EXCELLENT SCIENCE IN ASEAN

BEST SELECTED PAPERS AND POSTERS FROM YOUNG **ASEAN SCIENTISTS** ON **WATER**, **FOOD** AND **HEALTH**

Ágota Dávid, Ádám Dávid, Béla Kardon PhD (Eds.)

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ISBN: 978-963-12-1839-8

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Published by:
Regional Centre for Information and Scientific Development (RCISD)
Hűvösvölgyi út 54. V/1.,
H-1021 Budapest
www.rcisd.eu

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Edited by: Ágota Dávid, Ádám Dávid & Béla Kardon PhD (RCISD)
Cover design: Maria João Maia (SPI)
Printed in Hungary by Stanctechnik Kft.
www.stanctechnik.hu

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The SEA-EU-NET 2 project is funded under the 7th Framework Programme for RTD under the Capacities Programme — International Cooperation.
Grant agreement no.: 311784

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Introduction

SEA-EU-NET

The main aim of the first phase of the Framework Programme 7 (FP7) funded SEA-EU-NET project – which started on 1 January 2008 – was to establish science, technology and innovation cooperation between the EU and ASEAN regions. One of the primary targets of the second phase of the project, running until September 2016, is to stimulate deeper and more productive cooperation in three selected global societal challenges of mutual interest: Health, Food and Water. These thematic areas are of complementary interest for both regions: they account for the largest share of co-publications as well as for the joint participation in the Research and Development Framework Programmes of the European Union.

STI Days

In 2014, SEA-EU-NET 2 initiated a forum like event, the so called ASEAN-EU Science Technology and Innovation Days in order to bring together researchers, scientists, science policy makers, innovative companies and other stakeholders from ASEAN and EU countries for a yearly three-day conference on science, technology, and innovation issues. The ASEAN-EU STI Days 2015 are the second edition of this event, which is focusing on presenting ASEAN research excellence to European stakeholders as well as on raising awareness for the current developments in S&T in Southeast Asia.

The Publication

The publication „Excellent Science in ASEAN – Best selected papers and posters from young ASEAN scientists on Water, Food and Health" is a result of a paper and poster competition, closely linked to the ASEAN-EU STI Days 2015. The main aim of this call for papers was to provide a possibility for young ASEAN researchers to publish and to introduce their scientific research to a wide European audience. The authors of the three best papers and two best posters were invited to Paris to take part and present their fields of research to the participants of the event. Further highly evaluated papers and poster abstracts on Food, Water and Health are to be read in this conference paper.

Future Plans

A next book is planned to be published back-to-back with the next ASEAN-EU STI Days in Hanoi, in 2016. Hopefully both the series of STI Days and the connected publications will be successful and popular enough to experience further editions.

……....PART I

Papers on Water……....

Climate change impacts on hydrology and rice yield in Northeast of Thailand

I PUTU SANTIKAYASA[1, 2], SANGAM SHRESTHA[2]

[1] Water Engineering and Management, School of Engineering and Technology, Asian Institute of Technology (AIT), Thailand
[2] Department of Geophysics and Meteorology, Bogor Agricultural University, Indonesia

Abstract

The impact of climate change on rice yields in Thailand was investigated. The SWAT basin scale, continuous time model was used as the hydrological and crop model under four climate models (CGCM3, CSIRO-KM3, ECHAM5, HADCM3) based on two IPCC's A1B and A2emision scenarios for the period 2011 – 2040 (2020s), 2041 – 2070 (2050s), and 2071-2099 (2080s). The mean value of ensemble model for the temperature was +0.8, +1.7 and +2.7 ^{o}C under A1B, on the period of 2020s, 2050s and 2080s, respectively. Under A2, the temperature was +0.8, +1.7 and +3.3 ^{o}C on the period of 2020s, 2050s and 2080s, respectively. The respective value for precipitation was +2.1, +8.7 and +3.0 %, under A1B and +1.7, +8.3 and +13.0 % under A2. The SWAT model was calibrated for the period of 1998 – 2000 and validated for the period of 2001 – 2006. The SWAT model shows the good performances to simulate the stream flow and the rice yield. On the impact assessment of the future climate to the stream flow, the stream flow is expected to increase about 4.3, 18.1, 6.2 % under A1B and about 1.1, 14.1, 28.1 % under A2, for the period of 2020s, 2050s and 2080s, respectively. The impact of future climate on the rice yields shows that the rice yields is expected to decrease about 0.43, 4.07 and 4.99 % under A1B in the period of 2020s, 2050s and 2080s, respectively. Under A2, the rice yield is expected to decrease about 0.71, 4.06, and 6.52 % on the period of 2020s, 2050s and 2080s, respectively. This study provides a useful input to effective planning of water resources of the study area. The study is focusing on the impact assessment of climate change to rice yield using the low-cost assessment tool and in the low level area.

Keywords: climate projection; climate change; rice yield.

Introduction

Thailand has a strong tradition of rice production. It has been known as the world's second largest exporter of rice and over 80% of the Thailand population eats rice as their main meal. Nonetheless, Thailand suffered more than USD $1.75 billion in economic losses related to natural disaster during the period of 1989-2002 in which about USD $1.25 billion (71%) was from crop yield losses (Asia 2009). Climate change was indicated as the causes of the increasing the intensity of natural disaster such as water deficit, flood, storm, pest and diseases in Thailand. Climate change directly affected precipitation and temperature, with rise in temperatures leading to water deficit and floods in the future, changing soil moisture status, and pest and disease incidence (Chinvanno, Suppakorn, and Center 2010). In Thailand, the drier spells in the middle of the wet season causes the damage of plants on the earlier growth stages and floods at the end of the wet season affects the plant on the

harvesting stages. Furthermore, increasing temperature causes the increasing of spikelet sterility in rice and reducing the yield (Wassmann and Dobermann 2007).
The impacts of climate change on rice production in Thailand have been assessed by previous researches. It was estimated that the yield of Thai rice was expected to decline about 18% in the 2020s because of alterations in temperature and rainfall cycle and through changes in soil quality, pests and diseases as the impacts of climate change (Babel et al. 2011). Results from the Mekong Wetlands Biodiversity Conservation and Sustainable Use Programme (MWBP) (2005) indicated that many rice growers in the basin area faced the risk of losing paddy fields from floods and droughts due to climate change. The government of Thailand prepared an action plan on global warming mitigation and raised public awareness on the impacts of climate change (Setsiroot 2007).
Analysing the crop production in a watershed needs the information of the water availability which is used in the agriculture on the production processes. These components are challenging because it is needed to evaluate the water availability and design the infrastructure of the agricultural system. As the natural process, the complex processes in the watershed scale are difficult to understand and simulate. For the past decade, the model approach was used to predict the hydrological processes. Hydrology models have the capability of predicting the complex nature of processes as well as the powerful tool to understanding the process. The tools are also effective to assess the effect of change on land use, climate, and management in the future.
Among the commonly used hydrological watershed models is the “Soil and Water Assessment Tool” (SWAT), a robust hydrologic model successfully employed in a number of watersheds. SWAT is a public domain watershed scale model developed by the Agricultural Research Service of the United States of America’s Department of Agriculture (USDA). The model was developed to predict the effects of land management on water, sediment, nutrients, pesticides, and agricultural chemicals in small to large complex basins.
Applications of SWAT have expanded worldwide over the past decade, especially in the United States and the European Union on predicting the stream flow. There is little research on the use of SWAT to predict the crop yield especially in the tropical climatic conditions of Thailand. The reason may be scarce data, not only temporal but also spatial scale for modelling in watershed hydrology. However, accurately assessing the hydrological processes in Thailand is a very important task because clearly understanding and predicting them is essential for appropriate watershed management as well as the agricultural system.
This study analysed the impact of climate change on the rice yield in Northeast Thailand. Three different future time period, 2011–2040 (2020s), 2041–2070 (2050s) and 2071–2099 (2080s), were chosen for the climate change impact assessment at three provinces of Northeast Thailand.

Materials and methods

Study area and data description

This study was conducted in northeast of Thailand. The basin comprises almost 19 provinces located in Northeast area. These provinces lie between latitude 14.5° to 17.5° North and longitude 102.12° – 104.9° East (Figure 1). The area lies in the Mekong River Basin (MRB). Mekong River flows along the borders of Lao-PDR; run along Thailand border in eastern part of the country and then runs to Cambodia. MRB covers the area in Thailand about 184 x 10^3 km^2 (MWBP 2005). In the Thailand area, about 80% of the population in the MRB have agriculture as their main activity and sources of life hood and rice is the most predominant crop in the area.

In the study area the climate is characterized as a sub humid tropic. The wet season ranges from May to October and the dry season range from November to April. The mean annual precipitation in the study area is 1134 mm. The average temperature in the region ranges from 19.6 ^{o}C – 30.2 ^{o}C. Maximum daily evapotranspiration is about 10.94 mm day^{-1} in April. The minimum evapotranspiration is 0.34 mm day^{-1} in June.
In this study, several data were used to assess the impact of climate change on rice yield in the North East of Thailand. The data includes the spatial, meteorological, hydrological and agronomic data. The spatial data includes the Digital Elevation Model (DEM), soil, and land use. The meteorological data includes the precipitation and temperature. The hydrological data includes the stream flow and the agronomic data includes the cultivar and yield production. The detail of data is summarized on the Table 1 and Table 2.

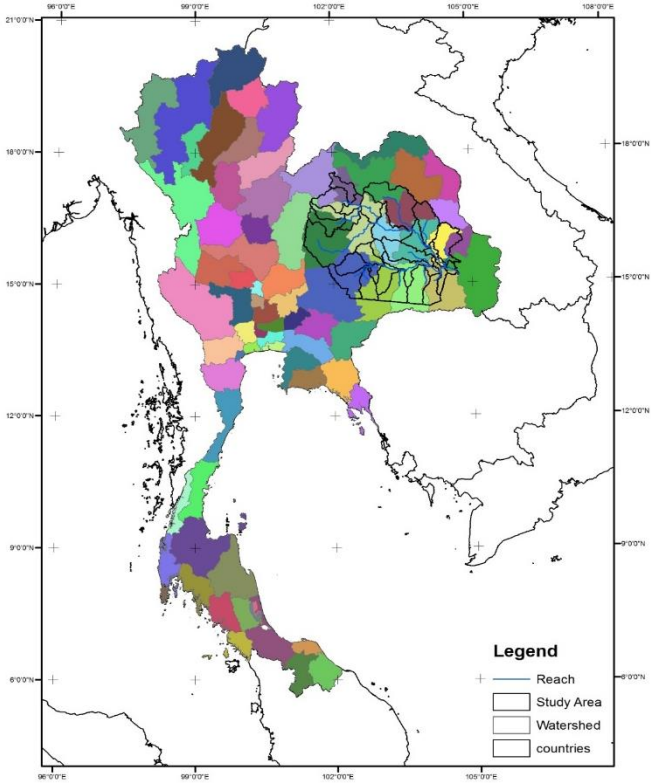
Table 1: Summary data used on the assessment
Source: Babel et al. (2011)

Data	Component	Sources
Spatial	Digital Elevation Model (DEM) 90 m x 90 x	U.S. Geological Survey
	Soil Map	FAO database
	Land use Map	
Meteorological	Precipitation (1980 – 2007)	Thai meteorological Department
	Temperature (1980 – 2007)	
Hydrological	Stream flow (1980 – 2007)	Thai meteorological Department
Agronomic	cultivar	
	yield	

Table 2: Crop growth characteristics and other input data for model calibration and validation, Rice Research Centre, Ubon Ratchathani. NPK: relative concentrations of nitrogen, phosphorus and potassium; DAS: days after sowing
Source: Babel et al. (2011)

Data	Calibration	Validation
Seeding date	8 Aug 1996	28 Aug 1996
Transplanting date	5 Sep 1996	26 Sep 1996
Plant density (m-2)	27	27
Flowering date	3 Nov 1996	18 Nov 1996
Treatment	Rainfed	Rainfed
Fertilizer application	(Mixed NPK 16:16:8) 96 kg ha^{-1} 31 DAS	(Mixed NPK 16:16:8) 96 kg ha^{-1} 31 DAS

Figure 1: The location map of study area



Climate scenario

This study used the future climate scenario constructed by Southeast Asia START Regional Centre at Cualongkorn University - Thailand, as the input to the SWAT model for forecasting future rice yield. The future climate model was predicted using the Global Circulation Model (GCM) ECHAM5 (Table 3). To increase the resolution from global to local extent, the dynamic downscaling was applied. The GCM output was further downscaled at the regional level using the regional climate model (RCM) PRECIS (providing regional climates for impact studies) for the study area at 25 × 25 km.

The predicted future climate scenario was applied to the calibrated SWAT model for the study sites to determine the impacts on rice yield during the 3 future periods. The impacts were then determined by computing the changes in the yield averaged for each of the 3 future decades (2020–2029, 2050–2059 and 2080–2089), with respect to the yield as obtained for the actual daily weather data collected for 10 consecutive years from 1997 to 2006 for each of the 3 sites.

Table 3: Global climate model used in this study
Source: IPCC 2007

Model	Research center	Resolution	IPCC scenario	Period
ECHAM 5	MPI, Germany	Atmospheric (1.87° x 1.87°) Ocean (1.5° x 1.5°)	A1B[1], A2[2]	(2011-2040) (2041-2070) (2071-2099)
HadCM3	Hadley Center, UK	Atmospheric (2.5o x 3.75o) Ocean (1.25o x 1.25o)	A1B, A2	(2011-2040) (2041-2070) (2071-2099)
CSIRO-MK3	CSIRO, Australia	Atmospheric (1.9o x 1.9o) Ocean (1.9o x 1.9o)	A1B, A2	(2011-2040) (2041-2070) (2071-2099)
CSIRO-MK3	CSIRO, Australia	Atmospheric (1.9o x 1.9o) Ocean (1.9o x 1.9o)	A1B, A2	(2011-2040) (2041-2070) (2071-2099)
CGCM3	CCCMA, Canada	Atmospheric (2.8o x 2.8o) Ocean (1.4o x 1.0o)	A1B, A2	(2011-2040) (2041-2070) (2071-2099)

[1] "Rapid economic growth with balance between fossil and non-fossil energy sources"
[2] "Regionally oriented economic development"

Soil and Water Assessment Tool

Soil and Water Assessment Tool (SWAT) is a basin-scale, continuous-time model that operates on a daily time step with the objective of predicting the impact of water/land and agronomic management measures on hydrologic cycles and the accompanying sediment, nutrient and pollutant loadings in un-gauged watersheds (Arnold et al. 1998). The model is physically based, computationally efficient, and capable of continuous simulation over the long time periods. SWAT comprises the sub-modules of each component such as weather, hydrology, plant growth, nutrients, pesticides, bacteria, and land management.

The SWAT model was preferred over other models for this study because its emphasis on water management. Simulation of irrigation water on cropland can be simulated on the basis five alternative sources: stream reach, reservoir, shallow aquifer, deep aquifer, or water body external to the watershed. The irrigation applications can be simulated for specific dates or with an auto-irrigation routine, which triggers irrigation events according to a water stress threshold. SWAT deals with irrigation as a water input as well as uses the precipitation as natural water inputs into a specific hydrological unit.

The successful application of SWAT under irrigated cropping systems can be widely found around the world. Santhi et al. (2005) used SWAT to investigate the irrigation water demands and water savings in irrigation projects in Texas, and the results proved that SWAT model is a useful tool for regional planners and irrigation

managers, the successful experiences of which can be applicable in other irrigation systems. Ficklin et al., (2014) was applied SWAT on the assessment of the impact of the climate to the irrigation on agriculture area in San Joaquin Valley, California, USA. SWAT was also used to investigate the impacts of climatic changes on the hydrological cycles and agricultural water supply and demand. For example, Rosenberg et al. (2003) used SWAT and HUMUS model to examine the potential influences of climatic change have on agricultural productivity and irrigation water supply in the coterminous area, in USA. Numerous studies have shown the application of SWAT in heavily irrigated areas under either humid (Immerzeel, Gaur, and Zwart 2008, Gosain, Rao, and Basuray 2006) or arid climates (Bouraoui et al. 2005). However, in this study, both volume and timing of irrigation were assumed to be uniform within the study sub-basins.

The SWAT model divides the area as the sub watershed which is spatially implicit simulation unit as the Hydrological Response Unit (HRU). HRU consists of a set of homogeneous combination of land use, soil and management features, being determined by users based on either dominant area or user-specified criteria. The soil water content of each HRU is calculated using the equation 1.

$$SW_t = SW_0 + \sum_{i=1}^{t}(P_{day} - P_{surf} - ET_a - D_{seep} - Q_{grw}) \quad \text{..... (Equation 1)}$$

where SW_t is the final soil water contents; SW_0 is the initial soil water content; t is the time (day); P_{day} is the precipitation; R_{surf} is the surface runoff; E_{ta} is the actual evapotranspiration; D_{seep} is the soil percolation and Q_{grw} is the return flows.

The hydrologic balance is simulated for each HRU, including canopy interception of precipitation, partitioning of precipitation, snowmelt water, and irrigation water between surface runoff and infiltration, redistribution of water within the soil profile, evapotranspiration, lateral subsurface flow from the soil profile, and return flow from shallow aquifers. SWAT simulates surface runoff volume and peak runoff rates by using daily or sub-daily precipitation. In this study, the modifying Soil Conservation Service (SCS) curve number method is used.

The potential ET was calculated using the Penman-Monteith approach. Recharge below the soil profile is partitioned between shallow and deep aquifers. Return flow to the stream system and evapo-transpiration from deep-rooted plants was calculated from the shallow aquifer. Furthermore, the water recharges to the deep aquifer was assumed as the water lost from the system.

In SWAT, the phonological plant development was calculated based on daily accumulation of heat units. The potential biomass is based on the method developed by Monteith and Moss (1977). A harvest index is used to calculate the crop yield. The plant growth was estimated based on the combination factors of temperature, water and fertilizer stress. However, the detailed of root growth, micronutrient cycling and toxicity responses were not calculated by SWAT model.

The plant growth in SWAT is controlled using the heat unit concept. Each degree of the daily mean temperature above the base temperature is one heat unit. Therefore, to reach one growth cycle, SWAT used the accumulated of the heat unit. The heat unit accumulation for a given day is calculated by following the Equation 2, as:

$$HU = \bar{T}_{ave} - T_{base} \qquad when \qquad \bar{T}_{ave} > T_{base} \quad \text{..... (Equation 2)}$$

where HU is the accumulated heat unit in a given day, T_{ave} is the mean temperature (°C) and T_{base} is the plant's base or minimum temperature for growth.

To estimate the biomass production, SWAT model uses the leaf area development, light interception and conversion of intercepted light into biomass assuming a plan

species-specific radiation- use efficiency. The amount of daily solar radiation intercepted by the leaf area of the plant is calculating using the Beer's law as Equation 3, as:

$$H = 0.5H_{day}(1 - \exp(-k_l LAI)) \quad \text{(Equation 3)}$$

where H is the amount of intercepted photo-synthetically active radiation on a given day (MJ m^{-2}), H_{day} is the incident total solar (MJ m^{-2}), $0.5H_{day}$ is the incident photo-synthetically active radiation (MJ m^{-2}), k_l is the light extinction coefficient, and LAI is leaf area index.

The potential increase of biomass then calculated as the function of H and radiation uses efficiency (RUE) as Equation 4:

$$\Delta bio = H.RUE \quad \text{(Equation 4)}$$

where Δbio is the potential increase in total plant biomass (kg/ha) and RUE is the radiation use efficiency (kg/ha. $MJ/m^2)^{-1}$.

Model calibration and validation

SWAT was design to apply on the large un-gaged basin and can be used without calibration (Srinivasan, Zhang, and Arnold 2010). However, the calibration processes is used to increase the reliability and the uncertainty of the model. The validation was used to test the validated model using the data which is not used in the calibration process. In this study, a traditional split-sample technique was conducted against observed stream flows of the watershed outlet gauging station.

For the stream flow, the data from 1998 - 2000 is used for model calibration and data from 2001-2006 for model validation. The model parameters used for calibration consist of two sub-modules that are the base flow module and the surface runoff module. The base flow module parameters include the threshold water level in shallow aquifer required for return flow to occur (GWQMN) and the ground water revap coefficient (GW_REVAP). The surface runoff module parameters include the available water capacity of the first soil layer (SOL_AWC), SCS runoff curve number (CN2), and the Manning's "n" for the main channels (CH_N2). For the crop yield, there are no calibration was utilized for the model. However, the performance of the model was evaluated based on crop yield of the model and compared with yield of year 1996 from previous research (Babel et al. 2011).

Two statistic were used to evaluate the model performance: Nash–Sutcliffe Efficiency coefficient (NSE) and the correlation coefficient (R) as well as visual comparison of hydrographs. The NSE indicates how well the model output compared with the observed data, which is determined as:

$$NSE = 1 - \frac{\sum_{i=1}^{n}(X_i - Y_i)^2}{\sum_{i=1}^{n}(Xi - X_{mean})^2} \quad \text{(Equation 5)}$$

where X_i is the observed data; Yi is the simulated output; X_{mean} is the average observed data.

Coefficient of correlation (R) is one statistical measurement widely used to test the linear relation between two variables. The R is computed as:

$$R = \frac{\sum_{i=1}^{n}(X_i - X_{mean})(Y_i - Y_{mean})}{\sqrt{(\sum_{i=1}^{n}(X_i - X_{mean})^2)(\sum_{i=1}^{n}(Y_i - Y_{mean})^2)}} \quad \text{(Equation 6)}$$

where X is the observed data; Y is the model simulated output for the time period entered for evaluation.

Results and discussion

Model performance

Performance of SWAT model to simulate stream flow

The calibrated value of the SWAT parameter was summarized in the Table 4 while the statistic indicator is presented in Table 5. By applying the calibrated parameters, the model was simulated the stream flow with the accuracy about 78% compared with the observed stream flow. The simulated flow was substantially underestimated for the months September – October. However, the model was simulated the stream flow with good performance for the remaining months. The model shows the NSE value of 0.85 indicates that the model was good performance to simulate the stream flow during calibration period.

During the validation period, the predicted peaks flows and the time to peak matched well with the observed value. However, the peak flow on the month October was underestimated while it plotted using the scatter diagram. The simulated flow shows good agreement with the observed data with the value of r^2 =0.82 and NSE=0.89. In the most case, monthly stream flow were reasonably predicted by SWAT for the study area during calibration and validation period. However, stream flows were quite underestimated in the wet month through the period of study.

Table 4: The value of parameters on calibration process

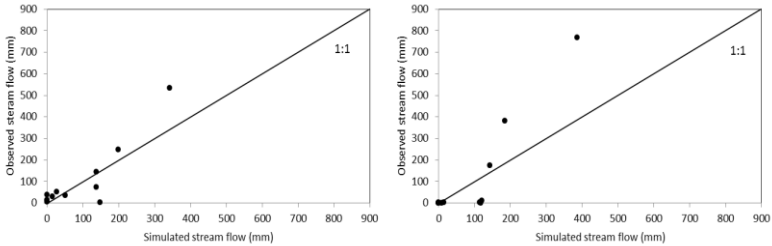
Parameters	Range (unit)	Calibrated
GWQMN	0-100 (mm)	100 (mm)
GW_REVAP	0.02 -0.20	0.1
SOL_AWC	-	0.5
CN2	± 10%	10%
CH_N2	0.01 - 0.3	0.05

Table 5: The statistical value of calibration and validation

Parameters	Calibration		Validation	
	Observed	Simulated	Observed	Simulated
Average (m^3/s)	32.6	29.9	36.8	30.9
Peak flow (m^3/s)	220.8	204.5	333.2	302.9
Volume (10^6 m^3)	86.3	87.6	97.38	90.8
R^2	0.78		0.81	
NSE	0.85		0.89	

A plot of observed and simulated stream flow shows that the SWAT model as managed to reproduce monthly stream flow relatively well. Overall, the results were relatively pleasing on the period of calibration and validation.

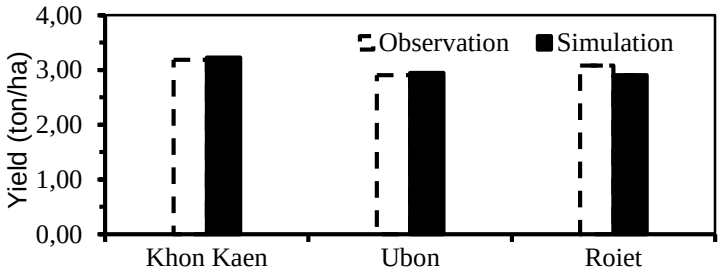
Figure 2: Observed versus simulated stream flow during calibration (left) and validation (right) period.



Performance of SWAT model to simulate rice yield

In this study, the rice yield was simulated using the SWAT model. Since there are no parameter was calibrated related to the crop yield, the statistical was calculated to assess the SWAT performance on simulated the crop yield especially rice yield. The rice yields at three different locations (Khon Kaen, Ubon, Roi Et) were compared with the simulated yield. The results show that rice yield was simulated with r = 0.71. However, the model slightly over predicted the yield in Khon Kaen and Udon provinces where models slightly underestimated the yield in Roi Et province.

Figure 3: Observed and simulated rice yield in Khon Kaen, Ubon and Roi Et Provinces



The observed and simulated yield at Ubon Ratchathani, indicated the model results are in agreement with the observation data. The yields simulated with the bias of 1.3% in Khon Kaen, 1.24% in Ubon and -5.7% in Roi Et. However, the error is still acceptable and the model can be used to project future rice yield under climate change.

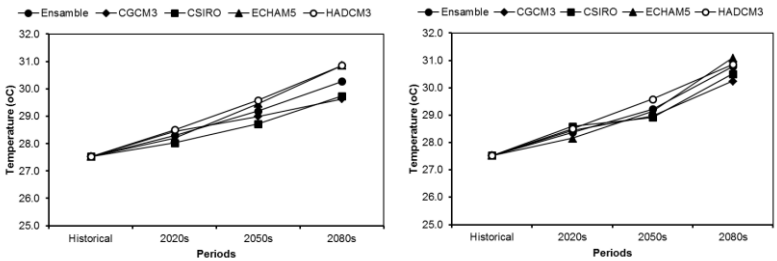
Future climate

In this study, the future climate scenarios are projected based on the four Global Circulation Models (GCMs), namely: CGCM3, CSIRO, ECHAM5 and HadCM3 under two climate scenarios (A1B and A2) and for two climate variables (temperature and precipitation).

Projection of future temperature

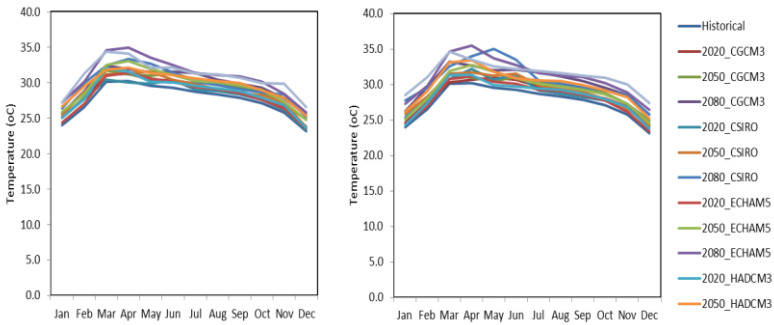
In general, the future temperature is projected to increase by all GCM. As shown in Figure 4, the temperature for the period of 2020s, 2050s and 2080s is projected to increase about 0.8, 1.7 and 2.7 °C under A1B scenario. Under A2 scenario, the temperature is expected to increase 0.8, 1.7 and 3.3 °C for the period of 2020s, 2050s and 2080s, respectively. This result shows that in both scenarios, the temperature is projected to increase in the future, however, the increment will less under A1B scenario than A2 scenario. This is due to the fact that A1B represents medium low scenario which produces less CO2 as compared to A2 scenario which is high scenario.

Figure 4: Projection of temperature from the ensemble GCM under A1B (left) and A2 (right) climate scenarios



On the monthly trend, the future temperature is projected to increase in all of the months (January – December), however, the increment is expected to doffer among the GCM (Figure 5). The highest projection temperature will occur during June under both A1B and A2 scenarios for the period of 2020s. Under the periods of 2050s, the highest projection temperature will occur during May under A1B scenario and during April under A2 scenario. For the period of 2080s, the highest projection temperature will occur during April under A1B scenario, and during April and May under A2 scenario.

Figure 5: Projection of monthly temperature from the ensemble GCM under A1B (left) and A2 (right) climate scenarios

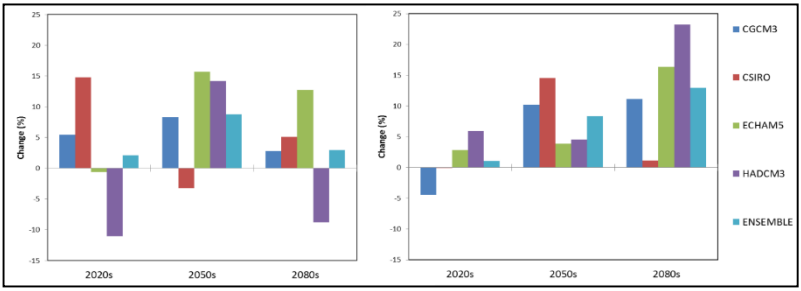


Projection of future precipitation

The projection of precipitation in the study was expected to have the increasing trend for all three periods for both scenarios A1B and A2. Comparing with the baseline period (1970 – 2000), the 2020s, 2050s and 2050s periods shows the increasing trend. However, the precipitation is projected to decrease in 2080s compared with the precipitation in 2050s in A1B scenario. On the A1B scenario, the precipitation was expected to increase about 2.1, 8.7 and 3.0 % on the period of 2020s, 2050s and 2080s, respectively. Under A2 scenario, the future precipitation was expected to increase about 1.7, 8.3 and 13.0 % on the period of 2020s, 2050s and 2080s, respectively.

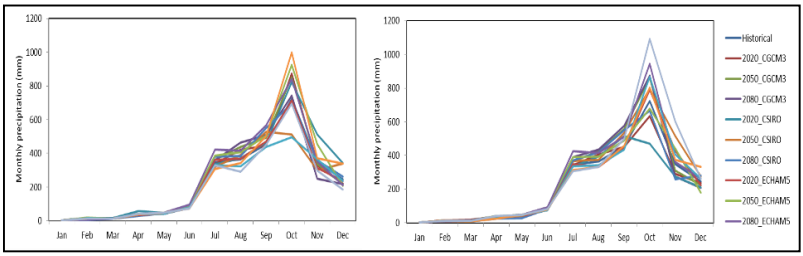
However, the future precipitation was projected to decrease by several GCM under A1B scenarios. For example, HADCM3 was projected the decreasing precipitation about 10% in the period of 2020s period under A1B scenario. Similarly, in the period 2080s, the HADCM3 was projected the decreasing of precipitation about 7.8% compared with the historical period. On the other hand, under A2 scenarios, all the GCMs were projected the increasing of precipitation on all three periods, 2020s, 2050s and 2080s. However, under A2 scenarios, the precipitation projection by the CGCM3 is expected to decrease about 4% in the 2020s period.

Figure 6: Projection of precipitation from the ensemble GCM under A1B and A2 climate scenarios



For the monthly trend, the future precipitation is projected to increase in the period of July – December for all the GCM and all the scenarios (A1B and B2). However, the future precipitation will increase higher under A2 scenario than A1B scenario (Figure 7).

Figure 7: Monthly projection of precipitation from the ensemble GCM under A2 climate scenarios



Impact of climate change on stream flow

The stream flow is affected by the amount of precipitation falling on the area. Similarly, the soil water availability is largely depends to the how much water input i.e. precipitation to the watershed and the output, i.e. evapotranspiration, release into the atmosphere. Therefore, the change on the precipitation and also temperature can significantly affects the soil moisture content and distribution and the finally impact to the stream flow of the watershed.

The variation of the stream flow was indicated by the variation of the precipitation and temperature in the study area. The variation on the temporal and spatial extent is highly influenced by the variability of the hydrological variables. That variability is aggregated on the variability of soil water. Therefore, the variability of the parameters is expected to influence the variability of soil water in any time horizon.

The result indicated that, the future stream flow is expected to increase for the period of 2020s, 2050s and 2080s under both climate scenarios, A1B and A2. Under A1B scenario, on the 2020s period, the future stream flow is expected to increase about 14.1 and 22.7 % by the CGCM3 and CSIRO GCM and is expected to decrease by 0.7 and 18.7 % by the ECHAM5 and HADCM3 GCM. However, as the ENSEMBLE, the future stream flow under A1B scenario on the 2020s period is expected to increase about 4.3 % compared with the historical period. On the 2050s periods, the future stream flow is expected to increase by 16.7, 30.5 and 33.2 % by the CGCM3, ECHAM5 and HADCM3 GCM and is expected to decrease about 8.3 % by the CSIRO GCM. As the ENSEMBLE, the stream flow on the 2050s period is expected to increase about 18.1 % compared with the historical period. On the last period of this century 2080s, the stream flow is expected to increase by 5.6, 7.0 and 23.0 % by the CGCM3, CSIRO and ECHAM5, however, is expected to decrease by 11.0 % by the HADCM3 GCM. In the ENSEMBLE, the stream flow is expected to increase about 6.2 % compared with the historical period.

