

7,12-dimethylbenz(a)anthracene induced breast cancer in Sprague Dawley rats proved by ultrasound imaging

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7,12-Dimethylbenz(a)anthracene induced breast cancer in Sprague Dawley rats proved by ultrasound imaging

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Abstract. Many studies have shown that some of cancer animal models have been established to study various aspects of cancer biology, including breast cancer. Substantial evidence has shown that administration of 7,12-dimethylbenz(a)anthracene (DMBA) in animal models can mimic human breast cancer in many ways. This preliminary research aimed to develop rat breast cancer models that will be used for a further study on phytotherapy as the alternative medicine for breast cancer. Four young Sprague Dawley female rats, aged 60 days, received a single subcutaneous injection of 25 mg DMBA mixed with 0.5 mL sunflower oil and 0.5 mL sodium chloride 0.9%. Control group received 0.5 ml of sunflower oil and 0.5 ml of saline only. Tumor detection by palpation and rats' body weight measurement were performed every week after DMBA induction. At 10-weeks after DMBA induction, the tumor development was examined by ultrasound imaging. No significant differences were observed in the body weight between DMBA-induced rats and non-induced rats during 8-weeks after induction. Starting from week ninth, the body weight of the DMBA group significantly decreased ($p=0.02$) compared to the counterpart. A solid mass on the breast area was firstly detected at week seventh in one rat. Three weeks later the solid masses were detected in all DMBA-induced rats that proved by the ultrasound images showing hypoechoic lesions with irregular margin and calcification indicating malignancies. DMBA induced malignant lesions in female rats that demonstrated by ultrasound imaging.

Keywords: Animal model, breast cancer, DMBA, rats, ultrasound imaging

INTRODUCTION

Cancer is a cell disease characterized by a disorder or failure of the multiplication regulating mechanism and the function of another homeostasis in multicellular organisms resulting in uncontrolled body cell growth, and the ability to invade other tissues [1,2]. One type of cancer that has a high prevalence is breast cancer. Data from GLOBOCAN (IARC) in 2012 found that breast cancer in the world has a new percentage of cases that is equal to 43.4% and the percentage of deaths from breast cancer by 12.9% [3,4]. Currently, cancer therapy is applied radiotherapy, chemotherapy, surgery, or a combination. However, the therapeutic method has proven not to provide a satisfactory safety effect and can provide quite severe side effects experienced by patients. Medicinal plants are thought to have the potential as anticancer with minimal side effects when used with appropriate dosage and time of use and accuracy of use [1,5]. Here, we report the development of breast cancer animal model using a carcinogenic agent, 7,12-

dimethylbenz(a)anthracene (DMBA), as the starting step to assess the efficacy of potential chemopreventive herbal against breast cancer.

Materials and Methods

Animal

The animals used in this study is eight white rats (*Rattus norvegicus*) strains of healthy Sprague Dawley strain with a weight of 125-170 grams which were eight-week-old with the distribution of four rats in the DMBA-induced, two rats in the normal control group and two rats in the negative control group. They were kept in an animal room with 22–23°C air temperature. The care of laboratory animals followed accepted standards in Indonesia.

Carcinogen

Mammary tumors were induced by DMBA (7,12-Dimethylbenz(a)anthracene) prepared according to the method previously described [6]. DMBA was dissolved immediately before use in 0.5 mL sunflower oil and 0.5 mL sodium chloride 0.9%. Fresh solutions were prepared before application.

Experimental Design

Sprague Dawley rats aged eight weeks were divided into three groups. Group 1 received DMBA subcutaneous injection. Group 2 as normal control which was only given 0.5 mL sunflower oil and 0.5 mL 0.9% NaCl. Group 3 as the negative control group. Induction of DMBA subcutaneously in the breast area with a single dose of 25 mg DMBA dissolved in 0.5 mL sunflower oil and 0.5 mL 0.9% NaCl. Observations were made on changes in body weight, cancer incidence, number and type of cancer every day for 14 weeks. After tumor development in the expected group, confirmation was made by ultrasound imaging. After 14 weeks maintenance, all rats were terminated and dissected for removal of tumor tissue, both breast and tumor outside the breast for histopathological examination. In this article, we only report the result of ultrasound imaging.

