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“Study on Medicinal Chemical Compound and Toxicity of Wanet(*Bambusa vulgaris* Schrad. Ex J.C, Wendt.) and Kyakhat-wa(*Bambusa bambos* Munro) Species”



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Abstract

The second most useful and reliable non-wood plant in Myanmar is bamboo. There are 21 genera and 102 species of bamboo in Myanmar. (Kress et.al. 2003) and it is about 2170000 hectares (U Khin Maung Sint, 2008). Bamboo is a fast-growing plant and can growing up to 60 cm in height per day (Dinesh Bhonde et.al. January-2014). Therefore, it is being used all over the country as a substitute for wood to solve the problem of shortage of wood. In addition, medical uses are also widely found. Thus, it is very important to study and present the various uses of bamboo. In this research paper, the kinds and rates of bio-beneficial organic compounds and its toxic substances contained in **Wanet (*Bambusa vulgaris* Schrad. Ex J.C, Wendt.) and Kyakhat-wa (*BambusabambosMunro*)**, which grow abundantly in Myanmar, will be study and present by analyzing the Phytochemical Test, Antimicrobial Activity, Antioxidant Activity and Cytotoxicity Activity of leaf and stem. Moreover, the medical properties and uses of Wa-net and Kya-khat-wa will be express in detail. Thus, the sample was collected from the Research Station No. (9), Mawbi, Yangon, Myanmar (17°08'26.4"N 96°00'05.1"E) in randomly. According to the test results (**phytochemical Test**), Alkaloids, Saponins, Tannins, Flavonoids, Glycosides, Steroids, Terpenes, Phenolic, Polyphenol, Lipophenol and Phytochemicals (10) have been at the extract of bamboo (Wanet and Kyakhat-Wa) leaf and stem. But the Reducing Sugar Phytochemical compound was found only in Wanet bamboo stem's extract and not found in Wanet leaves and in both of leaf and stem of Kyakhat-Wa extract. The significant result dealing with **antimicrobial**, the leaf of Wanet is strongly against to *Staphylococcus Aureus* that can cause Bloodstream infections, pneumonia, bone and joint infection and *Bacillus cereus* that can cause Brain abscess, cellulitis, endocarditis, Meningitis, Pneumonia, Osteomyelitis. In fact, the leaf of Kyakhat-wa extract is different with others, it has no strong capacity of against to *Staphylococcus Aureus*. In the **antioxidant activity**, there is high antioxidant activity in all extract except Wanet leaf extract. The evidence result is at Kyakhat-wa stem extract, it is show that the antioxidant activity is included almost testing of different concentration test sample. The **cytotoxicity activity** of Wanet leaf is 84.43 ± 8 mg/mL and the Wanet stem is 79 ± 25 mg/mL at the extract concentration of 1mg/mL. This mean that the cytotoxicity content of Wanet leaf and stem are much included. If under the standard control lysis is over 30%, it can assume that there is enough cytotoxic activity to the red blood cells. This mean of cytotoxicity content is that it can destroy some bacteria, the cancer cell and it can be used as medicine ingredient.

Keywords ; *Bambusa vulgaris* Schrad. Ex J.C, Wendt, *BambusabambosMunro*, Antioxidant Activity, Antimicrobial Activity

ဝါးနက်နှင့်ကြခတ်ဝါးတို့တွင်ပါဝင်သောဆေးဘက်ဝင်ဓါတုဒြပ်ပေါင်းများနှင့် အဆိပ်အတောက်များကိုဆန်းစစ်လေ့လာခြင်း

လွင်လွင်အောင်၊လက်ထောက်သုတေသနအရာရှိ
သစ်တောသုတေသနဌာန

စာတမ်းအကျဉ်း

မြန်မာနိုင်ငံတွင် ဒုတိယအသုံးဝင်ဆုံးနှင့် အားအထားရဆုံးသော အပင်မှာဝါးပင်ဖြစ်ပါသည်။ မြန်မာနိုင်ငံတွင် ဝါးမျိုးစု (၂၁) မျိုးနှင့် မျိုးစိတ် (၁၀၂) မျိုးရှိပါသည်(Kress et.al. 2003)။ အကျယ်အဝန်းအနေဖြင့် ၂၁၇၀၀၀၀ ဟက်တာခန့်ရှိသည်(UKhinMaungSint, 2008)။ ဝါးသည် ကြီးထွားမှုမြန်သော အပင်တစ်မျိုးဖြစ်ပြီး တစ်နေ့လျှင်အမြင့် ၆၀စင်တီမီတာထိ ကြီးထွားနိုင်ပါသည် (Dinesh Bhonde et.al. January-2014)။ သို့ဖြစ်ပါ၍ သစ်ရှားပါးမှုပြဿနာကို ဖြေရှင်းရန်အတွက် သစ်အစားထိုးအနေဖြင့် နိုင်ငံတဝှမ်းအသုံးပြုလျက်ရှိပါသည်။ ထို့အပြင် ဆေးဘက်ဝင် အသုံးပြုမှုများကိုလည်း တွင်ကျယ်စွာ အသုံးပြုလျက်ရှိပါသည်ကို တွေ့ရပါသည်။ ထို့ကြောင့် ဝါး၏အမျိုးမျိုးသော အသုံးဝင်မှုများကို လေ့လာဖော်ထုတ်တင်ပြရန်မှာ လွန်စွာ အရေးကြီးလှပါသည်။ ဤစာတမ်းတွင် မြန်မာနိုင်ငံတွင် ပေါများစွာပေါက်ရောက်လျက်ရှိသည့် ဝါးနက် နှင့် ကြခတ်ဝါး (ဝါးအလုံးနှင့်အရွက်)တို့အတွင်း အော်ဂဲနစ်ဒြပ်ပေါင်းများအား လေ့လာ ဖော်ထုတ်ရန်နှင့်၎င်းတို့၏ ဆေးဘက်ဝင်ဂုဏ်သတ္တိများကို အသေးစိတ်ဖော်ပြပေးပြီး ယင်းတို့၏ အသုံးပြုမှုများကို ကျယ်ကျယ်ပြန့်ပြန့် ပိုမိုသိရှိနိုင်စေရန်အတွက် ဆောင်ရွက်ခဲ့ခြင်း ဖြစ်ပါသည်။ သို့ဖြစ်ပါ၍ ရန်ကုန်မြို့၊ မော်ဘီသုတေသနစခန်း အမှတ်(၉)မှ စုဆောင်းထားသောဝါးနက် (*Bambusa vulgaris* Schrad. ex J.C, Wendt.)နှင့် ကြခတ်ဝါး (*Bambusa bambos* Munro) (၂)မျိုးမှဝါးအလုံးများအားကျပ်နစ်ဖြင့် ရွေးချယ်ခုတ်ယူစုဆောင်းခြင်းနှင့် အရွက်များခုတ် ယူစုဆောင်းရယူ၍ပါဝင်သော အော်ဂဲနစ်ဒြပ်ပေါင်း (phytochemical) အမျိုးအစားများ၊ အဏု ဇီဝပိုးမွှားများအားကာကွယ်နိုင်မှု (Antimicrobial activity)၊ ဓါတ်တိုးဆန့်ကျင်နိုင်စွမ်းဂုဏ် သတ္တိ(Antioxidant activity) နှင့် အဆိပ်အတောက်ပါဝင်မှု (Cytotoxicity Activity)စသည့် စမ်းသပ်မှု (၄)မျိုးဖြင့် Standard Method များအတိုင်း စမ်းသပ်ဆောင်ရွက်ခဲ့ပါသည်။ ဝါးနက် နှင့် ကြခတ်ဝါး(ဝါးအလုံးနှင့်ဝါးအရွက်)တို့တွင် အော်ဂဲနစ်ဒြပ်ပေါင်း(phytochemical) အမျိုးအစား များပါဝင်မှုရှိ/မရှိစမ်းသပ်ခဲ့ရာတွင် Alkaloids၊ Saponins၊ Tannins၊ Flavonoids၊ Glycosids၊ Steroids၊ Terpenoids၊ Phenolics၊ Polyphenols၊ Lipophenolsစသည့် ဆေးဘက်ဝင်ဓါတုဒြပ်ပေါင်း (၁၀)မျိုး အသီးသီးပါဝင်နေကြောင်း စမ်းသပ်တွေ့ရှိရပြီး (Reducing Sugar Phytochemical)

ဓါတုဒြပ်ပေါင်းမှာ ဝါးနက် (ဝါးအလုံး)တွင်သာ ပါဝင်မှုကိုတွေ့ရှိရပြီး ဝါးနက် (အရွက်)နှင့် ကြာခတ်ဝါး(ဝါးအလုံးနှင့်ဝါးအရွက်)တို့တွင်ပါဝင်မှုမရှိသည်ကိုတွေ့ရှိရပါသည်။ စမ်းသပ်တွေ့ရှိချက်များအရ ဝါးနက် (ဝါးအလုံးနှင့်အရွက်) extract နမူနာသည် ဆီးလမ်းကြောင်းပိုးဝင်ခြင်းများ၊ နမိုးနီးယား၊ အရိုးနှင့်အဆစ်များ ပိုးဝင်ခြင်းကို ဖြစ်စေသောပိုး (*Staphylococcus Aureus*) နှင့် ဦးနှောက်အမြှေးရောင်ခြင်း၊ တင်ပါးဆုံအဆီခဲတွင် ပိုးဝင်ခြင်း၊ အရေပြားပိုးဝင်ခြင်း၊ နှလုံးခန်းနှင့် နှလုံးပြွန်ရောင်ခြင်း၊ ကျောရိုးအရွတ်မျှင်နှင့် ဦးနှောက်သွေးမျှင်အမှေးပါးရောင်ခြင်း၊အရိုးတစ်ရှူးများရောင်ရမ်းခြင်းဖြစ်စေသောပိုး (*Bacillus cereus*) ကိုထိရောက်စွာကာကွယ်ပေးနိုင်သော ဂုဏ်သတ္တိရှိကြောင်းတွေ့ရှိရပြီး ကြာခတ်ဝါးအရွက် extract နမူနာမှာ (*Staphylococcus Aureus*)ပိုးအား ဆန့်ကျင်နိုင်စွမ်းမရှိပေ။ ဓါတ်တိုးဆန့်ကျင်နိုင်စွမ်းဂုဏ်သတ္တိ(Antioxidant activity)စမ်းသပ်မှုတွင် ဝါးနက်အရွက် extract နမူနာမှလွဲ၍ စမ်းသပ်မှုအားလုံးအပေါ်တွင် ဓါတ်တိုးဆန့်ကျင်နိုင်စွမ်းဂုဏ်သတ္တိ ရှိကြောင်းတွေ့ရှိရပါသည်။ ဖျော်ရည်ပြင်းအား 1mg/mL အသုံးပြုထားသော စမ်းသပ်မှုတွင် ဝါးနက်အရွက်၏ cytotoxicity သည် 84.43 ± 8 mg/mLဖြစ်ပြီးဝါးနက်အလုံး၏ cytotoxicity သည် 79 ± 25 mg/mL ဖြစ်သည်။ ဆိုလိုသည်မှာ ဝါးနက်အရွက်နှင့် အလုံးတွင် cytotoxicity များစွာပါဝင်နေကြောင်းတွေ့ရှိရပါသည်။ အဆိပ်အတောက်ပါဝင်မှုမှာ စံချိန်စံညွှန်းသတ်မှတ်ချက်အရ lysis တန်ဖိုးသည် ၃၀%အထက်ရှိပါကသွေးနီဥများကို ချေဖျက်ရန်လုံလောက်သော cytotoxic ပါရှိနေသည်ဟု ယူဆရပါသည်။ ထို့ကြောင့် ဝါးနက်အရွက်နှင့် ပင်စည်အတွင်း cytotoxic ပါဝင်နေခြင်းသည် အချို့သော ဘက်တီးရီးယားများနှင့် ကင်ဆာဆဲလ်များကို ဖျက်ဆီးနိုင်ပြီး ဆေးဝါးဖော်စပ်ရာတွင်ပါဝင်ပစ္စည်းအဖြစ်အသုံးပြုနိုင်ပါသည်။

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LIST OF ABBREVIATIONS

Hydrochloric Acid (HCl), Calcium (Ca)
 Ethanol Alcohol (EtOH), Copper (Cu)
 Sulfuric Acid (H₂SO₄), Iron (Fe)
 Argente (Ag), Mercury (Hg)
 Aluminum (Al), Potassium (K)
 Arsenic (As), Molybdenite (Mo)
 Gold (Au) Lead (Pb)
 Oxygen (O)
 Sulfides (S)
 Antimony (Sb)
 Silicate (Si)
 Zinc (Zn)
 2,2-diphenyl-1-picryl-hydrazyl-hydrate (DPPH)

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1. Introduction

Bamboo is one of the fastest growing woody plants in the world and it is also a potential source of bioactive substances. Bamboos are a group of woody grasses, which gives us a versatile material for different purposes. They belong to the subfamily Bambusoideae of the family Poaceae. There are more than 1250 species and 75 genera in the world. They are distributed in tropical, subtropical and temperate regions of the world, with a great abundance in the tropical Asia, Africa and Latin America, extending as far north as the southern United States or Central China, and as far south as Patagonia. They also grow in northern Australia (Flint, 2008). Bamboo is a fast-growing plant and can grow up to 60 cm in height per day (Dinesh Bhonde et.al. January-2014). The second most useful and reliable non-wood plant in Myanmar is bamboo. There are 21 genera and 102 species of bamboo in Myanmar. (Kress et.al. 2003) and it is about 2170000 hectares (U KhinMaungSint, 2008). The weather of Myanmar is very suitable for many kinds of bamboos the complex landscape with varying altitude. Bamboo is one of the natural resources of the tropics, and because of its wide distribution, availability, rapid growth, easy handling and desirable properties, it has been used widely in the daily life of the local community as a sustainable resource.

