2, respectively. The lower cohort P/B (2 - 5) usually occurs in hemimetabolous and holometabolous aquatic insects which as part of their life history, leave the aquatic habitat for emergence (Waters, 1987). The cohort P/B ratios were half of the corresponding annual P/B ratios because we used the CPI value for half a year (182.5).

Secondary production analysis is a powerful tool for assessing ecosystem degradation (Carlisle & Clements, 2003). Pollution can cause increased production or act as a physiological stressor and cause decreased production (Benke & Huryn, 2010). Several studies have used invertebrate production to assess effects of heavy metal contamination. Our result showed that elevation of concentration of particulate Hg and TSS in water cause a decrease in the secondary [production of *Cheumatopsyche* spp. In Curug Bitung where the Hg and TSS concentrations were the highest among the other locations, has the lowest secondary production. TSS concentration in the three locations was high (> 10 mg dm⁻³). Higher TSS loading can cause dilution of organic content of periphyton, which is a significant component of the energy (food) base in the stream habitat, because of higher siltation (Runck, 2007).

Conclusions

Mercury concentration in Cikaniki River was so high that it exceeded the tolerable value. Secondary production of *Cheumatopsyche* spp. in Cikaniki River was relatively high for a tropical stream. Secondary production of *Cheumatopsyche* spp. in Cikaniki River was affected by mercury pollution that

comes from gold amalgamation activities at the river bank and elevation of TSS loading which cause decreasing of the secondary production.

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References

- Benke AC, Huryn AD. 2006. Secondary production of macroinvertebrates. In: Hauer FR, Lamberti GA. (Eds). *Methods in Stream Ecology*. 2nd edition. Academic Press. Elsevier: Burlington, MA. Pp.691-710
- Benke AC. 1984. Secondary production of aquatic insects. In: Resh VH, Rosenberg DM. (Eds). *The Ecology of Aquatic Insects*. Praeger, New York, Pp.289-322
- Benke AC, Wallace JB. 1980. Trophic basis of production among net-spinning caddisflies in a southern appalachian stream. *Ecology* **61**(1):108-118
- Benke AC, Wallace JB. 1997. Trophic basis of production among riverine caddisflies: implications for food web analysis. *Ecology* **78**(4):1132-1145
- Benke AC, Huryn AD. 2010. Benthic invertebrate production - Facilitating answers to ecological riddles in freshwater ecosystems. *Journal of North American Benthological Society* **29**(1):264-285
- Boening DW. 2000. Ecological effects, transport, and fate of mercury: a general review. *Chemosphere* **40**(12):1335-1351
- Bowles DE, Allen RT. 1991. Secondary production of net-spinning caddisfly (Trichoptera: Curvivalpia) in an Ozark Stream. *Journal of Freshwater Ecology* **6**(1):93-100
- Carlisle DM, Clements WH. 2003. Growth and secondary production of aquatic insects along a gradient of Zn contamination in rocky mountain streams. *Journal of North American Benthological Society* **22**(4):582-597
- Coquery M, Cossa D, Sanjuan J. 1997. Speciation and sorption of mercury in two macro-tidal estuaries. *Marine Chemistry* **58**:213-227
- Desrosiers M, Planas D, Mucci A. 2006. Total mercury and methyl mercury accumulation in periphyton of boreal shield lakes: Influence of watershed physiographic characteristics. *Science of the Total Environment* **355**:247-

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- Dudgeon D. 1997.** Life histories, secondary production and microdistribution of hydropsychid caddisflies (Trichoptera) in a tropical forest stream. *Journal of Zoology* **243**:191-210
- Eaton AD, Clesceri LS, Rice EW, Greenberg AE. 1995.** *Standard Methods for the Examination of Water and Waste Water*. 19th ed. American Public Health Association (APHA), American Water Works Association (AWWA), Water Environment Federation. Washington D.C. Pp.1085
- Entrekin SA, Tank JL, Rosi-Marshall E, Hoellein TJ, Lamberti GA. 2009.** Response of secondary production by macroinvertebrates to large wood addition in three Michigan streams. *Freshwater Biology* **54**:1741-1758
- Fleeger JW, Carman KR, Nisbet RM. 2003.** Indirect effects of contaminants in aquatic ecosystems. *The Science of the Total Environment* **317**:207-233
- Hurn DA, Wallace JB. 2000.** Life history and production of stream insects. *Annual Review of Entomology* **45**:83-110
- Jacobsen D, Cressa C, Mathooko JM, Dudgeon D. 2008.** Macroinvertebrates: Composition, life histories and production. In: Dudgeon, D. (Ed). *Tropical Stream Ecology*. Elsevier Inc. Pp.65-105
- Kondratieff BC, Bishop RJ, Brasher AM. 1997.** The Life cycle of an introduced caddisfly, *Cheumatopsyche petiti* (Banks) (Trichoptera: Hydropsychidae) in Waikolu Stream, Molokai, Hawaii. *Hydrobiologia* **350**:81-85
- Lughart GJ, Wallace JB. 1992.** Disturbance on Benthic Functional Structure and Production in Mountain Streams. *Journal of the North American Benthological Society* **11**(2):138-164
- Parker CR, Voshell JR. 1983.** Production of filter-feeding trichoptera in an impoundment and a free flowing river. *Canadian Journal of Zoology* **61**:70-87
- Runck C. 2007.** Macroinvertebrate production and food web energetics in an industrially contaminated Stream. *Ecological Applications* **17**(3):740-753
- Sanchez RM, Hendricks AC. 1997.** Life history and secondary production of *Cheumatopsyche* spp. in a small Appalachian stream with two different land uses on its watershed. *Hydrobiologica* **354**:127-139
- Shieh SH, Ward JV, Kondratieff BC. 2002.** Energy Flow through Macroinvertebrates in a Polluted Plains Stream. *Journal of the North American Benthological Society* **21**(4):660-675
- Waters TF. 1987.** The effect of growth and survival patterns upon the cohort P/B ratio. *Journal of North American Benthological Society* **6**(4):223-239
- Whiles MR, Wallace JB. 1995.** Macroinvertebrate Production in a Headwater Stream during Recovery from Anthropogenic Disturbance and Hydrologic Extremes. *Canadian Journal of Fisheries and Aquatic Sciences* **52**(11):2402-2422
- World Health Organization. 1989.** *Mercury-environmental aspects*. WHO, Geneva, Switzerland

Yustiawati, Syawal MS, Terashima M, Kimura T. 2003. Speciation analysis of mercury in river water and sediment in West-Jawa and Central Kalimantan, Indonesia. *Annual Report JSPS March 2003*:210-218