Result

Rats growth

Until the week eight, there was no difference in rat growth ($p = 0.582$) which was assessed from the average weight gain of 17.50 ± 5.44 g and 21.50 ± 4.21 g, in DMBA-induced and control rats, respectively. Entering the week nine the rats that received DMBA began to experience weight loss and became increasingly evident at week 10 with an average weight loss of 10.75 ± 6.21 g. The weight of the control rats was constant when they entered the week 8 and decreased slightly at week 10 (Figure 1).

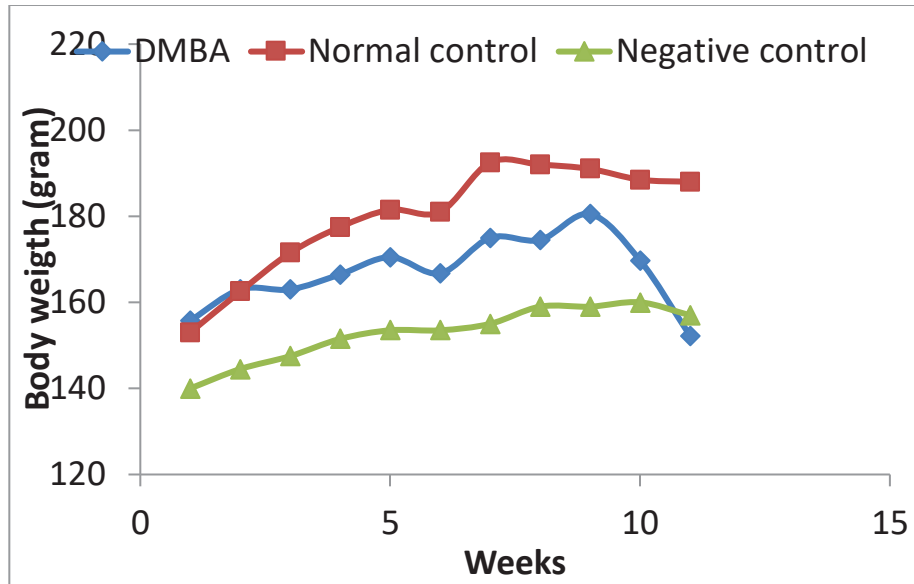


FIGURE 1. Rats growth monitored by gain or loss body weight in gram.

Tumor development

The palpable mammary tumor was firstly detected at week seven on 1 of 4 DMBA-induced rats. Three weeks later, all rats had palpable tumors which were characterized by nodules in the area around the mammary gland (Figure 2). Necrosis occurred in two rats and also hardened to the thigh area which might have the effect of the spread of cancer cells.

Ultrasound imaging

Ultrasound imaging was conduct to ensure tumor formation caused by DMBA-induction. The procedure was performed on all groups. The results revealed that DMBA-induced group have the irregular, heterogeneous, hypochoic mass with calcification, spiculations, and angular margins (Figure 3). In contrast, the results of ultrasound imaging from the normal control group and the negative control group had smooth and isochoic apperance.