Under A2 scenario, in the period of 2020s, the stream flow is expected to increase about 6.8 and 13.9 % by the ECHAM5 and HADCM3, however, is expected to decrease by about 7.9 and 17.5 % by CGCM3 and CSIRO GCM. The ESEMBLE analysis shows that on the 2020s period, the stream flow is expected to decrease about 1.1 % compared with the historical period. In the period of 2050s, the stream slow is expected to increase about 20.3, 22.6, 4.6, and 8.9 % by the CGCM3, CSIRO, ECHAM5 and HADCM3 GCM, respectively, and it is expected to increase about 14.1 % on the ENSEMBLE compared with the historical period. In the period of 2080s, the stream flow is expected to increase about 22.8, 1.9, 32.6 and 55.0 % by the CGCM3, CSIRO, ECHAM5 and HADCM3 GCM, respectively. Similarly, while assessed by the ESEMBLE, the stream flow is expected to increase about 28.1 % compared with the historical period.

For the monthly bases, the stream flow is projected to increase during July – November in both A1B and A2 scenarios. However, the projection of stream flow during October for the period of 2050s is expected to higher then period 2080s under A1B scenario.

Figure 8: Projection of precipitation from the ensemble GCM under A1B and A2 climate scenarios

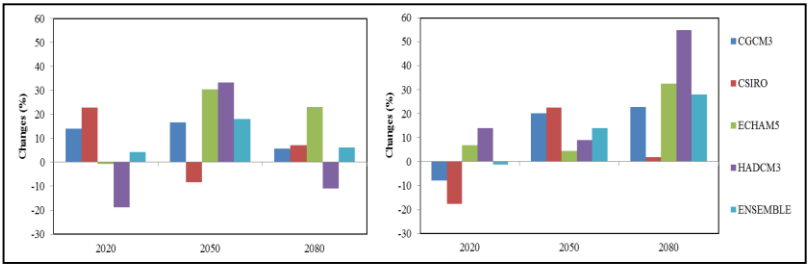
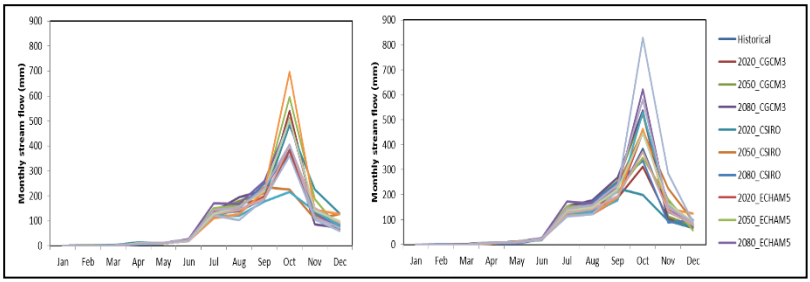


Figure 9: Monthly projection of stream flow from the ensemble GCM under A2 climate scenarios



Effect of climate change on rice yield

According the data from previous research for the planting and harvesting date, this research used the similar parameters as used in the previous research (Babel et al. 2010). In this research, the result shows that during the period of 2020s, the crop yield will decrease about 0.43 % under A1B scenario and 0.71% under A2 scenario. For the period of 2050s, the future crop yield is expected to decrease about 4.70 and 4.06% under A1B and A2 scenario, respectively, compared with the historical period. For the period 2080s, the future crop yield is expected to decrease about 4.99 and 6.52% under A1B and A2 scenarios, respectively, compared with the historical period.

Table 6: Historical and projection of crop yield under A1B and A2 climate projection using ensemble GCM at three locations

		Rice Yield (ton/ha)			
		Historical	2020s	2050s	2080s
Khon Kaen	Baseline	3.186			
	A1B		3.172	3.036	3.027
	A2		3.164	3.056	2.978
Ubon	Baseline	2.908			
	A1B		2.895	2.771	2.763
	A2		2.887	2.790	2.718
Rioet	Baseline	3.081			
	A1B		3.068	2.936	2.927
	A2		3.059	2.956	2.880

Table 7: Historical and projection of crop yield under A1B and A2 climate projection using individual GCM in Khon Kaen

		Rice Yield (ton/ha) in Khon Kaen			
		Historical	2020s	2050s	2080s
		3.186			
CGCM3	A1B		3.089	2.976	2.898
	A2		3.110	2.889	2.837
CSIRO	A1B		3.156	3.009	2.982
	A2		3.026	3.026	2.912
ECHAM5	A1B		3.150	3.016	2.895
	A2		3.262	3.068	2.990
HADCM3	A1B		3.293	3.145	3.333
	A2		3.256	3.243	3.173

Table 8: Historical and projection of crop yield under A1B and A2 climate projection using individual GCM in Ubon

		Rice Yield (ton/ha) in Ubon			
		Historical	2020s	2050s	2080s
		2.908			
CGCM3	A1B		2.820	2.716	2.646
	A2		2.839	2.637	2.589
CSIRO	A1B		2.881	2.747	2.722
	A2		2.762	2.762	2.658
ECHAM5	A1B		2.875	2.753	2.642
	A2		2.978	2.800	2.729
HADCM3	A1B		3.006	2.870	3.042
	A2		2.972	2.960	2.896

Table 9: Historical and projection of crop yield under A1B and A2 climate projection using individual GCM in Roi Et

		Rice Yield (ton/ha) in Rio Et			
		Historical	2020s	2050s	2080s
		3.081			
CGCM3	A1B		2.988	2.878	2.803
	A2		3.008	2.794	2.743
CSIRO	A1B		3.052	2.910	2.884
	A2		2.926	2.926	2.816
ECHAM5	A1B		3.046	2.916	2.799
	A2		3.155	2.967	2.891
HADCM3	A1B		3.185	3.041	3.223
	A2		3.149	3.136	3.068

As shown in the Table 6 that the crop yield in period 2020s is expected to decrease in the future, however, the future crop yield is projected to increase under HADCM2 climate projection for both A1B and A2 scenario. The A2 climate scenario is also expected to impact on increasing of crop yield for the 2020s period. Similarly, for the period 2080s, the A1B climate scenario under HADCM3 projection will increase the future crop yield about 4.6% compared with the historical period.

As shown in the Table 6 - 9, the decrease in paddy yield at three locations of study area in the future period is mainly and increasing the temperature during the future period.

Furthermore, the increasing of precipitation during August and September which is also impact on the decreasing of the crop yield because in those month is the harvesting period of paddy in all areas. The increasing of the temperature will impact on the decreasing of the soil moisture and affect the decreasing of the crop yield.

Conclusion

The present study assessed the future climate under two IPCC-SRES scenarios, A1B and A2, and the impact of climate change on the stream flow and rice yield in Northeast area of Thailand using SWAT model. The SWAT model simulates the stream flow in the study area and the rice yield using the Erosion-Productivity Impact Calculator (EPIC) plant growth model. It is indicated the SWAT model shows the good performance to simulate the stream flow. Similarly, the SWAT model shows the good performance on simulating the rice yield in the study area.

The future temperature was projected to increase in all of the future time period; 2020s, 2050s and 2080s; under both climate scenarios, A1B and A2. Similarly, the precipitation was expected to increase in the future period, 2020s, 2050s and 2080s; under both climate scenarios, A1B and A2. The stream flow is projected to follow the pattern of the precipitation in the future period. The increasing of precipitation is expected to impact to the increasing of the stream flow in the future period.

The assessment of future crop yield under future climate indicates the decreasing of rice yield in the study area for the period of 2020s, 2050s and 2080s. The rice yield is expected to decrease about 0.43- 0.71% in the 2020s period and about 4.99 – 6.52% in the end of the century period. The decreasing of the crop yield is expected because of the change of the pattern of precipitation during the early stage and the harvesting stages in the future. Increasing the precipitation on the harvesting stages, increase the possibilities of the flood, in which causes a failed harvest of rice in the area.

It can be conclude that the results of this study provide a useful input to effective planning of water resources of the study area. The study is focusing on the impact assessment of climate change to rice yield using the low-cost assessment tool and in the low level area.

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Uncertainties in SWAT extreme flow simulation under climate change of the Bago River Basin in Myanmar

AUNG YE HTUT, SANGAM SHRESTHA

Water Engineering and Management, Asian Institute of Technology, Thailand

Abstract

Uncertainty in climate change impact analysis has been widely recognized. Analyzing it becomes an important task particularly when impact analysis results are used for adaptation purposes. A methodology aiming to investigate the impact of climate change due to impacts of uncertainty sources (the emission scenarios and General Circulation Models) on future extreme flows in the Bago River Basin is proposed. Delta change approach is applied to bias correct GCM outputs and the extreme flows are simulated by the SWAT (Soil and Water Assessment Tool) model. The SWAT model calibration and validation results indicate that the model has a good performance in the Bago River Basin. In the whole basin, the monthly stream flows are observed in increasing from May to September and peaked in September relative to heavy rains under two scenarios during future periods of 2020s, 2050s and 2080s. The uncertainty analysis results show that the uncertainty introduced from GCM structures is much larger than those from emission scenarios at Zaungtu and Bago stations. A majority of the GCMs can project an increase in stream flow for all the three future periods in the Bago River Basin. MIROC5 and BCCCSM1.1 project maximum increased stream flow changes for all the two scenarios in the whole basin. The highest range of uncertainty for annual stream flow is projected under RCP4.5 scenario for the 2020s and 2050s (300 m^3/s to 450 m^3/s) and under the RCP8.5 scenario for the 2080s (350 m^3/s to 450 m^3/s). An average value of all GCMs indicates a positive all seasons as well as in annual stream flow changes except summer season in the Bago River Basin.

Keywords: climate change; GCMs; RCPs; SWAT; uncertainties.

Introduction

Industrialization and urbanization are increasing dramatically along with the population growth all over the world. Increased emission of greenhouse gases especially CO_2 causes global warming. Global and regional climate have been changing as evidenced by increase in temperature and rainfall intensity. The impacts of climate change primarily driven by global warming are highly extensive, complicated and uncertain. Global surface temperature has risen by 0.74 ˚C during the twentieth century and the warming trend accelerated in the last 50 years (Arnell, 2004). Climate change impacts, such as changes in precipitation and temperature, are leading to more flood and drought events each year. Climate variability and change have a huge effect on the general well-being of communities across the world. Also many living organisms are sensitive to changing climate patterns. Availability and variability of water resources have been affected by climate change effects. Climate change has affected water resources globally and regionally in the last 30 years (Boyer et al., 2010). Particularly rainfall, known as the main driver of hydrological cycle, has been varying in the most parts of the world.

The changes in climate variables such as rainfall and temperature has distributed the public health, industrial, irrigation and municipal water demands and the ecosystem (Zhang et al., 2011).

Representative Concentration Pathways (RCPs) are four greenhouse gas concentration (not emissions) trajectories adopted by the IPCC for its fifth Assessment Report (AR5). The pathways are used for climate modeling and research. They describe four possible climate futures, all of which are considered possible depending on how much greenhouse gases are emitted in the years to come. The basic concept is that of RCPs which are expressed in terms of watts per square metre of radiative forcing (W/ m^2). Fifth phase of the Climate Model Intercomparison Project (CMIP5) is the principal framework for coordinated climate modeling experimentation supporting the preparation of the t AR5 to be released in 2013. About 20 modeling groups from around the world are undertaking the CMIP5 experiments and model data is being hosted on the Earth System Grid which consists of international data nodes and gateways.

The impact analysis of climate change on hydrology or water resources is a wide issue. The hydrological cycle has been substantially influenced by climate change and human activities. It is therefore of utmost importance to analyze the impact of climate change on hydrology, particularly on a regional scale, in order to understand potential future changes of water resources and water-related disaster, and provide support for regional water management (Xu et al., 2013). Hydrological models provide a framework to conceptualize and investigate the relationships between climate, human activities and water resources (Huang, 2014). Wang et al (2006) stated that choosing of hydrological model is dependent on the purpose of study and data available. Several studies have attempted to quantify the uncertainty arising from a variety of sources, including future greenhouse gas (GHG) and aerosol emissions, GCM structure and initial conditions downscaling method and hydrologic model structure and parameters (Schwank et al., 2014). Boe et al., (2007) investigated the uncertainties associated with low-flow change, stemming from a combination of emission scenarios, GCM structures, statistical downscaling methods, and hydrologic model parameters and structure. Their results indicated that the low flow change was most sensitive to uncertainty in the GCM structure and downscaling method, but it was less affected by uncertainties due to hydrological model parameters and emission scenarios. Hansson et al., (2008) explained that investigation of uncertainty within climate change hydrological impact assessments has often focused on GCM uncertainty. A more common approach to investigating GCM uncertainty is to use a range of projections for the same emissions scenario derived from an ensemble of GCMs (Gardner, 2009). Countries like Myanmar, which are dependent on stream flow may face greater problems as uncertainty in the amount, timing and frequency of flows increase.

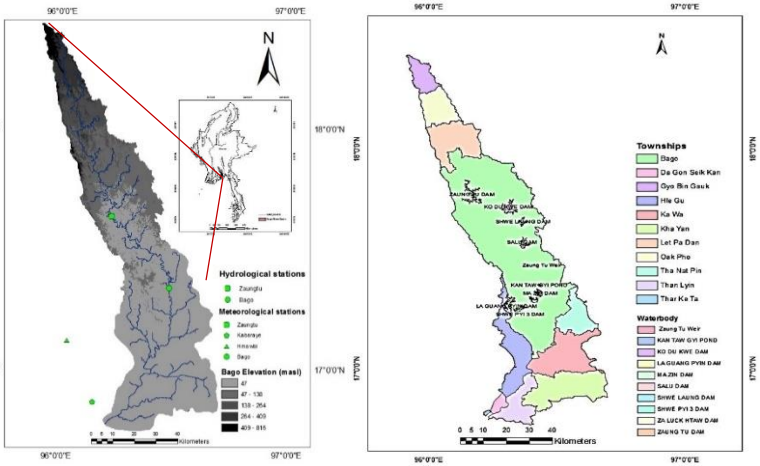
Study area and data

Study area

This paper is conducted in the Bago River Basin. The Bago River Basin is the smallest river basin among the top ten basins of Myanmar, covering 91% of Bago district and its area is 4,883.1km^2. The basin area lies within latitudes 16° 40′ 30″ and 18° 25′ 48″ N, and longitudes 95° 54′ 39.6″ and 96° 44′ 38.4″ E. This river basin is one of the most important and useful river basins in lower Myanmar for hydropower generation, irrigation use, fisheries and navigation use. In this study basin, a hydropower dam for electricity and a diversion weir for irrigation use were constructed near Zaungtu village in 1996 and 1998, respectively. Total number of 2,1498 stream flowing into the Bago River from start to end. For the purpose of flood control during the rainy season and the irrigation water use for summer paddy cultivation and the water supply for green project around Yangon area during the dry season, three earthen dams namely Kodukwe, Salu and Shwelaung were constructed in 2011 and opened in May, 2012

and the flood diversion channel from Zaung Tu weir to Moeyongyi lake was also completed in 2012 (Bago City Administrative, 2013) seen in Figure 1. A large part of the Bago River Basin almost 60% is below 47m elevation, followed by the elevation range of 47 to 138 (14%). The highest elevation of the sources of the Bago River is about 800 meters above sea-level in southern Bago Mountain Range. The area around Bago Mountain Range is mountainous and has steep slopes but most of the area is flat plain with good nutritious farm land. The river valley is mostly steep in the mountainous area of southern Bago Mountain range, but it decreases in elevation towards the south (Hlaing et al., 2008). This study area has four meteorological stations (Bago, Zaungtu, Kabaraye and Hmawbi) and only two hydrological stations (Bago and Zaungtu) locating beside the main river for stream flow data (Figure 1).

Figure 1: Elevation ranges, meteorology and hydrology stations and location of the Bago River Basin



Hydro meteorological data

According to Koppen's climatic classification scheme, the climate of the Bago River Basin is the tropical monsoon (Am) with a heavy seasonal rainfall, high temperature, and distinct wet and dry seasons (Hlaing et al., 2008). The mean monthly rainfall and temperature regimes of all the meteorological stations used in this study for the period of 1975-2005 are as shown in Table 1. According to the meteorological records, January and December are the coldest months whereas April is the hottest month. The basin scale average annual values for maximum temperature, minimum temperature and mean temperature are 33 °C, 21 °C and 27 °C respectively (Figure 2). Hence, the annual range of temperature is about 6.8 °C. Within the baseline period, Bago River Basin has had an average annual rainfall of 2,980 mm with 130 rainy days per annum in average. The monthly distribution of precipitation is directly related to the southwest monsoon. The monsoon precipitation (May to September) comprises 91% of the yearly cumulative. With 46% of annual precipitation being shared between them, July and August are the wettest months for the basin. On the other hand, January and February are the driest months shown in Figure 3. Figure 4 shows high flows occurring in August at both stations. The low flows can be seen in

January, February, March, April and December. Comparison of the two stations, Bago gives peak of flow in rainy season than Zaungtu except in summer and winter of Zaungtu (Figure 4). Seeing the distribution of observed annual average stream flow that Bago is higher in stream flow than Zaungtu until 2007. But the stream flow has been increasingly changed in Zaungtu station after 2007 (Figure 4). The period of stream flow data availability for the two stations is mentioned in Table 1.

Table 1: Statistics of meteorological and hydrological parameters at DMH stations in the Bago River Basin

Station	Avg annual precipitation (mm)	Mean Tmax (°C)	Mean Tmin (°C)	Tmean (°C)	Avg annual Stream flow (m^3/s)	Lat_N (Degree)	Lon_E (Degree)	Elevation (masl)
Bago	3185	32.6	20.6	26.6	1597.9	17.3	96.5	19
Kabaraye	2607	33.1	21.2	27.2	-	16.9	96.2	31
Zaungtu	2746	33	20.1	26.6	1412.8	17.6	96.2	37
Hmawbi	2541	33.1	21.7	27.4	-	17.1	96.0	22

Tmean= mean temperature, *Tmax=* maximum temperature, *Tmin=* minimum temperature
Meteorological parameters are between 1975 to 2009
Hydrological parameters are between 1990-2009

Figure 2: Distribution of average monthly Tmax, Tmin and Tmean for the whole Bago River Basin for 1975-2005 (the baseline period)

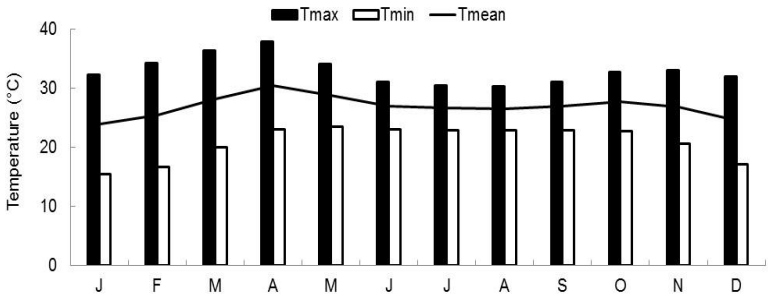


Figure 3: Distribution of average monthly precipitation for all the meteorological stations and the whole Bago River Basin

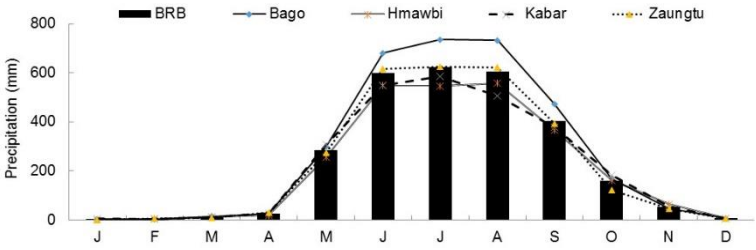
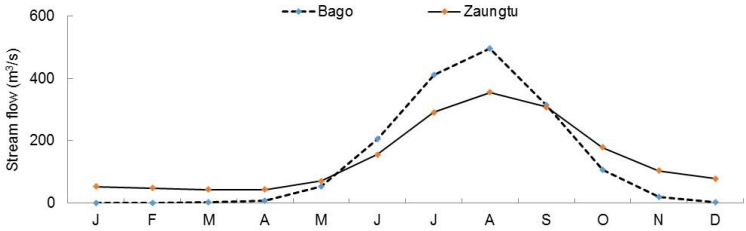


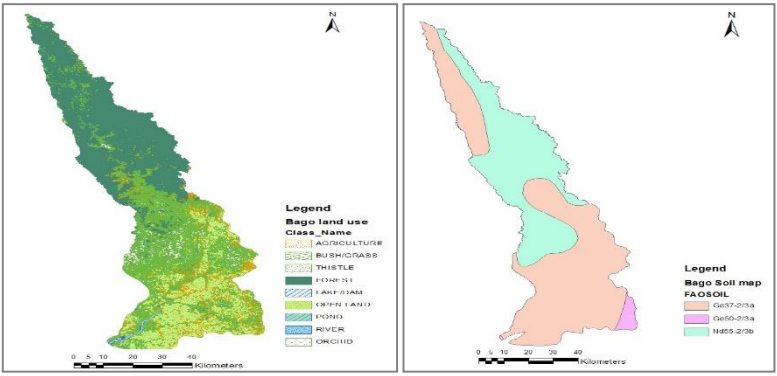
Figure 4: Distribution of average monthly stream flow at all the stream flow stations in the Bago River Basin for 1990-2009 (the baseline period)



Spatial Data of the study area

The spatial data required for developing a physically based hydrological model are: Digital Elevation Model (DEM) (Figure 1), Land use and Soil types (Figure 5).

Figure 5: Land cover and soil types of the Bago River Basin in the year 2010

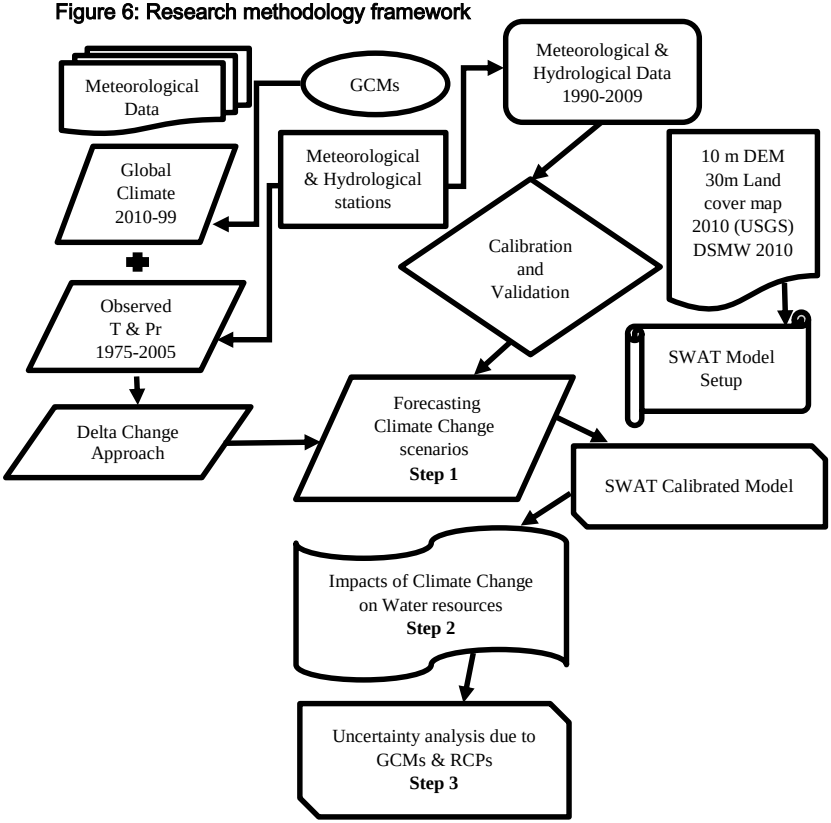


This study is used 10m resolution DEM of the whole basin created from topographic maps and channel survey maps by using some steps. The land cover and land use

map used is developed from 30m resolution raster data with projection of WGS_1984_Zone_47N from USGS with the use of image processing for the year of 2010. In this basin, grass is the most dominated land cover type contribution more than 32% of the total area. Open land is almost 28% of the study area. The other major land cover types in the basin are agriculture 18% and forest 10% of this area in 2010 year. Lake, River and Thistle are less percentage contributions in this area. For soil map, Digital Soil Map of the World (DSMW) is produced for the whole basin soil map. This soil map is a resolution of 1: 5000,000 scales and good quality as well as soil maps from SOTER website. As per DSMW, only three major classes of soils are found in the Bago River Basin shown in Figure 5. The dominate soil types are Eutric Gleysols (Ge37-2/3a), Eutric Gleysols (Ge50-2/3a) and Nitosols (Nd55-2/3b) seen in Figure 5.

Methodology

Figure 6: Research methodology framework



Forecasting climate change scenarios of the Bago River Basin

In this section, to understand the variations in temperature, the projected changes in both variables are analyzed for the three spells- the 2020s (2010-2039), the 2050s (2040-2069) and the 2080s (2070-2099) relative to the 1975-2005 climatology under the RCP4.5 and 8.5 scenarios. The future climate data in this study are downloaded from ESGF (Earth System Grid Federation) and IS-ENES, Climate4impactportal websites. Six GCMs that are included in CMIP5 are considered in this study shown in Table 2.

Table 2: A list of the climate models used in this study with a brief indication of their origin, resolution and the number of realizations available for each climate change scenario

Model	Resolution (long by lat)	Scenarios	Origin
BCC-CSM1.1	2.815˚×2.815˚	RCP 2.6, 4.5, 6 & 8.5	Beijing Climate Center, China
CCSM4	1.25˚ ×0.9˚	RCP 2.6, 6 & 8.5	National Center for Atmospheric Research, USA
MIROC5	1.40˚×1.40˚	RCP 2.6, 4.5, 6 & 8.5	Atmosphere and Ocean Research Institute, Japan
MRI-CGCM3	1.125˚×1.125˚	RCP 2.6, 4.5 & 8.5	Meteorological Research Institute, Japan
BNU-ESM	0.75˚ × 0.75˚	RCP 2.6, 4.5 & 8.5	College of Global Change and Earth System Science
FGOALs-g2	1.9˚ × 1.9˚	RCP 2.6, 4.5 & 8.5	Chinese Academy Science

Time series of GCMs show a bias in simulating 20th century precipitation and temperature fields and, therefore, cannot directly be used to force hydrological models in order to assess the impact of the projected climate change on certain components of the hydrological cycle. In this study, the delta change/ratio method uses observed climate as a baseline and the capability of the GCM to produce simulations that are comparable to observed climate is less crucial. It is stable and always gives results that can be related to the present conditions. In this technique, the observed climate time series are adapted with estimated monthly climate changes from the GCM output. The future monthly temperature shifts (T_f) and precipitation (P_f) scaling factors are calculated by Equations 1 and 2 respectively. And Equations 3 and 4 are then used in predicting the future meteorological parameters.

$T_f = T(GCM\ simulated)_f - T(GCM\ simulated)_p$ .....(Equation 1)

$P_f = PPT(GCM\ simulated)_f\ /\ PPT(GCM\ simulated)_p$ .....(Equation 2)

Where, $_P$ is used for the present and $_f$ is used for the future time period. The future scenarios are then generated using Equation 3 and 4.

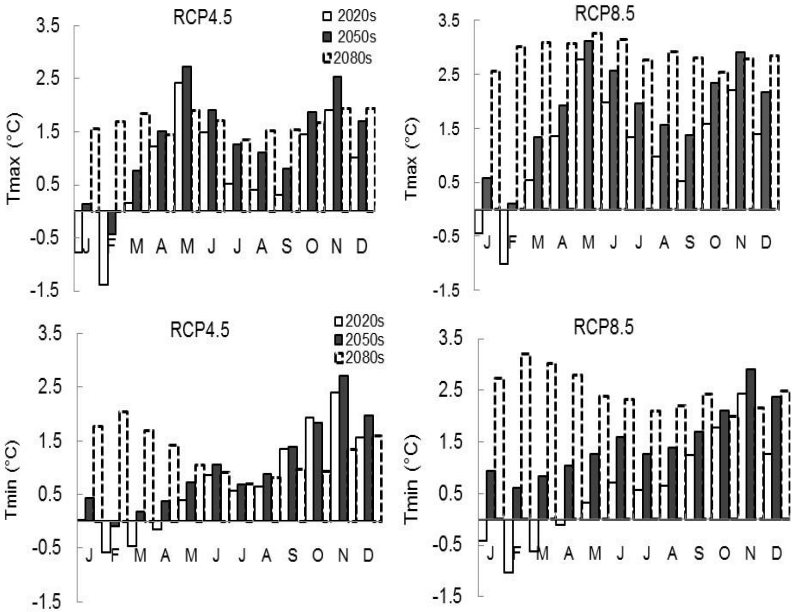
$F_s\ (T)\ = T\ (observed\ ) + T_f$ .....(Equation 3)

$F_s\ (PPT)\ = PPT\ (observed\ )\ \times\ P_f$.....(Equation 4)

In the whole study area for the future, the simulated average temperature will be higher than the observed temperature with peak values in the month of April under

both scenarios. The mean temperature is projected to be at 38.4˚C (2020s), 38.8˚C (2050s) and 39.3˚C (2080s) in April under RCP4.5 and at 38.5˚C (2020s), 39.4˚C (2050s) and 40.8˚C (2080s) under RCP8.5 seen in Figure 7.

Figure 7: Future average monthly changes in Tmax and Tmin relative to the baseline period (1975-2005) under RCP4.5 and RCP8.5 in the Bago River Basin



Compared to the present-day condition, Tmax over the basin is projected to increase by 2.9 °C by 2080s period under the high emission scenario RCP8.5 and 1.7 ºC under RCP4.5. Strong temperature change for all three periods (>1.8°C) appears along the whole basin in May and November under RCP4.5, whereas under RCP8.5, it is observed that strong temperature change will occur for all three periods (>2.7ºC) only in May. Moderate temperature change for all three periods (>1°C) appears in April, June, October and November under RCP4.5 and only in November under RCP8.5 (>2ºC). Although the observed Tmax is high in April in the whole basin, the temperature change for April is quite moderate. However for August, the observed Tmax is low, but its temperature change is high in 2080s period under RCP8.5. For minimum temperature, except for the months of January, February, March and April of 2020s, all projections under both RCPs indicate an increase in Tmin. Tmin over the whole basin was projected to increase by 2.5°C by 2080s under RCP8.5 and 1.3ºC under RCP4.5. Strong minimum temperature change for all periods (>1.3°C) appears in November under RCP4.5 and also (>2ºC) under RCP8.5 scenario. Moderate temperature change for all three periods (>1.5°C) appears in December under RCP4.5 and October under RCP8.5 (>1.8°C).

It is also observed that mean temperature will undergo the greatest change (rise) in 2080s period as most of the high temperature changes are observed in this period.

The simulations of average monthly precipitation under RCP4.5 and 8.5 scenarios can be compared with the baseline period of the whole basin as shown in the Table 3. The peak of observed precipitation in the whole basin can be seen in July, but the simulated precipitation peaks are changed according to the scenarios. September is observed as a peak of precipitation amount (934 mm) in 2020s, 855 mm in 2050s under RCP4.5 and 826 mm in 2020s under RCP8.5. The other months of peak precipitation are August (691 mm) in 2080s under RCP4.5 and 806mm in 2050s under RCP8.5 and July (868 mm) in 2080s under RCP8.5. Under RCP4.5 scenario, the simulated annual precipitation amounts are observed to be 4,085 mm (37% increase from observed annual precipitation) in 2020s, 4,185 mm (40% increase) in 2050s and 3,157 mm (6% increase) in 2080s. For RCP8.5 scenario, the simulated annual precipitations are predicted 4,116mm (38% increase from the baseline data) in 2020s, 4,263mm (43% increase) and 4,178mm (40% increase) in 2080s. The annual precipitations under RCP 4.5 and 8.5 do not show a constant trend but rather fluctuate throughout 2010-2099 period. Rainfall has the greatest drop of 70mm in May in Bago River Basin for both scenarios. The highest rise in precipitation fluctuation is projected for the month of September. The months of December, January and February showed comparatively insignificant fluctuations. The changes in precipitation can play important role in causing the shift of weather patterns in 2010-2099. There can be seen the same pattern of the average monthly changes in precipitation for the whole basin under both scenarios that May has decreasing precipitation changes and September is the peak of precipitation change. The peak of precipitation changes of RCP4.5 is greater than those of RCP8.5 and 2020s period is the affected period for precipitation changes.