The rural people of Myanmar have been using bamboos as rafters and purlins, walling material, and flooring in building houses, in making agricultural implements, as cloth lines, for fencing, and for making various kitchen utensils, and furniture. Myanmar people make bamboo toys as present. Among the musical instruments coppers, pipe and xylophones are made of bamboo. Immature bamboos are used for tying material, and bamboo shoots for food. Today, the demand for bamboo is not limited to such a small scale, but extended from the rural small industrial sector to the large industries. As medicinal usage, the health benefits of bamboo include treating wounds and ulcers due to its antibacterial properties. Bamboo juice is particularly helpful for ulcers. Bamboo shoots are good for the stomach and can cure mild symptoms like indigestion and diarrhea. Studies have indicated that consuming bamboo shoots can reduce cholesterol and thus, ensure a healthier life (Raymond, 1982). Advantages of bamboo include its ability to control blood pressure due to the abundance of potassium. Potassium helps to lower blood pressure and keep the body healthy. A lot of cosmetic products integrate bamboo ingredients for facial and hair care. In addition, uses of bamboo plants are not only limited to dietary benefits. (Soltys,1963). Thus bamboo plays an important role in our daily life. Among them, about the kinds and rates of bio-beneficial organic compounds and toxicity of Wa-net and Kyakhat-wa, which grow abundantly in Myanmar, will be present and study in this research paper.

The present was analysis the two kinds of bamboo species especially leaf and stem of Wanet (*Bambusa vulgaris* Schrad. Ex J.C,Wendt.) and Kyakhat-wa (*Bambusabambos* Munro) to know their content of Phytochemical Compound, Antioxidant Activity, Antimicrobial Activity and the main one is the Toxicity Activity with the follow experiment methods. For the antioxidant activity; the DPPH radical scavenging assay testing method, for the Toxicity; Hemolysis Assay's way with the healthy person's Red Blood cell specimen and the Agar well diffusion method was used in Antimicrobial Activity analyzing by using 5 bacteria species. From the current research, the exception is to get the useful data for the medical field and to spread out the knowledge of utilization of bamboo in variety fields and

to promote the mind-set dealing with bamboo, is that bamboo is not only useful in construction field and food but also in others various field like medical fields.

2. Objective

- 1) To analysis the toxicity activity and the organic compound of bamboo (Wa-net and Kyakhat-wa) to know in detail.
- 2) Based on the systematically researched data, it aim to support to the related fields to develop in effective way of utilizing.
- 3) To promote the usage of bamboo in medical fields.
- 4) To spread out the knowledge dealing with bamboo utilizing without wasting.

3. Literature Review

To increase the self-sufficiency of developing countries, indigenous materials must be exploited to the full (Jules J.A. Janssen, 1998). Among them, bamboo is a familiar material with a long history of usefulness. The ever increasing demand for raw material due to the ever increasing population of the world, the increasing decline of productivity of forests, the imposing problems arising from destruction of forests, and the short production cycle and rapid growth of bamboos make a great interest in bamboo as a wood substitute. The diminishing wood resource and restrictions imposed on felling in natural forests, particularly in the tropics, have focused world attention on the need to identify a substitute material which should be renewable, environmentally friendly and widely available. In view of its rapid growth, a ready adaptability to most climatic and edaphic conditions and properties superior to most juvenile fast growing wood, bamboo emerges as a very suitable alternative (D L Jayanetti, et. al., 1998).

3.1. Overview of Bamboo in Myanmar

According to FAO estimates in 2014, Myanmar's land area was estimated at 65.3 million hectares, of which 29.6 million hectares or 45.3 % is forest area. (FAO Country Profile 2017). In another report (GFRA 2010 Country Report Myanmar), FAO claimed that 2.7% (or 859,000 hectares) of the total forest area are bamboo resources. Myanmar ranks fifth in the world in terms of bamboo forest area, with China as the major producer, followed by India, Indonesia and Lao People's Democratic Republic. As stated in the Book of Bamboo Species in Myanmar (2007), there are 118 identified bamboo species belonging to 18 Genera in Myanmar. In a related study conducted by Dransfield and Widjaja (1995) on the distribution of woody bamboo genera, there are two (2) genera of bamboo species found in Myanmar. These are Genus *Melocalamus* and *Pseudostachyum*. Related to this, a study conducted by the Business Innovation Facility Project entitled "Myanmar Bamboo Sector Competitiveness Study" (2015) indicated that Tanintharyi Region has some of the best bamboo natural forests. In the paper presented by the Myanmar Survey Research and Bamboo Products, Marketing and Extension Section in a forum jointly organized by ITTO and the Forest Department of Myanmar in 2006, the document showed that there are 48 identified major bamboo species habitat in Myanmar, and only three (3) bamboo species (*Bambusa arundinacea*, *B. binghami* and *Dendrocladistans*) are listed to be thriving in Tanintharyi Region. (Table 1).

No.	MyanmarName	ScientificName	Habitat/Locationin Myanmar
1	KyaKhatWa	<i>Bambusaarundinaceae</i>	Bago, Yoma and Tanintharyi Division
2	KyathaungWa	<i>Bambusapolymorpha</i>	Bago Yoma
3	KyarWa	<i>Bambusapallida</i>	Naga Mountain Region and Kachin State
4	NagaChatWa (WaKyap)	<i>Bambusabinghami</i>	Bago Yoma and Tanintharyi Division
5	PilawPinanWa	<i>Bambusa nana</i>	Lower Myanmar
6	SinthanaWa	<i>Bambusasinthana</i>	—
7	Ta PinTine Net/ ThaikWaGyi	<i>Bambusaburmanica</i>	—
8	Ta Pin Tine Wa	<i>Bambusalongispiculata</i>	Mogoke and Thanwin Valley
9	Thaik-wa	<i>Bambusatulda</i>	Bago, Sittaung and Dawei Region
10	Thaik-wagyi	<i>Bambusaburmanica</i>	—
11	ThaikWabo	<i>Bambusakingiana</i>	Kachin and Northern Shan State
12	Thar Law Wa	<i>Bambusatharlawwa</i>	—
13	WaByet	<i>Bambusaoffinis</i>	—
14	Wa Min	<i>Bambusathalawwa</i>	—
15	WaMyinn	<i>Bambusagriffithiana</i>	—
16	WaPhyuKalay	<i>Bambusaoliveriana</i>	Some Regions in Lower Myanmar
17	Tin-wa	<i>Cephalostachyumpergracile</i>	Bago Yoma and Upper Myanmar
18	Kyatwa	<i>Cephalostachyumburmanicum</i>	Various Parts of Myanmar
19	HmyinWa	<i>Dendrocalamusstrictus</i>	Central and Lower Regions of Myanmar
20	KyaloWa (Wa Bo)	<i>Dendrocalamusbrandissi</i>	Hilly Regions
21	WaGyi	<i>Dendrocalamuscalostacyus</i>	Kachin, Shan State and Upper Chindwin Region
22	Wa Net	<i>Dendrocalamuslongispathus</i>	Ayeyarwaddy and Yangon Region

23	WaPhyu	<i>Dendrocalamusmembrenaceus</i>	Various parts of Myanmar
24	WaPyaw	<i>Dendrocalamuslongifimbriatus</i>	–
25	Wabo Aye	<i>Dendrocalamushookeri</i>	Banmaw and Kathar Districts
26	Wabo Gyi	<i>Dendrocalamusgiganteus</i>	Shan State, Upper Chindwin and Some Parts of Lower Myanmar
27	Wabo-Myet-San-Gye	<i>Dendrocalamushamiltonii</i>	Upper Myanmar
28	Wani	<i>Dendrocalamuslatiflorus</i>	Hilly Regions
29	HtamyinWa	<i>Dendrochloadistans</i>	Myeik and Thanintharyi Regions
30	KamyinWa/Tamyin Wa	<i>Dendrochloadistans</i>	Thanintharyi Region
31	Sin Nin Wa/Wa New/ WaTha Put	<i>Dinochloamaclellandi</i>	–
32	Wanwe	<i>Dinochloamilellandi</i>	Some Regions in Lower Myanmar
33	Wa New Goat	<i>Dinochloacampactiflora</i>	Some Regions in Lower Myanmar
34	WaPhyuGyi	<i>Gigantochloamacrostachya</i>	–
35	Wa toe	<i>Gigantochloaapus</i>	–
36	KayinWa	<i>Melocanabambusoldestrin</i>	Rakhine Yoma and Upper Chindwin
37	Tha Put Wa	<i>Neohouseauastricta</i>	Kachin and Northern Shan State
38	Wa Soon	<i>Neohouseauadulloo</i>	Kachin and Northern Shan State
39	WaTha Put	<i>Neohouseauahelferi</i>	Nago Yoma and Lower Myanmar
40	KyaungWaAme'	<i>Oxythenantherathwaitesii</i>	–
41	ThaikTuMyinTuWa	<i>Oxythenantheraparviflora</i>	–
42	WaPyaut	<i>Oxythenantheranigrociliata</i>	Kachin State and Kathar District
43	Watoke	<i>Oxythenantheraalbociliata</i>	–
44	Wa Ni Par	<i>Pseudosorghumpolymorphum</i>	–
45	WaKhar	<i>Pseudostachyumwakha</i>	–
46	ThanutWa	<i>Thyrsostachysoliveri</i>	Kachin and Shan States
47	Htiyowa/HteeWa	<i>Thyrsostachyssiamensis</i>	Various Parts of Myanmar
48	WaKauk	–	-

Table 1. Source: The Botanical Name and Genus of Main Plants in Myanmar Agricultural Services6

3.2 Utilization of Bamboo

Bamboo is an important non-timber forest product material in rural communities as it has a wide range of uses. Among its major uses is for construction for almost all parts of the house, including fences and scaffolding. Bamboo is also used for agricultural implements such as bins, carry baskets, trays, sieves, crates, etc. for on-farm and off-farm use for arts and handicrafts, furniture, traditional toys, musical instruments and furniture chopsticks, charcoal and food. Bamboo has the potential to create livelihood opportunities for the rural communities and generate income along the chain of various production activities from plantation to marketing of finished bamboo products. In the food sector, bamboo shoots are becoming popular, and sold in raw form and as preserved and being exported to different countries. Bamboo has developed as a valuable substitute for wood over the past years. With new applications, industrial products have emerged as bamboo-based panels, parquet tiles, modern laminated furniture and floorboards.

3.3. Market Situation of Bamboo

Based on the study conducted by BIF (Myanmar Bamboo Sector Competitiveness Study, December 2015) the total bamboo market amounted to US\$ 1.369 billion with a significant increase of 20% from 2013 to 2014. As shown in Figure 4, Global Bamboo Exports 2007-2014, the overall performance dipped in 2009 due to the global financial crisis in 2007-2008. The largest market were woven bamboo products, which consists of bamboo basketwork, bamboo mats and screens, plaits and plaiting materials.