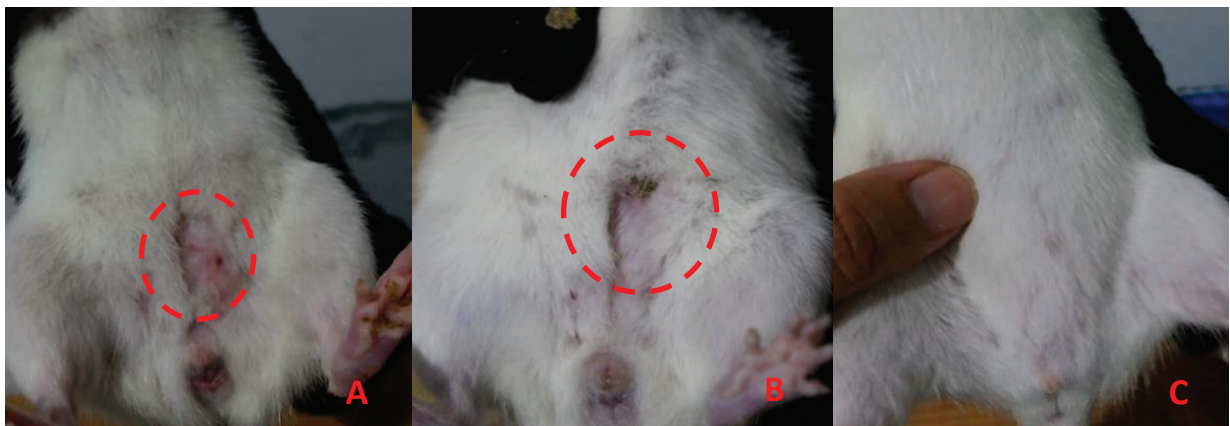


FIGURE 2. Tumor development in DMBA-induced rats at week eight (A) and week ten (B). Palpable tumors are marked by dotted circle. C is the control rats without any tumor development.