Table 3: Average monthly precipitation (mm) in the Bago River Basin during the baseline period (1975-2005) and the three future periods under RCP4.5 and 8.5 scenarios

Month	Average monthly precipitation (mm)								
	Baseline			RCP4.5			RCP8.5		
	2020s	2050s	2080s	2020s	2050s	2080s	2020s	2050s	2080s
Jan.	2.2	2.2	2.2	2.1	2.8	3.7	2.0	3.3	5.2
Feb.	3.9	3.9	3.9	1.9	1.8	3.2	1.2	1.6	2.8
Mar.	11.0	11.0	11.0	7.2	22.4	25.4	7.1	16.6	19.9
Apr.	25.1	25.1	25.1	41.9	71.0	76.4	39.0	84.6	71.1
May	283.1	283.1	283.1	224.6	242.5	326.8	216.7	262.8	309.5
Jun.	597.8	597.8	597.8	687.6	716.3	649.2	741.6	801.6	738.9
Jul.	622.2	622.2	622.2	723.5	689.9	689.1	759.8	736.6	867.6
Aug.	604.1	604.1	604.1	770.7	798.6	691.2	818.0	852.1	833.7
Sep.	402.6	402.6	402.6	933.8	855.0	421.7	825.9	805.7	834.8
Oct.	159.2	159.2	159.2	538.8	516.1	163.9	476.3	519.1	362.8
Nov.	52.2	52.2	52.2	148.5	261.1	97.0	225.7	172.1	121.0
Dec.	6.1	6.1	6.1	4.6	7.7	10.0	3.3	7.0	10.3

Hydrological modelling

Of particular concern in this study is the Soil and Water Assessment Tool (SWAT) model which is widely used distributed hydrological model. Only the two hydrological stations (Zaungtu and Bago) are used in this study area.

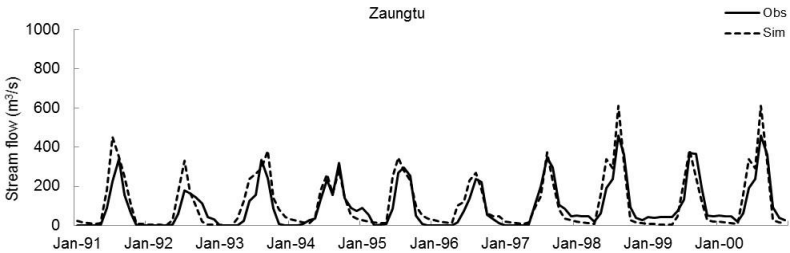
The stream flow data of 1990-2009 at these two stations are used in comparison with observed and simulated stream flow in calibration and validation. The outlet of the whole basin is selected at the entrance of Yangon River (end of the Bago River

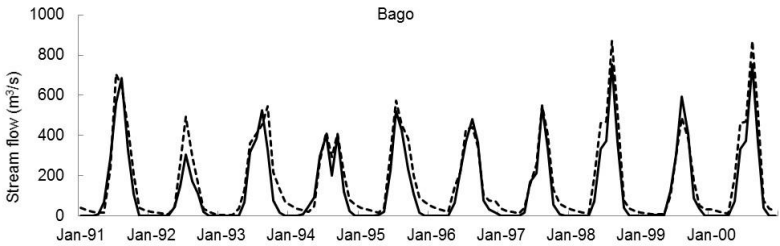
Basin). The whole basin is divided into 19 sub-basins with total basin area of 4883.1 km^2. DEM calculates good watershed characteristics such as elevations, longest flow path, reach etc. at sub-basin level. This study uses 10% threshold for land use, soil and slope (limitation is 5-20% for SWAT) to reduce the number of HRUs following the guideline of a research evaluated by Strauch et al., (2012). The land use map of 2010 and soil map (described in Figure 5) are loaded in ArcSWAT 2012 for land use and soil definition. With the combination of unique land use, soil and slope 71 HRUs for 19 sub-basins are generated within the Bago River Basin. Before actual SWAT calibration and validation, various input parameters are required are required for sensitivity analysis. In this study, the parameters controlling the stream flow are tested and changed. Total 22 sensitivity parameters are analyzed for sensitivity analysis. The three error parameters: R^2, PBIAS and NSE are used for the model performance. Table 3 shows the error parameter values using daily stream flow data for comparison. The validation is taken for 2001-2008 periods after good results of calibration. Checking these error parameters using monthly stream flow can be seen in the Figure 8. All sensitivity parameters are satisfactory at both Zaungtu and Bago stations in using daily and monthly data. R^2 determines the agreement between predicted and observed variable. NS shows the goodness of fit of observed and simulated data with 1:1. PBIAS evaluates whether the simulated data tends to be larger or smaller than the observed values (Zhang et al, 2013). In this study, sensitivity analysis was checked by using NSE, $R^2$and PBIAS for satisfactory of the calibrated parameters. Zhang et al., 2011 stated that NSE and R^2 values are greater than 0.6 means perfect match. On the other side, PBIAS should be less than 15% for good predicted efficiency (Ercan et al., 2014).

Table 4: SWAT performance to simulate stream flow using daily and monthly data for baseline period of 1991 to 2008

Station	Data	Calibration (1991-2000)			Validation (2001-2008)		
		R^2	NS	PBIAS	R^2	NS	PBIAS
Zaungtu	Daily	0.726	0.710	-21	0.725	0.706	-19
	Monthly	0.812	0.815	-15	0.824	0.811	-18
Bago	Daily	0.787	0.732	-24	0.708	0.701	-22
	Monthly	0.934	0.858	-16	0.934	0.814	-14

Figure 8: Comparison between observed and simulated monthly stream flow at Zaungtu and Bago for calibration period 1991-2000





Uncertainty in Stream Flow Projections

Uncertainty in seasonal and annual stream flow projections under two scenarios

The range of change in seasonal and annual projections under RCP8.5 from their respective GCMs for Zaungtu station is shown in Figure 9. The range is presented using box and whiskers plot. The box represents the middle 50% range, while the bar represents the median value. The whiskers at the two ends represent the extreme projections. The small size of the box indicates more agreement among the GCMs seen in the following figures. According to these results, under RCP8.5 scenario at Zaungtu, the annual stream flow is predicted to increase 53 m^3/s (37%) in 2020s, 57 m^3/s (40%) in 2050s and 39 m^3/s (27%) gradually. The summer is also the affected season with decreasing changes of 35 m^3/s in 2020s, 23 m^3/s in 2050s and 32 m^3/s in 2080s with average 64% decreasing change. During the rainy season, the average stream flow changes are higher than other seasonal flows in all periods. In the winter season, an increased change is varied from 80 m^3/s in 2020s, 65 m^3/s in 2050s and 42 m^3/s in 2080s with average percentage of 37 under RCP8.5 (Figure 9). In Zaungtu station, annual flow changes of RCP8.5 is always greater than RCP4.5 showing that 30m^3/s in 2020s, 41 m^3/s in 2050s and 34 m^3/s in 2080s respectively by checking the upper ends of the box boxes shown in Figure 10. All GCMs under two scenarios indicate increase in annual stream flow for both RCPs of all periods except RCP4.5 of 2080s with decreasing stream flow change. During 2020s in Zaungtu, the annaul flow change can be increased up to 420 m^3/s for RCP4.5 scenario means uncertainty. And RCP8.5 will be reached at high point of 440 m^3/s in 2080s period.

Figure 9: Changes in seasonal and annual stream flow at Zaungtu station under RCP8.5 for the three future periods relative to the baseline period

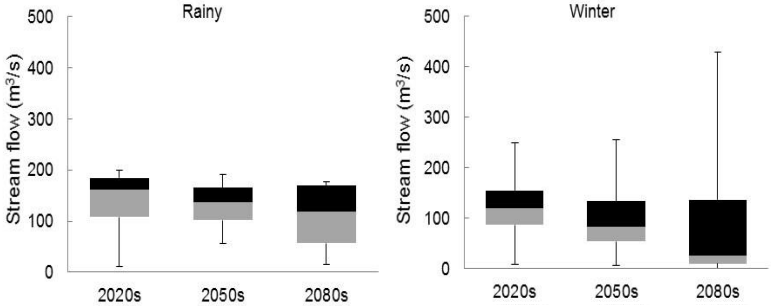
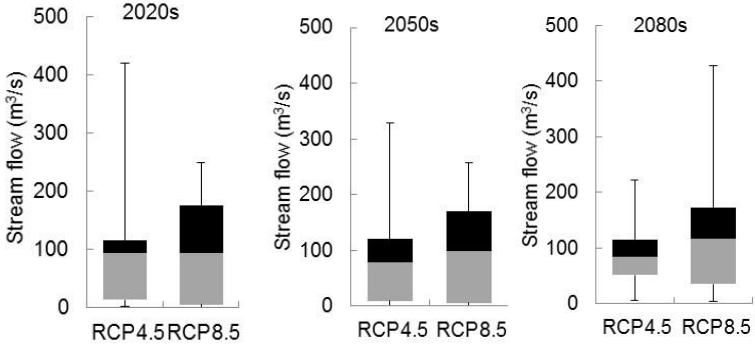


Figure 10: Changes in annual average stream flow for the 2020s, 2050s and 2080s relative to the baseline period (1990-2009) at Zaungtu



The relative change in annual and seasonal stream flow for the three future periods relative to the baseline period under 8.5 scenario at Bago station is shown in Figure 11. It is estimated to be maximum stream flow change with 86m^3/s (64%) for annual stream flow. Summer has the lowest change of flow in all periods only average change 19m^3/s. For the rainy season, the seasonal flow is increased in average 77m^3/s (26%) change in all periods. There can be seen increased amount of change 157m^3/s in 2020s, 145m^3/s in 2050s and 123m^3/s in 2080s period with averagely increase of 128% in winter season in resulting of the upper limits of box plots. In contrast, at Bago station under both scenarios, winter is the most affected season. For uncertainties of projections, there can be projected nearly 350 m^3/s in stream flow changes of annual and winter in 2080s period. On the other hand, 2050s period can be observed to be high in the summer and the rainy season by using cluster plots. In contrast, Zaungtu gives high annual stream flow changes than Bago station. The change in annual stream flow for RCP4.5 and 8.5 relative to the baseline period at Bago station is shown in Figure 12. This figure shows both scenarios and GCMs affect the annual stream flow change in all periods. The median value indicates the increase in all periods under both RCP4.5 and 8.5 scenarios. In all periods, RCP8.5 scenario is greater than RCP4.5 that is 2m^3/s in 2020s, 4 m^3/s in 2050s and 1 m^3/s in 2080s period. Results indicate that the increase change in stream flow can be reached at the highest point (350 m^3/s) in 2020s period under RCP4.5 scenario and the same level in 2080s under RCP8.5.

Figure 11: Changes in seasonal and annual stream flow at Bago station under RCP8.5 for the three future periods relative to the baseline period

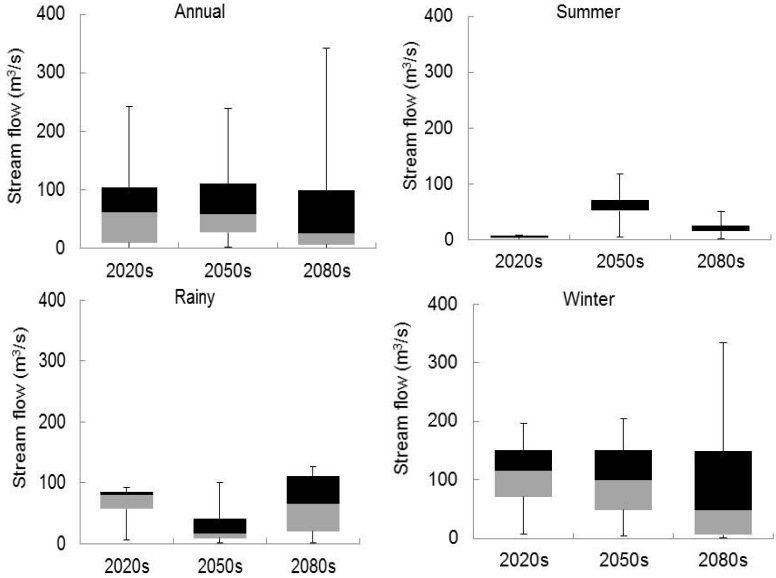
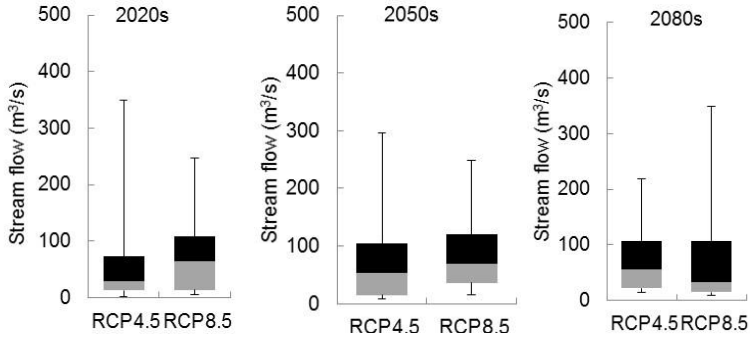


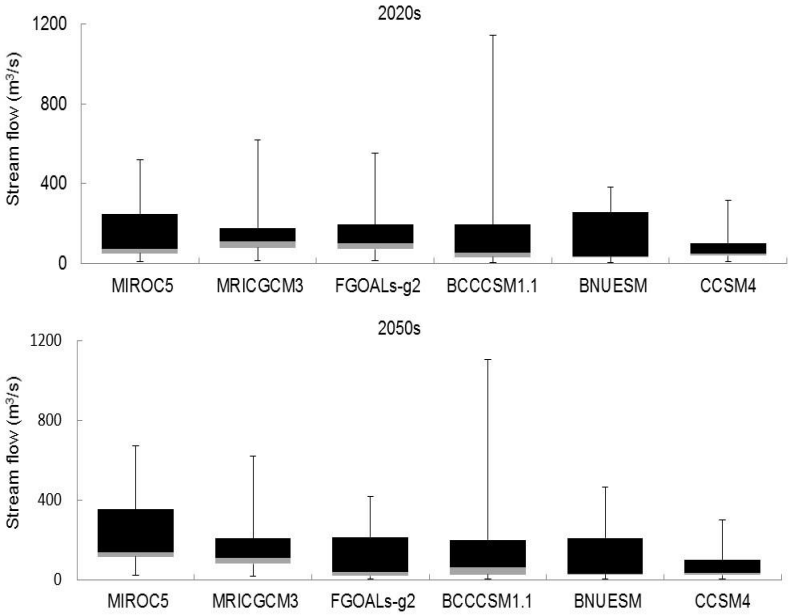
Figure 12: Changes in annual average stream flow for the 2020s, 2050s and 2080s relative to the baseline period (1990-2009) at Bago

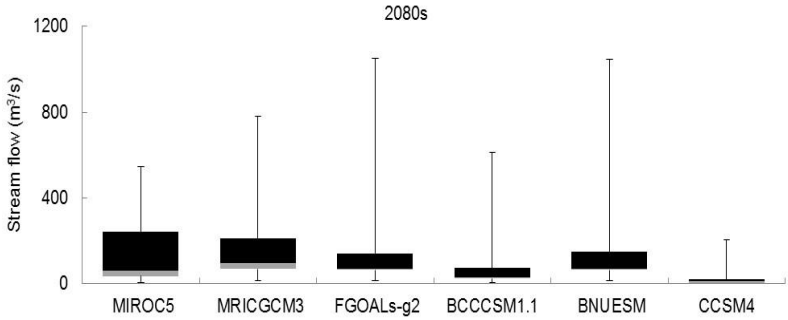


Uncertainty in Annual Stream Flow Changes due to GCMs

This section aims to outline the range of uncertainty arising from difference in projections from different GCMs under RCP8.5 scenario. The uncertainty range is estimated as the cluster plots. All GCMs indicate increase and decrease in stream flow changes in annual projections seen the figures below. At Zaungtu station, the changes in average annual stream flow due to 6GCMs under RCP 8.5 for 2020s, 2050s and 2080s relative to the baseline period are shown by checking their uncertainties in Figure 13. By comparing the upper limits of box plots of all GCMs, all projection from all GCMs are similar in 2020s period. It is observed that the projection of stream flow changes is high in MIROC5 model and low in CCSM4 for all 3 periods by using box plots. The stream flow changes of all GCM are not too high by their median values of box plots. But it can be projected to be high over 1000m^3/s in BCCCSM1.1 for 2020s and 2050s periods and in BNUESM for 2080s period because of uncertainties checked by the cluster plots seen in the below. Checking the maximum changes in stream flow (the upper ends of the box plots), BNUESM model can project the highest steam flow change in 2020s period, BCCCSM1.1 in 2050s and MIROC5 in 2080s. On the other hand, MRICGCM3, FGOALs-g2 and CCSM4 models are the least projected models for stream flow in 2020s, 2050s and 2080s periods. But in uncertainty, BCCCSM1.1 model can project over 900 m^3/s stream flow change in 2020s and 2050s periods and MIROC5 with 650m3/s stream flow change in 2080s period by observing the maximum level of cluster plots.

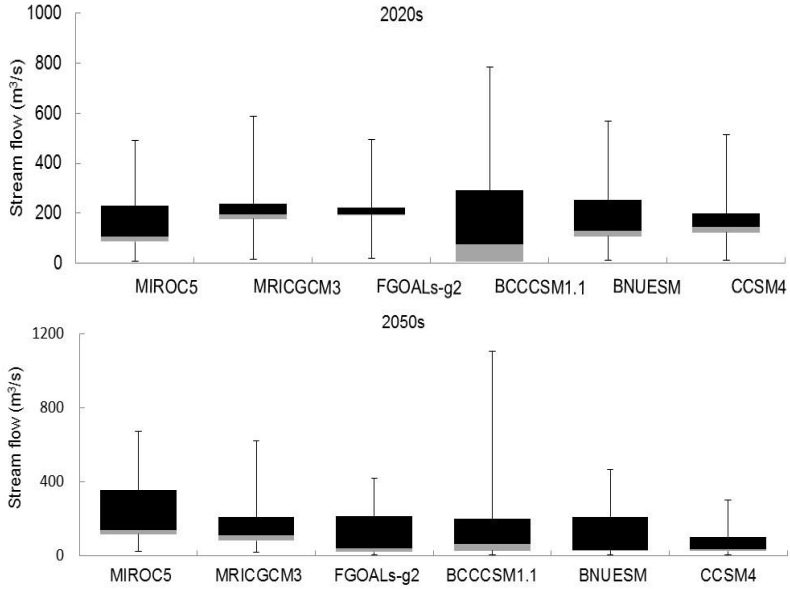
Figure 13: Changes in average stream flow due to GCMs under RCP8.5 for 2020s, 2050s and 2080s relative to the baseline period at Zaungtu station

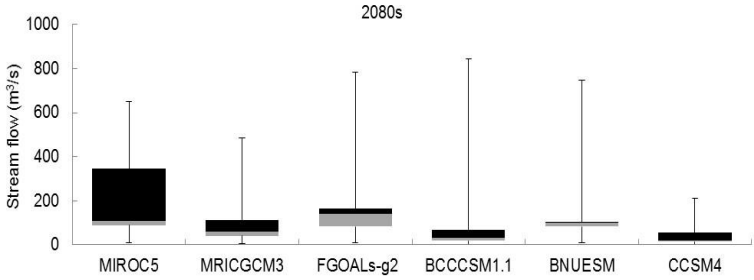




The uncertainty range for all GCMs at Bago station under RCP8.5 is presented in Figure 14. The figure below presents the average annual stream flow changes due to 6GCMs under both scenarios for three periods relative to the baseline at Bago station. In 2020s, all GCMs project the similar stream flow changes. MIROC 5 is the highest projected model for stream flow changes in 2050s and 2080s. It is proved that only BCCCSM1.1 model can be projected in maximum level of stream flow changes in all 3periods due to the cluster plots of this model.

Figure 14: Changes in average stream flow due to GCMs under RCP8.5 for 2020s, 2050s and 2080s relative to the baseline period at Bago station





Conclusions

This paper investigates the impact of climate change on future extreme flows through use of a distributed hydrological model SWAT in the Bago River Basin of Myanmar. Future extreme flows are simulated and their uncertainties are investigated by considering the emission scenarios and GCM errors for three future periods: an early-century period from 2010–2039 (2020s), a mid-century period from 2040–2069 (2050s) and a late-century period from 2080–2099 (2080s). This study forecasts the change in maximum, minimum temperature and precipitation under two Regional Concentration Pathways (RCP) 4.5 and RCP 8.5. The outputs of six General Circulation Models (GCMs) are bias corrected using delta change method to construct the future climate scenarios at basin level. It is found that the performances of GCMs are increased after bias correction. It is observed that average annual maximum and minimum temperatures are projected to rise in the entire basin under both scenarios — most significantly in the 2080s. Average summer temperature is projected to decrease by approximately 0.25°C in the first century period under both RCP4.5 and 8.5 scenarios. However winter season witness an increase in average temperature of 1.5-2.5 °C, following by the rainy season with increase of average temperature of 0.9–2.6 °C in future. The average annual precipitation in basin is projected to increase by 90 mm under RCP4.5 and 115 mm under RCP8.5. The winter season is expected to receive higher increase in precipitation: 175 mm under RCP4.5 and 210 mm under RCP8.5 scenarios. The wettest month keeps shifting over the three periods and is in contrast from the baseline months of observed data. The July, August, and September are projected to be the wettest months and the 2050s the wettest period as per the projections.

The SWAT model demonstrates a good performance in the study area, as all three evaluation indices (NS, R^2 and PBIAS) reaching satisfactory values in both calibration and validation periods, for all two hydrological stations. The performance of SWAT model during calibration and validation is satisfactory at the monthly and daily scales. This model is used to simulate the impact of climate change on stream flow of the basin. Under the climate change scenarios, the average stream flows and peak flows in winter season are projected to be higher in comparison to other seasons during 2010 to 2099. It is observed that monthly stream flows are projected to increase at maximum rate in September of all future period. Similarly the annual stream flows are projected to increase approximately 57% in the whole basin for all three periods of 2020s, 2050s and 2080s. By checking the upper limits of whiskers, the highest range of uncertainty for annual stream flow changes is projected under RCP4.5 scenario for the 2020s and 2050s (300 m^3/s to 450 m^3/s) and under the RCP8.5 scenario for the 2080s (350 m^3/s to 450 m^3/s). Large increase of extreme changes can be projected approaching 130m^3/s at Zaungtu station and 122m^3/s at Bago station for rainy season

flow under both scenarios. The winter season is the affected with large projection of flow changes of 286m^3/s at Zaungtu station and 254m^3/s at Bago station under both scenarios in the future period of 2010-2099.

Based on the results demonstrated in this paper, however, it is hard to tell the extent to which these uncertainty sources contribute to future extreme flow uncertainty. But it is widely acknowledged that GCM structure and GCM initial conditions will also cause uncertainty in future extreme flow simulation and point out that GCM structure is the largest uncertainty source. The projection of BCCCSM1.1 model can be over 900 m^3/s stream flow change in 2020s and 2050s periods and MIROC5 with 650m^3/s stream flow change in 2080s period by checking the upper limits of cluster plots at Zaungtu. On the other hand, only BCCCSM1.1 model can be projected in maximum level of stream flow changes in all 3periods due to the cluster plots of this model at Bago. It can be concluded that the large uncertainties are found in extreme flow projections. The results obtained in this study can provide further insight into the availability of future stream flow and aid in water resource management planning in the watersheds. For example, if there is a flood, many infrastructures and people will be affected. Decision makers are proposed to take the possibility of increasing frequency of flood into account during disaster management and infrastructure construction. Therefore the findings of this study can contribute to effective planning aimed at flood and drought risk management and adaptation.

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The Ways to Improve the Water Environmental Information Management in Vietnam

NGUYEN THI ANH TUYET[1], HUYNH TRUNG HAI[1],
TRAN THANH CHI[1], SAORI USHIMI[2]

[1] Hanoi University of Science and Technology
[2] Japan International Cooperation Agency

Abstract
The paper addressed the environmental information management issues related to water resources and proposed the ways to improve the information collection system - supported to water environmental management in Vietnam. The formats of 3 information kinds related to "the status of the damage caused by water pollution in public water bodies", "water pollution monitoring data in the public water bodies" and "the status of the enforcing water environmental policies" are also developed based on the current Vietnamese reporting formats and referred to the Japanese examples. In term of environmental management, the target information selected will fulfill the purpose effectively for the evaluation of the effectiveness of policies, for making now policy and for public disclosure.

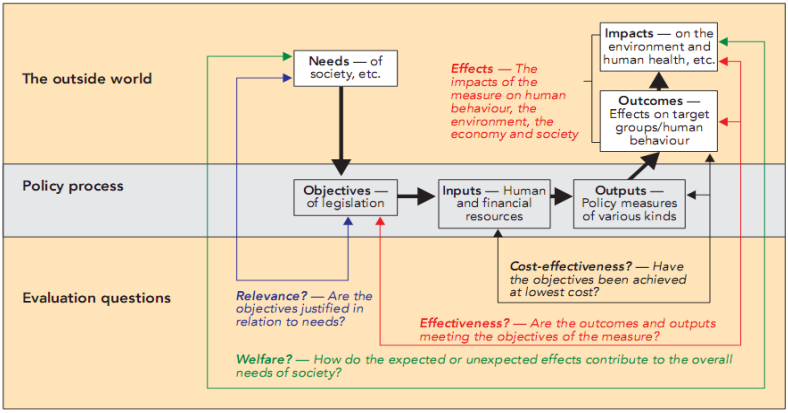
Keywords: environmental information management; policy evaluation; information formats.

Introduction
The bases of water environmental policies are "to set the water quality environmental standards (WQES) and to implement the countermeasures to the pollution sources to achieve the WQES in order to prevent the damages caused by water pollutions". Three of the key legal frameworks relevant to water environmental information management in Vietnam are Law on Environmental Protection, Environmental Status Report and Circular of the Collection, Management, Exploitation and Use of Resources and the environment data. Subsequently EIA data, Monitoring data and Inspection data are picked up to look at the details of its legal framework and the actual status of each department's environmental information management system.

- EIA data: EIA reports play an important role in environmental protection by forcing all organizations and individuals to commit to the activities for environmental protection. According to circular 29/2011/ND-CP, in the process of making EIA report, *governmental sector has an authority to give approval, check or instruct to the target project based on legal documents*. This administrative procedure is very important to secure the environmental report to be effective.
- Monitoring data: according to the Decree 102/2008/ND-CP on *"The collection, management, exploitation and use of natural resources and environmental data"*, it is defined the information to be collected and the responsibility of Provincial People's Committee (PPC) over the data.
- Inspection data: In Decree 86/2011/ND-CP *"Detailing and guiding the implementation of a number of articles of the law on inspection"* stipulate the reporting of the inspection results. It is defined the obligation of publishing the inspection conclusions as well as the reporting the information to the central government.

Currently, in Vietnamese policies, the main countermeasures to the industries focus on forcing industries to comply with "waste water effluent standards" and "payment of waste water discharging fee". In the reality, even if these policies are brought into the implementation, there are many cases [1] happening such as "the policies cannot be effective because of certain problems in polices themselves or in the ways of implementation", or "the policies cannot catch up with the spread of the water environmental pollution caused by the rapid economic growth". Therefore, it is very important to evaluate water environmental policies, namely, to observe whether the water environmental policies are addressing the problems happening in the fields.
Figure 1 describes the various measures to evaluate the effectiveness of policies [2]. In order to evaluate the policy effectiveness correctly, a central government needs to answer the questions described in the Figure and, therefore, in order to answer these questions, the information from the local governments are very needed. In Vietnam, Ministry of Natural Resource and Environment (MONRE) plays a role to evaluate the existing environmental policies and its effectiveness as a responsible central government, it requires the various information which is gained from all the processes of the enforcement. Thus MONRE must to organize the mechanism of collecting, managing and utilizing the environmental information from Departments of Natural Resource and Environment (DONREs).

Figure 1: Policy evaluation framework EEA [2]



It is ideal if the evaluation of the policies can be conducted from a broad set of perspectives. However, considering the current capacity and feasibility of water environmental information management (EIM) in Vietnam, it is quite difficult to evaluate the policies comprehensively. Therefore, it can be started with the basic steps known as "determining basic information to evaluate and to improve the water environmental policies".

The status and obstacles of water environmental management in MONRE

The authors have conducted the survey to grasp the current environmental information management system in Vietnam by focusing on environmental information collection, management and utilization. Hereby the facts found through the survey are shown as following as well as the current legal frameworks regarding environmental information management systems.

The actual status of the environmental information management in MONRE

As seen so far, the regal frameworks clearly define the obligation of the information sharing and reporting system. The water environmental information is mainly collected from other departments in Vietnam Environmental Agency (VEA/MONRE), from the local management agencies (DONREs), other sources (supplied from manufacturing facilities or industrial parks) or themselves through measurement, inspection, testing as the legal documents stipulate...

Based on the survey results in a number of departments in the VEA/MONRE and evidences obtained through the research, the consultant team found that at present it is lack of an appropriate mechanism for environmental information sharing/provision/ management. This problem leads to that the environmental management is not really effective or consuming time and labor.

The following is reasons that have caused the situation:

- The relationship between the current mechanism and responsibility-duties of related officers/units is unclear. In fact today we still have some legal documents for the environmental information submission/sharing but the implementation is not effective. The reason may be lack of defining specific responsibilities and obligations and lack of regime of rewards and punishments to encourage or remind.
- Unclear assignment leads to difficultly distinguish responsibilities of officials.
- Administration has a lot of clues, such as DONREs are basically under the management of provinces/cities, while the professional issues resolved by MONRE needs the contribution of DONREs, so the information reporting must not be simple.
- In most cases awareness and responsibilities of assigned officials are not serious and strict in information provision because they can have no benefit.
- Another cause is the habit of passive working (derived from coercive methods of education) and inefficient work organization leading to that a parts of Vietnam officials has not really concentrate on the job if their obligation is not required.

It can be said that according to the survey result, the departments in MONRE which have the regular reports from DONRE are basically Centre for Environmental Monitoring (CEM) only. Other departments in MONRE are not receiving the regular report. Regarding the reporting formats, only CEM and Inspection Department (ID) have the unified formats for reporting.

Currently, VEA is assigned to draft circular on the management, exploitation and sharing of information, known as Circular on submission, storage, supply, exploitation and use of environmental data. If this circular is issued, the environmental information collection/management/use serving the state management on environmental protection is expected to be more effective.

The actual status of the environmental information management in DONRE

Five DONREs have been surveyed are Ha Noi, Hai Phong, Hue, Ho Chi Minh City, and Ba Ria – Vung Tau. In order to assess the status of the water environmental information collection and management in these DONREs, the number of EIA approved, discharge permits, inspections and violations sanctioned are collected.

In term of authorizing environmental profile at DONRE level, Environmental Protection Department controls EIAs. Enterprises and industrial zones periodically report to DONREs about their environmental protection activities and environmental quality monitoring results. Periodically the DONRE's inspector team checks and takes samples of wastewater for analyzing. When detecting the unusual case in term of water pollution, the team will coordinate with the relevant units of the district-level People's Committees to conduct field surveys and sampling. Main steps are 1) coordinating with the actual survey functional units at the scene of an incident and information; (2) sampling (if necessary); (3) synthesizing, evaluating, and determining the cause of the incident. Monitoring Centers of DONREs yearly monitors water environmental quality parameters of the corresponding provinces. Table 1 summarizes the status of river water quality monitoring report in the 5 DONREs surveyed.

For managing environmental information, the functional units of DONREs themselves store and process the environmental information. General records must be stored in the archives of the Center for Information Resources and Environment (CIRE) in two types (text/paper and digitalization). All the information is used for developing database and making reports.

As regulated, DONREs report the environmental status of the corresponding provinces to VEA/MONRE every 6 months. The report is formed. The reporting procedure is as followings: MONRE send an administrative document (in letter format) to 63 DONREs to remind the environmental reports. This administrative document is also sent to 63 Province People Committees (PPCs) at the same time. The responding time is about a week. Then, each DONRE sends the environmental report (in the forms) to both PPC and MONRE by post. The environmental information related to industrial zones is not included in these reports. The Management Divisions of industrial zones is responsible to report the environmental information to both PPCs and MONRE. Besides, the functional units of DONREs are compelled to periodically report to the higher levels – it means the corresponding functional units of MONRE. For example, the status of importing waste materials is compelled reported to PCD – MONRE; the information related to EIAs is compelled reported to VEA – MONRE; the status of hazardous waste management is compelled reported to DWMEP – MONRE. The report templates are provided (e.g., sample of an inspection report, sample of reporting monitoring results).

In general, the DONRE reports meet the requirements of MONRE except for cases. Sometimes, the DONREs' reports cannot meet entirely the requirements of MONRE because of a number of objective factors of the reporting process between levels.