3.4. Potential Health Benefits of Bamboo Leaf Extracts

Traditional Chinese medicine distinguishes bamboo leaves into two kinds. (1) Kuzhuye (“Ku” means bitter, “Zhu” means bamboo, and “Ye” means leaf), dried leaves of *Pleioblastus amarus* (Keng) Keng f. “It is pungent, sweet, slightly bitter, and cold, and enters the heart and lung meridians. It is often used to treat febrile diseases when there is heat in the heart, lung, or chest” (Yang, 2002). (2) Danzhuye (“Dan” means bland), which are dried leaves and stem of *Lophatherum gracile* Brongn. “It is less strong in clearing heat than the bitter form, but is good at promoting urination, thereby leaching out the heat from the heart and the small intestine. In clinical practice, it is used to treat urinary dysfunction, which starts or worsens in stressful situations. It is also used to treat eczema due to damp heat” (Yang, 2002). According to Dr. S. Dharmananda, the leaves of the black bamboo [*Phyllostachys nigra* (Lodd.) Munro] are used similarly as “Danzhuye” in Japan.

3.5. About the *Phyllostachys edulis* (Carrière) J.Houz

This species of bamboo is known as “Maozhu” in Chinese or “Moso” in Japanese, and it is one of the largest and fastest-growing bamboo species in the world. Studies carried out in the author’s laboratory have shown that an ethanolic extract from the leaves of *Phyllostachys edulis* (Carrière) J.Houz contains high levels of polyphenols and flavonoids (Lin et al., 2008). This bamboo extract (termed as BEX) protects a variety of cells against lipotoxicity induced by high levels of palmitic acid (Panee et al., 2008). Specifically, BEX inhibits palmitic acid-induced overproduction of proinflammatory cytokines, such as interleukin 6 (IL-6) and monocyte chemo attractant protein 1 (MCP-1), and this anti-inflammatory effect is partially mediated by flavonoids such as tricin and 7-O-methyltricin (Higa et al., 2012) through NF- κ B and AP-1 pathway inhibition (Higa and Panee, 2011). In addition, BEX also seems to affect the central nervous system and cancer development. For example, BEX as a dietary supplement ameliorated anxiety-like neurobehaviors in mice fed high fat diet (Del Rosario et al., 2012), and decreased the incidence of DMBA-induced mammary tumors in rats (Lin et al., 2008).

Further protective effects have been reported on 3-O-caffeoyl-one-methylquinic acid, an antioxidant isolated from the leaves of *Phyllostachysedulis* (Carrière) J.Houz. This compound was found to upregulate the expression of heme oxygenase-1 (HO-1) and other Nrf2- dependent phase II detoxifying genes, and protect against peroxide-induced cytotoxicity (Kweon et al., 2004; Kweon et al., 2006). This compound also prevents photocarcinogenesis via apoptotic elimination of tumor protein 53 (p53) mutant and DNA-repair defective cells (Kweon et al., 2007).

Furthermore, it has been documented that a butanol-soluble extract from the leaves of *Phyllostachysedulis* (Carrière) J.Houz contains derivatives of chlorogenic acid and has antioxidant activity (Kweon et al., 2001). Essential oils of *Phyllostachysedulis* (Carrière) J.Houz extracted by steam distillation contain cis-3- hexenol and have antioxidant and antimicrobial activities (Jin et al., 2011).

3.6. About the *Sasasenananensis* (Franch. & Sav.)Rehder

This species is known as Kumaizasa bamboo, found in Hokkaido, and is used in traditional medicine in Japan. Between 1998 and 2004 several articles documented a wide range of biological functions of an alkaline extract made from the leaves of this species of bamboo (the product was named “Sasa Health”). When the diet of mice was supplemented with this extract, the incidence and growth of mammary tumors in mice with spontaneous or Her2/NeuN-induced mammary tumorigenesis was inhibited (Ren et al., 2004; Tsunoda et al., 1998), spontaneous motor activity of both intact and gonadectomized mice was influenced (Nagasawa and Hattori, 2001), and some of the deleterious effects of restricted feeding were alleviated (Nagasawa et al., 2001).

Some anti-tumor effects of a new hot water extract from Sasa leaves were published in 2010. This extract inhibited the incidence and growth of 7,12 -dimethylbenz[α]anthracene (DMBA)-induced mammary tumors and the growth of inoculated sarcoma and melanoma cells (Seki and Maeda, 2010). The anti-tumor efficacy of the extract was attributed to immunopotential. 1,3- β -glucan has been identified as the probable primary immune potentiating factor in this extract (Seki et al., 2010).

3.7. About the Ethanolic Extract from the Leaves of *Phyllostachysedulis* (Carrière)

J.Houz

It has been shown in the author’s laboratory that the aforementioned ethanolic extract from the leaves of *Phyllostachysedulis* (Carrière) J.Houz (BEX) has regulatory effects on phase I and phase II enzymes in mouse liver. For example, it upregulates the activities of cytochromes P450 (CYP) enzymes CYP1A2 and CYP3A11, and uridine diphosphate glucuronosyltransferase (UGT), and slightly downregulates the activity of glutathione-S-transferase (GST). In Obese/diabetic mice the activities of CYP1A2 and CYP3A11 are upregulated, and supplementation with BEX further up-regulates these phase I enzymes (Koide et al., 2011). Although these effects are not toxic per se, potential interactions between BEX and drug metabolism should be considered, especially in obese/diabetic subjects.

3.8. About the Abortifacient Effect of the Leaves of *Bambusa vulgaris*Schrad

In contrast to the aforementioned antioxidant-rich extract from the leaves of the *Phyllostachysnigra* (Lodd.) Munro, an extract from the leaves of *Bambusa vulgaris*Schrad is used to induce abortion in Nigerian folk medicine. An aqueous extract of *Bambusa vulgaris*Schrad leaves, containing alkaloids, tannins, phenolics, glycosides, saponins, flavonoids and anthraquinones, was found to dramatically increase abortion frequency and decrease fetal survival rate in pregnant rabbits at doses of 250–500 mg/kg body weight per

day (Yakubu and Bukoye, 2009). This dose was associated with increased restoration index and post implantation loss, decreased serum progesterone, follicle-stimulating and luteinizing hormones, and decreased alkaline phosphatase activity and glucose concentration in the uterus (Yakubu and Bukoye, 2009; Yakubu et al., 2009). Interestingly, leaves of *Bambusa vulgaris* Schrad (boiled or raw) are also used to relieve labor pains or as a postpartum cleanser for live stock (Lans et al., 2007).

3.9. Potential Health Benefits of Bamboo Shoots

In traditional Chinese medicine, bamboo shoots are used to ease labor and the expulsion of the placenta by inducing uterine contractions. A poultice of the shoots is often used for cleaning wounds and healing infections. Bamboo shoot decoction taken along with honey is used to treat respiratory disorders. However, to most people bamboo shoots are best known as food. Fresh, dried, or fermented bamboo shoots are used in numerous Asian recipes. Nutrient analysis on freshly emerging juvenile bamboo shoots has shown high contents of amino acids, proteins, carbohydrates, vitamins, and minerals, and a low content of fat. As bamboo shoots age, the dietary fiber and moisture start to increase while vitamin and mineral contents decrease (Nirmala et al., 2007).

Consumption of bamboo shoots (360 g per day for 6 days as a dietary fiber, the species of bamboo used in this study was not disclosed by the authors) reportedly decreased serum total cholesterol, low-density lipoprotein (LDL), and the atherogenic index, and increased fecal volume and bowel movement frequency in healthy young women when compared with controls on a dietary fiber-free diet (Park and Jhon, 2009).

Bamboo shoot oil extracted from *Phyllostachysedulis* (Carrière) J.Houz (250–1000 mg/kg body weight per day) has been documented to decrease the same aforementioned serum parameters (total cholesterol and LDL), as well as serum triacylglycerol, phytosterol, and lipoprotein lipase. This same oil extract ameliorated fatty liver and increased the cholesterol content in feces in rats challenged with a high fat diet. These lipid-lowering effects have been attributed to inhibition of cholesterol absorption and increase of cholesterol excretion (Lu et al., 2010).

A high level of acetylcholine, an important neurotransmitter in the cholinergic nervous systems of vertebrates and insects, has been found in the upper portion of bamboo shoots (Horiuchi et al., 2003). Nutritional and clinical applications of bamboo shoots based on the presence of this compound have not been explored.

3.10. Impact on Male Fertility of Bamboo Shoot

It has been reported that an ethanolic extract of the tender shoots of *Bambusa arundinacea* Willd (dose of 300 mg/kg body weight per day) changed the structural and functional integrity of the epididymis and reduced fertility of male rats. A 7-day treatment markedly reduced the count and motility of sperm (Manonayagi et al., 1989), and decreased the fertility index by 85 percent (Vanithakumari et al., 1989). Mating activity was significantly compromised after 4 days of treatment. After withdrawal of the treatment for approximately one week, although the mating activity completely recovered, the fertility index only increased by 8 percent (Vanithakumari et al., 1989). The serum profile of protein and oxaloacetic/pyruvic transaminase activity suggested that this extract had no systemic toxicity (Vanithakumari et al., 1989).

Consumption of bamboo shoots was positively associated with hyperuricemia – a risk factor for cardiovascular disease – in an epidemiological study in Taiwan (Chuang et al., 2011). In a study done in three hospitals in Thailand, dietary intake of bamboo shoots was associated with aggravation of dyspepsia (Premgamone et al., 2010).

Although arsenic is predominantly found in inorganic forms in most terrestrial plants, a significant amount of organic arsenic was found in shoots of *Phyllostachys edulis* Carrière) J. Houz despite an absence of such compounds in the soil, suggesting a possible arsenic methylation process in the shoots, especially in the core section (Zhao et al., 2006). The toxicological aspect of this finding has yet to be further explored.

3.11. Potential Health Benefits of Products Derived from Bamboo Stems

In traditional Chinese medicine, bamboo stems can be processed into varied forms as summarized below. (1) Zhuru (bamboo shavings), dried, long and thin slices shaved from the intermediate layer of the bamboo stalk after the outer skin is removed. It is “sweet and slightly cold”, and used for stomach heat syndromes that produce incorrect flow of qi, commonly causing nausea, loss of appetite, hiccups or vomiting (Yang, 2002). (2) Tianzhuhuang, also known as tabashir, is a dried resinous secretion with very high silica content (up to 85 percent silica) from the knots of certain female species of bamboo (Jones et al., 1966). It is “sweet and cold”, clears heat, resolves phlegm, relieves convulsion, especially used in remedies for children’s feverish disorders and epilepsy (Yang, 2002). (3) Zhuli, is the liquid sap obtained from the ends of freshly cut bamboo pieces with outer surface removed and exposed to heat. Zhuli is “colder” than Zhuru and Tianzhuhuang, enters the heart, lung, and stomach meridians. It has a lubricating nature, strongly eliminates phlegm-heat, and is used to treat epilepsy, schizophrenia, hemiplegia, facial paralysis, and numbness and tingling or cramp of the limbs (Yang, 2002). (4) Bamboo vinegar, a liquid condensed from the vapor generated by heating bamboo at very high temperature in an airless vessel (a process to make bamboo charcoal). It has high content of acetic acid, accompanied by phenols. Bamboo vinegar is largely produced in Japan and is used to treat varied skin diseases. Recent biomedical research has been mainly focused on the biological function of bamboo shavings, bamboo vinegar, and ethanolic extracts of bamboo culms, as summarized below.

3.12. About the Bamboo Shavings

Recent studies have been conducted on tri-terpenoid-rich extracts prepared using carbon dioxide supercritical fluid extraction from the shavings of different bamboo species.

An extract from the shavings of *Phyllostachys nigra* var. *henonis* (Mitford) Rendle (100–300 mg/kg body weight per day) decreased serum total cholesterol and total triglyceride levels in hyperlipidemic rats (Jiao et al., 2007). The same extract also reduced systolic pressure in rats with spontaneous hypertension, primarily by a vasodilatory effect (Jiao et al., 2007). *In vitro* results suggested that friedelin, a main triterpenoid compound in the extract, probably contributed to the vasodilator effect (Jiao et al., 2007).

An extract from the shavings of *Bambusa uldoides* Munro (40–250 mg/kg body weight/day) had an anti-fatigue effect in mice (i.e., it helped prolong the duration of weight-loaded swimming and climbing), also increased hepatic glycogen content, and decreased serum urea nitrogen and lactic acid levels (Zhang et al., 2006).

4. Material and Methods

4.1 Reagents and Chemical

All the chemicals, reagents, and solvents used in the assay protocols were of highest analytical grade. Methanol, Ethanol, Dimethylsulfide (DMSO), Sodium chloride (NaCl), Tripton, Ascorbic acid, Chloramphenicol were bought from Able (Teaching Aids), Mandalay.