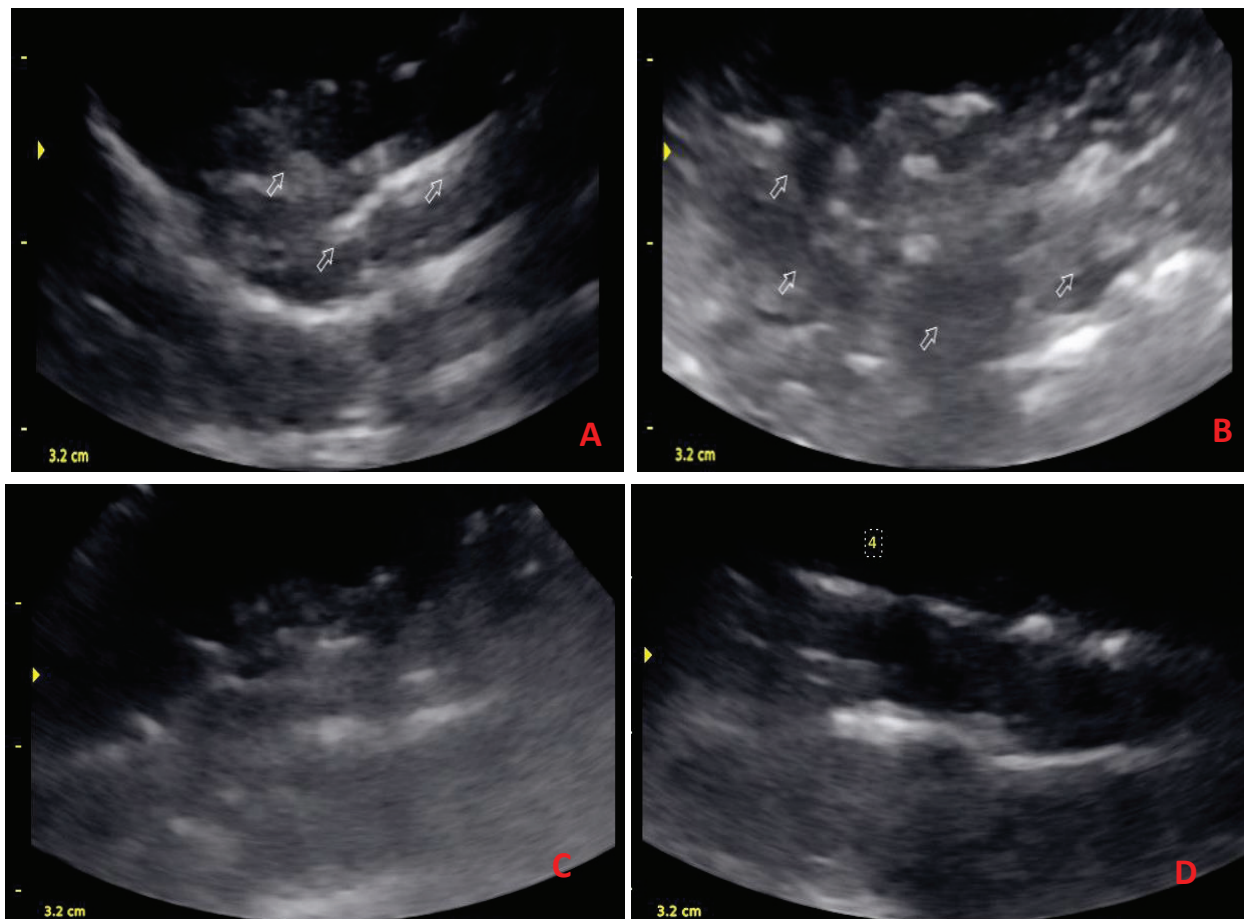


FIGURE 3. Ultrasound imaging of the DMBA-induced rats (A and B), normal control (C) and negative control (D)

DISCUSSION

Rats have been chosen as animal model compared to other animals in experimental terms to study mammary carcinogens because the mammary glands of rats have neoplasms that are very similar to humans [6]. DMBA is also a highly carcinogenic material depending on the DMBA injection method itself [7]. Skin that is in direct contact with DMBA could have papillomas [8]. DMBA also could cause leukemia when injected directly into a vein [9]. During puberty, pregnancy, and lactation, the mammary gland undergoes a very significant change because the mammary gland is one of several organs that always experience changes and development from birth [10].

There have been many studies regarding DMBA induction testing of tumor formation using rat model animals. However, our group wants to prove the effect of DMBA on tumor formation itself to be able to detect the stages of tumor development. We have conducted research and proved that DMBA induction could cause tumor development which is characterized by a lump. The area around the breast became hard and necrosis and proved that the formation of tumors through ultrasound imaging.

This report has not been finalized yet because at the end of the study the animal model will undergo the termination process that it will remove the tumor and expected organs to have metastasis for the histopathological examination. After this first step of research finished, it will proceed to the next stage by testing herbal extracts.

CONCLUSION

Induction of DMBA in female rats caused mammary tumor formation that confirmed by ultrasound imaging. The procedure of animal model development will be applied in the intended study on chemopreventive herbal against cancer.

ACKNOWLEDGMENTS

We acknowledge the Veterinary Clinic Medical School of Universitas Hasanuddin and drh. Waode SM, M.Si who performed the ultrasound procedure to the animal models.

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2. Blood chemistry profiles of DMBA-induced mammary tumor in female Sprague Dawley rats

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Windy Y.B

[Quoted text hidden]

ASMICBS 2018 Malang <asmicbs2018@gmail.com>

Tue, Oct 30, 2018 at 3:39 PM

To: Ika Yustisia <ikayustisia@gmail.com>

Dear Ika Yustisia

Bersama dengan ini, kami ingin menginformasikan bahwa Abstrak yang saudara(i) kirimkan pada kegiatan yang kami selenggarakan **DITERIMA** dan saudara(i) akan mengikuti Kegiatan **ORAL PRESENTATION** sesuai jadwal. Adapun jadwal Oral presentation akan diumumkan pada tanggal **12 November 2018** di website kegiatan www.asmicbs2018.fk.ub.ac.id . Abstrak dengan persetujuan author akan kami publikasi pada **AIP proceeding yang terindeks Scopus** dengan biaya tambahan sebesar **Rp 900.000,-**.

Jika masih ada hal kurang jelas, silahkan kontak kami secara langsung. Beritahukan kami, jika saudara(i) telah membaca email ini. Sekali lagi, kami ucapkan terimakasih atas partisipasi saudara(i) dengan mengirimkan abstrak pada kegiatan yang kami selenggarakan.

Best Regards
Windy Yuliana B

[Quoted text hidden]

ASMICBS 2018 Malang <asmicbs2018@gmail.com>

Sun, Nov 11, 2018 at 6:27 PM

To: Ika Yustisia <ikayustisia@gmail.com>

Dear Ika Yustisia

Bersama ini kami kirimkan **LoA** dari abstrak yang telah diterima. Adapun LoA tersebut sebagaimana terlampir. Informasi tambahan yang dapat kami sampaikan terkait pelaksanaan **Oral Presentation** yaitu setiap peserta akan diberikan waktu **selama 10 menit (7 menit Presentasi dan 3 menit Tanya Jawab)**. Dimohonkan agar setiap peserta dapat mempersiapkan **powerpoint presentation** dalam **bahasa inggris**.

