



Freshwater Stream



/		/	
School Name	Student Name	Group	
/ DD-MM-YYYY		/	
Site	Date	Time	Recent Weather Conditions

Freshwater Stream

INTRODUCTION

Background

In Hong Kong, we do not have large freshwater habitats like rivers or lakes, but we do have small streams, ponds, reservoirs, abandoned paddy fields and marshy areas distributed throughout the territory. Many freshwater systems have been polluted by agricultural, industrial and domestic wastes. It is said that natural water systems free from pollution are getting less and less.

Aims and Objectives

- To appreciate the wonders of the living world.
- To familiarize structure, flora and fauna in a freshwater stream community.
- To observe, compare and contrast the ecology of different freshwater stream habitats.

Equipment

For plant and animal sampling			For measurement of physical factors		
☐	Aquarium net	x2	☐	Compass	x1
☐	Brush	x2	☐	Light meter	x1
☐	Clip board	x1	☐	pH cum thermometer	x1
☐	Quadrat	x2	☐	Dried & weighted filter paper	x2
☐	Spoon	x2	☐	Filter funnel	x1
☐	Trap	x1	☐	Measuring tape	x1
☐	Plastic bag	x2	☐	Metre rule	x1
☐	Plastic basket	x2	☐	Sampling bottle	x2
☐	Plastic box	x1	☐	Stop watch	x1
☐	Plastic tray	x2	☐	Table tennis ball	x1
☐	Plastic vial	x6	☐	Towel	x1
☐	Fresh water streams identification kit	x2			
☐	Life buoy with a rope	(Share)			

Remarks

- Dress in shorts and canvas shoes with adequate tread. Sandals are not recommended. Be careful of broken glass and abandoned construction material etc.
- Walk slowly and try footing for the stability of substratum. Be careful of loose gravel and slippery rock surface.
- Never chase after any fast-moving animals to avoid accidents and habitat destruction.
- Never pollute/damage the environment in all sense. Minimize trampling.
- Minimize disturbance to the local people.
- Team leader should organize members to work in a serious and efficient way. Members should co-operate with the leader.

Since time is limited, you should work efficiently. If you do have extra time, you are highly recommended to carry out your own investigations, provided that it is safe to do so.

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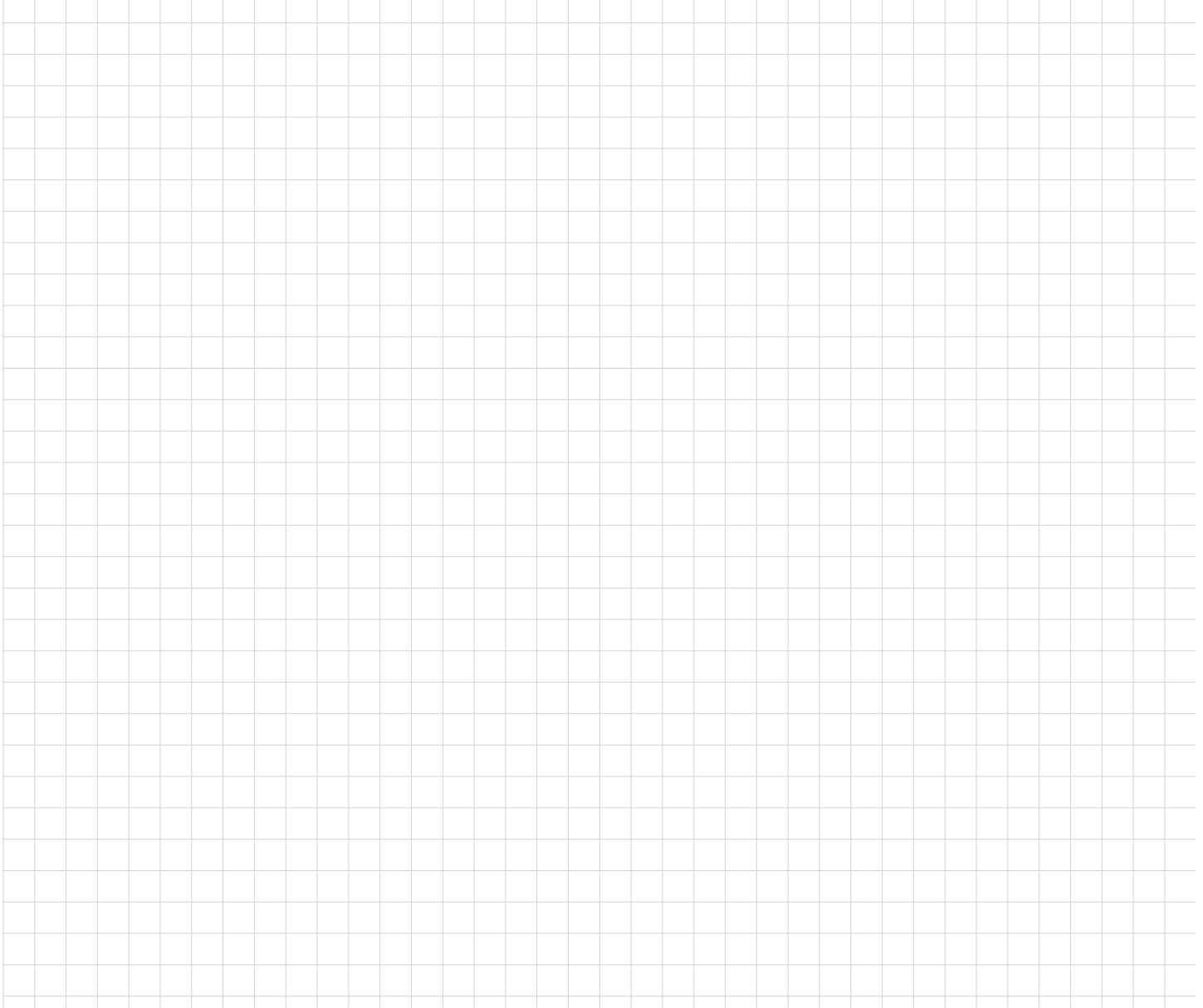
Site Observation