4.2. Bamboo Sample Collecting and Preparing

For this research paper, the sample of Wanet and Kyakhat-wa leaf and stem was randomly collected from the Research Station No. (9), Mawbi, Yangon, Myanmar ($17^{\circ}08'26.4''\text{N}$ $96^{\circ}00'05.1''\text{E}$) for the tested. Firstly, the leaf and stems of bamboos that collected with suitable length was dried naturally under suitable sunlight at the Wood Preservation Section, Forest Research Institute, Yezin, NayPyi Taw.



Figure 1. Sample Collecting from Mawbi Research Station



Figure 2. Sample Preparing; Drying the Stem and Leaf of Wanet and Kyakhat-wa

And the next step of sample preparing is cutting the bamboo piece into little bamboo bar that is about 2-5 centimetre long, 1.5-3 centimetre wide. And the small pieces of bamboo stem and leaf was measured the weight and burst and pressed to be soften.



Figure 3. Sample Preparing; Cut the Dried Bamboo Stem and Leaf



Figure 4. Sample Preparing; Weighting the Cut Bamboo Leaf and Stem

4.3. Preparation of Bamboo Leaf and Stem Extracts

The extracting of Wanet and Kyakhat-wa leaf and stem for Phytochemical and Toxicity Activity Testing was processed at the Wood Preservation Section, Forest Research Institute (FRI), Yezin, NayPyi Taw. For extraction, Ethanol was used according to the normal standard solution. To make extraction the measured bamboo sample block was burst and it was got the fluid of bamboo extract by mixing with ethanol. That fluid extract was measured and filtered and extracting by mixing with n-hexane, ethyl acetate and acetone organic solvent for about 1 month. After making extracting, the extract sample was dried in room temperature about 3 months.



Figure 5. Making Extracting the Burst Bamboo Stem and Leaf with Ethanol



Figure 6. Filtering the Extraction Mixture Solution to Get Extract Liquid



Figure 7. Drying the Filtered Extract Fluid at Room Temperature about 3 Month



Figure 8. Extracting and Measuring the Weight of Extract and put in glass bottle

4.4. Preparing and Method of Phytochemical Analyzing

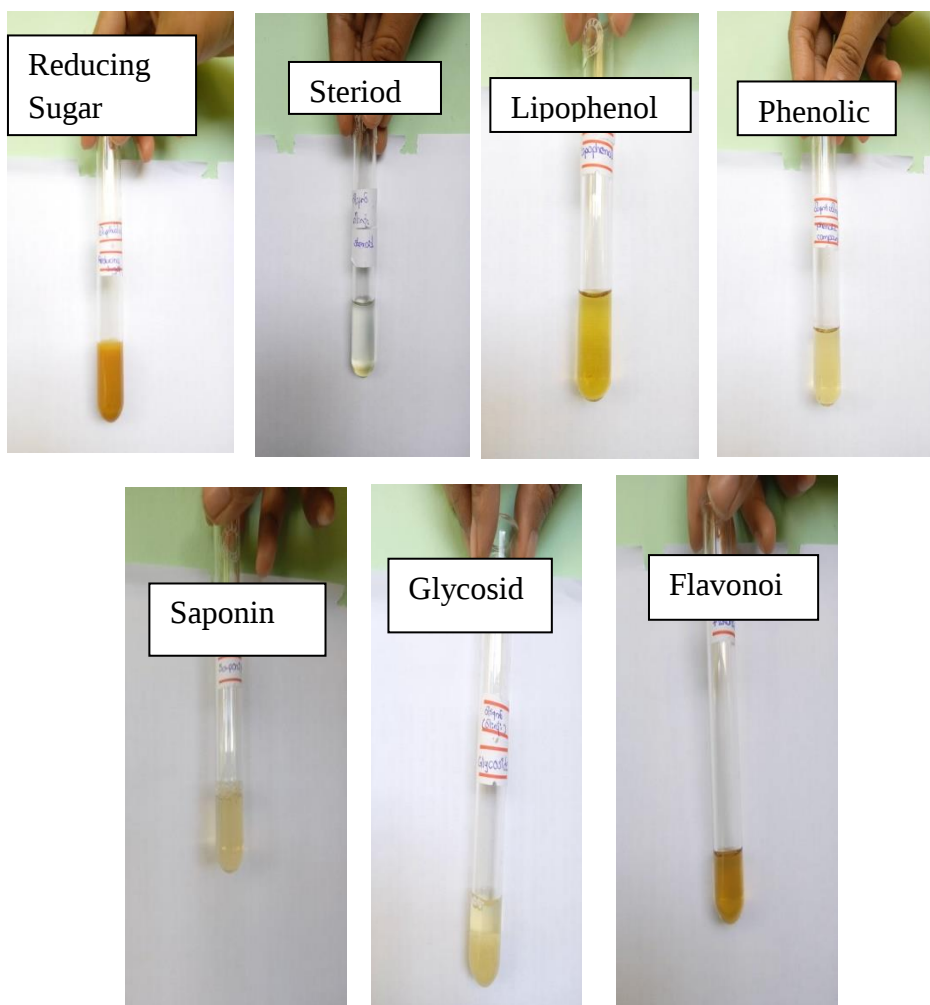
For this test, the required reagent was solving with the standard method and the experiment was followed by the Harborne JB method.





Figure 9. Making the reagent for the Phytochemical Analysing

According to the Harborne JB method, Alkaloids content was measured using Dragendroff's and Wagner's Reagent and the content of Saponins was measured by Distilled Water, Shake (sipping) test. Tannin content was determined by 10% FeCl_3 , (Dil.) H_2SO_4 reagent. Flavonoids content was measured with a small amount of magnesium and HCl reagent and then, 10% Lead acetate test material was used for Glycosides content measurement. Acetic Anhydride, (Conc.) H_2SO_4 (2 Drops), Chloroform + Acetic Anhydride + Conc. HCl (2 Drops) test materials was utilized for the test of content of Steroid and Terpene. The Phenolic content was determined using 10% FeCl_3 reagent. Polyphenol content was determined using 1% $\text{K}_3\text{Fe}(\text{CN})_6$, 1% FeCl_3 reagent, Lipophenol content was determined using 0.5 N KOH reagent. Reducing sugar content was also tested using Benedict's solution test kit.



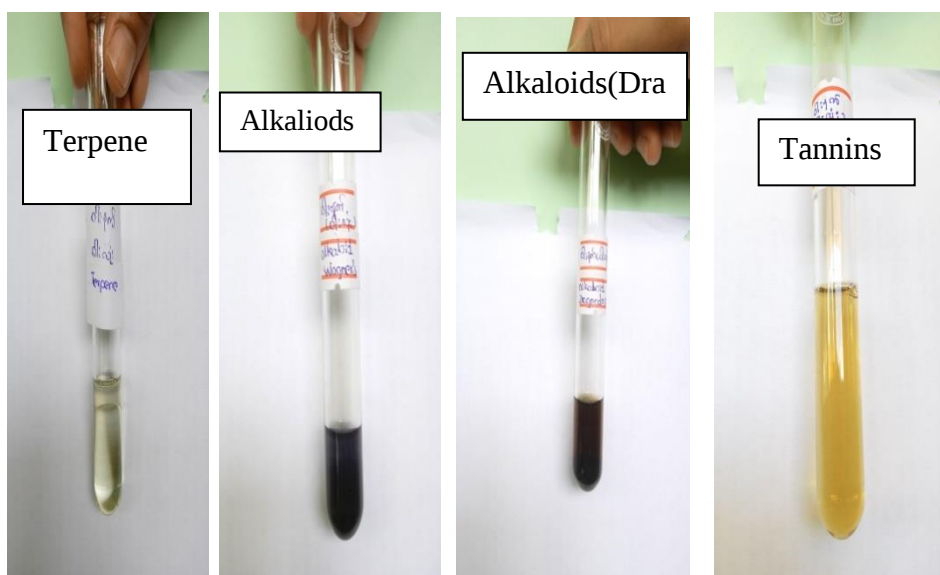


Figure 10. The Sample Bamboo Extract that Showing the Phytochemical Compound that Find out in Extract

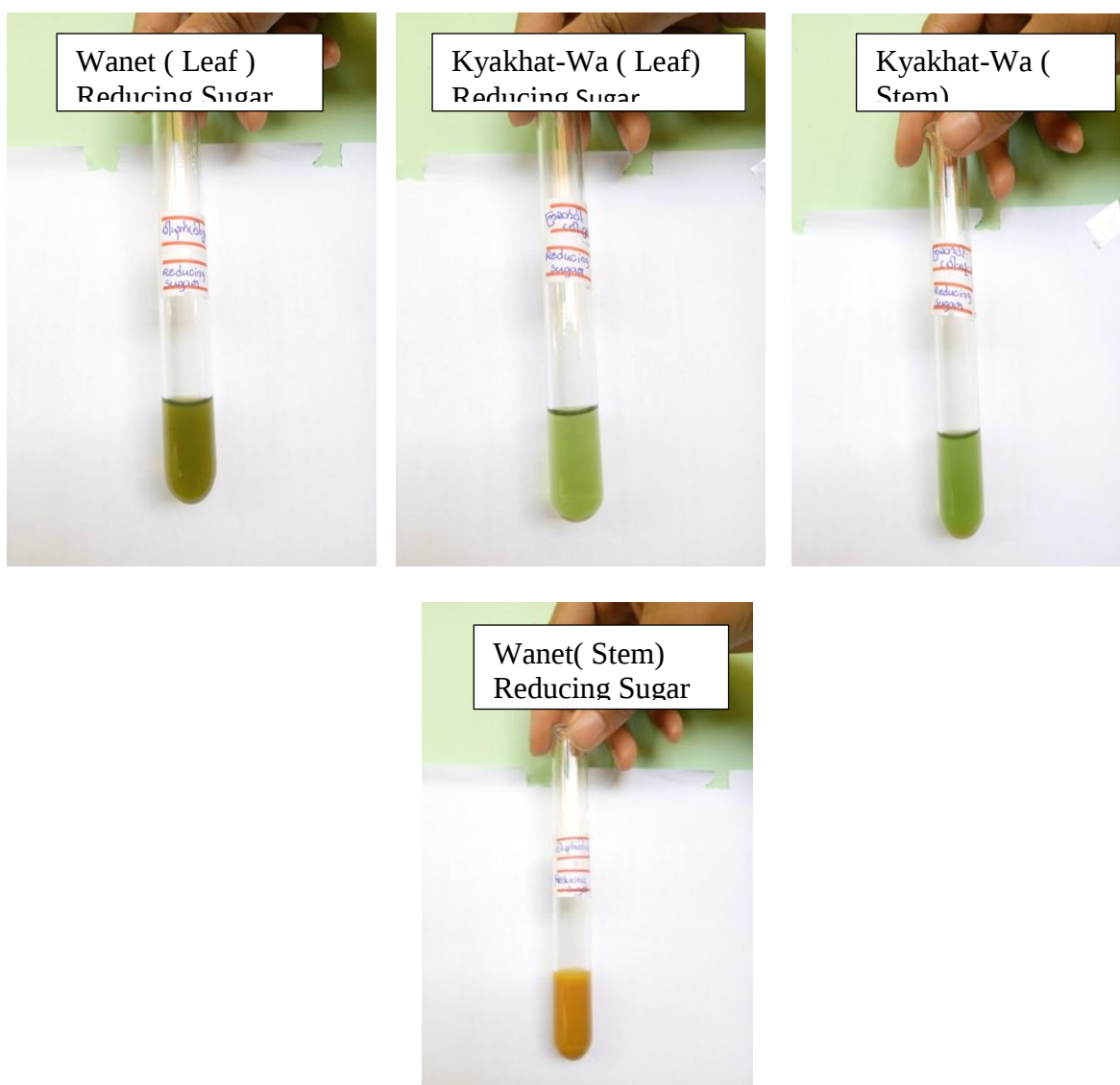


Figure 11. The Sample Bamboo Extract that Showing the Phytochemical Compound that cannot Find out in Extract

4.5. Preparing and Method of Antioxidant Analyzing

The antioxidant activity of the different concentration of extracts were determined by the DPPH radical scavenging assay according to Lee *et al.* Briefly, the reaction mixture containing 5 μ L of test sample with different concentration (diluted with DMSO) and 95 μ L of DPPH (0.3 mM) dissolved in methanol, was taken in a 96-well micro-titer plate and incubated at 37 °C for 30 min in the dark. The absorbance was measured at 517 nm by using 96 well micro plate reader (SPECTROstar Nano micro plate reader (BMG, Labtech, Germany)). Ascorbic acid was used as a standard. Percent Radical Scavenging Activity (% Inhibition) was calculated by using the following formula:

$$\% \text{Inhibition} = [1 - (\text{OD test compound} / \text{OD control})] \times 100$$

The following table is the positive control standard (Ascorbic Acid) to compare the result of the bamboo extracts in analyzing the results.