The targets should be tackled by MONRE and DONREs regarding the information management in MONRE

The issues which MONRE needs to tackle

1. About the scheme of reporting environmental information:

- There are many clues in MONRE receiving reports. Tasked mechanisms between the management levels are not really clear. It leads to the lower-level units confused. For example, there are a number of requirements (in an administrative document) unexpectedly sent by MONRE to DONREs for providing information.
- But they, sometimes, do not understand the mission, feedback is integrated to allow time for DONREs to follow up; or there are some official subordinate units (information received from PPC and DONRE at the same time).

2. About the legal base for reporting environmental information:

- Lack of legal documents regulated on managing and using environmental information. Lack of defining by policy for every cadre's work on environmental information. For example, according to the regulation on information keeping in Hai Phong (issued by Hai Phong PPC), an original profile of the information is compelled to keep.
- There is an overlap issues between the Environmental Protection Law and the Water Resources Law in term of allowing “wastewater discharge into water sources”. At the moment, if your project wants to discharge into water sources, you must apply for two permits (permits for discharge of wastewater into water sources and certificate of completing environmental protection works in the formal operational stages). This causes difficulties for enterprises.
- There are no unified formats established. Therefore, each DONRE has to make own format to manage the information. It makes it to difficult to collect, manage and utilize all the information from each province.

Table 1: The status of river water quality monitoring report in the 5 DONREs

Item	HNI	HPG	TT-HUE	HCM City	BR-VT
Frequency of monitoring	2/year	6/year	4/year	12/year	6/year
Report making	1) Report of the first half of the year 2) Report of the last half of the year 3) Annual report (summary)	1) Fixed term monitoring report 2) Annual report	1) Report of the first half of the year 2) Report of the last half of the year	1) Monthly report 2) Annual report	1) Fixed term monitoring report 2) Annual report
Maker	HNI CENMA	HACEM	TT-HUE Monitoring Station	HCM CEMA	CEMAB

Submit to	A relevant director of DONRE	Relevant Dept. in DONRE	Director of DONRE	Relevant Dept. in DONRE	DONRE (Director, EPA)
Content of the monitoring report	1) Rivers in HNI city and lakes、2) Red river 、3) Nhue-Day river (irregular)	1) 3 prioritized rivers in HPG 2) lakes and canals in HPG	1) Report of rivers, lakes, underground water and	1) Report of air, river (water level and flow velocity included), canals, underground water, and sea monitoring	1) Report of air, noise, river and urban domestic water, sea water, lakes, underground water and soil monitoring
Structure of the monitoring report	1) Purpose 2) Sampling/analysis methods 3) Monitoring points 4) Results of analysis 5) Data analysis 6) Conclusion 7) Suggestion	1) Monitoring plan 2) Sampling/ analysis methods and the results of analysis 3) Data analysis 4) Conclusion 5) Suggestion	1) Purpose 2) Summary of Monitoring plans 3) Data analysis and conclusion	1) Summary of Monitoring plans 2) Data analysis and evaluation 3) Conclusion	1) Summary of Monitoring plans 2) Data analysis 3) Evaluation and conclusion
Monitoring data management method	MS Excel	MS Excel	The tables of the analyzed results from outsourcing laboratory (MS Word)	MS Excel	MS Excel
Reporting to VEA	As per a request from VEA	As per a request from VEA	As per a request from VEA	As per a request from VEA	As per a request from VEA

The issues which DONRE needs to tackle

1. About the quality of environmental information:

- The cities and provinces have no general provision for managing and reporting information. The information is mainly kept in text format. Therefore, the quantity of information is not sufficient and satisfied the needs of management mission.
- The quality of the information is limited and the database is still very sparsely. Data related to effluent discharges from industrial and service activities, those had been reported by inspectors, have not been updated as database. The status of out-of-dated data leads to gaps in looking up information services.
- In big cities, such as Ha Noi and Ho Chi Minh City, the basic statistical sources on water pollution have been gradually collected. However, the pollution control is still ineffective. Most of production facilities have basically processing system, but the relative nature of treatment, no effective control of the processing system.
- From the MONRE's requests, a number of unexpected written requests do not have specific guidelines and the responding time is usually too short. Therefore, the information does not entirely meet the requirements.

2. About the scheme of reporting environmental information:

- The collaboration among PPC, DONRE and other departments in the certain province is usually not close. It leads to duplicating and overlapping the tasks.
- In DONREs, the coordination process (related to environmental information) is, sometimes, inconvenient due to the long distance between the functional leading units and the status of keeping information (e.g. keeping in text format or papers - it makes a difficulty to access, usually).
- Time required to provide the information of central agencies in a number of cases was too short. Therefore, except for the information controlled by DONREs, other environmental information, those are controlled by industrial zone's management divisions (via by PPC) or PPC, are not responded in time.
- The procedure of environmental information keeping is still sparsely. Once you require for providing information, it usually causes time-lost.
- CIREs receive the environmental data/reports in dossiers filed from other units of DONREs. However they do not response back to the units. Submission dossier does not provide the standard report form. In the case of self-reported, not only the form but the reliability of the repots is usually not very in good quality, still need to be checked.

3. About the infrastructure for collecting, keeping and releasing information:

- Information collected are still missing or discrete, hard chain service management, decision-making. Furthermore, the procedure of environmental information keeping is still sparsely. It usually causes time-lost whenever you require information providing.
- Lack of human resource for keeping and managing environmental information. "A staff holds a purity of offices" is popular. A behaving policy for those staffs is still weak.
- Sometimes, human resources to perform the tasks of state management in a local area (DONRE, PPC, wards and townships) have not been fully arranged or have not been properly trained professional. The environmental information obtained from inspection and wastewater controls in the area, therefore, have not yet been fully implemented effectively.

4. About the funding for collecting, keeping and releasing information:

- Financial recourse is limited - do not satisfy the needs of management mission.
- At DONRE's level, Inspection Departments and Departments of Environmental Protection do not have funds to carry out periodic inspection which only checked when the plaintiff, making the information collected can only serve a specific purpose in a particular time.
- Technologies of outstanding work are not completely met the demand set because the budget for operating is very low. In some units, this budget must be taken from the regular business fund.

The background of the problems

As discussed previously, providing information from DONREs to MONRE is not successfully conducted. Also the information sharing within MONRE is not effective. There are various reasons for these facts. The main reason why information management has not been successful within MONRE can be the followings:

1. The importance of environmental information management is not commonly understood among relevant people: in particular, the importance of following-up after issuing the regulations (the PDCA cycle[1]) or the importance of environmental information management for the following up the regulations.
2. As a problem of the administrative agencies, the structure of the cooperation between MONRE and DONREs as well as within MONRE is not sufficient (a problem of double controls, lack of the legal structure to rule all the policies, lack of administrative/organizational structure to coordinate all the policies).
3. The positioning of the environmental standard in water environmental policy is not clear. It is causing the lack of incentives of "making a connection between the environmental standard and monitoring data and between the environmental standard and the pollution source control (setting the environmental standard etc.)". As a result, it is difficult to analyze comprehensively the environmental information and to utilize the results to evaluate the current policies.
4. There is a lack of human resources and budgets to manage environmental information.

Suggestion for the improvement of the water environmental information management in Vietnam

Determining the target information

What kind of environmental information should be collected and utilized?

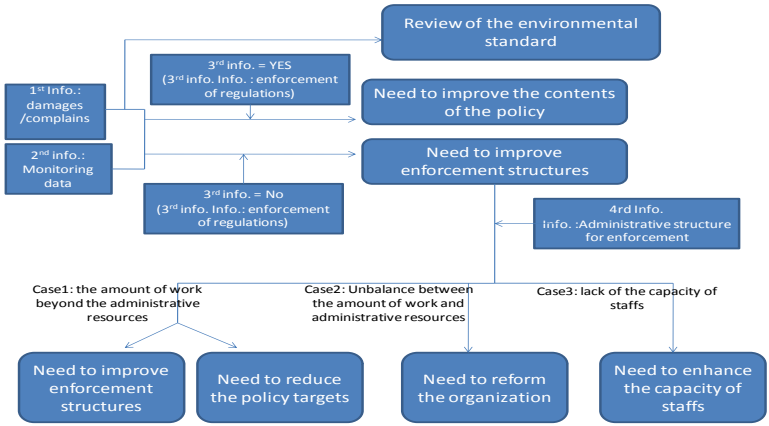
As basic information to evaluate/improve the policies, the following 4 kinds of information (Figure 2) should be grasped.

In this research, we focused on 3 kinds of information, those are "the status of the damage caused by water pollution in public water bodies (1st information)", "water pollution monitoring data in the public water bodies (2nd information)" and "the enforcement is correctly brought into practice by the procedures stipulated in the legal documents (3rd information)". It is because by referring the first two kinds of information, "the necessity of improvements of water pollution control policy in public water bodies" can be comprehended. "Public water bodies" here means river water, sea water, lake water and underground water. If the improvements of the pollution control policies are required, it is possible to identify which polluting substance should be controlled at the same time. Then, when the necessity to improve the water

[1] PDCA (Plan–Do–Check–Act). It is an iterative four-step management method for the control and continuous improvement.

pollution control policy is figured out, the targets of improvement can be divided into 2 categories; that is, "the improvement of the enforcement" and "improvement of policy itself". The 3rd information is used to identify which improvement is required. If the environmental problems in the field are not being addressed, it is highly possible that the problems lie not in the way of the policy-enforcement, but in the contents of the policy.

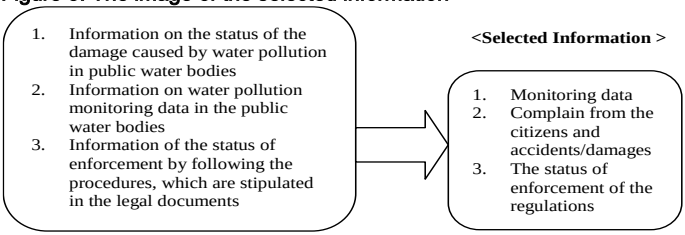
Figure 2: The utilization of the environmental information for the improvement of the policies



Selection of the target information

The most common information among the 3 kinds of information should be selected to maximize the results with relatively easy data processes. It is because showing "how to select the prioritized environmental information" and "how to manage it" can mean "demonstrating the importance of environmental information management". We select the target information (as presented in Figure 3) to fulfill the purpose effectively for the evaluation of the effectiveness of policies, for making now policy and for public disclosure.

Figure 3: The image of the selected information



In addition, the selected 3 information kinds can be used not only for policy evaluation and improvement, but also for raising awareness of the people, environmental education and public participation on the policy making. More concretely, by disclosing the environmental information, it can be contributed to the partnership between the government and the people such as "finding and reporting the water quality accident in a river etc.", "reflecting the people's opinions to policy making" or "Reducing the dumping of solid wastes and discharging of domestic wastewater into rivers.

According to our survey of the current status of the environmental information management in Vietnam, it was revealed the departments in MONRE which have the reporting format for DONREs are only Inspection Department (ID) and Center for Environmental Monitoring (CEM). Other departments do not have the unified formats for reporting to MONRE. In order to gasp the whole picture of the environmental status, it is necessary to use the same (or at least compatible) formats in nationwide.

Proposed formats to send the selected information from DONREs to MONRE

In this part, the annual reporting formats from DONREs to MONRE are illustrated for the selected 3 information above. Regarding the formats of ID and CEM, the current reporting formats of ID and CEM are referred and then some formats are suggested for the improvement by referring the Japanese examples [3,4,5].

Monitoring Data

Considering the importance of environmental monitoring data, the information below are focused:

1. Water Environmental Monitoring Data – DO/BOD/COD
- Changes over the year by measuring points
- Water quality changes in upstream and downstream

2. Data on Toxic substances in water bodies
- Items, which exceed the environmental standards and degree of the excess

In Vietnam, the water quality monitoring data is assessed based on the technical regulations of surface water, ground water and costal water. In this research, we mainly focus on surface water. Each regulation defines the types of water body, the parameters to be assessed and the limit values. The Vietnamese government stipulated in 2008 "Technical Regulation on surface water quality 08:2008/BTNMT". It shows 32 parameters to be assessed in surface water bodies. This regulation defines the limit values for each parameter depending on the purposes of use as below as a type of water body:

- A1: Good for domestic water supply purpose and other purpose as of A2, B1 and B2
- A2: For domestic water supply purpose, but suitable treatment technology must be applied; conservation of aquatic lives, or other purpose of B1 and B2
- B1: For irrigation purpose or others with demand for similar quality water or other purpose of B2
- B2: For water transportation and other purpose with demand for low-quality.

The parameter and limitation values in coastal water are defined in QCVN 10:2008 /BTNMT depending on the purposes of use as:

- Aquaculture/Conservation
- Beaches/Sports
- Others

Therefore, in order to develop the monitoring formats, these regulations should be the base.

a. Changes over the years by a water body type

One of the most useful aspects of collecting the data is to give us quick view of the trend of the measuring items. The format is proposed and shown in Table 2.

Table 2: Water quality changes over the years by water body type

Note: it should be calculated by using annual average rate at the environmental monitoring points.

		2001	2002	...	2012
River	Total				
	A1				
	A2				
	B1				
	B2				
Cannel, Ditch	Total				
	A1				
	A2				
	B1				
	B2				
Lake	Total				
	A1				
	A2				
	B1				
	B2				
Sea	Total				
	Acuaculture/ Conservation				
	Beach/ Sports				
	Others				

In this format, as a typical parameter, BOD is applied to measure for river/cannel/ditch water, COD_{Cr} is applied to measure lake water (by QCVN08/2008), COD_{Mn} is applied to measure costal water (by QCVN10/2008), and DO is applied to measure for all water bodies.

Each DONRE submits this format to MONRE and then, MONRE can sum up all the data collected from the provinces. It can show the trend of the water quality of the country by a water body type.

b. Water quality changes in up and downstream

Grasping the water quality in upstream and downstream shows in which point the water quality becomes deteriorated. By grasping the point, it makes it possible to identify the possible pollution sources. Thus it helps a responsible authority to make the countermeasures.

If all the samplings are performed in the same dates for each point, the annual average can be inputted. In addition, the changes by years can be also shown by linking the data to the other sheet as below. It means that if the administrator input the data into each cell, the data from each parameter links to “changes by years’ sheet” automatically. Thus it makes easier to make the graphs as well as easier to understand the trends over years. Table 3 shows the format of monitoring results with 8 typical parameters. It is noted that the parameters can be added or reduced based on the situation of the region.

Table 3: Water quality changes in upstream and downstream over years

Note: Points A, B, C are the environmental monitoring points located from upstream to downstream.

		2001	2002	...	2012
Point A	DO				
	BOD				
	SS				
	Ammonia				
	Total N				
	Total P				
	E. Coli				
	Coliform				
Point B	DO				
	BOD				
	SS				
	Ammonia				
	Total N				
	Total P				
	E. Coli				
	Coliform				
Point C	DO				
	BOD				
	SS				
	Ammonia				
	Total N				
	Total P				
	E. Coli				
	Coliform				

c. Degree of the achievement of environmental standard

Understanding the degree of the pollution and its substances are critically important since these materials can harm the human health. In order to grasp the degree of the pollution and its substances, it is helpful to show the exceeded items and its ratio.

Table 4. Items, which exceed the environmental standards and degree of the excess

STT	Parameters	Unit	Number of Exceed of the environmental standard and Analysis										
			2012										
			A1		A2		B1		B2		Total		
			No. Exceed	No. Analysis	No. Exceed	No. Analysis	No. Exceed	No. Analysis	No. Exceed	No. Analysis	a. No. Exceeded	b. No. Analysis	a/b (%)
1	pH												
2	DO	Mg/l											
3	Total of SS	Mg/l											
4	COD	Mg/l											
5	BOD5 (20°C)	Mg/l											
6	Ammonia (NH+4) (calculating based on N)	Mg/l											
7	Clorua (Cl-)	Mg/l											
8	Florua (F-)	Mg/l											
9	Nitrit (NO-2)	Mg/l											
10	Nitrat (NO-3)	Mg/l											
11	Phosphat (PO3-4)	Mg/l											
12	Xiannua (CN-)	Mg/l											
13	Asen (As)	Mg/l											
14	Cadimi (Cd)	Mg/l											
15	Lead (Pb)	Mg/l											
16	Crom III (Cr3+)	Mg/l											
17	Crom VI (Cr5+)	Mg/l											
18	Bronze (Cu)	Mg/l											
19	Tin (Zn)	Mg/l											
20	Niken (Ni)	Mg/l											
21	Steel (Fe)	Mg/l											
22	Mercury (Hg)	Mg/l											
23	Surface active material	Mg/l											
24	Oil and grease	Mg/l											
25	Phenol (Total)	Mg/l											
26	Pesticide chemical-organic Clo	µg/l											
	Aldrin +dieldrin	µg/l											
	Endrin	µg/l											
	BH	µg/l											
	DDT	µg/l											
	Endosunfan (Thiodan)	µg/l											
	Lindan	µg/l											
	Chlordane	µg/l											
	Heptachlor	µg/l											
27	Pesticide chemical-organic	µg/l											
	Paration	µg/l											
	Malation	µg/l											
28	Herbicide chemical	µg/l											
	2,4D	µg/l											
	2,4,5T	µg/l											
	Paraquat	µg/l											
29	Total of radioactive activity α	Bq/l											
30	Total of radioactive activity β	Bq/l											
31	E.Coli	MPN/1											
32	Coliform	MPN/1											

In many countries, there are two different types of items 1) Environmental items (BOD, COD, pH, etc.), and 2) Health items (Toxic Items). Health items are toxic items, which can harm human health. Thus, those are applied as the nationwide items and values for all the water bodies as well as underground water. Those items should not to be exceeded, and in case of the excess, the pollution sources should be identified promptly and countermeasures should be addressed. In Vietnam, all those items are mixed. Thus, in the reporting formats, specific items can be highlighted or selected to focus on the toxic items excesses.
In order to fill in this format, the breakdown information is needed. For example, if the monitoring (sampling and analysis) are done in 3 of A1 rivers each month at 2 points. "No. analysis" should be written as: 3 (river of A1) *2 (sampling points in each river) *12 (times (month)) = 72. Therefore, the number of analysis in A1 river in the year = 72.
Grasping how much degree the environmental standards are achieved shows the whole picture of the water quality in the regions and the country. If the water quality cannot be met with the environmental standard, the responsible authority can identify its reason and plan the concrete countermeasure promptly.

Accidents and complains

Environmental Protection Agency (EPA) in DONRE is in charge of dealing with accidents relevant to water pollution as well as complains from citizens. This information can be critically important to find possible polluting activities. Therefore, the number of the events should be recorded and depending on the necessity, MONRE can ask the further information from DONRE. This format is to make a graphic chart to show the trends.

Table 5: The status of accidents and complains relevant to water pollution

Accident	2001	2002	...	2012
Total				
River				
Lake				
Sea				
Underground				
Complains				
Total				
River				
Lake				
Sea				
Underground				

Status of enforcement

a. Information on the results of inspection

The reporting format of inspection activities has been established by MONRE (Inspection Department) and it is transmitted from DONRE to MONRE as periodic report. Based on our survey in 5 DONREs, the periodic report is being sent to MONRE every six month. Table 6 shows a new format based on the current inspection reporting format.
It shows all the relevant activities which the inspection department is in charge of. Also in this way, the changes over years can be shown as a graph.

Table 6: Results of inspection activities

No	Item	2001	2002	...	2012
1	Order of improvement				
	improvement				
	Order of temporary suspention				
2	Checking conducted				
3	Inspection conducted				
	during day time				
	during night time				
4	Admonistrative advice				
5	Emergency countermesures				
6	Request for countermesures				
7	Penalty applied				
	discharging standard				
	Violation of order of improvement				
8	Other violation				
9	Pecuniary administrative sanction				
10	Money recovered				

b. Information on the results of the administrative procedure

EIA reports are critical information to prevent any possible pollution as well as the inspection activities. However, in terms of information necessity for MONRE, not all the information is needed to be sent to MONRE. As MONRE, the whole picture of EIA activities should be understood. In that case, the following information would be satisfying for MONRE. If there is any special case, which MONRE could be interested, MONRE can ask DONREs to submit the detailed or specific information each time.

Table 7: The status of EIA reporting (SEA/EIA/EPC/EPP)

	2001	2002	...	2012
No. of SEA applied				
Approved				
Order of change of the plan				
No. of EIA applied				
Approved				
Order of change of the plan				
No. of EPC applied				
Approved				
Order of change of the plan				
No. of EPP applied				
Approved				
Order of change of the plan				

c. Discharging permit and fee

Water Recourse Management Department (WRMD) is in charge of issuing the waste water permit and collecting the fee. Knowing the waste water discharging point is critical information to identify the pollution sources if any accident happens. In addition, the permit is linked with the fee collection. Therefore, it must be shown how much it was collected and how much it was sent to “Environmental Protection Fee” within the collected amount.

Table 8: The status of discharging permits and discharging fee

No.	Item	2001	2002	...	2012
1	Total no. of discharging facilities (have been registed)				
2	No. discharging permit newly applied				
3	Toal Fee collected				
	Environmental Protection fee collected				

Conclusion

The above proposed formats are the sum-up of the breakdown information and made for the annual reporting from DONRE to MONRE. In order to fill in these formats, it is necessary to have the breakdown information, which DONRE is collecting periodically as a routine work.

For the improvement of the information management system as proposed, it is crucial to organize the basement on the water environmental information management in parallel. One of the reasons why the collection/management of environmental information is not yet established is because of the factor that structure to management of the environmental information is not organized yet.

Acknowledgments

The authors wish to thank Dr. Nguyen Quoc Khanh (Director of Center for Information and Environmental Documents, Ministry of Natural Resources and Environment) for his valuable discussion.

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Rapid Assessment of Flood Prone Areas of Selected Critical Rivers in Mindanao, Philippines: An initial step of MinDANOW

EINSTINE M. OPISO[1], GEORGE R. PUNO[2], VICTORIA T. QUIMPANG[3], ROSE ANGELICA L. AMPER[1], JORGE A. B CIPRIANO[1], ABIGAIL J. LABADAN[1], ALJEM O. BONGHANOY[1], MELEA LOUIESE C. LEDRES[1]

[1] Geo-environmental Engineering Group, College of Engineering, Central Mindanao University, Philippines
[2] Environmental Science Department, College of Forestry and Environmental Science, Central Mindanao University, Philippines
[3] Biology Department, College of Arts and Science, Central Mindanao University, Philippines

Abstract

Several critical rivers in Mindanao which are outside the jurisdiction Mindanao river basin councils were rapidly assessed to come up with their current status and degree of impairment which were used as basis for the formulation of intervention recommendation and rehabilitation plan under the MinDANOW program. The critical rivers namely Dipolog, Iponan, Mandulog, Padada, Cabadbaran, Surigao, Tandag and Tago were identified based on their impaired roles to agricultural, industrial, tourism/recreation and domestic use development; susceptibility to flooding; and large potential threat of flooding to socio-economic development such as population, agriculture and infrastructure. Geostatistical analysis was employed for the geomorphologic characterization of watersheds. Actual field survey was conducted with the use of interview questionnaire, generated rapid assessment instrument, and multi-parameter probe (HORIBA U-52G) for the determination of the water quality among the selected sampling sites for abiotic and biotic assessment. Ranking results based on the actual field survey and geomorphological analysis revealed Tago River to be the most deteriorated with severely scoured banks and channelized stream. The increasing degree of impairment of rivers is as follows: Padada< Tandag < Dipolog < Mandulog < Iponan < Cabadbaran < Surigao < Tago. Several stressors were identified to have influenced each river's integrity which includes major economic activities along the rivers such as unregulated physical resource extraction and intensive agricultural cultivation. Upstream areas of the watersheds and riparian zones lack vegetation which makes surrounding built up and residential areas more vulnerable to flooding. It is recommended that greenbelt establishment and environmental protection ordinances by the LGU must be implemented. The conduct of hydrologic analysis is recommended which will include biophysical characterization activities. Moreover, comprehensive watershed assessment studies must be employed for further rehabilitation strategies and remedial activities. For an effective coordination of government policies, an establishment of an institutional governing board is recommended.

Keywords: watershed; assessment; characterization; geomorphologic; abiotic; biotic.

Introduction

Mindanao's rich watershed resources play prime roles in boosting the economy effectively irrigating thousand hectares of farmlands and support socio-economic activities. In the past years however, Mindanao was repeatedly visited by typhoons which brought along calamities that caused destruction of properties, immobilization of economy, and thousands of mortality. It seems to become the new norm eliminating the idea of a typhoon-free Mindanao. The continuous denudation of the upland forests even made the area more vulnerable to disasters. Under the Mindanao Nurturing Our Waters Program (MindaNOW) implemented by Mindanao Development Authority (MinDA), a rapid assessment of the selected critical rivers in Mindanao was conducted. As one of the many projects and programs to help address aforementioned problems, this study was formulated to come up with the current status and degree of impairment for each river which are outside the jurisdiction of the Mindanao river basin councils. These are useful for the formulation of intervention recommendations and rehabilitation plans for the selected rivers.

Methodology

Geomorphological Assessment of the Selected Critical Rivers

Assessment areas were described using GIS and RS image by illustrating the geomorphic features, soil type, geology, vegetative cover surrounding the selected critical rivers and hazard map. Remotely sensed generated information on the ground was validated using a global positioning system receiver device together with a survey questionnaire. Geostatistical analysis was employed for geomorphologic characterization of the watersheds.

Abiotic Assessment of the Selected Critical Rivers

Current water quality condition of the river was evaluated using a multi-parameter probe (HORIBA U-52G) to determine the turbidity, pH, temperature, dissolved oxygen (DO), total dissolved solids (TDS) and the total suspended solids (TSS) of the river measured in a laboratory. It was compared to DENR Administrative Order 34 (DAO 34) for the qualification of Philippines' water quality standards. Hydrologic connectivity and various physical, landscape and hydrologic condition stressors in the selected critical rivers were evaluated.

Biotic Assessment of the Selected Critical Rivers

Present condition of the riparian vegetation including the biological condition stressors at the study sites was evaluated.

Demographic Study of the Assessment Area

Socio-economic profile of the surrounding residents on the selected critical rivers was assessed with the use of a survey questionnaire and the secondary data gathered from the concerned government offices.

Ranking of Selected Critical Rivers

The different factors of river components (biotic and abiotic) were evaluated and categorized into four classifications: optimal, sub-optimal, marginal and poor with corresponding ratings. Rivers were ranked based on the different factors and were given assigned ratings.

Population along the river, slope, stream order, soil type and land cover are the parameters considered as contributory factors, another component considered in

ranking the rivers. Each factor were rank, given assigned ratings and were summed for the each river's final ranking of contributory factors.
Overall ranking of rivers was done by summing the assigned ratings on the final rankings of each river's components which were then summed.

Results & Discussions

Description of the Study Sites

The selected critical rivers which were identified as susceptible to flooding with potential threats to socio-economic development are located within highly industrialized areas. Two assessment stations, around the midstream and downstream areas, were established in each river. Locations, coordinates and number of surrounding barangays are shown in Table 1.

Table 1: Location, latitudes and longitudes and No. of barangays traversing the river

Region	Rivers	Locations	Latitudes (N) and Longitudes (E)	No. of Baran-gays traversing the river
Region IX	Dipolog	Dipolog City, Polanco, Piñan and Dapitan City	8˚25'0"- 8˚35'0" N lat. and 123˚18'0" to 123˚30'0" E long.	26
Region X	Iponan	Cagayan de Oro City (CDOC) and Municipality of Opol, Northern Mindanao	8˚8'0"- 8˚32'0" N lat. and 124˚24'0" to 124˚40'0" E long.	18
	Mandu-log	Bubong, Lanao del Sur, Iligan City, Tagoloan II and, Kapai, Lanao del Norte	8˚0'0"- 8˚20'0" N lat. and 124˚15'0" to 124˚35'0" E long.	22
Region XI	Padada	Municipalities of Matanao, Padada and Hagonoy Davao del Sur	6˚30'0" - 6˚50'0" N lat. and 125˚00'0" - 125˚24'0" E long.	13
Region XIII	Cabad-baran	Municipalities of Cabadbaran Remedios T. Romualdez and Santiago, Agusan Del Norte	9˚3'0" – 9˚17'0" N lat. and 125˚33'0" – 125˚47'0" E long	14
	Surigao	Surigao City, Surigao Del Norte	9˚35'0"- 9˚45'0" N lat. and 125˚25'0" to 125˚35'0" E long	20
	Tandag	Tandag City, Lanuza Municipality, Surigao Del Sur	9˚1'30"-9˚10'0" N lat. and 126˚00'0" to 126˚8'30" E long.	8
	Tago	Tago, Cagwait, San Miguel, Tandag, Lanuza, Carmen, Surigao Del Sur and Sibagat, Agusan Del Sur	8˚44'30"-9˚10'0" N lat. and 125˚43'0" to 126˚43'0" E long.	27

Figure 1: Assessment stations 1 and 2 of rivers of (A) Dipolog, (B) Iponan, (C) Mandulog and (D) Padada

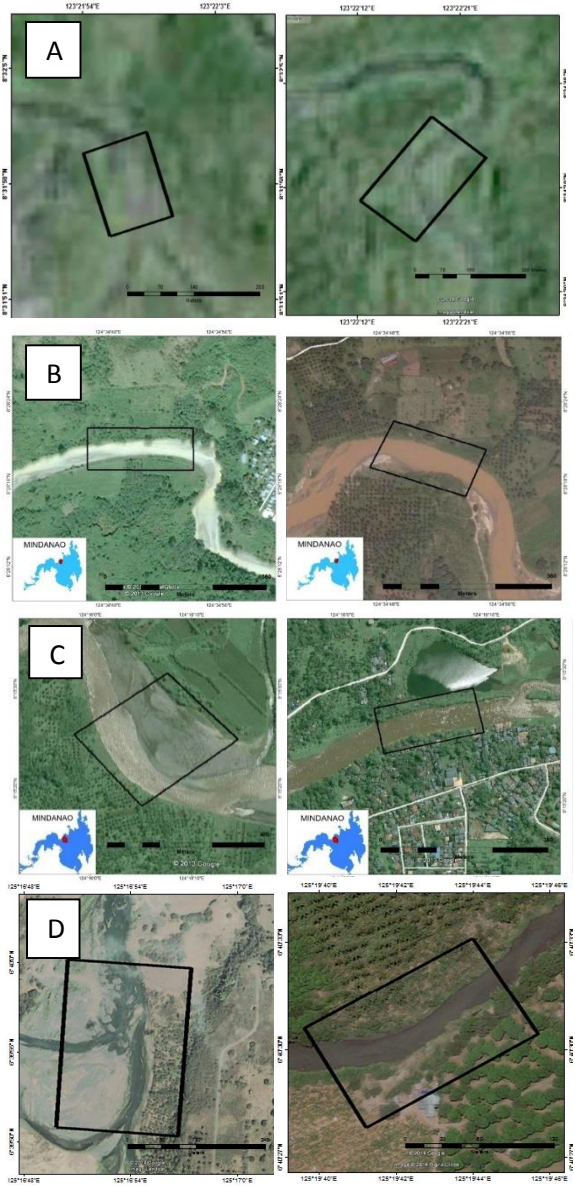
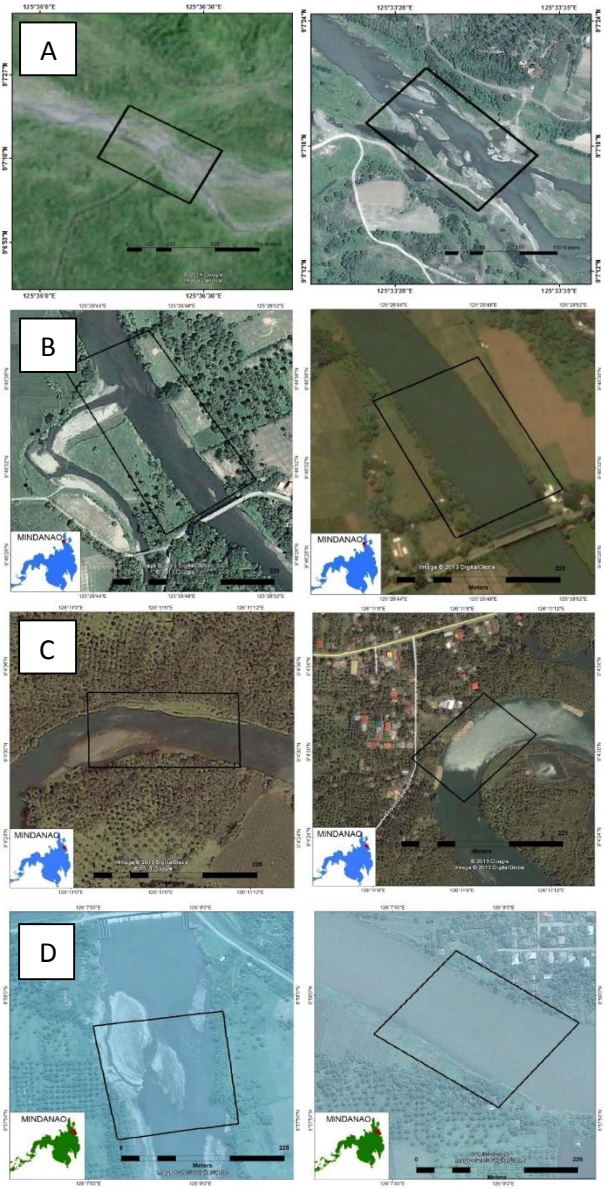


Figure 2: Assessment stations 1 and 2 of (A) Cabadbaran, (B) Surigao, (C) Tandag and (D) Tago rivers



Geomorphologic Description

Geomorphologic values reveal that all watersheds of selected critical rivers are elongated, hence easier to manage due to slow disposal of water and low discharge runoff. This may be caused by the small number of streams resulting to a watershed of high infiltration capacity. Watershed also has high water holding capacity in which may dangerously result to mudslide incidence when saturation capacity of soil has exceeded due to continuous rainfall. Drainage pattern of watershed is distorted caused by some disturbances like quarrying activities, presence of dam, and/or agricultural cultivation.