Choose 10m of the stream and Draw a sketch map top view of the surrounding area on Figure 1, indicating:

- your position in the study site (with a compass) ;
- direction of stream flow ;
- vegetation ;
- breadth, any dam/boulders/trees/fallen log/submerged log etc. ;
- other particulars of interest.

In order to proceed the sampling and measurement works at the same time, divide your group into 2 teams. One is responsible for biotic investigation while the other is to take abiotic measurement. However it is more important to understand the whole picture, so try to get involved in the work of your partners.

Figure 1. Site profile – Top view of the study area



Location of Field Site



Site A



Site B



Weather Information

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Study of Abiotic Factors

Record data on Table 1&2

2.1. Water sampling

Carefully collect 2 water samples by sampling bottles (500ml) provided (1 filtered and 1 non-filtered). Mark the sampling site on the sketch map.

2.2. Stream substratum

Use your naked eyes to identify the types of the stream substratum.

2.3. Dimensions of field area

Measure the maximum and minimum width of the area of study by a measuring tape.

2.4 Abiotic factors measurement in quadrats and traps

Use ruler to measure water depth of quadrats and traps.

Use lux meter to measure light intensity at the surface (S) and the bottom (B) of the stream, and calculate the light transmittance.

Use pH meter cum thermometer to measure water temperature of quadrats and traps.

Place ping pong ball at one end of the quadrats or meter ruler, and record the time needs for the ball to flow to another end, and calculate the speed of flow of stream.

Use dissolved oxygen meter to measure dissolved oxygen in quadrats and traps.

★ Do not drop the equipment into the water.

★ Do it prior to other studies!

★ Avoid generating air bubbles. Invert your bottle to check against trapped air and leakage.

Let's think...

Where should the bottle be placed during facing the water flow?

A. In front of your body. OR

B. Behind your body.

Which direction should the bottle mouth face?

A. Against the flow direction. OR

B. As the flow direction.

★ Remember to rinse the probe

Table 1.

Texture of substratum	Gravel / Coarse sand / Fine sand / Mud / Others
Distribution of substratum	Homogeneous / Heterogeneous / Patchy / Others
Water Colour	Clear / Less Clear / Brown / Black
Smell	None / Some / Moderate / Strong
Floating matters	None / Some / Plentiful / Abundant

Table 2.

Sample	Depth (m)	Light Intensity (lux)		Rate of light transmission (%)	Temp (°C)	pH	Drifting time of the ping pong (s)	Current Speed (ms ⁻¹)	Dissolved oxygen (mg/L)
		Above water surface (S)	10cm under surface (B)						
Sampling Area 1									
Sampling Area 2									
Sampling Area 3									
Sampling Area 4									
Trap 1									

The widest breadth (m)		The narrowest breadth (m)	
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Study of Biotic Factors

3.1. Plant and algae sampling

Search and identify freshwater plants at different microhabitats. Observe any special adaptive features and collect samples with plastic bags.  Also collect a full vial of decaying leaves for later investigation.

3.2. Animal sampling (record data on Table 3)

Randomly select 3 sampled area by a 0.5mX0.5m quadrats.