Jika masih terdapat hal yang kurang jelas, jangan ragu untuk menanyakan kami secara langsung. Sekali lagi kami ucapkan selamat dan terimakasih atas partisipasi saudara. Sampai bertemu di malang.

Best Regards
Windy Yuliana B

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2 attachments

 **Padlianah.pdf**
360K

 **Muhammad Arif.pdf**
362K

Ika Yustisia <ikayustisia@gmail.com>
To: mutmainahnina35.mn@gmail.com, padlianahana@gmail.com

Mon, Nov 12, 2018 at 6:59 AM

Assalamu'alaikum Ana dan Nina

Bersama ini terkirim surat penerimaan dari KIBI

Terima kasih
[Quoted text hidden]

2 attachments

 **Padlianah.pdf**
360K

 **Muhammad Arif.pdf**
362K

ASMICBS 2018 Malang <asmicbs2018@gmail.com>
To: Ika Yustisia <ikayustisia@gmail.com>

Wed, Nov 14, 2018 at 6:39 AM

Dear Ika Yustisia

Bersama ini kami kirimkan kan kembali LoA penerimaan Abstrak. Kami memohon maaf adanya kesalahan penulisan nama pada LoA sebelumnya. Mohon kiranya dimaklumi
Atas perhatiannya kami ucapkan terimakasih

Best Regards
Windy Yuliana B

[Quoted text hidden]

 **Mutmainnah Arif.pdf**
204K

Ika Yustisia <ikayustisia@gmail.com>
To: mutmainahnina35.mn@gmail.com

Wed, Nov 14, 2018 at 6:41 AM

----- Forwarded message -----

From: **ASMICBS 2018 Malang** <asmicbs2018@gmail.com>

Date: Wed, Nov 14, 2018, 6:40 AM

Subject: Re: Abstrak ASMICBS 2018 Malang_Prodi S2 Ilmu Biomedik PS Unhas

To: Ika Yustisia <ikayustisia@gmail.com>

[Quoted text hidden]

 **Mutmainnah Arif.pdf**
204K



**3rd Annual Scientific Meeting of Indonesia
Consortium for Biomedical Sciences
Post-Graduate Program, Faculty of Medicine**
Universitas Brawijaya, Malang
Jl. Veteran Malang 65145. Telp. (0341) 575790



No : 035/Pan-ASMICBS/X/2018
Lampiran : -
Perihal : Penerimaan Abstrak

Kepada Yth.:

1. **Mutmainnah Arif** (Master Program of Biomedical Science on Pharmacology Sekolah Pascasarjana Universitas Hasanuddin Makassar)
2. Padlianah (Master Program of Biomedical Science on Biochemistry and Molecular Biology Sekolah Pascasarjana Universitas Hasanuddin Makassar)
3. Ika Yustisia (Departemen of Biochemistry Faculty of Medicine Universitas Hasanuddin Makassar)

Dengan hormat,

Kami dengan senang hati memberitahukan bahwa abstrak saudara yang berjudul: **“7,12-Dimethylbenz(a)anthracene induced breast cancer in Sprague Dawley rats proved by ultrasound imaging”** telah diterima untuk sesi presentasi oral dalam **3rd Annual Scientific Meeting for Biomedical Sciences** tahun 2018 di Malang.

Sekali lagi, kami mengucapkan selamat dan berterima kasih atas partisipasi Saudara dalam kegiatan ini. Kami berharap dapat bertemu Saudara di Malang pada 22-23 November 2018.

Malang, 2 November 2018
Ketua Panitia



dr. Hidayat Sujuti, Ph.D, SpM
NIP. 196701231996011001



Ika Yustisia <ikayustisia@gmail.com>

Manuskrip Mutmainnah Arif dan Padlianah_Unhas Makassar

2 messages

Ika Yustisia <ikayustisia@gmail.com>

Tue, Oct 30, 2018 at 11:13 AM

To: ASMICBS 2018 Malang <asmicbs2018@gmail.com>

Yth. Panitia ASMICBS 2018

Bersama ini terkirim 2 manuskrip mahasiswa kami atas nama Mutmainnah Arif dan Padlianah dari Prodi Magister Ilmu Biomedik Sekolah Pascasarjana Universitas Hasanuddin Makassar.