Watersheds of Dipolog and Mandulog are largely covered with clayey, a type of soil with low infiltration capacity that tends to support runoff. This may influence discharge within the watershed by increasing runoff and thereby increasing the risk of flooding. Cabadbaran, Iponan and Surigao watersheds on the other hand are mostly covered with cultivation and open canopy with sparse and little vegetation. Decreased number of natural vegetation may increase discharge runoff, hence of flooding in the watershed. Other than the largely cultivated areas of Padada watershed which tends to increase runoff, other outlying parameters specifically the unique shape of Padada watershed significantly contributes to the incidence of flooding around the area. Not only largely covered with cultivated lands, Tandag and Tago watersheds are also dominated by clayey soil materials which tend to support and increase runoff.

Abiotic factors

Results revealed that hydrologic connectivity of the rivers ranges from sub-optimal to optimal conditions exhibiting sufficient inundations. This however excludes Cabadbaran River which is fully disconnected from its floodplain (Table 3). As defined, hydrologic connectivity is the ability of the water to flow into or out of the wetland or inundate adjacent areas of the river (Muldavin et al. 2011). Cabadbaran River displays a very ruined profile with no distinct banks. This has resulted from the indiscriminate and extensive quarrying along the river considered as a major stressor for landscape and physical structure condition of the river.

Rivers of Region 13 were evaluated to be poor in landscape and physical structure conditions due to the existing metallic extraction and extensive quarrying within the watersheds. The rivers of other regions were consequently evaluated as marginal in the same context wherein stressors are present in lesser area percentage. Other identified stressors present under rivers' premise are urban encroachment, cultivation of riparian areas, presence of industrial/commercial establishments and engineered structures, and occurring sediment deposition out of natural ecological process.

Considerable adverse impacts are consequently caused by the said stressors to each river's integrity. Results of physico-chemical water quality assessment revealed that four of the assessed rivers namely Dipolog, Iponan, Padada and Tago failed to meet the DAO 34 water quality standards (Table 4). Known as the Department of Environment and Natural Resources Administrative Order 34, the standard composing 7 parameters namely TDS, temperature, pH, turbidity, DO, TSS and Nitrate concentration was used as the basis (except for Nitrate) for water quality assessment of rivers. Increased amount of suspended solids in river water is mainly attributed to the occurring physical resource extraction and the increase of agricultural cultivation along river banks. According to Zalidis et al. (2002), pollution is one the many detrimental effects of agricultural practices. However the rest of the rivers are classified as sub-optimal to optimal in water quality.

Table 2: Abiotic Components Assessment Results

River	PARAMETERS									Total	Description
	Hydrologic Connectivity	Landscape Condition Stressor	Hydrologic Condition Stressor	Physical Condition Stressor	Physico-chemical Parameters	Bottom substrate instream cover	Embeddedness	Channel Alteration	Bank Stability		
Dipolog	13	9.67	10	6.83	2	8.33	7.5	12	5	74.33	Marginal
Iponan	12.25	6.5	4.5	2.5	5.5	2.92	7.25	3.17	1.25	45.84	Poor
Mandulog	20	9	8	7	14.5	5.5	4.5	9	6	83.5	Marginal
Padada	13.5	6	7.25	9.5	2.25	8.25	3.75	9.5	4.75	64.75	Marginal
Cabadbaran	3.17	1	7.67	1	17.5	3.33	11.17	1.33	2.5	48.67	Marginal
Surigao	11.50	1	10.5	1	11.5	12	13.5	8.5	6	75.50	Sub-optimal
Tandag	15.5	1	13	1	17	10	8.5	10.5	5.75	82.25	Sub-optimal
Tago	18	1	2.5	1	12.5	3.5	4	4.5	0.75	47.75	Marginal

Table 3: Physico-chemical parameters

RIVERS	PARAMETERS						Description
	TDS	Tempe-rature	pH	Turbidity	DO	TSS	
Dipolog	Passed	Passed	Passed	Failed	Passed	Failed	Poor
Iponan	Passed	Passed	Failed	Failed	Passed	Failed	Poor
Mandulog	Passed	Passed	Failed	Failed	Passed	Passed	Sub-optimal
Padada	Passed	Passed	Passed	Failed	Passed	Failed	Poor
Cabadbaran	Passed	Passed	Passed	Failed	Passed	Passed	Optimal
Surigao	Passed	Passed	Passed	Failed	Passed	Passed	Sub-optimal
Tandag	Passed	Failed	Passed	Passed	Passed	Passed	Optimal
Tago	Passed	Failed	Failed	Failed	Passed	Failed	Poor

Four rivers namely Iponan, Mandulog, Cabadbaran and Tago are evaluated to have a poor bottom substrate condition implying absence of natural habitat and instead are composed mostly of gravels. While Dipolog, Padada and Tandag rivers are with less desirable habitat and only Surigao river was found to have an adequate habitat. Disrupted habitats affect diversity and abundance of aquatic organisms. Decreasing and even loss of natural habitats could be pointed to the modifications to the surrounding areas of rivers brought by human disturbances.

Embeddedness which refers to the extent which rocks are buried by fine sediment appears to be greatest in Padada, Tago and Mandulog Rivers with more than 75% fine sediments. Embeddedness by sedimentation affects river function through reduction of the diversity and abundance of aquatic life by the amounts of unnatural sediments that disrupt natural setting of habitat. Moreover, suspended sediment causes the water to be turbid affecting therefore water turbidity. An increased turbidity reduces light transmission, thereby reducing the growth of algae and aquatic plants, which can adversely affect the entire aquatic ecosystem.

Heavy deposits of sediment material, increased bar development and extensive channelization was observed at Cabadbaran River which could be pointed out to the large-scale quarrying activities along the river. Its banks are almost level to river bed. Iponan and Tago rivers are another two extremely channelized rivers.

Biotic factors

Biotic factors of river system include vegetation in the surrounding area which serves as the first defence against erosion. With grasses, ground covers, shrubs and trees that can effectively protect the soil, agricultural and forested areas have shown significant influence in controlling the water quality. Vegetation reduce flooding by slowing down the transit if rainwater in the catchment and by providing a “roughness” to the land cover in wooded floodplains which can hold back the flood peak in high flows. Preserving these areas as open space and greenbelt accomplishes the multiple goals of flood control water quality enhancement as well as habitat protection.

Lack of canopy and the decreasing vegetative cover among all rivers can be attributed to the increasing urbanization of areas under study leaving the rivers to be mostly covered with grasses to patches of trees and shrubs. Cabadbaran river in particular was found to be dominantly covered with soil and rock materials instead of plants. Stretch of riparian covered by vegetation is moderate range classified marginal to sub-optimal. There is insufficient amount of sapling and seedlings and invasive species found are in less amount indicating moderate disturbance in the vicinity of rivers. Only few stressors on the biotic condition were identified including low intensity ranching, lack of vegetative management and human disturbance. Vegetation patch structures are less to more suitable and less disturbed condition.

Table 4: Results of Assessment on Biotic Components of Selected Critical Rivers

River	PARAMETERS									Total	Description
	Canopy Cover	Bank Vegetative Protection	Streamside cover	Riparian Vegetative Zone Width	Native Riparian Regeneration Rating	Invasive Exotic Plant Species Cover	Biotic Condition Stressors	Vegetation Horizontal Patch Structure	Vegetation Vertical Structure		
Iponan	1.67	3.75	10.75	4.29	5.09	9.75	9.3	11.25	12	67.84	Marginal
Surigao	4	3	8.5	5.5	7.5	7.5	11.5	8	5	60.5	Marginal
Tago	2	6.25	9.5	6.25	9.5	5.5	13	5.5	8	65.5	Marginal
Tandag	1	3.5	14.5	6	5	9.5	12	7	12	70.5	Marginal
Mandulog	1	3	6.25	4.5	2.75	11.63	12	4.25	3	48.38	Marginal
Dipolog	3.5	6.92	11.83	6.5	13.33	12	14.33	10.83	9.83	89.08	Marginal
Cabadbaran	1	2.25	5	5.25	8.25	11.5	12.33	3.5	6.17	55.25	Marginal
Padada	3	3.88	10.5	3.88	9.5	13.5	16.25	8.75	7.75	77	Marginal

Based on the biotic condition of rivers mainly considering the composing vegetation along the rivers are classified as marginal, a less suitable and less disturbed river condition.

Contributory Factors

Secondary data collected and considered in ranking the rivers includes: number of population living along the identified critical rivers, river slope, percentage of the area cultivated within the watershed, stream order, and percentage of clayey and sandy soil types found within the watershed. These factors were considered in this study due to its significance upon the holistic view on the possible cause and effect of flooding towards the aspect of environment, society and economy.

Knowing the possible impact of flooding, it would be easier to come up with the appropriate recommendations that shall be enforced to help lessen any flooding incidence upon these rivers.

Population density was found to significantly affect river system through impacts of urbanization. In this study Iponan river was the most populated. According to Maya et al. (2012), rise in population and developmental activities are related to the high densities of mines/quarries. Average slope degree in watersheds was as well given significance on river ranking. Steepest slope is associated to more surface runoff and erosion (Harvey et al. 2009). Water surface with greatest slope was found to have the highest occurring velocities. Moreover, according to Jankauskas et al. (2007), erosion rates are found to increase with slope steepness. The watershed of Iponan has the highest degree of slope among other watersheds.

Agricultural cultivation has significantly been associated to river-shallowing and channelization. Most of the rivers in this study were observed with varying degree of chanel alteration which is similar with that of Lower Susquehanna Basin, Maryland where many miles of stream banks were destabilized and sand/silt bars formed in slow moving areas as lands within the basin were developed for agriculture (Millard et al. 1999).

Most of the rivers in this study are low in stream order. Lower stream orders imply greater possibility of flood occurrence. It is more prone to flash floods since precipitating water directly reaches mainstream and causes immediate flooding especially during heavy rainfalls. On the other hand, it takes more time for runoff to reach higher order streams delaying increase of water and hence of flooding.

Occurring erosions and vulnerability to runoff is also reflected to the soil type composition of watershed. The clayey and sandy were the two considered soil types in ranking since these were considered to have the most influence in flooding and erosion scenarios of a watershed. Tandag has greatest percentage of clayey material. Clayey type as a compacted soil is known for its poor drainage and infiltration capacity which tends to support runoff. Iponan on the other hand has the greatest composition of sandy soil. The non-cohesive nature of sandy soil type on the other hand easily detaches on the surface and is directly delivered upon commencement of runoff (Ahmed et al. 2009), hence prone to erosion.

Table 5: Ranking of Contributory Factors

Rivers	Population	Degree of Slope	Expanse of cultivation	Stream order	Amount of clayey soil	Amount of sandy soil
Dipolog	2	7	2	1	2	3
Iponan	6	8	1	1	7	1
Mandulog	7	4	4	2	8	7
Padada	1	1	8	1	6	7
Cabadbaran	4	2	7	2	5	6
Surigao	3	6	3	1	3	2
Tandag	8	3	5	1	1	4
Tago	5	5	6	1	4	5

Ranking of Rivers

Table 7 shows the final ranking of rivers based on abiotic and biotic components and the contributory factors which pose significance on the holistic view of possible cause and effect of flooding towards the aspect of environment, society and economy.

Among the eight assessed rivers, Tago river got the lowest rating implying as the most degraded river. Abiotic, biotic assessment and corresponding contributory factors, which relates to the vulnerability and susceptibility of the watershed to scenarios of flooding and erosion were the integrated aspects which come up to the current ranking.

Table 7. Final Ranking of Selected Critical Rivers

Rivers	Rating			SUM	RANK
	Abiotic Component	Biotic Component	Contributory Factors		
Tago	10	40	40	90	1
Surigao	150	20	5	175	2
Cabadbaran	20	10	200	230	3
Iponan	5	80	150	235	4
Mandulog	250	5	80	335	5
Dipolog	80	250	10	340	6
Tandag	200	150	20	370	7
Padada	40	200	150	390	8

Conclusion and recommendations

Ranking results based on the actual field survey and geomorphological analysis revealed Tago River to be the most deteriorated with severely scoured banks and channelized stream. The increasing degree of impairment of rivers is as follows: Padada< Tandag < Dipolog < Mandulog < Iponan < Cabadbaran < Surigao < Tago. Evidences of river profile modification are seen in the extreme river channelization, sediment deposition, poor bank stability, poor river bank vegetation and absence of canopy. The excessive and unregulated vast large scale quarrying and intensive agricultural cultivation along the river are considered as stressors causing deterioration that led to gradual loss of optimum river function. As stated by Maya et al. (2012), quarrying is one of the most recognized destructive human activities that affect land, water, air and biosphere. A degrading river makes it vulnerable to flooding scenarios that may cause negative impacts to domestic, agricultural and industrial aspects. Moreover, the insufficient density of vegetation along riparian zones and upstream areas of watersheds makes surrounding built up and residential areas more vulnerable to scenarios of flooding. The modifications of rivers affect ecological system hindering total environmental service.

To limit further river mutilation from indiscriminate and unsustainable resource extraction, it is recommended to have empowerment among Local Government Units for the strict implementation and reinforcement of laws concerning sustainable quarrying operations. An enstablishement of an institutional governing boards is suggested to which will be responsible on well coordinating government policies. It is recommended that greenbelt establishment and environmental protection ordinances by the LGU must be implemented which will cover series of agroreforestation along denuded areas in watershed. Hydrologic analysis which will include biophysical characterization activities and further comprehensive watershed assessment studies in rehabilitation strategies is recommended.

Acknowledgement

The researchers would like to thank the Mindanao Development Authority (MinDA) headed by Ms. Janet M. Lopoz, the Exectuive Director for the funding and CMU administration headed by Dr. Maria Luisa R. Soliven for the support.

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……....PART II

Papers on Food……....

gDisposable Acetylcholinesterase Biosensor for Pesticide Detection Based on Poly (1,5-diaminonaphthalene)/polypyrrole Nanowires Bilayer

LÊ-HUY NGUYEN

School of Chemical Engineering, Hanoi University of Science and Technology, Vietnam

Abstract

In this study, an acetylcholinesterase (AChE) biosensor was developed based on bilayer of poly(1,5-diaminonaphthalene) and polypyrrole nanowire (P1,5DAN/PPy NWs) structures modifying carbon screen-printed electrodes (SPEs). A polypyrrole nanowire inner layer was electrodeposited on the surface of SPEs to enhance conductivity and specific areas. A poly(1,5-diaminonaphthalene) outer layer was used for immobilizing AChE through glutaraldehyde cross-linking. The conducting polymer poly(1,5-diaminonaphthalene)/polypyrrole nanowires (P1,5DAN/PPy NWs) bilayer was electrosynthesized and characterized by Field Emission Scanning Electron Microscope (FE-SEM), Fourier Transform InfraRed (FT-IR) and Cyclic Voltammetry (CV) techniques. On the basis of the inhibition of organophosphate pesticides (OPs) on the enzymatic activity of AChE, the AChE-immobilized P1,5DAN/PPy NWs/SPEs were designed for electrochemical determination (cyclic vomtametry and chronoamperometry) of Trichlorfon as one of the popular OPs.

Introduction

Organophosphate pesticides (OPs) have been widely used in agriculture due to their low persistence and high insecticidal activity. However, the contamination of OPs present in the eco-systems creates a lot of pollution problem. Current analytical methods like gas or liquid chromatorgraphy and mass spectroscopy have been widely used for the determination of OPs [1]. However, control food safety problems and environmental pollution recently require the development of better devices for simple, quick and convenient analysis.

Meanwhile, electrochemical biosensors in general and enzyme sensors in particular, have received considerable attention regarding the detection of pesticide residues owing to the advantages of low cost, simplicity and high sensitivity, thus making them excellent candidates for portable detection devices. In living beings OPs bind irreversibly to the active site of the acetylcholinesterase (AChE) enzyme [2]. Thus acetylcholinesterase (AChE) biosensors based on the inhibition on AChE have shown satisfactory results for pesticides analysis, where the enzyme activity was employed as an indicator of quantitative measurement of insecticides. When AChE is immobilized on the working electrode surface, its interactions with the substrate of acetylthiocholine produce the electroactive product of thiocholine. The inhibition on the enzyme system can be monitored by measuring the oxidation current of thiocholine [3].

$$\text{Acetylthiocholine} + H_2O \xrightarrow{\text{AChE}} \text{Thiocholine} + \text{Acetic acid}$$

$$2\text{Thiocholine} \xrightarrow{\text{anodic oxidation}} \text{dithio-bischoline} + 2H^+ + 2e^-$$

The immobilization of enzyme to solid electrode surface still remains a great challenge for the fabrication of biosensors. The use of conducting polymepolypyrrole (PPy) matrix for entrapment enzyme has been received great attention in the past few years. PPy exhibits a high conductivity, easy preparation, and excellent environmental stability. However, it cannot attach enzyme directly due to the absence of functional groups such as carboxyl or amino-groups. To resolve this problem, a poly(1,5-diaminonaphthalene) (P1,5DAN) outer layer with functional amino groups was electrosynthesized onto polypyrrole nanowire (PPy NWs) which was previously template-free electrodeposited. The proposed bilayer configuration (labeled as P1,5DAN/PPy NWs) performed for covalent immobilization AChE in the design of electrochemical biosensors in rapid determination of Trichlorfon as one of the popular OPs.

Experiments

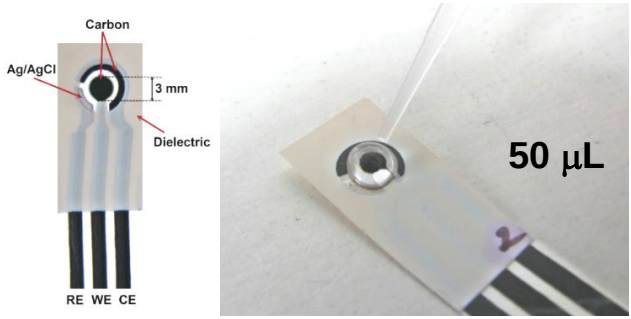
Reagents and Apparatus

Pyrrole monomer (Fluka) was purified by distillation under nitrogen atmosphere before using. Acetylthiocholine chloride (ATCl), AChE (VI-S, 1000 IU/mg from electric eel), Trichlorfon ([2,2,2- trichloro-1-hydroxyethyl]-phosphonic acid dimethyl ester), glutaraldehyde (25%), Bovine Serum Albumin (BSA) and $LiClO_4$ were purchased from Sigma–Aldrich.

The 0.1 M PBS solution (pH 7.4) was prepared by mixing the solutions of 0.1 M Na_2HPO_4, 0.1 M KH_2PO_4, 0.137 M NaCl and 2.7 mM KCl and de-ionized water was used throughout the experiments.

The electrochemical experiments were carried out with an Autolab PGSTAT302N potentiostat interfaced to a GPES 4.9 software (EcoChemie, The Netherlands) using the screen-printed electrodes (SPEs) at room temperature. SPEs were fabricated by thick-film technology with DEK Albany 247 printing machine (Weymouth, UK). SPEs (each 3 mm × 12 mm) with a standard three-electrode cell were printed onto polyethylene terephthalate film. The working and counter carbon-based screen-printed electrodes were prepared from the carbon ink Electrodag PF-407A. The diameter of the working carbon electrode was 3 mm. The reference electrode was printed from the silver ink Electrodag 418 S with $E_{ref} \approx +0.3$ V *vs*. standard hydrogen electrode (SHE) [4]. Finally, a last layer of a thick film curable dielectric was covered with PVC insulator, leaving a defined rectangular shaped (circular well of 5 mm diameter) working area (Figure 1).

Figure 1: The picture of the integrated screen-printed electrodes



Electropolymerization of poly(pyrrole/1,5-diaminonaphthalene) bilayer nanowires

Firstly, the electropolymerization of PPy NWs on SPEs was carried out by using 0.15 M Py monomer in 0.2 M Na_2HPO_4 in the presence of various concentration of $LiClO_4$ (1 mM – 15 mM) [5] under potentiostatic conditions at E = +0.75 V in 500 s. Then, P1,5DAN layer was covered onto PPy NWs/SPEs surface in 0.1 M $HClO_4$ solution containing 1.10^{-3} M DAN using cyclic voltammetry (CV). The parameters for CV: scan rate 50 mV/s; the potential range for electropolymerization is between –0.02 and +0.75 V. Afterwards, the produced electrodes (P1,5DAN/PPy NWs/SPEs) were washed with deionized water and incubated in PBS for 2 hours.
Fourier Transform Infrared (FT-IR) spectra were recorded with Nicolet 6700 FT-IR Spectrometer. The surface morphology was analysed by Field Emission Scanning Electron Microscope (FE-SEM) with Hitachi S-4800.

Fabrication of AChE sensors

A volume of five microliters of 2.5 % glutaraldehyde was coated on P15DAN/PPy NWs/SPEs to form cross-linked P1,5DAN with free CHO groups. The modified electrode obtained was treated with 6 µL AChE solution (300 mU, containing 5 g/L BSA to maintain the stability of AChE) and kept at room temperature for 1 h for covalent linkage of the AChE to the electrode surface [6]. Then, the obtained sensors (labeled as AChE-P15DAN/PPy NWs/SPEs) were washed with PBS (pH 7.4) to remove the excess of AChE. The sensor was stored in the refrigerator at 4 °C for further use.

Electrochemical measurement

As can be seen in Figure 1, 50 µL droplet deposited over the electrochemical microcell delimited by the circular dielectric layer. The AChE-P1,5DAN/PPy NWs/SPEs were tested by cyclic voltammetry (CV) and Chronoamperometry at +0.65 V in PBS solution, and the anodic current was recorded as I_0. After, 1.0 mM ATCI solution (optimized value was determined from [3]) was injected into the microcell, the anodic current was recorded as $I_{control}$. For the measurements of Trichlorfon, the PBS solution containing the different standard concentrations of Trichlorfon was injected into the microcell. The anodic current of ATCI on AChE-P1,5DAN/PPy NWs/SPEs with Trichlorfon inhibition is I_i. The inhibition of Trichlorfon was calculated as:

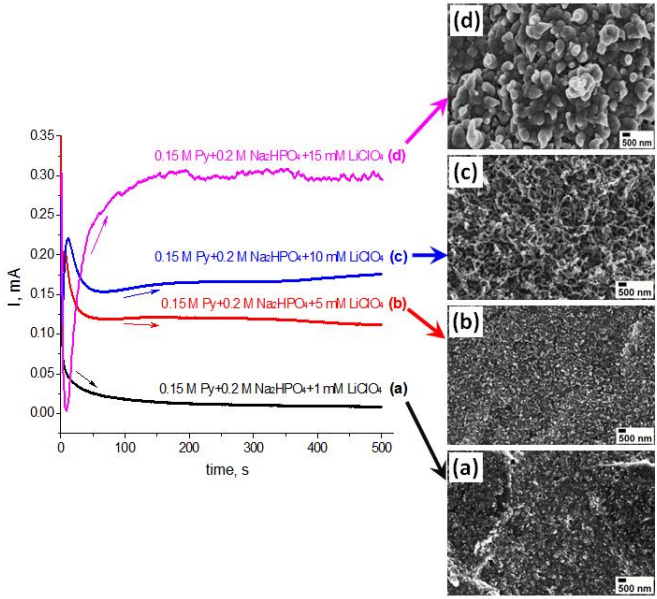
$$I\% = [(I_{control} - I_i)/(I_{control} - I_0)] \times 100\%$$

Results and Disscustions

Electropolymerization and characterization of the P1,5DAN/PPy NWs

The P1,5DAN/PPy NWs bilayer-modified electrodes were obtained as described in Section 2.2. First, the synthesis voltammograms and surface morphology of PPy NWs were given in Figure 2.

Figure 2: Current density versus time of polarization at 0.75 V in 0.15 M Py + 0.2 M Na_2HPO_4 +: 1 mM $LiClO_4$ (a); 5 mM $LiClO_4$ (b); 10 mM $LiClO_4$ (c); 15 mM $LiClO_4$ (d) and SEM images of PPy deposits obtained, respectively



The rate of polymerization increases on further increase of the $LiClO_4$ concentration. At 1 mM $LiClO_4$, the anodic current decreases versus time to a very low value (curve a). The PPy film on the electrode is so thin and incomplete (Figure 2a). When $LiClO_4$ concentration (5 mM and 10 mM) increases in monomer solution, first the anodic current response increase highly then it decreases rapidly. In observations on the voltammograms, the continuous current (curve b) is slowly decreased or saturated, but the continuous current (curve c) higher and it slightly increases with time. The morpholog in Figure 2b and Figure 2c respectively show that PPy NWs generated in 10 mM $LiClO_4$ is the best. 10 mM $LiClO_4$ concentration was selected for polymerization of PPy NWs.

As seen in Figure 2, curve d corresponding to 15 mM $LiClO_4$, the PPy cauliflower-like was observed (Figure 2d). This PPy NWs structure has uniform pattern, high effective surface area, and low diffusion resistance so it is a suitable candidate for the purpose of electrode surface modification [7].

In this work, a layer P15DAN was covered on PPy NWs in order to improve the ability to immobilize enzyme AChE onto the electrode surface via covalen binding.

Figure 3: Cyclic voltammograms (CVs) registered during electropolymerization of P1,5DAN on PPy NWs

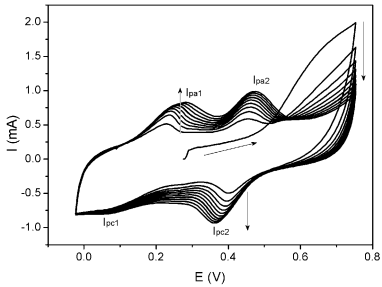


Figure 4: SEM image of P1,5DAN/ PPy NWs bilayer

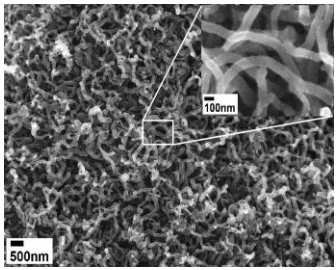


Figure 3 shows the cyclic voltammograms (CVs) taken during the course of electropolymerization of P1,5DAN on PPy NWs/SPEs. The anodic current increases at the first cycle due to the oxidation of 1,5DAN monomer. The appearance of two typical redox systems (from the second cycle, at 0.23 V/0.07 V and 0.45 V/0.40 V) and the current continuously increased during scans reflecting the growth of P1,5DAN film on the PPy NWs/SPEs surface. The SEM image showed a uniform nanowire structures of P1,5DAN/PPy NWs distribution on the electrode surface. The diameter of the nanowires ranges from 80 to 100 nm (Figure 4).

Figure 5 showed CVs behavior of PPy NWs/SPEs and P1,5DAN/PPy NWs/SPEs in 0.1 M PBS (pH 7.4) solution. Two observed redox couples are corresponding to the transformation of oxidation state of P1,5DAN [8] clearly seen when compared with CV of pure PPy NWs under the same conditions. The bilayer was formed by an inner layer of PPy NWs (high specific surface and good conductivity) and an outer layer of P15DAN (where immobilization AChE via amino group), thereby enhancing sensitivity for biosensing.

Figure 5: CVs responses of PPy NWs and P1,5DAN/PPy NWs film-coated SPEs in 0.1 M PBS (pH 7.4), scan rate: 50 mV/s

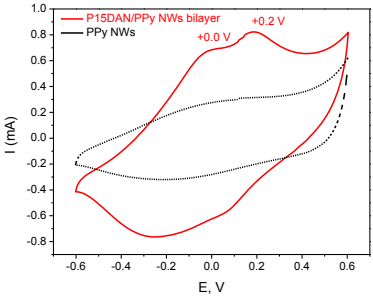
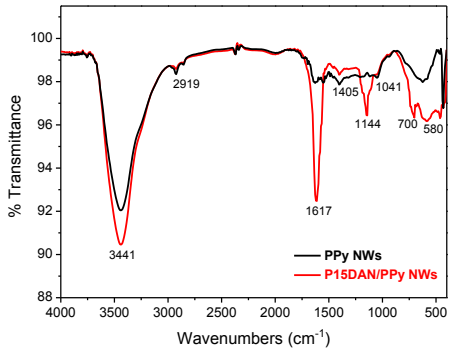


Figure 6 shows the FT-IR spectra of P1,5DAN/PPy NWs bilayer as compared to pure PPy NWs. As is observed, the spectra present the peaks at 3441, 1407 and 1041 cm^{-1} which are corresponding to the N–H, C–N stretching vibration and C–H in-plane vibration on the PPy ring. The peak at 2919 cm-1 is designated to the vibration of CH_2. The characteristic peaks of the bilayer P1,5DAN/PPy NWs showed the increases in intensities at 3441, 1617, 1144, 700 and 580 cm^{-1}. The peaks at 3441 and 1617 cm^{-1} represent N–H stretching and $–NH_2$ scissoring vibration, respectively. Further, a peak appearing at 1144 cm^{-1} is associated with the vibration of C–N stretching vibration. The stronger band at 500-700 cm^{-1} is associated with the $–NH_2$ wagging due to a presence of diaminonaphthalene in polymer chain. All suggests the successful incorporation of P1,5DAN onto PPy NWs matrix by two-step electrochemical polymerization process.

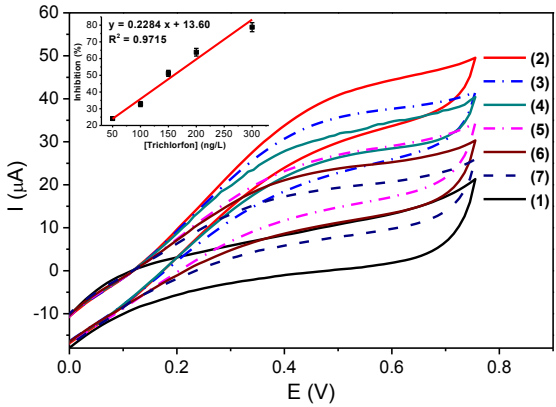
Figure 6: FT-IR spectra of PPy NWs and P1,5DAN/PPy NWs in KBr pellet at room temperature



Pesticide sensing based on AChE-P15DAN/PPy NWs

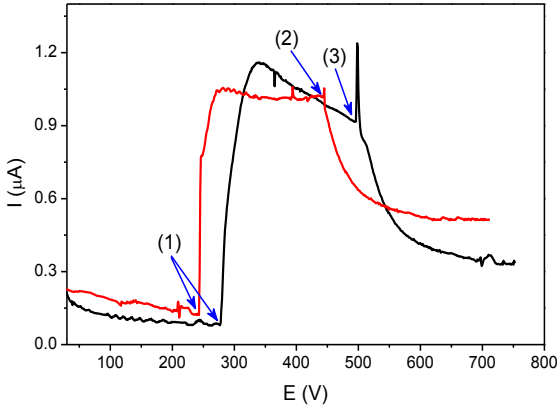
AChE-immobilized electrodes were studied by CV technique between –0.1 V and 0.75 V, scan rate 50 mV/s (Figure 8). The cyclic voltammograms of the AChE-P1,5DAN/PPy NWs/SPEs in PBS solution (curve 1) and in PBS containing 1.0 mM ATCl (curve 2). The AChE-P1,5DAN/PPy NWs/SPEs exhibited significant electrocatalysis to the oxidation of ATCl from *ca.* +0.2 V to +0.75 V. When AChE-P1,5DAN/PPy NWs/SPEs were injected with standard solutions of Trichlorfon at given concentrations, the currents decreased drastically (curve 3-7) compared with the case of no inhibitor (curve 2). The current values at +0.65 V was decreased to 24.12% when the concentration was 50 ng/L (curve 3). It is due to Trichlorfon acting as one of the OPs involved in the irreversible inhibition action on AChE, thus reduced the enzymatic activity to its substrate. At exposure to higher Trichlorfon concentration from 100 ng/L (curve 4) to 300 ng/L (curve 7), the anodic current decreased to 32.79% and 78.78%, respectively. The AChE activity inhibition of Trichlorfon (%) and Trichlorfon concentration (ng/L) have a certain linear relationship. The regression equation was I% = 0.2284×C + 13.60 with the coefficient of determination of 0.9715 (Figure 7, inset).