Collect, identify and count the animals at site with the help of nets, plastic tray and brushes. ☒ Place the trap in the stream for 30 minutes, and identify and count the animals trapped inside.

★To protect our wildlife and environment, do not collect unnecessary specimen and always put minimal disturbance. Walk slowly in water and always return stones back to original positions.

★Be careful of allergic plant species.

★Perform animal sampling after finishing all other works.

Table 3.

Species Name	Abundance				
	Sampling Area 1	Sampling Area 2	Sampling Area 3	Sampling Area 4	 Trap 1
Mayfly Nymph					
Damselfly Nymph					
Dragonfly Nymph					
Stonefly Nymph					
Caddisfly Larva					
Water Penny					
Bloodworm					
Mosquito Larvae					
Pond Skaters					
Long-armed Shrimps					
Predaceous Chub					
White Cheek Goby					
Sharphead Sleeper					
Snail					
OTHERS					
Species Richness					
Total Number of Organisms					

LABORATORY WORK

Table 4. Adaptations of organisms

Organism	Threats	Environmental Challenges or Stress	Corresponding Adaptations

LABORATORY WORK

Equipment

☐ 250ml beaker	x1	☐ Test tube rack	x1
☐ E. coli detection dish	x1	☐ Wash bottle	x1
☐ Compound microscope	x1	☐ Slides	(Share)
☐ Cuvettes	x2	☐ Cover slips	(Share)
☐ Dropper	x1	☐ Electronic Balance	(Share)
☐ Evaporating dish	x1	☐ Oven	(Share)
☐ Forceps	x2	☐ Heat resistant gloves	(Share)
☐ Glass rod	x1	☐ Dissolved Oxygen meter	(Share)
☐ Petri dishes	x5	☐ 70% Alcohol	(Share)
☐ Stereomicroscope	x1	☐ Spectrophotometer	(Share)
☐ 1 ml syringe	x1	☐ Refractometer	(Share)
☐ Test tube	x2		

Chemicals

☐	Solution A (Ammonium molybdate / H_2SO_4)	(Share)
☐	Solution B (5% Stannous chloride)	(Share)
☐	Solution C (Nessler's reagent)	(Share)
 ☐	Chemical Oxygen Demand detection kit	x1

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Biological Investigation

4.1. Plant and animal identification

Use the reference books, photographs and stereomicroscope provided to identify specimens collected from the streams.

4.2. Microscopic organisms in algal mass and plant debris

Use forceps to tear a decaying leaf into small pieces. Put the sample onto a slide, observe and record any protozoan and other microscopic organisms present under a compound microscope.

★ Transfer the animals in the glass chamber specified after identification and clean up the vials.

★ Put the used slides and cover slips at respective beaker/vial specified.

Remarks

- The equipment/chemicals are rather expensive. Please handle with care and consult technician when necessary.
- Discard all reagents/solutions in the chemical waste bottle.

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Water Sample Analysis

Record data on Table 4

5.1. Salinity

Read the salinity by placing 2-3 drops of filtered sample onto a refractometer.

5.2. Ammonia content

Pour 25ml of filtrate into a 100ml beaker, add 1ml of solution C and swirl the mixture. A yellow colour indicates the presence of ammonia. Fill a cuvette to the white mark and measure the ammonia concentration by spectrophotometer (425nm).

5.3. Phosphate content

Pour 25ml of filtrate into a 100ml beaker, add 1 drop of solution A and solution B respectively and swirl the mixture. A blue colour indicates the presence of phosphate ions. Fill a cuvette to the white mark and measure the phosphate concentration by spectrophotometer (506nm).

5.4. Total suspended solids (TSS)

Weigh a pre-dried filter paper and filter 500ml water sample into another 500ml plastic bottle. Put the filter paper into an evaporating dish, dry in a 105°C oven overnight and reweigh.

$$\text{TSS (mg/L or ppm)} = [\text{increase in mass (mg)}] \times 2$$

5.5. E. coli content

Use a syringe to spread 1ml water sample from unfiltered water sample onto the E. coli detection dish. Incubate overnight at 37°C.

5.6. Chemical oxygen demand (COD)

Remove the line to clear the aperture from the top of the tube. Press the sides of the tube to expel approximately half of volume. Immerse the tube in the sample. Release the sides to fill the tube up to the half. Shake the tube a few times. After the reaction time (4-6mins), put the tube on the color chart as shown and compare with the standard colors.

★ Do not easily discard any solution.

★ Remember to rinse the glass chamber before taking readings.

★ Fully fill the sample on the glass chamber.

★ Prevent generating air bubbles in the chamber.