Sample Concentration (μ g/ml)	500	250	125	62.5	31.25	15.625	IC ₅₀ (μ g/ml) $\pm$ SEM	Method
% Inhibition $\pm$ SEM	99.82 $\pm$ 0.15	100.83 $\pm$ 0.97	100.55 $\pm$ 0.13	99.03 $\pm$ 0.77	88.04 $\pm$ 3.27	27.00 $\pm$ 2.55	20.32 $\pm$ 0.71	DPPH Radical Scavenging Assay

Table 2. (%) Inhibition and IC₅₀ of Ascorbic acid

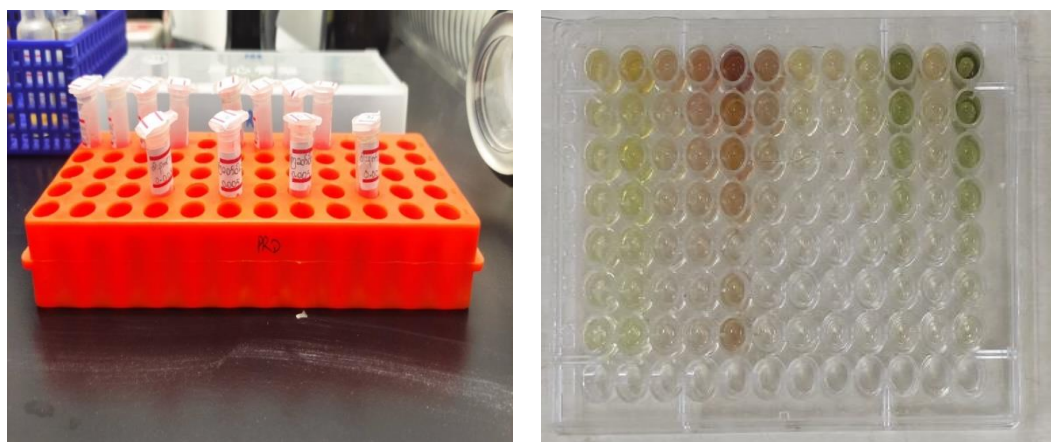


Figure 12. Sample Preparation and Dilution



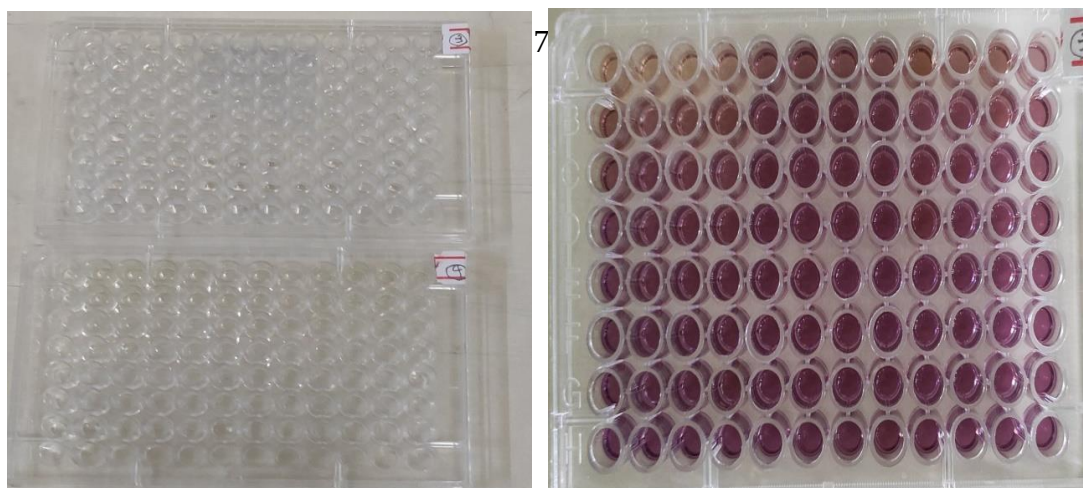


Fig 14. Reaction plates before and after incubation

4.6. Preparing and Method of Toxicity Analyzing

In testing the toxicity of leaf and stem of Wanet and Kyakhat-wa, operated the cytotoxicity assay following by the Hemolysis Assay using the healthy person's Red Blood cells (RBCs). Firstly the sample of blood from a healthy person was collected and placed in a hyperin tube. Then mix with 0.85 % NaCl solution and transfer to 15 ml Tube and centrifuge at 1500 rpm for 5 min continuously. After centrifuging, it was discarded the supernatant. That supernatant was washed with 0.2 M Chilled PBS 2-3 times till to get the clear supernatant. And the pellet was collected and made 5% blood suspension with 0.85% NaCl.

The hemolysis experiment procedures was follows,

1. Samples were made to the desired concentration with PBS or 0.85% NaCl
2. Add 5% blood suspension to the sample
Triton-X-100 was used as the positive control
PBS or 0.85% NaCl was used as the negative control
3. Incubated at 37°C at the incubator for 30 min
4. After incubation, centrifuge at 3000 rpm for 10 min
5. Take supernatant and measure at 540 nm
6. Calculation with following equation

Percent Lysis or hemolysis activity of sample = $(\text{Abs of sample} / \text{Abs of positive control}) \times 100$

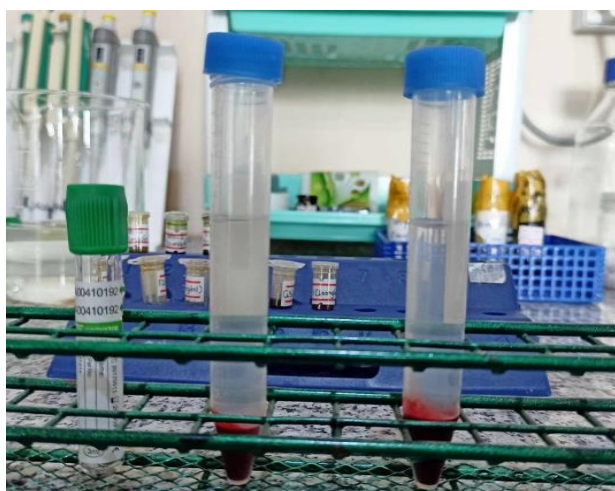


Figure 15. Blood Sample collection



Figure 16. Blood Sample preparation by doing suspension



Figure 17. Incubation and Centrifugation the blood sample

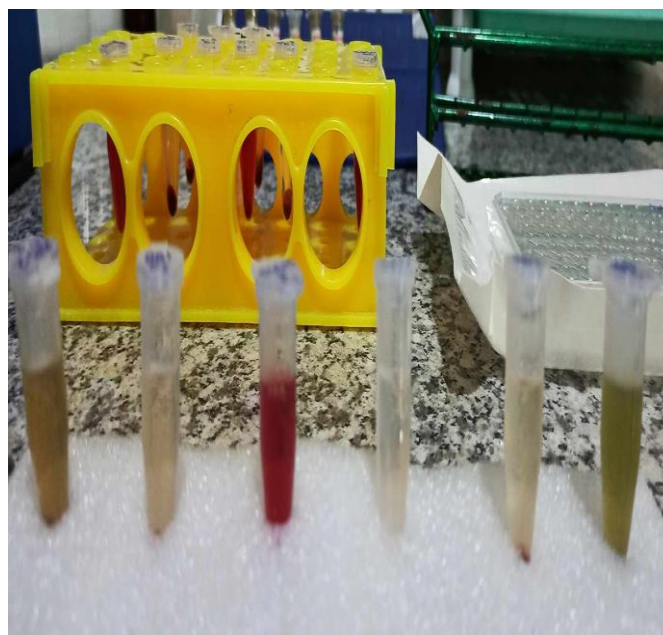


Figure 18. Samples ready to be tested after mixing with extract in different concentration

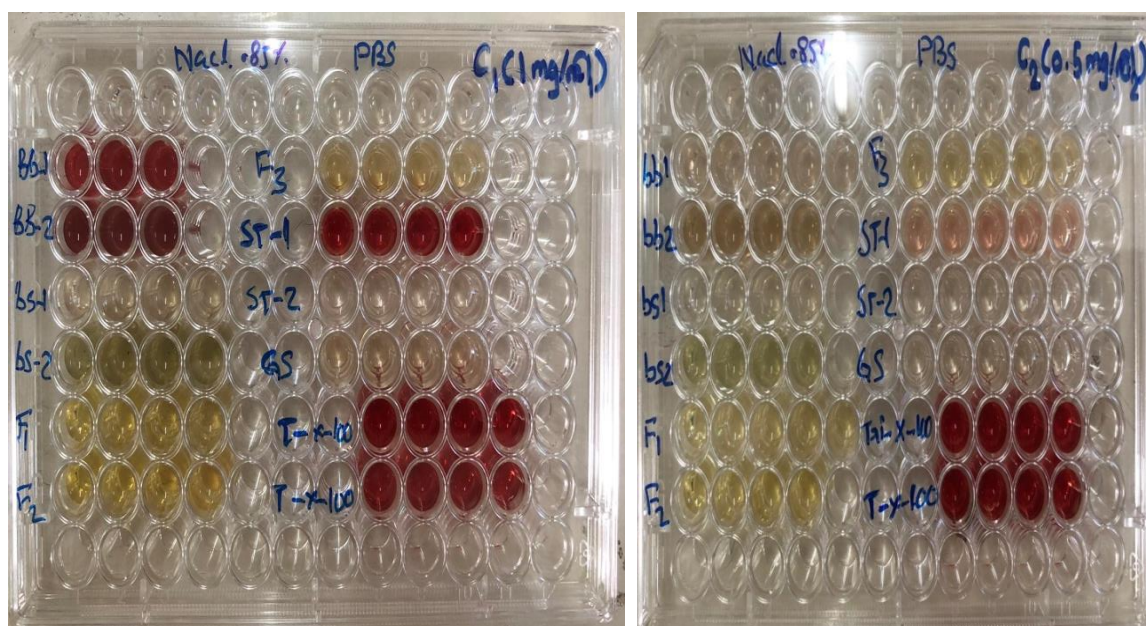


Figure 19. Measuring Absorbance at 540 nm (Sample concentration: 0.5 mg/mL) using 96-well Plate Reader (Sample concentration: 1 mg/mL)

4.7. Preparing and Method of Antimicrobial Activity Evaluation

The experiment to evaluate the antimicrobial activity of plant extracts was carried out at the Pharmaceutical Research Laboratory, Department of Biotechnology Research (DBR), Kyaukse District, Mandalay Region, Myanmar.

Glassware and Equipment used for this research experiment

Glassware and Equipment used for this research work is listed as follows:

- | | |
|-------------------------------|------------------------|
| 1. Balance: | Shimazu, Japan |
| 2. Incubator: | ESCO, USA |
| 3. Bio Safety Cabinet: | LabTech, Korea |
| 4. Digital Photo Colorimeter: | Apel, PD 303 |
| 5. Ultrasonic Cleanser: | UCP-10, GIOTECH, Korea |
| 6. Autoclave: | Jisico, Korea |
| 7. Cork borer | 8mm |
| 8. Petridishes | 100mm |

Tested microorganisms

Two Gram-negative bacteria (*Escherichia coli* and *Pseudomonas aeruginosa*), and three Gram-positive bacteria (*Staphylococcus aureus*, *Enterococcus faecalis* and *Bacillus cereus*) were used as the tested microorganisms for this experiment.