Figure 7: CVs of AChE-P1,5DAN/PPy NWs/SPEs in PBS (1) containing 1.0 mM ATCl (the control experiment) (2) with Trichlorfon concentrations of 50 ng/L (3), 100 ng/L (4), 150 ng/L (5), 200 ng/L (6), and 300 ng/L (7). Inset: The calibration curve show the relation between inhibitions and concentrations of Trichlorfon



Similarly, Figure 7 presents the results of typical inhibition experiments by chronoamperometry technique. Following stabilisation of baseline, 0.1 mM ATCI is injected in the solution provoking a fast response. Trichlorfon was then injected and a rapid decreasing of enzymatic activity occurs, reaching approximately a steady-state within 100 s. The current decreased to 50% and 65% respective with 100 ng/L and 200 ng/L Trichlorfon. The chronoamperometry technique shows higher sensitivity for detection. It is our key research field in the future for the application of portable biosensor fabrication in rapid and ultralsensitive identification of OPs residues.

Figure 8: Chronoamperometric curve of AChE-P1,5DAN/PPy NWs/SPEs in PBS injected: 1.0 mM ATCI (1); 100 ng/L Trichlorfon (2) and 200 ng/L Trichlorfon (3)



Conclution

This work demonstrated successfully two-step electrodeposition of P1,5DAN/PPy NWs bilayer film on SPEs. The positive combination of all the most outstanding aspects of P1,5DAN and PPy NWs shows the film with high specific surface area, relatively high conductivity, easy preparation and ability of covalent immobilization of biomolecules via amino groups. The disposable AChE biosensor base P1,5DAN/PPy NWs bilayer have been developed for the determination of Trichlorfon. These features provide scope for utilizing the methodology proposed in the present study to immobilize other biomolecules in the process of fabricating biosensors.

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Escaping from Rainfall Uncertainty During Oil Palm (Elaeis Guineensis Jacq.) Seedling Field Transplanting Period of by Pre- application of Biochar and Paclobutrazol at Main-Nursery Stage

TAUFIQ CAESAR HIDAYAT[1], SITI HAJAR AHMAD[1], IMAN YANI HARAHAP[2], HERI SANTOSO[2], EDY SIGIT SUTARTA[2], ROSENANI ABU BAKAR[1], SITI ZAHARA SAKIMIN[1], NOR ELLIZA TAJIDIN[1]

[1]Universiti Putra Malaysia, Malaysia
[2]Indonesian Oil Palm Research Institute, Indonesia

Abstract

Oil palm has become the most important business in many countries in Southeast Asia is sensitive to water stress during transplanting periods in the field. Thus, approximately, planters have to add cost 10% to 20% to cover the effect of water stress. There is lack information on how to face those of problems to decreasing the cost. Applying right doses of biochar and paclobutrazol (PBZ) will produce a tolerance seedling to water stress. The aim of this study is to determine the effect biochar and PBZ in escaping from uncertain rainfall during transplanting period of oil palm. Six months before transplanting to the Rain-Shelter for water stress simulated field condition, oil palm was treated seedling (TS) with a combination of 50 g biochar/seedling and 100 mg PBZ/seedling (treated) and compared to 0 g biochar/seedling and 0 mg PBZ/seedling (control). Seedlings were treated with three Water stress (WS) condition by 100%, 75% and 50% of Field Capacity (FC). There were interactions between TS x WS and TS x SA (seedling age) for seedling height. Treated oil palm showed a rapid growth by 10 cm/months compared to control only 3 cm/months. Control seedling leaf area comparatively greater to treated seedling and WS significantly reduce the leaf area, but it was significantly increased linearly to SA. Treated seedling chlorophyll content was higher than control and it did not affect by WS. On the other hand, chlorophyll content was significantly decreased by water stress 75 and 50 FC in control up to 6% and 9%, respectively (r^2=0.62). Treated seedling showed better in photosynthetic rate compared to control even at 50 FC. Even WS decrease all seedling photosynthetic rates, but treated seedling showed better recovery than control by SA and transpiration rate either. Proline content in control was significantly higher compared to treated seedling as indicating of water stress by 75 and 50 FC. WS treatments significantly decreasing dry shoot production in control seedling, but it did not affect to treated seedling. However, WS treatments were significantly decreased dry shoot all seedling. Seedlings were treated with a combination of biochar and PBZ showed a better growth during drought simulated conditions.

Additional index words: oil palm seedling; water stress; biochar and PBZ.

Introduction

Water is one of the most common and most important substances in plant (Kramer and John 1995). Water is present in every part of the plant and plays a significant role to control growth and yield (Kirkham 2005) of oil palm (Sun, Cao et al. 2011).

Until now, rainfall is the only source of water in oil palm plantation. However, amount and seasonal distribution of precipitation are not the same for each region. Areas with a little rainfall could trigger water deficit and causes water stress to the plant. Water deficit can cause death of transplanted seedlings and reduce yield of mature oil palm. Oil palm plantations in Indonesia and Malaysia, which often suffer from drought are Lampung, South Sumatera, West Java, Borneo and Eastern Indonesia, and Perlis and Kedah in Malaysia (Siregar, Amir et al. 1995).

Water deficit is a major factor limiting the productivity and geographical distribution of oil palm (Sun, Cao et al. 2011). Predominantly, water stress triggers many reactions in the plant that lead to decrease growth rate, relative water content, change of biomass partition, and nutrient distributions. Another effect of these stresses are increasing in the root /shoot ratio, leaf relative conductivity, low rate of photosynthesis, closure stomata, chlorophyll rate and causes death in advance. Some research has been conducted to investigate the causes of morphological and physiological changes of plants in water deficit condition. They found that paclobutrazol (Wang, Shiow et al. 1985; Latimer 1992; Smith 1992; Abou, El-Khashab et al. 1997; Carvajal 1998; Abdul Jaleel, Manivannan et al. 2007; Ali and Shawn 2010) as a plant growth inhibitor and biochar (Laird 2010; Anderson, Condron et al. 2011; Barrow 2011; Galinato, Yoder et al. 2011; Mulcahy, Mulcahy et al. 2012) as a soil amendment could overcome these problems.

Paclobutrazol is a triazole derivative and has been shown to inhibit shoot growth on apple (Wang, Tung et al. 1986), Maple (Marshall, Tannis et al. 2000), mango (Salomon and Reuveni 1994) and oil palm (Hashim 1991). Paclobutrazol, with its active triazol derivate, blocks with high specificity, the oxidative steps leading from ent-kaurene to ent-kaurenoic acid in the biosynthetic pathway of gibberellic acid (GA). One of the main roles of GA in plant is to stimulate cell elongation. When GA production is inhibited, cell division still occurs, but the new cell does not elongate. It produced a plant with the same number of leaves, internodes and stems, all compressed into a compact form. In a compact form it is easy to maintain and to transport oil palm seedlings to the field. The compact seedlings also produced less leaf and stem surface area, but with more fine roots to absorb water (Ali and Shawn 2010). The chlorophyll contents and mineral element concentrations in leaf tissue have also been reported to increase while structural changes occured in the leaves that provide physical barriers to moisture loss. Paclobutrazol was also shown to change the source and sink of photoshyntate from leaves to root and increase the root-shoot ratio. In addition to block ing GA biosynthesis, paclobutrazol is known to increase the production of hormone abscisic acid with the same pathway as GA. One of the functions of abscisic acid is to close stomatal mechanism and reduce water loss from the leaves through transpiration. Paclobutrazol can enhanced tolerance to water stress and improve water use efficiency (Wang, Tung et al. 1986).

A number of studies have been conducted to investigate the effect of biochar application in its potentials role in carbon sequestration (Galinato, Yoder et al. 2011), reducing greenhouse gas emission, renewable energy, waste mitigation, soil amendments (Kookana, Sarmah et al. 2011; Lehmann, Rillig et al. 2011; Zhang, Bian et al. 2012), increasing soil nutrient level and yield (Quilliam, Marsden et al. 2012). Zhang et.al. (2012) found that soil pH, soil organic carbon, and total nitrogen were increased, and soil bulk density decreased under biochar amendment. Adding only 1% (w/w) of biochar into the soil increased soil pH from 4.2 to 5.9 and influences the bioavailability of toxic elements such as Al (Kookana, Sarmah et al. 2011).

However, application of biochar can change soil bulk density and affect the soil water relation, rooting pattern, soil fauna and the most important thing to this research is soil holding capacity (Lehmann, Rillig et al. 2011).
This occurs because the density of biochar is lower than that of soil and because biochar contains macro and microspores (Downie, Crosky et al. 2009). Kookana et al. (2011) found that specific surface area of biochar range from 8 m^2/g to 600 m^2/g with microspores diameters <2 nm. Such extraordinary sorption ability of biochar comes from their high specific surface area, aromaticity, and micro porosity. Meanwhile, other research found that biochar had a significant effect on soil quality with increased level of soil nutrient, soil moisture, organic matter, and a higher of cation exchange capacity (Quilliam, Marsden et al. 2012).
Even though the benefits of paclobutrazol and biochar to overcome water stress in ornamental plants have beeb well studied, there is lack of information about effect of water stress on growth performance of oil palm seedling in response to paclobutrazol and biochar under simulated field condition. The aim of this study was to determine the effects of combining PBZ and biochar in controlling vegetéative growth and proline contents as a method to escape from uncertainty of rainfall (water stress) during field transplanting period.

Materials and Methods

Seedling materials

Experiment was conducted at the IOPRI Marihat – Substation (02^{0}55" NL, 99^{0}05" EL, 369 meters asl). The 3-month old oil palm seedlings from pre-nursery were transplanted to a polybag that contain 20 kg air-dried top soil + 50 g biochar as a soil amendment. Those seedlings were maintained by standard agricultural practices in the open field grown. At 6-months old, seedlings were treated with growth retardant 95% (PP333-TC) PBZ by 100 mg/seedling. PBZ was applied by sowing above the media. The combination of biochar + PBZ was 50 g biochar and 100 mg PBZ/seedling and seedlings without treated with biochar + PBZ were used as a control. At 12-months old, seedling and it's grow media were transplanted to a pot that contain 150 kg air-dried top soil. At 13-months old seedlings were transferred under rain-shelter for simulated water stress condition.

Seedling height, leaf area and leaf chlorophyll contents

Seedling height was measured from the ground to the tip of the highest leaf with measuring tape. Rachis length (RL) and number of fronds (NF) were measured at selected standard leaf (Frond number 9). RL was measured from the point of insertion of lowest rudimentary leaflets to the tip of the rachis. Number of fronds was calculated from frond number one at the end of fronds. Six leaflets were cut each side (at the longest leaflets about 1/4 from the leaf tip) and measured the length and mid-width of each leaflet with ruler or tape. Calculate length x mid-width for each leaflet, and then the mean of the six leaflets was called as b. Relative leaf area (LA) are 2n x b, where b is the mean length x width and n was the leaflet number on one side of rachis (Thomas and Hardter 2010).
For chlorophyll contents determination, each seedling was analysed for the chlorophyll meter (CM) values, which measured the leaf greenness by using Conica-Minolta SPAD 502 as a chlorophyll meter.
Actual chlorophyll content in the leaf was determined based on the CM value and leaf chlorophyll content standard curve. Chlorophyll contents was obtained by follow an equation that was described by Hamzah (2011):

$$Total\ leaf\ chlorophyll\ content\ (y) = 0.0092X + 0.0858$$

The total leaf chlorophyll content was shown in mg chlorophyll/mg leaf fresh weight.

Photosynthetic and transpiration rate

Photosynthesis and transpiration rate was measured with non-destructive using a photosynthesis system, and these instruments measure PAR and control PAR at set intensities. PAR was normally quantified as µmol photons/m2/second, which was a measured of the photosynthetic photon flux (area) density or PPFD by using the Licor LI-6400 Nebraska, USA.

Leaf proline detection

Proline assessment proceeded according to Marin, Andreu et al. (2009) based on proline's reaction with ninhydrin acid. For proline calorimetric determinations, at 1:1:1 solution of proline, ninhydrin acid and glacial acetic acid, was incubated at 100 ºC for 1 hour. The reaction was arrested in an iced bath and the cromophore was extracted with 4 ml toluene and its absorbance at 520 nm was determined in a BioMate 5 spectrophotometer-thermo spectronic, New York, USA. The method was calibrated for each determination with standard proline solutions within the detection range of the method (0-39 µg. ml-1).

Shoot and root dry weight

Dry matter was an important growth indicator that indicated the fixation of carbon dioxide by photosynthesis. Dry matter was determined by a destructive method at the end of the experiment. Shoot fresh weight was measured using an analytical balance after separated it with the root of the root neck (data not supply). All parts of the shoot were taken for dry matter weight for each sample in every treatment and replications. The shoot was chopped into small pieces and was put into a cloth bag and dried in an oven at 70±5 ^{0}C for 48 hours. Then, the dry matter weight was taken and per cent dry matter was calculated using the following formula:

$$Percentage\ of\ shoot\ dry\ weight = \frac{Dried\ weight\ of\ shoot}{Fresh\ weight\ of\ sample} X100$$

Root fresh weight was measured using an analytical balance after removing all the sticky soil on the fibrous root. All parts of the roots were taken for dry matter weight for each sample in every treatment and replication. Root was chopped into small pieces and was put into a cloth bag and dried in an oven at 70±5 ^{0}C for 48 hours. Then, the dry matter weight was taken and per cent dry matter was calculated using the following formula:

$$Percentage\ of\ shoot\ dry\ weight = \frac{Dried\ weight\ of\ root}{Fresh\ weight\ of\ sample} X100$$

Experimental design and Statistics

The experimental was conducted in a randomized complete block design (RCBD) with factorial arrangement of treatments, in four replications. The treatments consisted of two seedling transplant of treated (50 g BC + 100 mg PBZ/seedling) and control (0 g BC + 0 mg PBZ/seedling) and three water stress level of 100%, 75% and 50% FC. The data were analysed using the analysis of variance (ANOVA), and significant means were separated by least significant difference (LSD) at P≤0.05 (SAS 9.3). When interactions between treatments were significant, pooled LSD or regression analysis were carried out separately for each treatment with dependent variable. However, if the interactions were not significant, only main effects comparisons were performed.

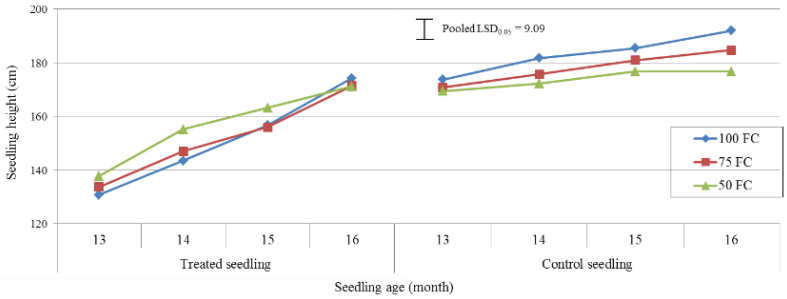
Results

The 3-month old oil palm seedlings from pre-nursery were transplanted to a polybag that contain 20 kg air-dried top soil + 50 g biochar as a soil amendment. Those seedlings were maintained by standard agricultural practices in the open field grown. At 6-months old, seedling were treated with growth retardant 95% (PP333-TC) PBZ by 100 mg/seedling. PBZ was applied by sowing above the media. The combination of biochar + PBZ was 50 g biochar and 100 mg PBZ/seedling and seedlings without treated with biochar + PBZ were used as a control. At 12-months old, seedling and its grow media were transplanted to a pot that contain 150 kg air-dried top soil. At 13-months old, seedling that grown in the big pot were transferred under rain-shelter for simulated water stress condition. Parameters evaluation in this study were seedling height, leaf area, chlorophyll content, photosynthesis and transpiration rate, proline content, shoot and root dry weight.

Seedling height

There were significant interaction effects between seedling transplant x water stress x seedling age on height of oil palm seedlings grown under rain-shelter (Figure 1). Average initial height of treated seedling at 13 months old was significantly lower by 22% compared to control seedling, as the effect of PBZ application in decreased seedling height when it was in the main-nursery. Three months after water stress treatment, height of PBZ100+BC50 seedlings increased rapidly compared to the control seedlings. Control 50 FC seedling height was significantly lower compare to control seedling height at well watered 100 FC at 16-months old. However, control 50 FC seedling height was no significant different compare to all water stress treated seedling at the same age. It showed that water stress did affect on seedling height of control seedling. In contrary, water stress did not effect on seedling height of treated seedling.

Figure1: Interaction effects between seedling transplant x water stress x seedling age of oil palm seedlings grown under rain-shelter. The vertical bar indicates pooled LSD bar at P=0.05. FC = field capacity. The average height of seedling transplant at 12-months old was 120 cm for treated (PBZ100+BC50) and 150 cm for control (PBZ 0+BC 0).



Leaf area and chlorophyll contents

There were no interaction effect between seedling transplant x water stress x seedling age on leaf area of oil palm seedlings grown under rain-shelter (Table 1). Leaf area was increase 3 times bigger at 16 months old compare to initial age at 13 month old. However, PBZ100+BC50 seedlings were significantly decreased leaf area by 15% lower compared to controls. Water availability in soil significantly decreases leaf area by 19% and 24% at 75 and 50 FC, respectively.

There were significant interaction effects between seedling transplant x water stress x seedling age on leaf chlorophyll of oil palm seedlings grown under rain-shelter (Figure 2). Initial control chlorophyll contents at 13 months old was significantly lower compared to treated seedling, but there was not significantly different between treated and control at 1 months after treated with waters tress. From figure 2 showed that water stress did affect in decreasing chlorophyll content, both of treated and control seedling. However, treated seedling showed a better recovery of chlorophyll contents compare to control except for well watered (100 FC). At limiting water supply (75 and 50 FC) chlorophyll contents decrease with time, it was severed at 50 FC. Chlorophyll contents of treated seedling was not significantly difference compare to control at 100 FC, but significantly higher by 15% and 25% compare to control at 75 and 50 FC, respectively.

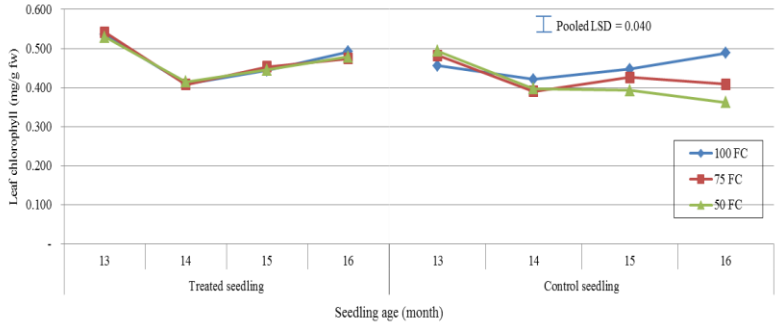
Table 1: Main and interaction effects of seedling transplant x water stress x seedling age on leaf area and leaf chlorophyll of oil palm seedling grown under rain-shelter.

Factor	Leaf area (m^2)	Leaf chlorophyll (mg/g fw)
Seedling transplant (ST)		
Control (PBZ0 + BC0)	7.92 a	0.430 b
Treated (PBZ100 + BC50)	6.73 b	0.469 a
Water stress (WS) (% field capacity)		
100	8.55 a	0.461 a
75	6.93 b	0.448 ab
50	6.50 b	0.440 b
Seedling age (SA) (months)		
13	4.01 c	0.506 a
14	6.50 b	0.407 c
15	6.50 b	0.435 b
16	12.29 a	0.451 b
TS x WS	ns	*
TS x SA	ns	*
WS x SA	ns	*
TS x WS x SA	ns	*

Means within a column followed by the same letter are not significantly different by Least Significant Different test (LSD) at P≤0.05. ^{ns}non-significant at P≤0.05, *significant at P ≤ 0.05.

PBZ = paclobutrazol, BC = biochar. Control treatment was grown without PBZ and BC, while treated seedling was grown with 100 mg PBZ/seedling + 50 g BC/seedling. BC was applied during transplanting and PBZ was applied to soil at 3 months after transplanting.

Figure 2: Interaction effects between treated seedlings and water stress with seedling age on leaf chlorophyll contents of oil palms in rain-shelter. The vertical bar indicates pooled LSD bar at P=0.05. FC = field capacity.



Photosynthetic and transpiration rate activities

There were interactions effect between seedling transplant x water stress x seedling age on photosynthesis (Figure 3) and transpiration rate (Figure 4) of oil palm seedlings grown under rain-shelter. Figure 3 showed there were no significant different between treated and control seedling at initial stage. At one month after water stress treatment, photosynthesis rate was significantly decreasing both for treated and control compare to initial stage. Photosynthesis rate was significantly severe at control seedling with decreasing water contents to 50 FC. On the other hand, treated seedling at 75 and 50 % FC was not significantly different compare to control seedling at 100 and 75 FC, respectively. But, photosynthesis rate of treated seedling at 100 FC was significantly higher by 25% compare to control on the same level of water contents at one month after water stress treatment. At two months after water stress treatment there were increasing of photosynthesis rate for all seedlings except at control treated with 75 FC and revers to slow down at three months after water stress treatment. However, photosynthesis rate treated seedling was significantly higher compare to control seedling at 16 months old except for control with well watering. Indeed, photosynthesis of treated seedling at well watered was significantly higher by 20% compare to control at the same level of watering.

Figure 3. Interaction effects between treated seedlings and water stress with seedling age on photosynthesis rate of oil palms in a rain-shelter. The vertical bar indicates pooled LSD bar at P=0.05. FC = field capacity.

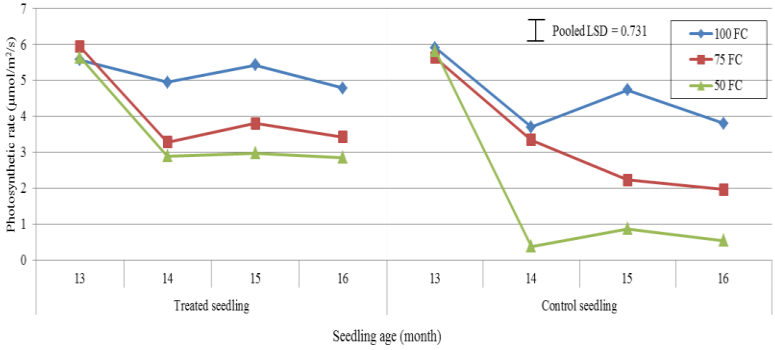
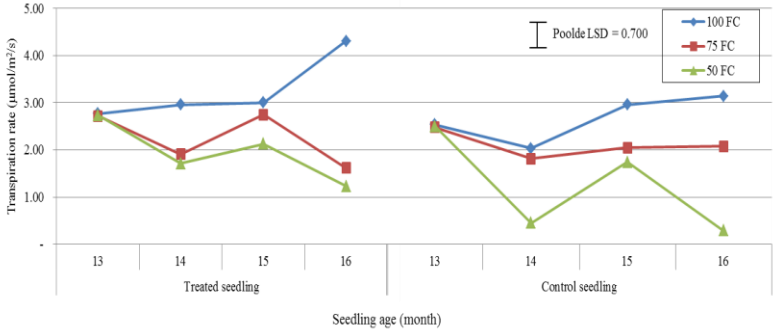


Figure 4: Interaction effect between treated seedlings and water stress with seedling age on transpiration rate of oil palms in rain-shelter. The vertical bar indicates pooled LSD bar at P=0.05. FC = field capacity.



At first month water stress treatment, all transpiration rate in all treated seedling were significantly lower compare to initial stage except at control treated seedling. Water stress at the level of 50 FC affect control seedling severely. It transpiration was reduced almost at the 0 level to survive from drought. It was clearly shown there are transplanting shock during the first month of transplanting, similar pattern like photosynthesis rate.

During two months water stress treatment, all seedling starts to recover and adopt the environmental condition. However, during three months water stress treatment, transpiration rates of treated seedling at 100 FC was significantly higher compared to all seedling. Transpiration of control at 50 FC was significantly severe compare to all treatments. On the other hand, treated seedling at 75 FC was not significantly different compare to control at the same level of water treatment, but significantly different compare to treated and control by water stress 100 FC and 50 FC.

From figure 3 showed that there were not decreasing value of treated seedling during water stress treatment due to transpiration rate and the lowest transpiration rate was found on control seedling at 16 months old and 50 FC water stress condition.

Leaf proline detection

There were no significant interaction effects of treatment on proline contents of the oil palm leaflet (Table 2). The result indicated that there was significantly different ($P \leq 0.05$) in proline contents when seedling was treated with a combination of PBZ and biochar compared to control.

Proline content at treated seedling significantly lower by 16% compare to control. Also, the result of this study showed that water stress treatment did affect proline production of oil palm in rain-shelter. Proline contents was significantly higher by 74% when its water contents in the soil was decreased by 75 and 50 % FC compare to 100 FC water conditions. However, there was no significant difference in proline content when seedling was treated at water stress by 75 and 50 FC.

Table 2: Main and interaction effects between seedling transplant x water stress on proline contents of oil palm seedling grown under rain-shelter.

Factor	Proline (mg/g fw)
Seedling transplant (ST)	
Control (PBZ0 + BC0)	4.67 a
Treated (PBZ100 + BC50)	3.90 b
Water stress (WS) (% field capacity)	
100	2.90 b
75	4.97 a
50	4.98 a
P x W	ns

Means within a column followed by the same letter are not significantly different by Least Significant Different test (LSD) at $P \leq 0.05$. ^{ns}non-significant at $P \leq 0.05$, *significant at $P \leq 0.05$.
PBZ = paclobutrazol, BC = biochar. Control treatment was grown without PBZ and BC, while treated seedling was grown with 100 mg PBZ/L/seedling + 50 g BC/seedling. BC was applied during transplanting and PBZ was applied to soil at 3 months after transplanting.

Shoot and root dry weight

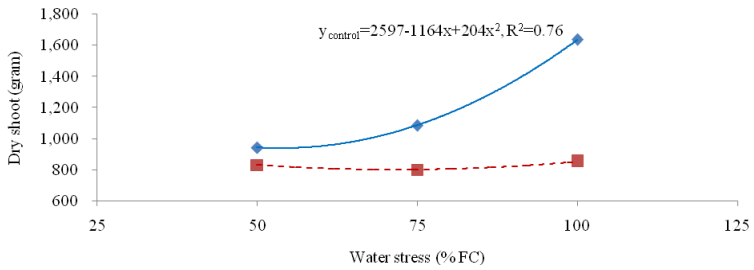
The result indicated that there was interaction between treatments on dry shoot. However, there were no interactions between treatments on dry root (Table 3).

Table 3. Main and interaction effects between seedling transplant x water stress on dry shoot and dry root of oil palm seedling grown under rain-shelter.

Factors	Dry shoot	Dry root
	----------gram----------	
Seedling transplant (ST)		
Control (PBZ0 + BC0)	1,221 b	637 a
Treated (PBZ100 + BC50)	828 a	606 a
Water stress (WS) (% field capacity)		
100	1,245 a	971 a
75	943 b	508 b
50	886 b	384 b
P x W	*	ns

Means within a column followed by the same letter are not significantly different by Least Significant Different test (LSD) at P≤0.05. ^{ns}non-significant at P≤0.05, *significant at P ≤ 0.05.
PBZ = paclobutrazol, BC = biochar. Control treatment was grown without PBZ and BC, while treated seedling was grown with 100 mg PBZ/L/seedling + 50 g BC/seedling. BC was applied during transplanting and PBZ was applied to soil at 3 months after transplanting.

Figure 5: Relationship between dry shoot and water stress when treated with combination of PBZ and biochar at control (PBZ0 + BC0) and treated (PBZ100 + BC50) of oil palm in rain-shelter. Solid line indicates significant quadratic responses.



For dry shoot biomass of 16 months old oil palm (at harvest), result indicated that there were significantly interaction effect of dry shoot and water stress at control transplant seedling. There were negative quadratic relationship between dry shoot and water stress at control seedling (R^2=0.76) (Figure 5). These relationships indicated that dry shoot was high initially and tended to decrease as water stress increase to 50 FC. When control palm was treated with water stress by 75 and 50 FC, the dry shoot was reduced by 34% and 42% compared to well watering 100 FC, respectively. However, there was no significant different of dry shoot biomass when treated seedling was treated with water stress by 75 and 50 FC compare to well watering 100 FC.

The result indicated that there were no significant interaction effects between the treatments on dry root (Table 5). Dry root was significantly decrease and affected by water stress. It was decrease by 48% and 60% when palm treated with water stress by 75 and 50 FC, respectively. However, treated seedling with combination of PBZ and biochar did not affect the dry shoot at harvest.

Discussions

This paper analysed the response of treated oil palm with combination of PBZ and biochar compare to control over time with simulation water stress condition (during transplanting period) in term of its effects on seedling height, leaf area, leaf chlorophyll contents, photosynthetic rate, transpiration rate and leaf proline contents. It was assumed, those treatments did effect on dry shoot and dry root biomass.

Plant height

The timing of transplanting has customarily been influenced principally by the availability of soil water to promote growth. In fact, climate change did affect the uncertainly rainfall that directly affect to set back the seedling growth. Set back in growth should be avoided in oil palm after transplanting to the field (Basiron, Jalani et al. 2000), because it could prolong the immature period (Turner 2003). One of growth parameter in oil palm is seedling height. Treated seedling with combination of PBZ and biochar significantly stimulate the seedling height over time. It was clearly showed that in respective of water stress treatment by 100, 75 and 50 FC, there were increasing in seedling height at treated seedling by 10 cm/month compare to control only by 3 cm/month. Those were evidences that PBZ together with biochar can over comes uncertainly rainfall in the field at transplanting period. PBZ inhibited GA production, decreased leaf area and stimulated other intermediary compound i.e. abscisic acid and the chlorophyll component phytyl, both beneficial to growth and health. Indeed, biochar can increase the water holding capacity to retain water longer than control soil. Both those characteristic did effect to over comes the uncertainly rainfall in the field. The same result was found that paclobutrazol improved drought tolerance in apple (Wang, Shiow et al. 1985). In the other hand, biochar as soil amendments increase tomato seedling resistance to drought in sandy soil (Mulcahy, Mulcahy et al. 2012). Even PBZ applied at 6-months before seedling transplanted under rain-shelter, the effect of PBZ was still present about 1-2 years after applied (Hashim 1991; Adriansen and Odgaard 1997; Anonimous 2007; Anonimous 2012).

Leaf area, leaf chlorophyll, photosynthesis and transpiration rate

One of the aims of this study was to find out at what time transplanting shock will be happen and at what time treated seedling start to diverse and how great they are over time. The experiment showed that during one month after transplant to the field, all seedling showed decreased on chlorophyll contents, photosynthesis and transpiration rate. All parameters was significantly lower compare to initial stage. It was evidence that there were transplanting shock at the beginning of transplanting period. On the other hand, even palms planted in ideal weather conditions, it will affected by transplanting shock and it getting worse by uncertainties drought period which stimulates the incidence of water stress in seedlings as effect of climate change, recently (Fairhurst and R. Hardter 2010). However, seedling that treated with a combination of PBZ and biochar showed the reverse effect of transplanting shock better compare to control. Indeed, the small leaf area in treating seedling effect to control the transpiration rate compare to control that has a bigger leaf area. Gilley and Fletcher (1998) found that PBZ can stimulate the level of some protein as

reaction to water stress. Other research found that seedling in drought simulation condition can maintain the levels of antioxidant and scavenging enzymes if treated with PBZ. These results suggest that the effect of water stress can minimized by the application of PBZ (Sankar, Jaleel et al. 2007). On the other side, biochar could increase water holding capacity and alkaline characteristic by improving the soil pH. Total pore surface area of biochar around 1.10 – 7.67 m^2g^{-1} is responsible to improving soil aeration (Case, McNamara et al. 2012) and provide protection against water stress in the field.

Effect of water stress on Proline contents

Proline was used as indicator that water stress was present in the seedling. It was found that proline contents was increased by water stress treatment over time (Toruan-Mathius, Wijaya et al. 2001). This similar result was found in this study, where proline contents were doubled when seedling treated with water stress 75 and 50 FC. However, proline contents were significantly lower at treated seedling compared to control. It was evident that water stress was present and stimulated proline contents when uncertainly rainfall occurrences. On the other hand, seedling when treated with a combination of PBZ and biochar showed a better response to water stress with still maintains the proline contents in low concentration.

Dry Shoot and root biomass

Photosynthesis produces assimilate that used to produce dry biomass of seedlings. Those, the decreasing of photosynthesis during water stress treatment did affect to dry biomass of oil seedling at harvested. A negative quadratic relationships between treatments on dry shoot showed that dry shoot was significantly lower when control seedling treated with water stress by 75 and 50 FC. In contrary, water stress treatments did not affect the dry shoot of treated seedling. It was clear, proven treated seedling showed a stable growth compared to controls.