★ Calibration can be made with deionized water.

★ Pick up the filter paper with forceps.

★ Maximize the surface area of the filter paper during filtration.

★ Invert the bottle several times before filtration.

★ Rinse the plastic bottle with the filtrate.

Table 4. Water sample analysis

Salinity (g/100g)	
NH_4^+ (ppm)	
PO_4^{3-} (ppm)	
TSS (mg/L or ppm)	
 E. Coli. Testing (cfu/100ml)	
 COD (ppm)	

Discussions and Conclusions

1. Evaluate the level of biodiversity of different quadrats or regions based on the species richness and the relative abundance of each species.
2. After pooling all information with other groups, can you draw any conclusions on our study?
3. Which factor(s) do you think is/are limiting to the community in the stream? Why?
4. Construct a cross sectional profile of your study area after integrating all your abiotic and biotic information.
5. Examine the external features of the animals collected, how do they adapt to the environment with respect to their:
 - particular microhabitats (beneath stones/free-swimming/on water surface etc.)
 - feeding habits (omnivorous/detritivorous/carnivorous/herbivorous etc.)
 - feeding relationships (competition / predation / commensalism / mutualism / parasitism etc.)
6. Based on the organisms collected or observed, try to construct food chains/web to show the trophic levels of these organisms.
7. Draw graphs to show the changes in abiotic factors in different locations along the streams. Do you think the streams have been polluted? Why?
8. State the limitations and drawbacks of the investigation. Suggest any improvements for further studies.

References

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POLLUTION INDEX

Table 1: Physical Factors Analysis (4 point Scale)

Assessment Score Physical Factors	0	1	2	3
Water Colour	Clear	Less Clear	Brown	Black
Smell	None	Some	Moderate	Strong
Floating matters	None	Some	Plentiful	Abundant
Temp. different from other groups (°C)	< 1	1 - 1.5	1.5 - 2	> 2
Mean rates of light transmission	> 70%	41% - 70%	10% - 40%	< 10%

Mean Assessment Score (A) on Physical Factors Analysis

Table 2: Chemical Factors Analysis (4 point Scale)

Assessment Score Chemical Factors	0	1	2	3
Dissolved Oxygen (ppm or mg/l)	> 7.0	5.1 - 7.0	3.0 - 5.0	< 3.0
pH	6.0 - 7.0	5.0 - 5.9 / 7.1 - 8.0	4.0 - 4.9 / 8.1 - 9.0	< 4.0 / > 9.0
Total Suspended Solid (ppm or mg/l)	< 20	20 - 35	35 - 50	> 50
Salinity (%)	< 3	3 - 6	7 - 10	> 10
NH ₄ ⁺ (ppm or mg/l)	< 2.1	2.1 - 5.0	5.1 - 7.0	> 7.0
PO ₄ ³⁻ (ppm or mg/l)	< 3.0	3.0 - 6.0	6.1 - 9.0	> 9.0
COD	< 5	5 - 13	14 - 50	> 50

Mean Assessment Score (B) on Chemical Factors Analysis

Table 3: Micro-organism analysis (4 point Scale)

Assessment Score Micro-organism	0	1	2	3
Escherichia coli (c.f.u./100ml)	< 180	181 - 400	401 - 610	> 610

Mean Assessment Score (C) on Micro-organism Analysis

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POLLUTION INDEX

Table 4: Biological Indicators

Organism Group	Indicator Species
i	Mayfly nymphs Stonefly nymphs Dragonfly nymphs Water pennies Snails with operculum Microscopic algae (Desmids, Hydrodictyon)
ii	Caddisfly larvae Mosquito larva Snails without operculum Freshwater shrimps Green algae (Cladophora)
iii	Leeches Blood worm (Chironomus larvae) Protozoa (Paramecium, Vorticella) Water louse (Asellus spp.) Green algae (Spirogyra, Oscillatoria, Anabaena)
iv	Sludge worm (Tubifex) Sewage fungi

Table 5: Biological Factors Analysis (4 point Scale)

Assessment Score	0	1	2	3
Biotic Factors				
Indicator Organisms	Group i Dominant + ii,iii,iv	Group ii Dominant + iii,iv	Group iii Dominant + iv	Group iv or no organisms found

Mean Assessment Score (D) on Biological Factors Analysis	
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Table 6: Degree of Pollution

Average Assessment Score (A), (B), (C), (D)	Pollution Magnitude
0.00 - 0.75	Clean
0.76 - 1.50	Slightly Polluted
1.51 - 2.25	Moderately Polluted
2.26 - 3.00	Severely Polluted

Note: Assumed the Physical, Chemical and Biological factors are equally important.