No.	Test Organism	Infection	Bacteria
1.	<i>Escherichia coli</i>	Urinary tract infection, abdominal and pelvic infection, pneumonia and meningitis	Two Gram-negative bacteria
2.	<i>Pseudomonas aeruginosa</i>	Malignant external otitis, Endophthalmitis, Endocarditis, Meningitis, Pneumonia, Septicemia	
3.	<i>Staphylococcus aureus</i>	Bloodstream infections, pneumonia, bone and joint infection	Three Gram-positive bacteria
4.	<i>Enterococcus faecalis</i>	Urinary tract infections, prostatitis, intra-abdominal infection, cellulitis, Wound infection,	
5.	<i>Bacillus cereus</i>	Brain abscess, cellulitis, endocarditis, Meningitis, Pneumonia, Osteomyelitis	
6.	Ethanol 70% solvent	-	Negative Control
7.	Chloramphenicol	The agent of treatment for bacterial conjunctivitis and otitis externs	Positive Control

Table 3. Test Organisms Used in Antibacterial Activity (NITE 2004)

Agar well diffusion method

The agar well diffusion method was used for antimicrobial activity evaluation by modifying the method described by Schlegel [1]. Tested microorganisms were inoculated in Muller Hinton Broth at 37°C for overnight. On the next day, the overnight broth culture was diluted with Normal saline to obtain the OD₆₀₀ at 0.08 to 0.1 with the approximate cell density of 1.5×10^8 CFU/mL. Muller Hinton Agar plates were prepared and sterilized by autoclaving at 121°C for 15 min. The broth inoculums were evenly spread out with sterile cotton swabs on the Muller Hinton Agar plates to obtain the uniform inoculums. After the plate was inoculated, 8-mm diameter wells were made on the agar medium by using a sterile cork borer. Each 50 µl (0.2g/ml) of samples was introduced into each well labeled. Chloramphenicol 30µg/well was used as the positive control. Then, the plates were placed in an incubator at 37°C for 16 to 18 hours. After incubation, the plates were examined and zone diameters of complete inhibition were measured and recorded to the closest millimeter.

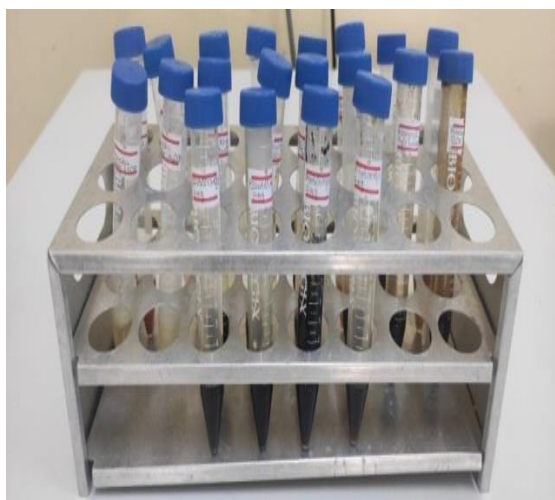


Figure 20. Sample Preparation to incubate



Figure 21. Broth culture preparation using shaking incubator



Figure 22. Absorbance reading for broth cultures dilution to obtain the OD 600 at 0.08 to 0.1



Figure 23. Prepared diluted broth cultures



Figure 24. Media sterilization using Autoclave

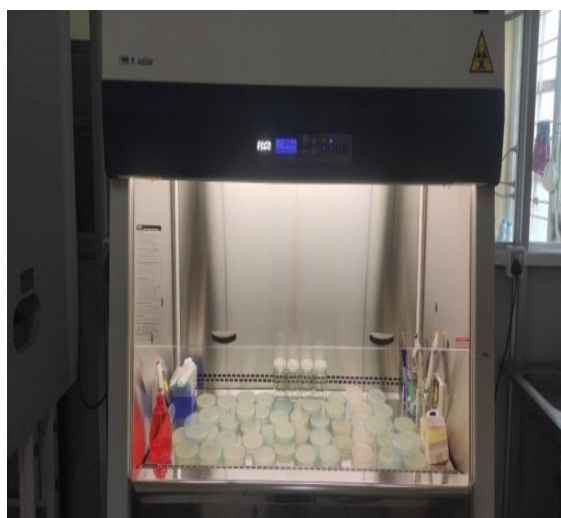


Figure 25. Media preparation under Biosafety Cabinet



Figure 26. Incubation the Sample



Figure 27. Antimicrobial experiment

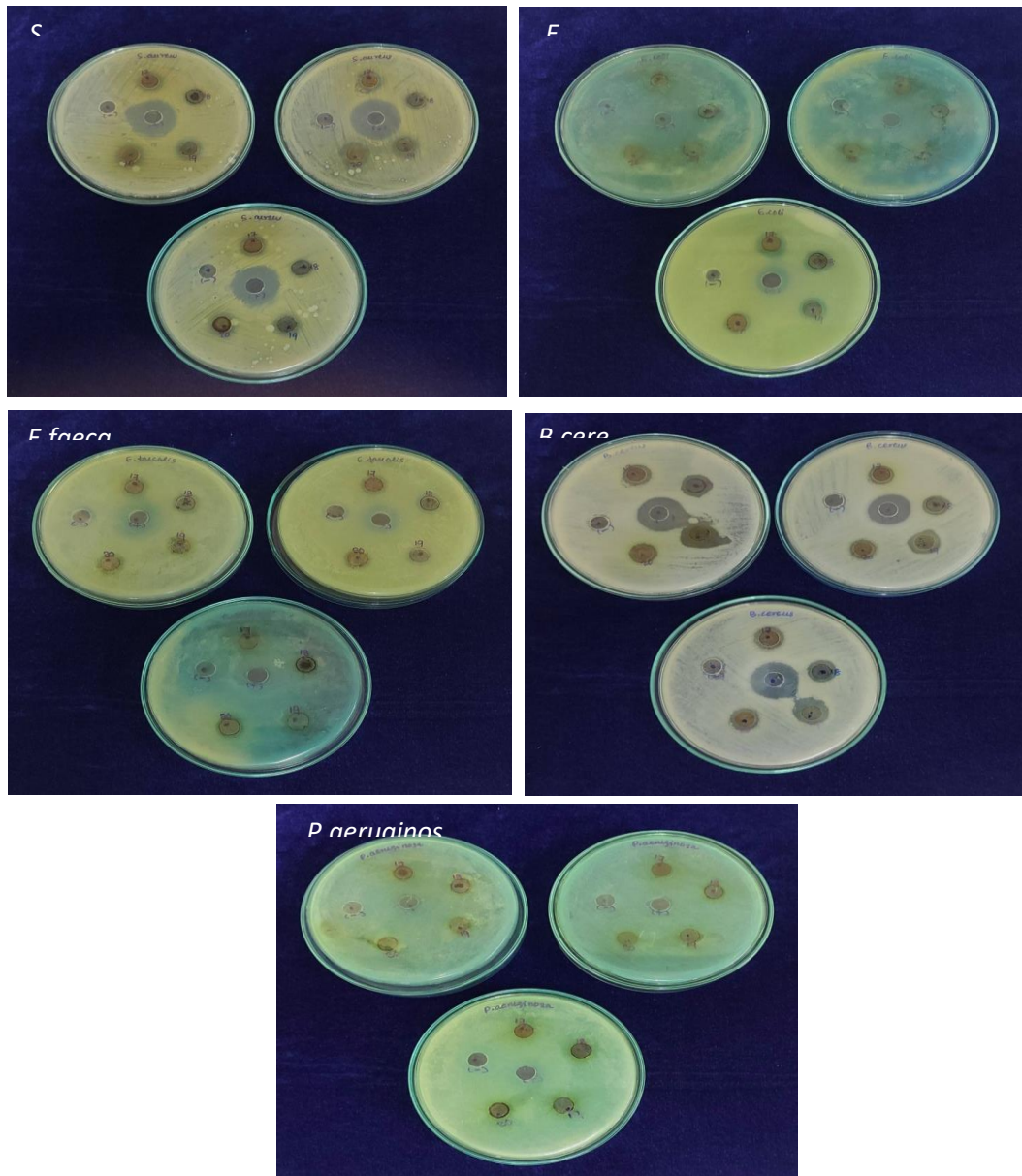


Fig 28. Culture plates showing zone of inhibition by plant extracts

5. Results

5.1. Phytochemical Activity

According to the Harborne JB method, the test result are as follow. According to the test results, Alkaloids, Saponins, Tannins, Flavonoids, Glycosides, Steroids, Terpenes, Phenolic, Polyphenol, Lipophenol and Phytochemicals (10) have been at the extract of bamboo (Wanet and Kyakhat-Wa) leaf and stem. But the Reducing Sugar Phytochemical compound was found only in Wanet bamboo stem's extract and not found in Wanet leaves and in both of leaf and stem of Kyakhat-Wa extract.

No	Test	Test Reagent	Preferences	Observation			
				Wanet (Stem)	Wanet (Leaf)	Kyakhat-Wa (Stem)	Kyakhat-Wa (Leaf)
1.	Alkaloids	Dragendroff's Reagent	Orange PPT	+	+	+	+
		Wagner's Reagent	Reddish Brown	+	+	+	+
2.	Flavonoid	Small Pieces of Mg Ribbon, HCl (Conc:), (5 Drops)	Yellow Color	+	+	+	+
3.	Glycoside	10 % Lead Acetate Solution	White PPT	+	+	+	+
4.	Lipophenol	NaOH Solution (4 Drops)	Pale Brown Color	+	+	+	+
5.	Phenolic Compound	10 % Ferric Chloride Solution	Brown Color	+	+	+	+
6.	Polyphenol	1% FeCl ₃ , 1% K ₃ Fe(CN) ₆ , (1 ml)	Greenish blue color	+	+	+	+
7.	Saponin	Distilled Water, Shake	Frothing	+	+	+	+
8.	Steroid	Acetic Anhydride, (Conc.) H ₂ SO ₄ (2 Drops)	Green Color	+	+	+	+
9.	Tannins	10% FeCl ₃ (Dil.) H ₂ SO ₄ (2 Drops)	Pale Brown PPT	+	+	+	+
10.	Terpene	Chloroform + Acetic Anhydride + Conc.HCl (2 Drops)	Pale Green Color	+	+	+	+
11.	Reducing Sugar	Benedict's solution	Brick red ppt	+	-	-	-

Table 4. Results of Phytochemical Compound in Two Bamboos Species

5.2. Antioxidant Activity

In the antioxidant evaluation of leaf and stem of wanet and kyakhat-wa, DPPH Radical Scavenging Assay method was used and the test was performed at Medical Biotechnology Laboratory, Department of Biotechnology Research (DBR), Kyaukse, Mandalay Division, Myanmar. The 7 kinds of different concentration were made to test antioxidant by comparing. All data were represented as Mean $\pm$ SEM from at least triplicate experiments. Ascorbic acid was used as a positive control for DPPH Radical Scavenging Assay. Ascorbic acid showed $99.82 \pm 0.15\%$ inhibition at 500 $\mu\text{g/ml}$ and IC_{50} of Ascorbic acid was $20.32 \pm 0.71 \mu\text{g/ml}$. The concentration of DPPH used for this experiment was 0.3mM. By comparing with standard control testing in terms of ascorbic acid, the results is pointing out, the bamboo stem extract has higher antioxidant activity than leaf extract. The significant result is at the Kyakhat-wa stem extract that was show the antioxidant activity is included at 5 different concentration testing among 7 kinds of concentration testing.