Conclusions

Oil palm responses to water stress as occurrences of uncertain rainfall in the field over time were shown at changing of seedling height, growth rate, leaf area, leaf chlorophyll contents, photosynthetic rate, transpiration rate and leaf proline contents, dry shoot and dry root biomass. From the data that were discussed above, it was clearly seedling material treated with combination of giving a better growth during drought simulated conditions. Thus, it recommended to applied 100 mg PBZ/seedling and 50 g biochar/seedling to oil palm seedling at 6 months before transplant to the field to overcome the uncertainly rainfall occurrences.

Acknowledgment

This work was supported by Indonesian Oil Palm Research Institute (IOPRI) funding and under work frame of Soil Science and Agronomy Division. The authors are thankful to all the management of IOPRI for they supports on fund, nursery, laboratory and technical assistance.

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Analytical Survey on Aflatoxin B1, Ochratoxin A, Fumonisin B1 and Fumonisin B2 of Cambodian Rice

DALIN LY[1], SAROM MEN[1], VIBOL SAN[2], NOEL DURAND[3], JEAN-CLAUDE MANEZ[3] AND DIDIER MONTET[3]

[1] Royal University of Agriculture, Cambodia
[2] Royal University of Phnom Penh, Cambodia
[3] Centre de Coopération Internationale en Recherche Agronomique pour le Développement, France

Abstract

An analytical survey of aflatoxin B1, ochratoxin A, and fumonisin B1&B2 was carried out by collecting 40 paddy rice samples during harvest and storage stage from two large rice producing provinces named Prey Veng and Takeo of Cambodia. In addition, 20 milled rice samples were also selected from four local markets such as Kandal, Oreusey, Beungkengkang and Chbar-ampov in the capital city of Cambodia (Phnom Penh). Moisture content of all samples was measured during sample collection and then samples were dried until moisture content reached about 13%. The dried samples were exported to CIRAD in France to analyse aflatoxin B1, total aflatoxins, ochratoxin A, and fumonisin B1&B2 using immunoaffinity column and HPLC with fluorescent detection column. The analytical results revealed that only one sample (1.67%) out of 60 rice samples was contaminated by fumonisin B1 and B2 at 0.8 µg/kg and 0.1 µg/kg, respectively. Furthermore, one milled rice sample was contaminated with aflatoxin B1 and total aflatoxins with the same levels of 0.75 µg/kg. Contamination levels of aflatoxin B1, total aflatoxins, and fumonisin B1&B2 were found in some samples, but the amounts of contaminations were below the European Union detection level (regulation No.1881/2006). The weak quantities of mycotoxins resulted probably of good pre- and post-harvest practices of farmers. Therefore, the result of this study demonstrated that Cambodia rice either paddy or milled rice has a good quality and is safe for consumption by local and/or international consumers.

Key words: Analytical survey, aflatoxin B1, ochratoxin A, fumonisin B1&B2, Cambodian rice.

Introduction

Rice (*Oryzaesativa L.*) is one of the most important staple foods worldwide. A rice-based farming system forms the backbone of Cambodia's agricultural sector. Rice, which is cultivated primarily through traditional farming practices, is grown by over 80% of Cambodian farmers, 60% of whom produce for subsistence needs (Yu and Fan 2009). Rice is grown in areas where necessary humidity and temperature are available. Kernels can be contaminated by moulds during cultivation and subsequent handling of rice. These conditions increase contamination of rice with fungi and some of them could produce mycotoxins (Gonzalez, Juan, and Soriano 2006). In stored rice, the fugal flora is different from that in newly harvested rice. *Aspergillus spp.* are common contaminants of stored rice (Kim et al. 1998;Park, Lee, and Kim 2005;Sales and Yoshizawa 2005). Rice and other agricultural commodities are susceptible to fungal damage before and after harvest. The environmental conditions in the Philippines and other countries in Asia such as Cambodia, characterized by their high temperature (26-39°C) coupled with high relative humidity (67-98%), are conducive for the growth of mycotoxigenic fungi and the contamination by mycotoxins in

agricultural crops. Staple cereals are usually harvested during the rainy season resulting in high moisture content of the grain, sub-optimal conditions for processing and storage, and potentially rapid accumulation of mycotoxins such as aflatoxins (Sales and Yoshizawa 2005). During cultivation and subsequent handling of rice, kernels could be contaminated by moulds, which can grow and produce mycotoxins if conditions are favourable. The fungi may later die due to increase of temperature or dry periods, but once produced; the stable mycotoxins will remain in the rice. Fungal activity depends on the moisture content and temperature, which can both vary significantly in a silo depending on its design and environmental factors. Based on Fredlund et al. (Fredlund et al. 2009), rough estimates showed that high rice consumers may have an intake of 2-3 ng/kg aflatoxin b.w/day from rice alone. Therefore to reduce the aflatoxin contents in rice, preventive measures such as improved farming systems, post-harvest handling, proper drying and storage must be taken into account. In addition, intensified internal and official control may be necessary to improve the quality of available rice on the market (Fredlund, Thim, Gidlund, Brostedl, Nyberg, and Olsen 2009). Post-harvest treatment of rice, including adequate drying and conditions of storage, are crucial factors that will determine storage stability (Fredlund, Thim, Gidlund, Brostedl, Nyberg, and Olsen 2009). Moisture content and storage condition probably affect mycotoxins production. Rice samples collected across India varied in their fungal distribution and alfatoxin B1 contamination in relation to stage and storage condition. The formation of mycotoxins under the conditions of experimental storage of rice grain was explained by the fact that aflatoxin B1 and G1 were accumulated in the rice grain when initial moisture was greater than or equal to 16% (Reddy, Reddy, and Muralidharan 2009). In addition, Qazi and Fayyaz (Qazi, I and Fayyaz 2006) found that the amount of aflatoxins reach a maximum when the temperature of the grain mass rose 35-45°C and that the optimal temperature for ochratoxin A production by *Aspergillus ochraceus* is 28°C; ochratoxin A production depends on humidity and temperature (Sangare-Tigori et al. 2006). Furthermore, seasons had an impact on the contamination by these mycotoxins. In the rainy season, the ratio of contaminated samples and average of quantifiable levels of alfatoxin B1 in rice were logically higher than in the dry season (Nguyen et al. 2007). In Cambodia as it is in Vietnam, conditions of rice storage vary and local commercial rice may be badly stored, and sold without packing. Rice is directly exposed to humid and warm air and then mould growth and mycotoxin contaminations are highly probable. It can seriously affect consumer health. Many of the fluctuations reported in amounts of aflatoxins on rice samples were caused by expected variations in the assay rather than in actual present quantities (Shotwell et al. 1966). After processing, rice grains are generally left for drying in an open place. These grains, if improperly dried, become ideal substrates for the growth of aflatoxin-producing fungi and subsequent aflatoxin production (Toteja et al. 2006). Mycotoxins have been associated with a number of human acute and chronic diseases. It has been estimated that 25% of the world's crops are affected by mould or fungal growth and as stable, natural contaminants of the food chain; mycotoxin reduction requires a multifaceted approach, including farmers, government agencies, food processors and scientists (Brydent 2007). Aflatoxins are of greatest concern as they are highly toxic, mutagenic, teratogenic and carcinogenic compounds that have been implicated as causative agent in human hepatic and extra hepatic carcinogenesis (Hussein and Brasel 2001). It was reported that aflatoxins are produced by several species of *Aspergillus* including *A. flavus* (aflatoxin B_1 and B_2) and *A. parasiticus* (B_1, B_2, G_1, and G_2). An intake of 1 ng/kg bodyweight (b.w)/day, corresponding to a life-time cancer risk of one extra cancer case for 10^5 individuals may be considered as an acceptable risk.

Furthermore, aflatoxins are genotoxic substances and therefore no tolerable daily intake (TDI) levels have been set for these toxins (Fredlund, Thim, Gidlund, Brostedl, Nyberg, and Olsen 2009). In addition, ochratoxin A is a nephrotoxin agent with immunotoxic, neurotoxic and teratogenic effects at higher dose levels and it has been reported that ochratoxin A and citrinin are produced by some Aspergilli (*Aspergillus ochraceus, Aspergillus carbonarius, Aspergillus niger*) and Penicillia (*Penicillium viridicatum, Penicillium verrucosum,* and *Penicillium cyclopium*) (Manderville and Pfohl-Leszkowicz 2006;Pfohl-Leszkowicz et al. 2002). Some studies have found the implication of ochratoxin A in certain epidemic nephropathies in animals and humans (Bennett and Klich 2003;Castegnaro et al. 2006;Pfohl-Leszkowicz, Petkova-Bocharova, Chernozemsky, and Castegnaro 2002). Approximately 4.5 billion persons living in developing countries are chronically exposed to largely uncontrolled amounts of aflatoxins which result in changes in nutrition and immunity (Williams et al. 2004). Fumonisins are phytotoxic mycotoxins which are synthesized by various species of the fungal genus *Fusarium* (Yazar and Omurtag 2008). The International Agency for Research on Cancer (International Agency for Research on Cancer 1993) has evaluated the cancer risk of fumonisin to humans and grouped them as group 2B (probably carcinogenic). The objective of this study was to determine rice contamination in Cambodia by aflatoxins, fumonisins and ochratoxin A using HPLC for mycotoxins quantification.

Materials and Methods

Materials

Samples were collected at three different stages such as at harvest, storage, and milled rice in the markets. Twenty paddy rice samples were selected from each province (Prey Veng and Takeo) and twenty milled rice samples were purchased from four retail markets (Kandal, Oreusey, Beungkengkang, and Chbar-ampov) in Phnom Penh. A minimum sample size of 1000g of paddy rice was collected, sun-dried and temperature was not over 40°C. All samples were dried until moisture content reached about 13%. Then 200g of samples was packed in poly-ethylene plastic bag before sending to CIRAD in France for analyse.

Methods

Sample preparation

All reagents, sodium chloride, nitric acid, potassium bromide, phosphate buffer (PBS), sodium dihydrogen phosphate, *o*-phthal aldehyde (OPA) were of analytical grade (Sigma). All solvents, methanol, acetonitrile, 2-Mercaptoethanol, and borate were of HPLC grade (Merck). Deionised water was used for the preparation of all aqueous solutions and for HPLC. Standard mycotoxins, aflatoxins (B1, B2, G1, G2), ochratoxin A (OTA), fumonisins B1 (FB1) and B2 (FB2) were supplied by R-Biopharm Rhone LTD.

Standard preparation of aflatoxins and OTA

Standard solution (SolutionAF_OTA1): 100 µL of 1 µg/mL total aflatoxins (TAFs) standard and 60 µL of 1 µg/mL of OTA standard made up to 2.5 mL with methanol then mixed with 2.5mL water. SolutionAF_OTA1 contained: total aflatoxins 20 ng/mL and OTA 12 ng/mL. The calibration curve was prepared on four points as shown in Table 1. Injection loop of 100 µL and isocratic flow rate of 0.8 mL/min were used. The mobile phase was a mixture of 50% distilled water, 25% acetonitrile and 25% methanol, 120 mg/L of KBr and 350 µL of nitric acid 4M for 1 L. Aflatoxin and OTA concentrations were determined by using a spectrofluorimetry detector at 0 to 20 min

with 362 nm λ excitation and 425 nm λ emission (Aflatoxins detection), 20 min to 50 min with 333 nm λ excitation and 460 nm λ emission (OTA detection). Control of chromatographic condition was achieved with a standard solution: Solution AF_OTA3.

Table 1: The calibration curve of aflatoxins and OTA

	Solution $_{AF\_OTA}$ 2 2.5 ng/mL Afla + 1.5 ng/mL OTA	**Solution $_{AF\_OTA}$ 3 1.25 ng/mL Afla + 0.75 ng/mL OTA**	**Solution $_{AF\_OTA}$ 4 0.625 ng/mL Afla + 0.375 ng/mL OTA**	**Solution $_{AF\_OTA}$ 5 0.25 ng/mL Afla + 0.15 ng/mL OTA**
Solution AF_OTA 1 20 ng/mL Afla+ 12 ng/mL OTA	Solution $_{AF\_OTA}$ 1 0.25 mL	Solution $_{AF\_OTA}$ 2 1 mL	Solution $_{AF\_OTA}$ 3 1 mL	Solution $_{AF\_OTA}$ 4 0.8 mL
Methanol: water 1:1	1.75 mL	1 mL	1 mL	1.2 mL

Standard preparation of fumonisins

Standard solution$_{Fumo}$1 : 500 µL of 20 µg/mL fumonisin B1 and B2 standard at a ratio of 2:1 (13.33 µg/mL FB1 and 6.66 µg/mL FB2 in 1 mL of 1:1 acetonitrile: water. Solution 1: 5000 ng/mL of fumonisin B1 and B2. The calibration curve was calculated on four points as shown in Table 2. The mixture was mixed and kept for 1 min to react before the injection of 100 µL of the derivatized eluate into the HPLC system. 100 µL injection loop and isocratic flow rate of 1 mL/min were used. The elution solvent was a mixture of 77 % methanol and 23 % 0.1 M sodium dihydrogeno phosphate. Concentrations of FB1 and FB2 stock solution were determined by using a spectrofluorimetry detector with λ excitation = 335 nm and λ emission = 440 nm. Control of chromatographic condition was achieved with a standard solution: Solution Fumo 3.

Table 2: The calibration curve of fumonisins

	Solution$_{Fumo}$ 2 666 ng/mL fumonisins (444 ng/mL FB1, 222 ng/mL FB2)	**Solution$_{Fumo}$ 3 333 ng/mL fumonisins (222 ng/mL FB1, 111 ng/mL FB2)**	**Solution$_{Fumo}$ 4 133 ng/mL fumonisins (89 ng/mL FB1, 44 ng/mL FB2)**	**Solution$_{Fumo}$ 5 66.5 ng/mL fumonisins (44.5 ng/mL FB1, 22 ng/mL FB2)**
Solution$_{Fumo}$ 1 5000 ng/mL of fumonisin B1 and B2	Solution$_{Fumo}$ 1 : 0.40 mL	Solution$_{Fumo}$ 2 : 1.5 mL	Solution$_{Fumo}$ 3 : 1 mL	Solution$_{Fumo}$ 4 : 1 mL
Methanol: water 1:1	2.6 mL	1.5 mL	1.5 mL	1 mL

Analysis of aflatoxin B1 (AFB1), total aflatoxins (TAFs) and ochratoxin A (OTA)

25 g of rice/paddy and 5 g of sodium chloride were poured into a warring blender and 100 mL of extraction solvent (methanol + water, 80+20) was added. Solution was mixed for 2 min at high speed, and then the extract was centrifuged at 3000 rpm for 10 min. Filtered extract of 2 mL was diluted with 18 mL of PBS buffer (R-Biopharm). Then diluted extract was poured into the reservoir and the flow rate was adjusted to 2 mL/min maximum on the column. The column was rinsed twice with 10 mL of PBS. The reservoir was removed and a silanized recovery flask was placed directly beneath the column. The column was eluted with 1 mL of methanol (VWR). The solvent was sucked up with a syringe during elution, and the same thing was done with 1 mL water into the glass syringe barrel, pass through the column and collect in the amber vial, to give a total volume of 2 mL.

Analysis of fumonisin B1 and B2

The same protocol as 2.2.4 was applied. 25 g of rice/paddy were mixed with 5 g of sodium chloride into a warring blender and 100 mL of extraction solvent (methanol + water, 80+20) was added. Solution was mixed at high speed for 2 min, and then centrifuged to separate the extract at 3000 rpm for 10 min. Filtered extract (10 mL) was diluted with 40 mL of PBS buffer. The immunoaffinity column was fitted to the vacuum manifold and the reservoir to the immunoaffinity column. The diluted extract was poured into the reservoir and the flow rate was adjusted to 2 mL/min maximum in the column. The column was rinsed twice with 10 mL of PBS. The reservoir was removed and a silanized recovery flask was placed directly beneath the column. The column was eluted with 1.5 mL of methanol. The solvent was suck up with a syringe during elution, and make the same thing with 1.5 mL water into the glass syringe barrel, pass through the column and collect in the amber vial, to give a total volume of 3 mL.

To prepare fumonisin B1 and B2 derivatives, about 200 µL of sample extract or standard solution was mixed with 200 µL of derivatization solution. The mixture was mixed and kept for 1 min to react before the injection of 100 µL of the derivatized eluate into the HPLC system. Derivatization solution was prepared as follows: 5 mL of 1 mg/mL of OPA reagent was mixed with 10 µL of 2-mercaptoethanol and 2 mL of borate buffer (pH 10). The pH was verified and adjusted to 10 if needed. This solution was kept in the dark at 4°C.

Results and Discussion

Socio-economic data

Different agricultural techniques were used by local farmers. According to the results obtained by interviewing farmers during rice grain sampling, 70-100% of total sampled households in Takeo and Prey Veng province commonly used chemical fertilizers such as DAP (Diammonium Phosphate) (18-46-0), Urea (46-0-0), NPK (20-20-25). Farmers in Takeo province also used much amount of cattle manure as organic fertilizer approximately at 90% higher than farmers in Prey Veng who used organic fertilizer at about 10-30%. In addition, results indicated that pesticides, imported from Vietnam, are being used by some households in Takeo and Prey Veng to kill insects or other pests. The amounts of pesticides are normally applied by farmers at 10-36% of the total sampled households. The average of moisture content, measured during sample collection in the fields, on the wet samples after immediate harvesting was approximately 20-25%, so these samples needed to be dried more to make sure that the moisture content was lower than 14% in order to prevent spoilage

or eliminate microbiological activity during storage. In Cambodia, rice grain is generally sun dried after harvesting in order to reduce moisture content and then stabilized for long term storage. Therefore, all studied samples had about 14% of moisture content.

Mycotoxins analysis

Aflatoxins (B1, B2, G1, G2), OTA, FB1 and FB2 were determined by an immunoaffinity column clean up method coupled with HPLC. As can be seen from Table 3, only 1 out of 20 paddy rice samples contented FB1 and FB2 in Prey Veng province at levels of 0.1 and 0.8 µg/kg, respectively. All levels of fumonisins contaminated in paddy rice were very low compared with the European Regulation on cereals products for direct human consumption. According to Katta *et al.*1997, FB1 and FB2 were also more concentrated in the germ, bran, animal flour, and fine than in the inner portions. The levels of TAFs contaminations in the study of milled rice at 0.75 µg/kg was more than 5 folds below the European legislative limits (4 µg/kg) (Table 4). The results showed very low contaminations in milled rice by AFB1 and TAFs because milled fractions were the result of the whitening process, where the bran layer was removed from the brown kernels. The fact that the levels of aflatoxins in those fractions were statistically lower than the concentrations found in the rest of the fractions provided strong evidence that whitening, as a physical process used to convert the grain in an edible food intended for human consumption, was effective in avoiding the transference of high aflatoxin quantities to those final fractions. Some other studies have reported that different mycotoxins were found in fractions extracted from the outer layers of kernels in higher levels than in fractions coming from the internal parts. They reported also that aflatoxins were removed in fractions intended for human consumption (milled broken grains and milled whole kernels) at rates up to 97 %. In addition to the main goal of removing the outer portions of the grains, rice milling incidentally also reduced initial contamination by more than 90%; therefore, dehulling and whitening processes could be considered as good processes to improve the safety of the final fractions. Castells *et al.*2007 also reported that hull and bran fractions were more heavily contaminated by aflatoxins than milled broken grain and milled whole kernel fractions. Moreover, the high content in fats of outer layers may be favourable to the attack of moulds; thus growth of *A. flavus* occurred mostly in surface layers. Mycotoxins contamination in rice is usually lower as in wheat or corn. However, there are some reports that rice has been contaminated with mycotoxins such as aflatoxin B1, B2, G1, G2 (AFS), citrinin, deoxynivalenol (DON), fumonisin B1, B2, B3 (FMS), fusarenon-X (Fus-X), nivalenol, ochratoxin A and zearalenone (Tanaka et al. 2007).

Table 3. Mycotoxins analysis of paddy rice from two provinces of Cambodia

Mycotoxins	Contaminated sample		Mycotoxins (µg/kg)	EU standard No 1881/2006 (µg/kg)
	Takeo	Prey Veng		
Ochratoxin A	0/20	0/20	nd	3
Aflatoxin B1	0/20	0/20	nd	2
Total aflatoxins	0/20	0/20	nd	4
Fumonisin B1	0/20	1/20	0.8	-
Fumonisin B2	0/20	1/20	0.1	-
nd: no detectable				
detection limit:	µg/kg		µg/kg	
Aflatoxin B1	0.05	Ochratoxin A	0.1	
Aflatoxin B2	0.05	Fumonisin B1	0.05	
Aflatoxin G1	0.1	Fumonisin B2	0.05	
Aflatoxin G2	0.1			

Table 4. Mycotoxins analysis of milled rice from markets in Phnom Penh

Mycotoxins	Contaminated sample		Mycotoxins (µg/kg)	EU standard No 1881/2006 (µg/kg)
Ochratoxin A	0/20		nd	3
Aflatoxin B1	1/20		0.75	2
Total aflatoxins	1/20		0.75	4
Fumonisin B1	0/20		nd	-
Fumonisin B2	0/20		nd	-
nd: no detectable				
detection limit:	µg/kg		µg/kg	
Aflatoxin B1	0.05	Ochratoxin A	0.1	
Aflatoxin B2	0.05	Fumonisin B1	0.05	
Aflatoxin G1	0.1	Fumonisin B2	0.05	
Aflatoxin G2	0.1			

In addition, Pande *et al.*1990 showed that rice samples were contaminated with aflatoxins more than wheat and corn. From the result of analysis, there was only one sample contaminated by fumonisins among all 60 samples. Therefore, we found the same results as Pitt *et al.* 1994 and Pacin *et al.* 2002 who showed that species of *fusarium* have been isolated from newly harvested paddy rice and low levels of *fusarium* toxins have been detected (Kim, Kim, Shon, Ryu, and Chung 1998;Park, Lee, and Kim 2005). However, *fusarium* toxins are not considered as a risk in rice (The Commission of The European Communities 2005). *Fusarium* mycotoxins found in food are produced mainly in the field, although some toxin synthesis may occur during storage. Temperature and moisture conditions are crucial factors affecting fungal infection and toxin synthesis (Yazar and Omurtag 2008). Table 4 illustrated that 1 out of 20 milled rice samples was contaminated by AFB1 and TAFs at the same level of 0.75 µg/kg. The Maximum Limit in cereals forAFB1 is 2 µg/kg, and for the total aflatoxins (B1, B2, G1, and G2), it is 4 µg/kg (Regulation No 1881/2006). Aflatoxin B1 belongs to the most studied mycotoxin family in the world. In an eastern African country, such as Kenya, rice may be contaminated at levels ranging from 294 to 1050 µg/kg. In the USA, aflatoxin B1 contamination in rice was ~280 µg/kg, in India 180 µg/kg, in the Philippines 16 µg/kg, in Thailand 98 µg/kg (Sangare-Tigori, Moukha, Kouadio, Betbeder, Dano, and Creppy 2006) and in Vietnam 29.8 µg/kg (Nguyen, Tozlovanu, Tran, and Pfohl-Leszkowicz 2007), at levels up to 96 µg/kg for aflatoxins in rice samples in Malaysia; and regarding

AFB1 and AFG1 for parboiled rice samples in Sri Lanka, levels up to 185 and 963 µg/kg, respectively, the maximum tolerated level (MTL) of AFB1 in rice in Iran (5 µg/kg) and TAFs (30 µg/kg) have been detected (Mazaheri 2009). Cambodia rice was less contaminated than in other countries, both developed and developing countries. Park *et al.* 2006 and 2005 showed that aflatoxin content was reduced by 30% during cooking (Park and Kim 2006;Park, Lee, and Kim 2005).

Conclusion

Contamination of paddy rice by these mycotoxins was studied in two provinces (Prey Veng and Takeo) on milled rice samples collected in retail markets in Phnom Penh. Among 60 samples analysed, 58 samples (96.7%) were not contaminated with AFB1, TAFs, OTA, FB1 and FB2. By the way, one sample of new harvest paddy rice in Prey Veng was contaminated by FB1 and FB2 with the level of 0.8 µg/kg and 0.1 µg/kg, respectively; and one sample of milled rice samples was contaminated by AFB1 and TAFs with the same level of 0.75 µg/kg. However, the level of contamination was lower than the European Commission legislative limits in cereal products for direct human consumption. The major staple food in many Asian countries, including Cambodia, is rice; hence our results showed that Cambodia rice both paddy and milled rice had a good quality and was safe for consumption as a food; Cambodian rice was then good for local and international markets. By consequence, the weak quantities of mycotoxins resulted probably of good pre- and post-harvest practices of farmers. However, it was an initiative survey done only on rice so these types of analysis could be extended to other food in Cambodia. It will be necessary to continue this survey in order to confirm the safe quality of Cambodian rice.

Acknowledgement

The authors wanted to thank the French Government for its financial support for this FSP project and the capacity building of the collaboration between Royal University of Agriculture (RUA) and CIRAD UMR Qualisud. A special thanks to Mrs. Chhoun Monyroth and Mrs. Hong Chanvibol for their assistance during mycotoxins analysis in CIRAD.

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……....PART III

POSTERS ON WATER……...

Microbial Fuel Cells: Innovation in Sustainable Energy Production and Wastewater Treatment

LUCY LAHRITA

Sanata Dharma University, Indonesia

Problem identification

World population and welfare are increasing rapidly, leading to a high demand for energy, specifically low-cost and sustainable electricity. However, about 1.3 billion of global populations living in remote areas of developing world still suffer from chronic energy poverty, while over 2 billion lack of clean water, including in Indonesia. Proper water treatment is required to prevent them from illness/ death. Nevertheless, they do not have economic means to afford it.

Maximizing unused and less economic resources

One man's trash is another man's treasure. Wastewater from local landfills contains high organic pollutants with no economic value. It is a trove if we know how to harvest it for generating electricity using bacteria. Our team has intensified our research to search for practical and novel technology that could address energy scarcity and water pollution problems simultaneously.

Smart Solution

Microbial fuel cells (MFC) that address two major issues at the same time-wastewater treatment and sustainable energy production-fits the United Nations' current mission in developing affordable and clean energy. Producing electricity using MFC would add economic value to wastewater and represent an attempt to fight environmental pollution against limited fossil fuels. The interests in MFC stem from its ability to operate at all weather conditions and pressure using inexpensive bacteria *Geobacter sulphurreducens* that form biofilms onto electrodes surface. MFC could produce electricity directly from organic contaminants within the wastewater without consuming conventional energy resources. Unlike activated sludge, MFC is practical for wastewater treatment. As wastewater has high organic concentrations, MFC will generate massive energy potentials. Organic matter in the wastewater has chemical energy that can fuel MFC when its bioreactor releases electricity in electrons flow from anode to cathode in an external circuit, generating power as by-product.

Anode: $C_6H_{12}O_6 + 6\ H_2O \rightarrow 6\ CO_2 + 24\ H^+ + 24\ e^-$

Cathode: $6\ O_2 + 24\ H^+ + 24\ e^- \rightarrow 12\ H_2O$ +

$$C_6H_{12}O_6 + 6\ O_2 \xrightarrow{\text{MFC}} 6\ CO_2 + 6\ H_2O$$

The bacteria get food from waste for survival, while we get electricity for energy,representing mutual symbioses between human and micro-organisms for environmental protection.

Our studies demonstrated that MFC could generate 3 Watts/m2 of electrode surface area as power outputs, while its continuous flow reaches 15 Watts/m3 of wastewater flowing through it. If an MFC is installed at a water treatment plant (WTP) for 10,000 inhabitants, it can produce about 0.8 MW, sufficient to power 500 homes.

Outcomes: Efficient, liveable, and sustainable

Currently renewable energy market grows to US$ 12 billion, while water technology market is about US$ 390 billion. Moreover, around 125 billion m3 of wastewater is treated with operational costs of US$25 billion annually, while the energy consumption takes 30% of total costs of water industry operations. The MFC reduces treatment cost by generating electricity on-site to power plants' operations. MFC may be linked to municipal waste streams, providing a sustainable system for waste treatment and energy production. With MFC, cities could make sustainable progress without damaging the earth.

Studies of Electrode Materials to Enhance Current Generation and Cod/Sulphate Removal in Microbial Fuel Cells

KORAKOT SOMBATMANKHONG, SUMITTRA CHAROJROCHKUL

National Metal and Materials Technology Center, Thailand

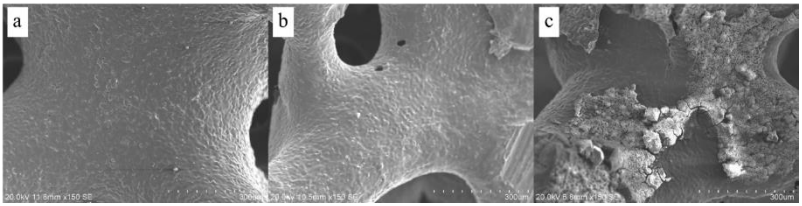
Abstract

Energy consumption has increased dramatically during the past half century due to the rapid increase in population and economic development. The majority of the energy needs have been met by combustion of fossil fuels leading to an increase in atmospheric carbon dioxide which is linked to the observed global warming trends. In addition to environmental concerns, a depletion of the world's limited fossil fuel reserves provides further motivation for the development of a novel technology which harnesses alternative sources of energy with no pollutant emissions. A microbial fuel cell (MFC) is considered to be both a promising sustainable energy source and a bioreactor which is fuelled by a priceless source of energy, wastewater. This not only helps cutting the energy costs but also accomplishes wastewater treatment simultaneously. Although up to 80% COD removal and Coulombic efficiency can be achieved in some cases, an amount of power generated by the MFCs is not sufficient to even power their pumping unit. In order to increase the level of power output, appropriate electrode materials should be employed in the MFCs because they are one of the key factors that help reducing the activation overpotentials. This work therefore focused on the studies of electrode materials for microbial immobilisation to improve current generation and COD/sulphate removal in MFCs.

The initial work investigated the electrochemical properties of various electrode materials to identify their suitability as electrode materials for MFCs which were primarily considered by their electrical conductivity and corrosion resistance along with the material cost and domestic availability. The electrical resistance was diagnosed using electrochemical impedance spectroscopy (EIS) whereas the corrosion rate per year was examined using both potentiometric and weight loss methods. Subsequently, the changes in surface morphology and elemental composition of those electrodes were studied using field emission scanning electron microscopy (FE-SEM) (see the representative FE-SEM images in Figure 1) and energy dispersive X-ray spectroscopy, respectively. All of the measurements were carried out in the environments which simulated conditions in the MFC. It was found that the ferric stainless steel (SS) coil type 409 exhibited the lowest electrical resistivity, in particular also with respect to the cost information; however, the corrosion rate was extremely high. Consequently, the austenitic SS type 316 wire mesh was employed in a MFC unit corresponding to the cost effectiveness and corrosion resistance. The MFC was built in house to develop an economical and commercially viable prototype. A continuous/upflow mode was performed without any requirements of mediator, noble metal catalysts nor proton exchange membrane. Instead, a mixed-culture biofilm immobilised on both anode and cathode could act as biocatalysts. A number of studies were conducted to investigate the effects of influent conditions (i.e. COD concentration and pH) and operating conditions (i.e. feed rate) on the MFC efficiency. These studies will provide parametric experimental data for an optimisation of the MFC operating conditions, resulting in an enhanced power output together with a great ability to accomplish wastewater treatment by the most economical MFC configuration.

Keywords: microbial fuel cells; wastewater treatment; microbial immobilization; electrode materials; biocatalysts.

Figure 1: The representative FE-SEM images of (a) the as-received austenitic SS type 316L foam and the austenitic SS type 316L foam immersed in (b) domestic wastewater and (c) 0.1M H_2SO_4 for 90 days



Risk Perception to the Decentralization of Water Management – A Case Study: in Rural-River Flooding Area of Eastern Indonesia

APOLONIA DIANA SHERLY DA COSTA, MSC

Institute of Social Geography, Faculty of Chemistry and Geoscience, Friederich Schiller University of Jena, Germany

Abstract

This study examines the risk perception to the decentralization of Water Management, A Case Study in Rural-River Flooding Area of Eastern Indonesia. The objectives of this study are to identify and analyze the risk perception to decentralization of water management, to analyze how does decentralization of water management influence risk perception of the community by concerning flood risk and to analyze do decentralization water systems support or achieve sustainable water for the households. The methods of this study are quantitative and qualitative analyses by surveying the 40 households who utilized water in two flood prone Villages are affected by the risk of river flooding in the downstream area of Benanain River. The result of this study shows that there are 32 huseholds who utilized water sourced from Benanain River to fulfill their daily neccesities. While, there are 8 households who have different perceptions. They apply the pattern of adjustment to the threat of flooding or suficient positively and how to use water as a basic need while keeping the sustainability of environment or sufficient, positively and effectively. The conclusions of this study are 32 households receiving government decentralization applied sufficient negatively, unsufficient positively, unsufficient negatively and sufficient positively and negatively. While, there are 8 households with different perceptions of using water showed a good pattern to blend with nature or sufficient positively and suficient, positively and effectively for a good sustainable. These findings have a good contribution to the development of community based on their risk perception to the decentralization of water management in rural-river flooding area in eastern part of Indonesia. Besides, by assessing the new formula or method, this study contributes a good new management system in multi-science scopes and multi-case scopes in every country in the world.