Sample Concentration($\mu\text{g/ml}$)	500	250	125	62.5	31.25	15.625	7.8125	IC_{50} ($\mu\text{g/ml}$) $\pm$ SEM	Method
DPPH radicals scavenging activity (%Inhibition $\pm$ SEM)	3.26 $\pm$ 1.96	0	0	0	0	0	0	>500	DPPH Radical Scavenging Assay

Table 5. The result of antioxidant in Wa-net Leaf Extract

Sample Concentration ($\mu\text{g/ml}$)	500	250	125	62.5	31.25	15.625	7.8125	IC_{50} ($\mu\text{g/ml}$) $\pm$ SEM	Method
DPPH radicals scavenging activity (%Inhibition $\pm$ SEM)	29.18 $\pm$ 5.01	6.58 $\pm$ 1.53	0	0	0	0	0	>500	DPPH Radical Scavenging Assay

Table 6. The result of antioxidant in Wa-net Stem Extract

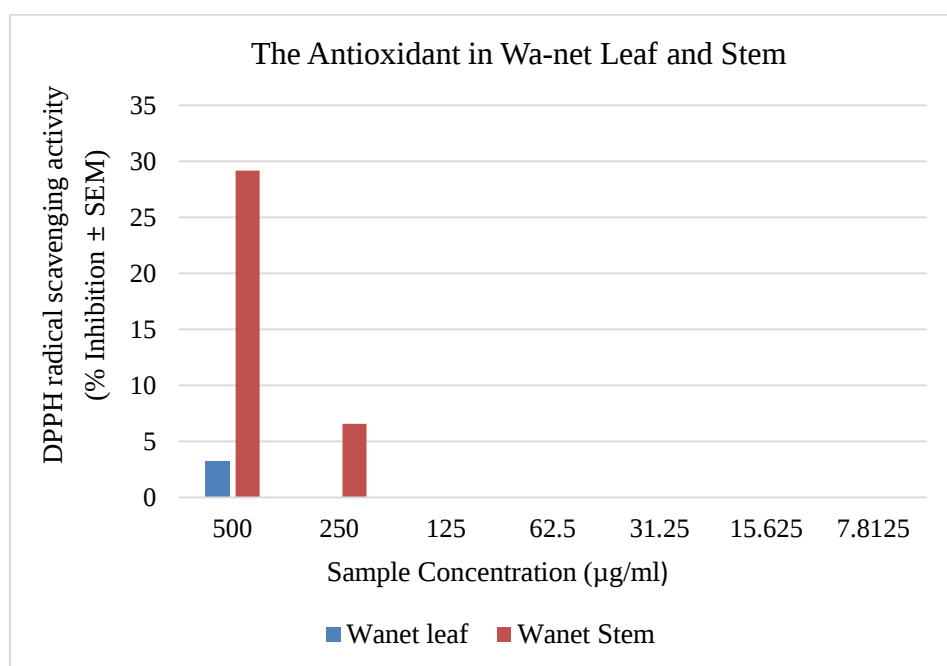


Figure 29. Result of Antioxidant Content of Wa-net Leaf and Stem

Sample Concentration (µg/ml)	500	250	125	62.5	31.25	15.625	7.8125	IC ₅₀ (µg/ml) ±SEM	Method
DPPH radical scavenging activity (%Inhibition ±SEM)	0	0	0	0	0	0	0	>500	DPPH Radical Scavenging Assay

Table 7. The result of antioxidant in Kyakhat-wa Leaf Extract

Sample Concentration (µg/ml)	500	250	125	62.5	31.25	15.625	7.8125	IC ₅₀ (µg/ml) ±SEM	Method
DPPH radical scavenging activity (%Inhibition ±SEM)	45.36±3.65	12.61±1.07	9.45±2.68	5.72±1.35	2.70±0.65	0	0	>500	DPPH Radical Scavenging Assay

Table 8. The result of antioxidant in Kyakhat-wa Stem Extract

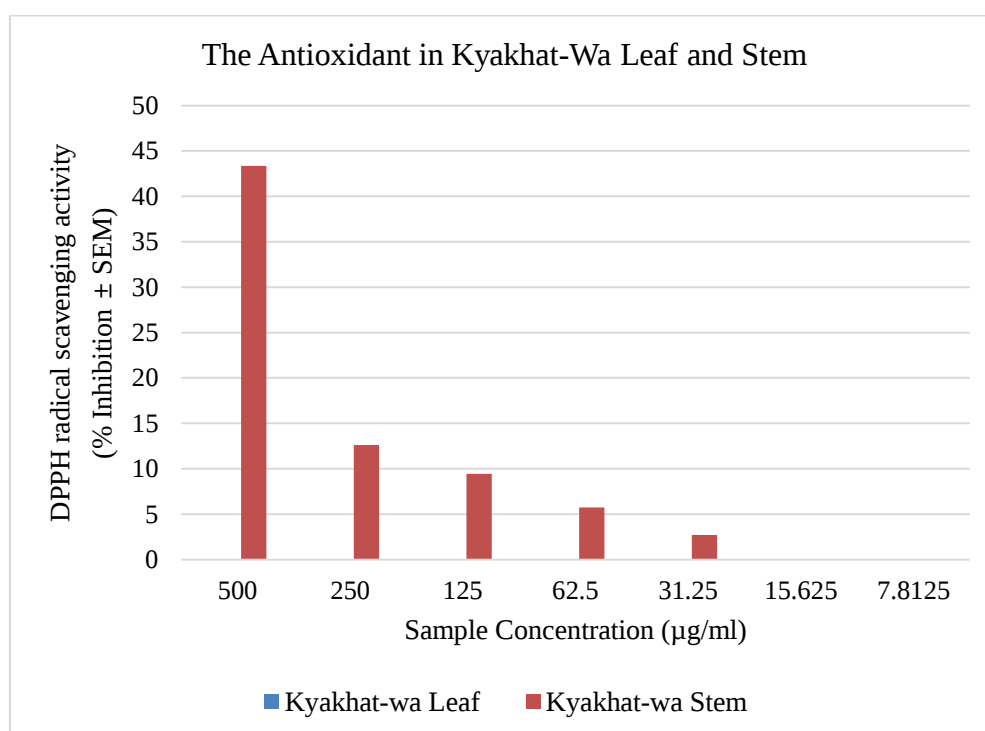


Figure 30. Result of Antioxidant Content of Kyakhat-wa Leaf and Stem

5.3. Toxicity Activity

The table show the result of toxicity of leaf and stem of Wanet and Kyakhat-wa together with the positive control sample (Triton-X-100) and negative control sample of the healthy person's blood red cells. The results was over 30% is mention the activity of toxicity (cytotoxic activity) is strong effect to the red blood cells. In this testing, the two different concentration solvent was used to get more detail result. Both of testing in different concentration 1 mg/mL and 0.5 mg/mL. As the whole result checking, the extract of Wa-net leaf and stem are showing the enough toxicity activity in the 1 mg/mL concentration sample testing but 0.5 mg/mL. When we compare the leaf and stem of Wa-net extract, the Wa-net extract have higher cytotoxic content than the Wa-net stem extract. And in Kyakhat-wa leaf and stem extract testing in both concentration, it was found that there is higher than 30% lysis and it can assume it is not enough to against red blood cells. Thus, this testing show that if we want to use the Kyakhatwa extract for medicine to against red blood cell, we should use more concentration solvent for it. Calculation with following equation;

Percent Lysis or hemolysis activity of sample = (Abs of sample/Abs of positive control)* 100

Sample	Code no.	Result	
		1 mg/mL	0.5 mg/mL
Wa-netwa Stem Extract	bb-1	79 ± 25	4.31 ± 0.18
Wa-net wa Leaf Extract	bb-2	84.43 ± 8	13.33 ± 0.2
Kyakhat-wa Stem Extract	bs-1	4.51 ± 0.15	3.58 ± 0.5
Kyakhat-wa Leaf Extract	bs-2	15.86 ± 1.71	7 ± 0.25
PBS (Negative Control)	PBS	1.39 ± 0.13	1.61 ± 0.1
Triton-X-100 (Positive Control)	T-X-100	100	100

***Percent Lysis ≥ 30 % is considered as cytotoxic activity to the red blood cells

Table 9. The result of cytotoxic activity of bamboo extracts

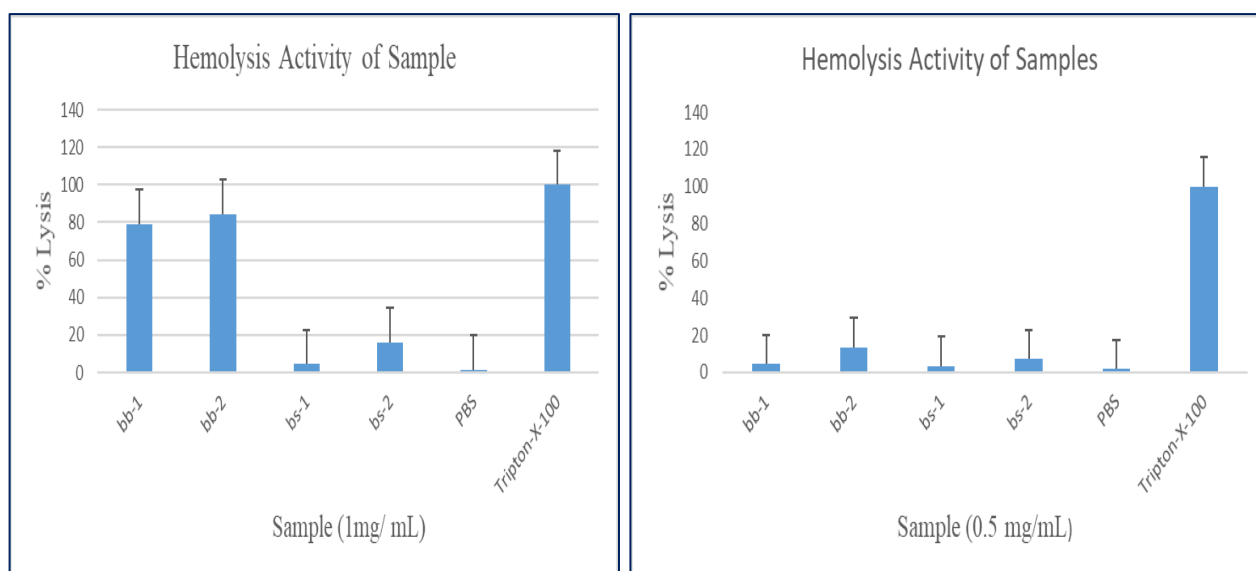


Figure 31. The Result of Cytotoxic Activity in chart (Test with 1 mg/mL and 0.5 mg/mL concentration)

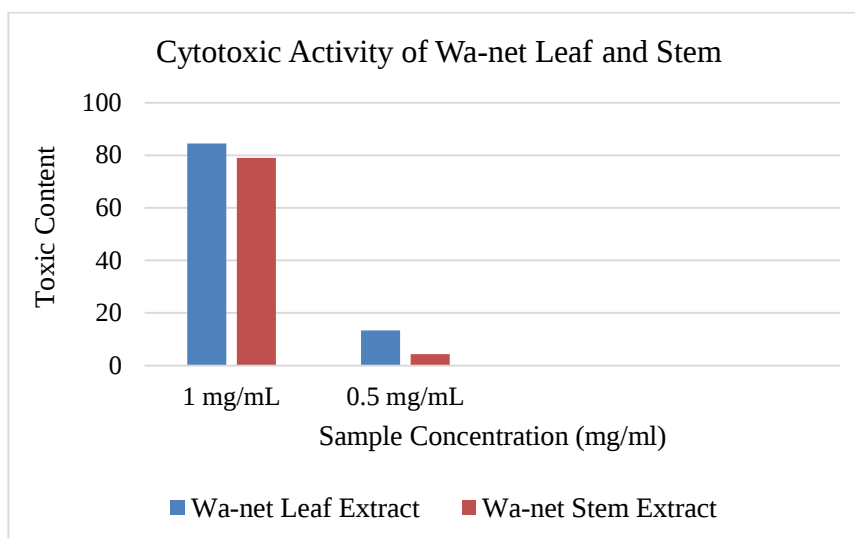


Figure 32. Result of cytotoxicity activity of Wanet Leaf and Stem

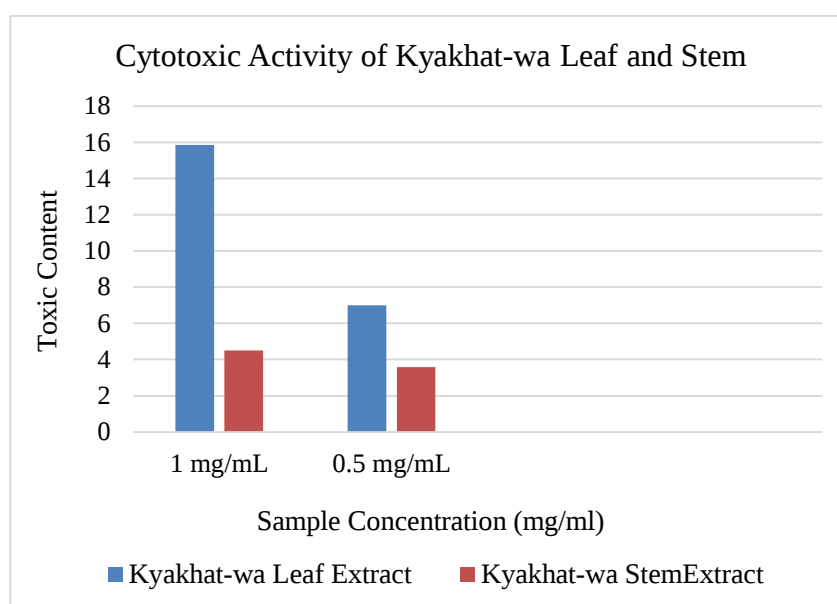


Figure 33. Result of cytotoxicity activity of Kyakhat-wa Leaf and Stem

The cytotoxicity activity of Wanet leaf was (84.43 ± 8 mg/mL) and the Wanet stem was found out 79 ± 25 mg/mL by using the 1 mg/mL extraction test. By seeing these two results, the result of Lysis is over 30% in both test, the toxicity is enough include and the more concentration was used the more cytotoxicity content was found.

5.4. Antimicrobial Activity Analysis

As the usual antimicrobial tests, the 5 bacteria species was used as specimen. The ethanol 70% solvent was negative control sample and chloramphenicol (the agent of treatment for bacterial conjunctivitis and otitis externa) was positive control sample. The result of InhibitionZoneDiameter present will represent it was against to bacteria. Overall results analysing is that the leaf of both bamboo species is stronger than the stem extract in againsting to bacteria. And the Wanet species has the high rate of antimicrobial activity in both leaf and stem, they can be against *Staphylococcus aureus* and *Bacillus cereus* with strong concentration. But Kyakhat-Wa leaf extract is weak in antimicrobial activity.

Sample	InhibitionZoneDiameter(Average(mm) ±SD)				
	<i>Escherichia coli</i>	<i>Staphylococcus aureus</i>	<i>Bacillus cereus</i>	<i>Enterococcus faecalis</i>	<i>Pseudomonas aeruginosa</i>
Kyakhat-Wa Leaf Extract	0	0	12.67±1.15	0	0
Solventcontrol (70%Ethanol)	0	0	0	0	0
Chloramphenicol	13.33±1.53	21.67±2.08	19.33±1.53	20.00±1.73	12.67±1.53

Table 10. The Result of Kyakhat-wa Leaf extract's antimicrobial activity

Sample	InhibitionZoneDiameter(Average (mm)±SD)				
	<i>Escherichia coli</i>	<i>Staphylococcus aureus</i>	<i>Bacillus cereus</i>	<i>Enterococcus faecalis</i>	<i>Pseudomonas aeruginosa</i>
Kyakhat-Wa Stem Extract	0	11.67±0.58	11.67±0.58	0	0
Solventcontrol (70%Ethanol)	0	0	0	0	0
Chloramphenicol	13.33±1.53	21.67±2.08	19.33±1.53	20.00±1.73	12.67±1.53

Table 11. The Result of Kyakhat-wa stem extract's antimicrobial activity

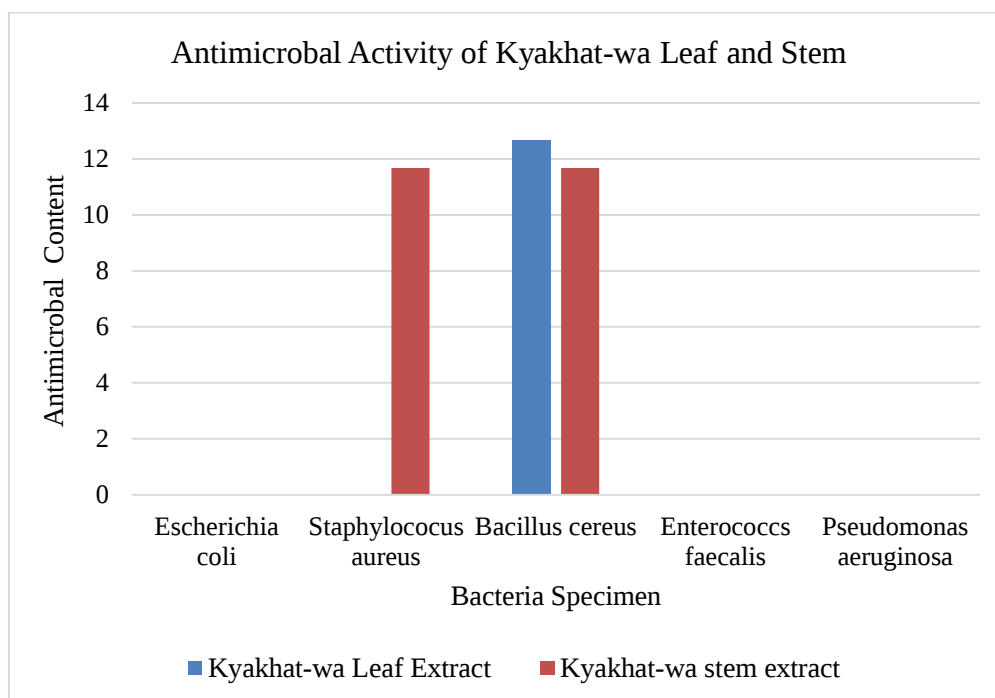


Figure 34. Result of antimicrobial activity of Kyakhat-wa Leaf and Stem

Sample	InhibitionZoneDiameter (Average(mm)±SD)				
	<i>Escherichia coli</i>	<i>Staphylococcus aureus</i>	<i>Bacillus cereus</i>	<i>Enterococcsf aecalis</i>	<i>Pseudomonas aeruginosa</i>
Wanet Leaf Extract	0	11.67±0.58	15.00±1.00	0	0
Solventcontrol (70%Ethanol)	0	0	0	0	0
Chloramphenicol	13.33±1.53	21.67±2.08	19.33±1.53	20.00±1.73	12.67±1.53

Table 12. The Result of Wanet Leaf extract's antimicrobial activity

Sample	InhibitionZoneDiameter(Average(mm) ±SD)				
	<i>Escherichia coli</i>	<i>Staphylococcus aureus</i>	<i>Bacillus cereus</i>	<i>Enterococcsf aecalis</i>	<i>Pseudomonas aeruginosa</i>
Wanet stem extract	0	12.00±1.00	11.33±0.58	0	0
Solventcontrol (70%Ethanol)	0	0	0	0	0
Chloramphenicol	13.33±1.53	21.67±2.08	19.33±1.53	20.00±1.73	12.67±1.53

Table 13. The Result of Wanet stem extract's antimicrobial activity

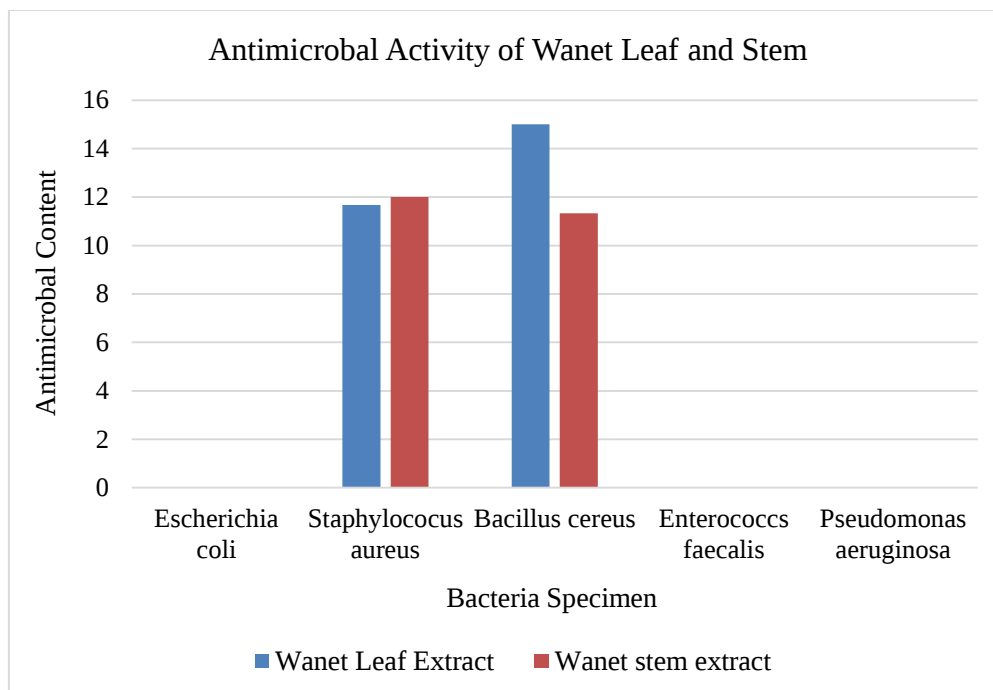


Figure 34. Result of antimicrobial activity of Wanet Leaf and Stem

6. Discussion

The basic analyzing of phytochemical compound was performed by the Harborne JB method. There were 10 phytochemical compounds found out respectively; Alkaloid, Saponins, Tannins, Flavonoids, Glycoside, Terpenes, Phenolic, Polyphenol, Lipophenol compound but the reducing sugar phytochemical compound was lacking in the Wanet Leaf and Kyakhat-Wa leaf and stem. Since it includes almost all kinds of phytochemicals, it can be assumed that the Wa-net and Kyakhat-Wa leaf and stem are rich in useful organic compounds. The antioxidant capacity was analyzed in terms of ascorbic acid ($99.82 \pm 0.15\%$ inhibition) as a standard comparing specimen and was best observed at the Kyakhat-wa stem extract. i.e. There are 7 different rates of concentration samples in this testing, the result of antioxidant activity occurred at 5 different rates of concentration sample in high rate of capacity till $45 \pm 3\%$ of inhibition at $500 \mu\text{g/ml}$ while the standard sample is 99% of inhibition. This result shows that the leaf of Kyakhat-wa was used enough as an antioxidant agent in production of medicine and other personal care products. Meanwhile, the Wanet leaf lacked antioxidant activity in this testing with 7 different rates of concentration sample. But this is not assumed, the Wanet leaf has no antioxidant activity. It just did not find out the capacity of antioxidants in this dilution solvent. Thus, the content of antioxidant activity is also dependent on the kind and concentration of reagent solvent. The other testing is antimicrobial activity analysis. Bamboo is also well known due to its abundant antimicrobial activity. This test was also analyzed with two standard control samples, 70% ethanol solvent as negative control and chloramphenicol as positive control. The test specimen was 5 species of bacteria and the testing method is agar well diffusion method. Among 5 species of bacteria, the test sample bamboo can be against the common two species (*Staphylococcus aureus* and *Bacillus cereus*). Wa-net leaf extract has the strongest capacity of against to *Bacillus cereus* till $15 \pm 1\%$ inhibition while the positive control sample was 19% inhibition and negative control was zero inhibition of against bacteria. The main important test of the present research is toxicity activity analyzing. For the testing, the lab procedure was following the method of Hemolysis Assay. It was used the blood from a healthy person as specimen. To know more detail result, two different rates of concentration of agent solvents; 1 mg/mL and 0.5 mg/mL were used. In the test procedure, it can be considered that if Lysis percent is over 30% , that extract includes toxicity (cytotoxic) activity. We found that it was present the cytotoxic activity just at Wa net leaf and stem at 1 mg/mL concentration. Among them, Wa-net leaf is the strongest one and Wanet stem is second. In here, when comparing the toxicity rate of stem and leaf, the leaf is higher toxic rate than stem. Overall analyzing on all testing, bamboo was rich in antioxidant compounds in advance rate and antimicrobial compounds to against bacteria. Moreover, it also has abundant toxicity rate that can be against to red blood cells. Thus, it has a really high range of utilizing in various fields.

7. Conclusion

The aim of this research is to evaluate the toxicity, antioxidant and antimicrobial activity of bamboo leaf and stem of Wanet (*Bambusa vulgaris* Schrad. Ex J.C, Wendt.) and Kyakhat-wa (*Bambusa bambos* Munro) Species for more advanced utilization in related fields. According to the objective, the research was performed with different methods and different concentration solvent and reagent. And the result was also shown to be significant. According to the result, the highest and significant antioxidant activity was found in the Kyakhat-wa stem, even the less concentration $31.25 (\mu\text{g/ml})$, the inhibition percent is 2.7% meanwhile the standard control test (ascorbic acid) was 88% . Overview analyzing the result of antioxidant activity testing, the stems of bamboo (Wanet and Kyakhat-wa) have more antioxidant than the leaf. Bamboo leaf and stem can be assumed to have a high antimicrobial effect due to the

result of this research. The evidence factor of bamboo is its toxic properties in stem and leaf. This research was also tested for toxicity of bamboo leaf and stem. Based on the evaluated results, Wa-net leaf and stem can be toxic to the red blood cell. This research result showed that the concentration rate can be determined on the cytotoxic activity, it was present or not. Thus, the researchers should think about the important fact of concentration rate in testing. Moreover, there are abundant phytochemical compounds in bamboo. Thus, bamboo can be used in medicinal fields to reduce the excess of red blood cells in blood. The exception of this research is to support the analyzed data to the related field to get a more effective utilizing method without wasting the resource, even the leaf of bamboo. Bamboo can be used not only as food and construction material but also in medicine and personal care products manufacturing due to its enriched antioxidant, antimicrobial and toxicity properties. This fact was also another aim of this research that is to appear and to know about the more effective and valuable utility of bamboo.

8. Acknowledgement

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