Hydraulic Model 1-2d Study of Forecasting, Warning Salt Water Intrusion in Downstream Area of Ma River System

NGUYEN THI HIEN

Asian Institute of Technology, Thailand

Abstract

Saltwater intrusion has become one of the biggest headaches in coastal areas. According to an annual survey in Vietnam, saltwater intrusion in the Central Part is gaining prevalent. Due to the effect of climate change and extreme events, saltwater intrusion has been one of the uncertainty and severe events which threatens agriculture and domestic over the years.

The study area is Ma river which is located in the North Central of Vietnam. It is situated on the geographic coordinates: North Latitude, East Longitude. Owing to the effects of rainfall and meteorology variables, the river flow is distributed irregularly while the change of salinity in space and time varies noticeably.

The objectives of the research are:

- Comprehensive study of hydraulic regime and effects of saltwater instrusion in Ma river downstream
- Modeling the process of saltwater instrusion and optimazation the parameters by using sensitive analyze
- Saltwater instrusion forecasting and warning scenarios

Methodology

From the observed data, salinity equation is applied to calculate salinity threshold (*1‰* and *4‰*) thereby have an overview of saltwater intrusion situation in Thanh Hoa Province over 15 years (Figure 4).

MIKE 11, MIKE 21 and MIKE FLOOD are applied to simulate the process of intrusion for the downstream area of Ma river. To specify:

- MIKE 11: simulate hydraulic regime (1D) in Ma river system (Figure 1)
- MIKE 21: simulate hydraulic regime (2D) in coastal zone with the first area is the Gulf of Tonkin and the second is only focused on 3 estuaries of Ma river system (Lach Truong, Lach Sung, Hoi estuary) (Figure 2, 3)

Figure 1

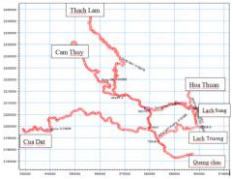


Figure 2

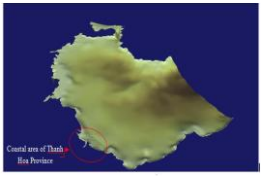


Figure 1

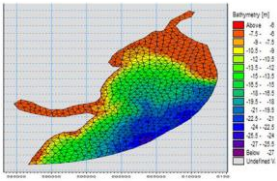
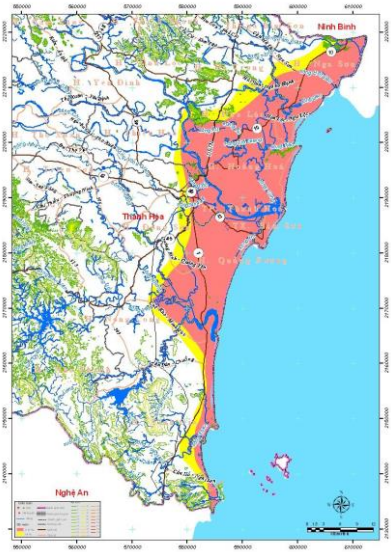


Figure 4: Maximum of 1‰ (yellow) and 4‰ saltwater intrusion threshold (observed 15 years)



– MIKEFLOOD: link river network (1D) and estuaries (2D) to simulate salinity process. Therefore, the forecasting and warning scenarios will be developed.

The research analyzes the sensitivity of parameters: *bed resistance – manning*, *advection - dispersion* which has direct effects on outcomes. After calibration process (observed data in 2010), the *bed resistance* (1D) and *advection - dispersion* as follow:

Table 1: Bed resistance – manning

River	Calibrate1	Calibrate2	Calibrate 3	Selected Value
Kenh De	0.035 - 0.06	0.03 - 0.06	0.025 - 0.05	0.025 - 0.05
Bao Van	0.035 - 0.06	0.03 - 0.05	0.02 - 0.04	0.02 - 0.04
Len	0.03 - 0.045	0.025 - 0.045	0.02 - 0.04	0.02 - 0.035
Buoi	0.045 - 0.05	0.045 - 0.045	0.04 - 0.045	0.04 - 0.045
Chu	0.03 - 0.06	0.025 - 0.06	0.025 - 0.05	0.025 - 0.05
Ma	0.03 - 0.055	0.25 - 0.05	0.02 - 0.04	0.02 - 0.04
Lach Truong	0.035 - 0.04	0.025 - 0.04	0.02 - 0.035	0.02 - 0.035

Table 2: Advection-dispersion parameters

Type	Coefficient	Initial	1st	2nd	3rd	4th	Selected Value
	Salinity	0-0.9	0-0.7	0-0.7	0-0.7	0-0.7	0-0.7
Dispersion (D)							
Single layer cohesive	Dispersion factor (a)	15	500	600	700	300	700
	Dispersion exponent (b)	0	5	5	5	5	5
	Minimum disp coeff.	0	500	400	250	300	250
	Maximum disp coeff.	50	800	900	850	1200	850
Multi layer cohesive							
Top layer	Dispersion factor (a)	15	500	450	550	300	800
	Exponent (b)	0	5	5	5	5	5
	Minimum disp coeff	0	500	400	150	300	150
	Maximum disp coeff	50	800	750	850	800	850
Bottom layer	Dispersion factor (a)	15	200	300	450	150	450
	Exponent (b)	0	5	4	4	4	4
	Minimum disp coeff	0	200	200	100	200	100
	Maximum disp coeff	50	700	650	650	600	650

Figure 4: Similarly with bed resistance – manning of MIKE21

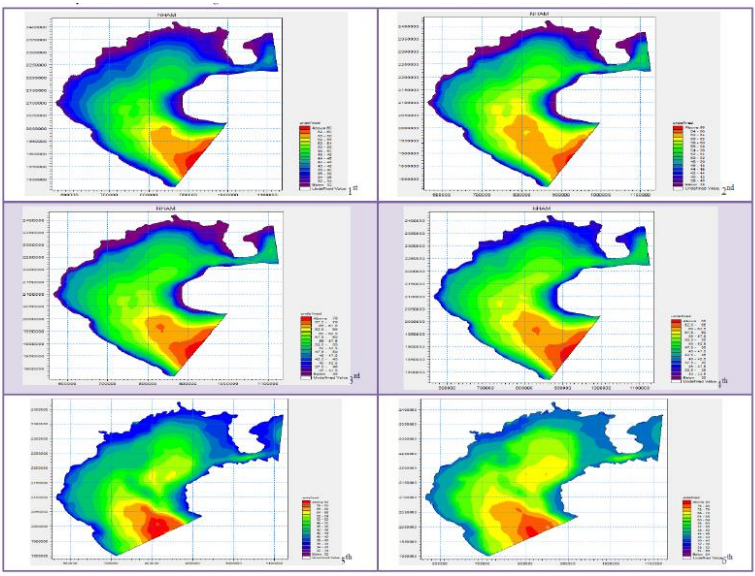
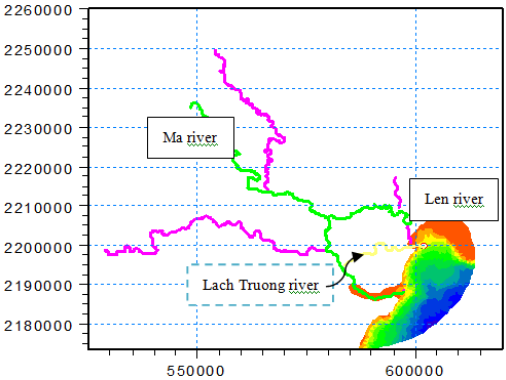


Figure 5: As a result, the research starts to calibrate and validate model for: 2003, 2005, 2009, 2011. Then, the next step is dynamic connection (MIKECOUPLE)



These paramaters are used for saltwater intrusion forecasting in 2012 (anticipated time: 24hours) and warning in the period of 2020 – 2100 (climate change scenarios) based on water demand and inflow (with different probability). Hence, presenting prospective scheme of saltwater intrusion for the next coming years in downstream area.

Water Management: to Utilize Water Through Management of Technology Dissemination

TIEN RAHMIATIN, S.IP, MPA

Ministry of Research, Technology and Higher Education, Indonesia

Abstract

Water is a vital element in human life. People cannot survive without water, because water is one of the pillars of life for humans. The availability of water in the world is so abundant, but that can be consumed by humans for the purpose of drinking water is very small.

Refer to the data, from the amount of water which is available in the world, only 5% are available as drinking water, while the rest is sea water. In addition, there is a tendency that the availability of clean water reduces every day. Based on data from World Health Organization, in Indonesia, it is predicted that around 60% of the river, especially in Sumatra, Java, Bali and Sulawesi are polluted by waste from organic materials and bacteria that cause diarrhea. According to the data from the Indonesia's Health Ministry in year 2002, there are 5.789 cases that cause of 94 deaths.

Deforestation and illegal logging that reduces the soil against water absorption participated also to add in reduced water intake. Besides, the unequal distribution of water to communities also contributes to this problem.

Various cases of environmental pollution and the deterioration of public health are the cases due to water waste from industrial activities, hospitals, markets, restaurants to households. To overcome these problems, we need a proper waste handling methods, targeted and sustainable.

According to the data, Indonesia is a country where is rich in water resources in which water availability reaches 15,500 cubic meter per capita per year which is still above the average of water availability in the world which is only 8,000 cubic meter per year. Nevertheless, Indonesia still has a shortage of clean water, approximately around 119 million people of Indonesia do not have access to clean water. As for people who have access, most of the clean water comes from the water distribution, the communities of water's business and water's wells. This condition is ironic considering that Indonesia includes in the 10 countries where rich source of fresh water is.

In some regions in Indonesia, there are the difficulities to access water because of environmental conditions around them. Environmental conditions were diverse, ranging from the structure of the soil in the form of limestone/coral so it is difficult to find a source of water, as well as the location of water sources that are difficult to be reached and far from residential areas. It is supported by the data from National Development Planning Agency, Republic of Indonesia that shows, more than 100 million people in Indonesia do not have access to clean water which is safe to be drunk. This is due to the unavailability of adequate facilities, in addition there is the low water supply budget priorities from the government.

Some cases are often found in the water supply are due to the difficulties of removal of water from the water source to the user. The process of removal of water from using the pump source needs a source of energy. Generally, the location of water source is far from residential areas and also far from the center of electricity. Therefore, the water pump with solar power is considered to be one of the ideal solutions to overcome some of the problems above.

Other water management system is how to process the water waste from Tofu product/industry to produce biogas. Tofu is one type of food source of protein with

soy-based material that is highly favored by the people in Indonesia. Most of its products in Indonesia are produced by small-scale industries that are mostly located on the island of Java. The industry is growing rapidly in line with the increase in population. On the other hand, this industry produces water waste which will pollute the environment. Industry requires water for processing, namely for process of sorting, reduction, skin peeling, washing, grinding, boiling and filtering.

Some programs of technology dissemination are conducted through some programs of incentive and technology specification program which are started by the identification of problem and attempt to propose the solution. Technology dissemination is covered by Regional Innovation System. The management program involves the capacity building program, strengthen the role of actor in coordination systems: research and development institution, universities, regional government, small and medium enterprises, and communities. Community's empowerment is a part of the goal as well.

……...PART IV

POSTERS ON FOOD……...

Bacillus spp., Plant-Derived Lactic Acid Bacteria and Shiitake Extract as a Novel Promising Biological Prevention Strategies for Mycotoxins

AWANWEE PETCHKONGKAEW[1], WONNOP VISESSANGUAN[2]

[1] Department of Food Science and Technology, Faculty of Science and Technology, Thammasat University, Thailand
[2] Food Biotechnology Research Unit, National Center for Genetic Engineering and Biotechnology (BIOTEC), National Science and Technology Development Agency, Thailand.

Abstract

Mycotoxin is a toxic secondary metabolites mostly produced by three genera of filamentous fungi (*Aspergillus* spp., *Penicillium* spp. and *Fusarium* spp.). It can contaminate various agricultural commodities either before harvest or under post-harvest conditions (FAO, 1991). The worldwide public health importance mycotoxins are: aflatoxins, ochratoxins, fumonisins, zearalenone, and trichothecenes (FAO, 2001). Once, mycotoxins contaminate into food and feed, they also are relatively stable to cooking and processing temperature, therefore, food preparation procedures cannot be expected to remove mycotoxins safely. Several mycotoxins in agricultural products and food cause health hazards to people and animals. Acute and even lethal poisoning by mycotoxins occasionally occurs; major concerns regarding mycotoxins exposure today are long-term effects of chronic exposure such as distortion of hormone balance, suppression of the immune system, and the ability of certain mycotoxins to cause cancer. Nowadays, methods for mycotoxins detoxification/degradation have been received increasing interest from both scientific communities and industries. Interestingly, the biological method provides and offers an attractive alternative for eliminating these mycotoxins. The extracellular fraction *Bacillus subtilis* IRU-1A showed aflatoxin B_1-degrading activity. The maximum activity condition was at 50°C and pH 8.0. The activity was stable in a wide range of pH (5.0-8.0) and temperature (25-60°C). The proposed aflatoxin B_1-degrading mechanisms of this strain may be possibly involved by a group of enzyme because the aflatoxin B_1-degrading activity was strongly inhibited by 1 mM sodium azide, 1 mM dithiothreitol (DTT) and 1 mM hydroxylamine. This extracellular fraction was not toxic at IC_{50} 4 mg/ml and it can be combined with water as a soaking agent for maize, which resulted in 54% of aflatoxin B_1 reduction of 120 min contact time. *Lactobacillus plantarum* BCC47723, a plant-derived lactic acid bacteria, was isolated from wild spider flower pickle)Pag-sian-dorng) had the highest zearalenone removal ability among thirty three isolates. The cell wall modification of viable cell of *Lactobacillus* spp. NB406 by heat, acid hydrolysis, and chemical substances were significantly enhanced the zearalenone binding ability (from 23% up to 32%). Moreover, the binding ability of inactivated cells was related to pH, temperature, incubation periods, and bacteria concentration. The maximum activity was at 70°C for 3 h, pH 5.0)bacteria concentration = 0.008 g ml^{-1}(. The activity was stable in a wide range of pH (3.0-8.0) and temperature (20-40°C). Shiitake (*Lentinus edodes*) extract (chloroform fraction) showed the preferential inhibition activity toward four fusariotoxin-producing filamentous fungi (*Fusarium graminearum* 1895, *F. verticilioides* 1641, *F. oxysporum* BCC 4977 and *F. moniliforme*). Surprisingly, the extract can also significantly inhibited the fumonisin B_1 production of F. *verticilioides* 1641 (50% decreased) since the seventh day of it growth (*in vitro* study).

Hence, the extracellular fraction of *Bacillus subtilis* IRU-1A, inactivated cell of *Lactobacillus plantarum* BCC47723 and Shiitake extract (chloroform fraction) might be further applied and also probably used to reduce the mycotoxins contamination in other foods and feeds commodities.

Keywords: mycotoxins; *Bacillus* spp. IRU-1A; *Lactobacillus plantarum*; Shiitake extract; Aflatoxin B_1; Zearalenone; Fumonisin B_1; *Fusarium* spp.

Bioactive Compounds, Human Health and Anti-Mycotoxin: Perspectives from Gac Fruit (Momordica Cochinchinensis Spreng)

TUYEN CHAN KHA

Faculty of Food Science and Technology, Nong Lam Unviersity, Vietnam

Abstract

Gac fruit, *Momordica cochinchinensis* Spreng, also known as "super fruit" or baby jackfruit or sweet gourd, is one of the traditional fruits in Vietnam. The fruit contains extraordinarily high levels of β-carotene, lycopene and lutein, a significant amount of unsaturated fatty acids, vitamin E, polyphenol compounds and flavonoids. Other potential medicinal compounds are reported to be present, including in the seeds. These nutrients have proved to be beneficial to human. However, Gac fruit is still underutilised, mainly used only in fresh form for local consumption in South East Asian countries. Therefore, it is desirable to extract and isolate the valuable active compounds, especially carotenoids for prevention and/or treatment of several diseases and mycotoxins in foods. It is generally agreed that natural bioactive components in plant extracts play an important role in human health. Therefore, it is interested in to test possible bioactivities and antioxidant activities related to carotenoids within a crude or pure Gac extract in order to allow further development of the compounds for use as nutraceuticals and/or pharmaceuticals for the treatment or prevention of numerous diseases.

This paper presents our recent publications on the bioactive compounds in Gac fruit, extraction and isolation of bioactives using several novel technologies, encapsulation and utilisation of Gac products for human health. Furthermore, the different evaluation techniques *in vitro* and *in vivo* for bioactivities and antioxidant activities of crude Gac extracts or isolated/purified carotenoids, and encapsulated bioactive compounds are proposed. Finally, the Gac extracts and their products rich in carotenoids possessed the potential for the discovery of new prevention strategy of mycotoxins are also highlighed, thereby warranting further investigations into their anti-mycotoxin properties. Therefore, more value can be added for the expansion of the Gac fruit industry, and importantly, novel prevention of several diseases and anti-mycotoxin can be identified.

Keywords: Gac fruit; carotenoids; bioactivity; antioxidant; anti-mycotoxin; extraction; encapsulation.

Preliminary Study of Plants Growing on Metalliferous Areas by Using Metabolite Profiling Approach

THI KIEU OANH NGUYEN[1], TIEN DAT NGUYEN[2], HOANG NAM PHAM[3], SERGE MICHALET[3]; SYLVIE NAZARET[3], MARIE-GENEVIÈVE DIJOUX-FRANCA[3]

[1] University of Science and Technology of Hanoi, Vietnam
[2] Institute of Marine Biochemistry, Vietnam Academy of Science and Technology, Vietnam
[3] Centre d'Etudes des Substances Naturelles, France

Abstract

Nowadays, an increasing number of microbes broadly distributed in the environment have involved in human and animals diseases, including opportunistic pathogens as etiologic agents of nosocomial infections. These latter microorganisms take advantage of the failure in the immune system of vulnerable people, as immunocompromised individuals to express their pathogenicity. Furthermore, several infectious agents become more and more resistant to the available biocides such as antibiotics, antifungal agents or antiparasitic agents presenting multi-drug resistance (MDR) phenotypes, which are mainly explained by the overexpression of MDR efflux pumps. Since the initial discovery of bacterial efflux pumps in the 1980s, many have been characterized in opportunistic bacterial pathogens with the capacity of extrusion structurally diverse compounds, leading to the therapeutically ineffective approaches. Anthropization largely contributes to the emergence and transmission of resistance genes in the environment, and highly contaminated soils and waters could be seen as hot spots for the expression of MDR phenotypes.

In recent years, public concerns relating to ecological threats caused by heavy metal have led to increasing attention paid for the research of new economical plants based on remediation technologies. Heavy metals are natural components of the Earth's crust, albeit in many ecosystems the concentration of several heavy metals reaching toxic levels due to the consequence of anthropogenic activities. Almost plants are significantly sensitive to metal ions, even at very low concentrations; however, certain plants have not only able to tolerate heavy metals but have also showed the ability defense against herbivore and pathogens due to the change of metabolism. The metabolite may play an important part on the expression or the dispersion of MDR phenotypes, and thus influence bacterial growth, especially rhizosphere bacteria. Since plant and rhizosphere share mutually symbiotic relationships, plant may have developed specific strategies to inhibit or cope with these mechanisms, thus representing a reservoir of the potential bioactive compounds, including antimicrobial agents. In this work, we aim to identify metabolites from hyperaccumulator plants active against MDR bacteria, and more specifically on pathogenic strains found in the environment, or which could be able to modulate the resistance properties of these MDR strains, and synthesize derivatives of the lead compounds selected, opening a new road in the field of drug discovery. In Vietnam, heavy metal pollution is increasing at the alarming rate caused by the massive development of the industry and mining. Heavy metals removal from contaminated areas by using phytoremediation, i.e hyperaccumulator plants, has been considered as an environmentally friendly, cost-effective and simple implementation solution.

To evaluate the impact of the polluted areas to an increase of MDR microorganism and the emergence and spread of new antimicrobial resistance mechanisms, we

used in the present study the plants selected on the basis of their contents in heavy metal and their soil-decontamination ability, growing in the highly heavy metal polluted environment in Thai Nguyen Province in Vietnam. We present here the first results obtained about the specific identification and quantification of the over-expressed metabolites derived from the plant growing in the polluted areas. Metabolite profiling of root extracts of the studied plants from the polluted surface were analyzed, using standard protocols for extraction, purification and characterization of natural compounds in combination with the statistical analysis. Parallel, the plants growing in the non-heavy metal polluted conditions were used as the control.

Keywords: *Pteris vitattae*; hyperaccumulator; multi drug resistance; heavy metals.

Free Range – A Vertical Chicken Farm Model in The City That Integrates and Showcases Energy Resource Systems

TAN ZI HUA

National University of Singapore, Singapore

Abstract

My study addresses Singapore's low food resilience problem due to a high reliance on external agricultural import. The issue is compounded by rising population and high food wastage in recent years. It aims to cultivate food consciousness and subsequently, inspire resource responsibility and initiative among the local community, by proposing a place for work-lifestyle immersion. As an anti-thesis to factory-like mass-food production, this is a place where users can be educated about the pleasures of growing and eating freshly-harvested organic food from an eco-centric, ethical farm.

The local project attempts to befit the title of a "Chicken Rice Capital of the World". It focuses on cultivating free-range chickens in a high-technology farm, supplemented by fish and vegetables grown by Aquaponics as feed for both chickens and humans. Sited near the city centre in a highly urbanized country, the strategy undertaken is to intensify land vertically and functionally, so it becomes an open roaming space for cultivating 3704 chickens per month and also function as a work-recreational space for willing participants from the community. The city farm will serve as a design prototype for showcasing and integrating resource systems, allowing for conventionally unthinkable free-range production to be seen as a possibility so that its limits can be assessed for future agricultural design ventures.

This poster will present on how the social and geographical dynamics of the country have resulted in the author's unique position of sustainability and food security, which condenses into the intention to educate citizens on resource production and usage. As energy can be saved by a good building design, and more energy can be actively produced to sustain a healthy food farm, the integrated use of various energy resources like rainwater, solar, biomass, human energy will be detailed in an architectural design proposal of the farm. The norm is to hide away the mentioned resources in auxiliary spaces that are usually reserved for utilities, but in my proposal, they become attractive showcases in the proposed building to counter a more fundamental problem for local food issues. A resource type is singled out to focus on-- rainwater harvesting, and its resulting water-sensitive approach to manage the entire food farm. Finally, a qualitative and quantitative evaluation will be shown to make known the limitations of such farm in local context, and also the infinite possibilities it holds globally, and for the future.

Sample images on next page

Keywords: free-range chicken; aquaponics; healthy food awareness; resource responsibility; vertical hi-tech farm.

Figure 1

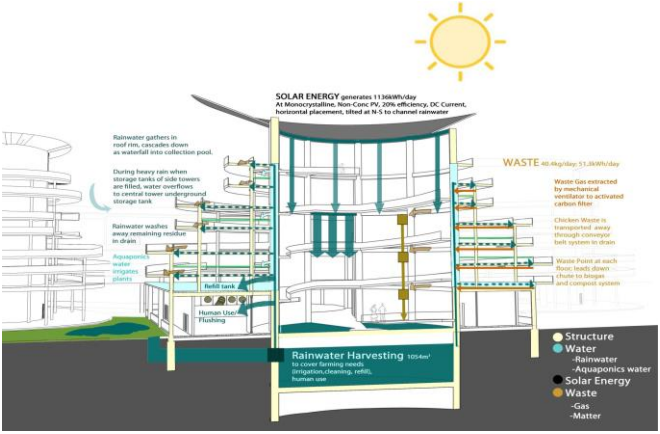
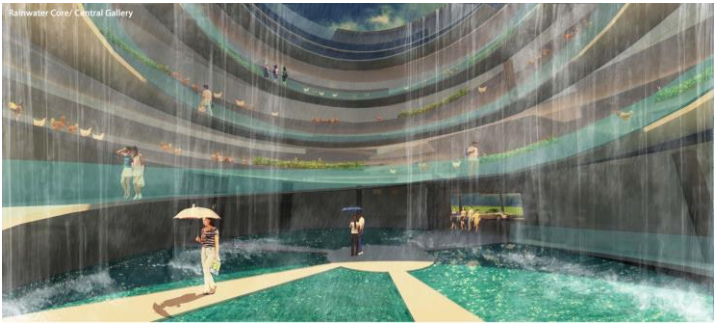


Figure 2

Figure 3



Effects of Fish Gelatin and Chitosan Coating on Quality Attributes and Nanostructural Changes of Fish Fillet

XIAO FENG[1, 2], HONGSHUN YANG[1, 2]

[1] Food Science and Technology Programme, Department of Chemistry, National University of Singapore, Singapore
[2] National University of Singapore (Suzhou) Research Institute, China

Abstract

Fish gelatin incorporating chitosan was applied as edible coating in fish fillet during cold storage to extend fillet's shelf life. Five edible coating groups including control, 0.4% (w/w) chitosan, 0.4% chitosan with 3.6% gelatin (w/w), 0.4% chitosan with 5.4% gelatin, and 0.4% chitosan with 7.2% gelatin were applied in fish fillet that was manually produced from farm-raised golden pomfret. Physicochemical properties including weight loss, pH, colour, electrical conductivity and total volatile basic nitrogen (TVB-N) were measured. The results revealed that the pH of control group increased significantly from 5.9 to 7.9 from day 0 to day 17 during 4ºC storage. On contrast, pH of gelatin/chitosan coated groups remained stable (approximately 6.1) at day 17. The TVB-N of control group increased from 3.59 to 93.52 mg/ 100 g, while for gelatin/chitosan coated groups TVB-N was below 13.56 mg/100 g at day 17, suggesting a preservation effect of antimicrobial coating. The pH and TVB-N of the control group increased since the microbes and endogenous enzyme might degrade the amino acid to ammonia, monoethylamine, dimethylamine, trimethylamine, and other volatile bases, leading to the unpleasant smell of the spoiled fish. The mechanism of the antimicrobial coating is that the positive charge of amino group of chitosan may interact with negative charge of protein on cell membrane of microbes to change the permeability of cell membrane, which may lead to microbe autolysis and inhibit the reproduction and metabolism of microorganisms. The microbial result was consistent with pH and TVB-N results. It was shown that the developed edible coating exhibited potent effect to suppress the growth of microbes, especially for bacteria. The total viable count (TVC) of control group increased dramatically to 5.99 log CFU/g at day 17, but the TVC for the four coating treated groups remained around 4 log CFU/g during the 17 day storage. The developed coating also showed effects on inhibiting the growth of yeasts and molds.

Interestingly, the weight losses of gelatin coated groups were dramatically less than those of control and 0.4% chitosan coated groups, which decreased the nutrient loss and preserved the fillet quality. Qualitative and quantitative nanostructural analysis of myofibril of fish fillet via atomic force microscopy provided evidence of delayed degradation of myofibril in chitosan/gelatin coated groups. It was found that the length of the control group decreased from greater than 15 μm at day 0 to 5.03 μm at day 17 during cold storage. While, the chitosan with 7.2% gelatin coating showed the best preservation on the length of myofibril, which was still greater than 15 μm at day 17. MALDI-TOF Mass Spectrometry results showed more compounds of greater molecular weight in chitosan/gelatin coated groups than control group. Overall, these results demonstrated that edible coating of chitosan combined with fish gelatin can delay the quality deterioration and nanostructural degradation of fish fillet during cold storage, and the developed coatings have significant effect to suppress the growth of microbes to preserve the fillet quality.

Microbiological Methods in Control of Fumonisin Mycotoxins

HUU ANH DANG

PhD student in Animal Sciences, Kaposvár University, Hungary

Abstract

The fumonisins are a group of mycotoxins produced mostly by several *Fusarium* species which are found mainly in maize and its products all over the world. These mycotoxins were proven that they can cause adverse effects on animal and human health such as porcine pulmonary, equine leucoencephalomalacia and human esophageal cancer. The physical or chemical methods have been applied to reduce toxicity of fumonisins. However it seems to be difficult to ensure the safety for animal and human health as well as apply in the process industry. Therefore, microbiological method was thought as the biocontrol of fumonisins production. Some impacts of microorganisms from various samples on *Fusarium* mycotoxins and fumonisins were demonstrated in the previous researches. This document represents some results of effect of microorganisms on fumonisins products and suggests using microbiological method to decrease fumonisin mycotoxins in food and feeding stuffs. The metabolism of fumonisin by gastrointestinal microbes was proven in *in vitro* experiments. The role of ruminal microflora from cow in the depletion of fumonisins B_1 (FB_1) was studied, FB_1 concentration was reduced by 18% after 72 h incubation (Caloni, Spotti, Auerbach, Camp, & Gremmels, 2000). After incubation of pig caecal content with FB_1 (72 hours), it was found that FB_1 (5 µg/ml) had been hydrolysis progressed, the molar concentration of partially hydrolyzed toxin was nearly equal with that of intact FB_1, while less that 1% was converted to aminopentol (Fodor et al., 2007).

FB_1 production can be inhibited by some bacterial strains. *Lactobacillus paracasie subsp. paracasie* can inhibit FB_1 production in 10-day incubation and the concentration of FB_1 was decreased by *Lactobacillus subsp. paracasie* after 20-day incubation (70.5 µl/ml compared with 300 µl/ml FB_1 in control group) (Gomah and Zohri, 2014). Some isolated rhizobacteria strains were also demonstrated to have biological effects on FB_1 produced by *Fusarium verticillioides*. In these bacterial groups, *Pseudomonas solanacearum* can inhibit FB_1 production completely and *Bacillus subtilis* also strongly inhibited FB_1 production in range from 70% to 100% (Cavaglieri, Passone, & Etcheverry, 2004).

Furthermore, FB_1 was hydrolyzed and deaminated by a bacterial strain isolated from soil after 3 hour period of incubation. There was a close phylogenetic relationship between this bacterium and the *Delftia acidovorans* or *Comanonas* group (Benedetti, Nazzi, Locci, & Firrao, 2006). *Streptococcus* and *Enterococcus* also have significant effect on FB_1 and FB_2, binding up to 24 and 62%, respectively (Niderkorn et al. 2007). In general, some bacterial strains which can metabolise, inhibit and degradate fumonisin mycotoxins were found. Therefore, the strategy of using microorganisms for control exposure of fumonisins can be established as a safe and effective method.

……....PART V

POSTER ON HEALTH……...

Genotyping of Multidrug-Resistant Streptococcus Pneumoniae Isolated from Indonesia

DODI SAFARI, PHD

Research Fellow Eijkman Institute for Molecular Biology, Indonesia

Abstract

Streptococcus pneumoniae is a leading cause of bacterial pneumonia, meningitis, and sepsis worldwide. Currently, epidemiological data on *S. pneumoniae* carriage and invasive disease is limited for the Indonesian population. Recently, we reported *S. pneumoniae* carriage was 46% in HIV-infected children in Jakarta, Indonesia with serotype 19F was most common isolates. We also found resistance to tetracycline and sulphamethoxazole/ trimethoprim were most common among isolates. In this present study, we investigated genotyping of multidrugs resistant *S. pneumoniae* isolated from Indonesia by multilocus sequence typing (MLST). We identified 10 of 42 isolates expressing lack of susceptibility to three or more of antimicrobial agents of different classes thus considered multi-drug resistant (MDR). Seven out of these ten MDR strains were either of pandemic Taiwan19F-14 MLST sequence type 236 (ST236, n=3) or single (ST271, n=1), or double (ST320, n=3) loci variants of it. Six out of these seven isolates were of serotype 19F. MDR strain of serotype 23F was a single locus variant of ST81 of pandemic Spain 23F-1 strain. In conclusion, the majority of multi-drug resistant strains identified in this study belonged to the Taiwan19F-14 *S. pneumoniae* epidemic clone.

The publication „Excellent Science in ASEAN – Best selected papers and posters from young ASEAN scientists on Water, Food and Health" is a result of a paper and poster competition, closely linked to the ASEAN-EU STI Days 2015. The main aim of this call for papers was to provide a possibility for young ASEAN researchers to publish and to introduce their scientific research to a wide European audience. The authors of the three best papers and two best posters were invited to Paris to take part and present their fields of research to the participants of the event. Further highly evaluated papers and poster abstracts on Food, Water and Health are to be read in this conference paper.

RCISD
Regional Centre for Information and Scientific Development
Hűvösvölgyi út 54. V/1.,
H-1021 Budapest
www.rcisd.eu

The SEA-EU-NET 2 project is funded under the 7th Framework Programme for RTD under the Capacities Programme — International Cooperation. Grant agreement no.: 311784

WWW.SEA-EU.NET

ISBN: 978-963-12-1839-8