Terima kasih

Salam hormat,

Ika Yustisia

2 attachments**Manuskrip Arif M_ASMICBS_301018.docx**

726K

**Manuskrip Padlianah_ASMICBS2018_301018.docx**

2326K

ASMICBS 2018 Malang <asmicbs2018@gmail.com>

Tue, Oct 30, 2018 at 3:15 PM

To: Ika Yustisia <ikayustisia@gmail.com>

Dear Ika Yustisia

Terimakasih telah mengirimkan abstrak pada kegiatan ini.

Informasi yang dapat kami sampaikan, yaitu manuskrip ilmiah secara lengkap dapat dikirimkan ke email asmicbs2018@gmail.com paling lambat tanggal **20 November 2018**. Abstrak dengan persetujuan author akan kami publikasi pada **AIP proceeding yang terindeks Scopus** dengan biaya tambahan sebesar **Rp 900.000,-**

Jika masih ada hal kurang jelas, silahkan kontak kami secara langsung. Beritahukan kami, jika saudara(i) telah membaca email ini. Sekali lagi, kami ucapkan terimakasih atas partisipasi saudara dengan mengirimkan abstrak pada kegiatan yang kami selenggarakan.

Best Regards

Windy Y.B

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Local Committee

***3rd Annual Scientific Meeting of Indonesian Consortium for Biomedical Sciences
(ASMICBS) 2018***

Master Program in Biomedical Sciences

1st Floor, Graha Medika (C) Building, Faculty of Medicine, Universitas Brawijaya

Jl. Veteran Malang, East Java, Indonesia, 65145



Ika Yustisia <ikayustisia@gmail.com>

Konfirmasi keikutsertaan kegiatan ASMICBS 2018 Malang

2 messages

Ika Yustisia <ikayustisia@gmail.com>

Fri, Oct 26, 2018 at 9:12 PM

To: ASMICBS 2018 Malang <asmicbs2018@gmail.com>

Yth. Panitia ASMICBS 2018

Dengan hormat

Bersamaan ini kami mengkonfirmasi keikutsertaan kami dalam kegiatan ASMICBS 2018 di Malang untuk abstrak kami yang berjudul:
yaitu:

- 1). 7,12-Dimethylbenz(a)anthraceneinduced breast cancer inSprague Dawley rats proved by ultrasound imaging
- 2). Blood chemistry profiles ofDMBA-induced mammary tumor in female Sprague Dawley rats

Kedua abstrak tersebut sudah kami kirim sebelumnya.

Adapun abstrak yang kami kirim terlebih dahulu dengan judul:

High expression of FBP1 in invasive breast carcinoma: a preliminary study on cancer gluconeogenesis, batal berpartisipasi karena yang akan presentasi berhalangan hadir.

Saat ini kami sedang mempersiapkan manuskrip kami. Insya Allah paling lambat tanggal 30 Oktober kami kirimkan berikut biaya pendukung (registrasi presenter dan prosiding).

Terima kasih

Hormat saya,

Ika Yustisia

ASMICBS 2018 Malang <asmicbs2018@gmail.com>

Fri, Oct 26, 2018 at 9:40 PM

To: Ika Yustisia <ikayustisia@gmail.com>

Dear Ika Yustisia

Terimakasih atas konfirmasi yang telah disampaikan ke pihak panitia.

Silahkan melakukan registrasi pada wensite asmicbs2018. Jika terdapat kendala dan terdapat hal yang masih belum jelas. Silahkan langsung menghubungi kami.

10/5/21, 3:04 PM

Gmail - Konfirmasi keikutsertaan kegiatan ASMICBS 2018 Malang

Best Regards

Windy Y.B

[Quoted text hidden]

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Local Committee

***3rd Annual Scientific Meeting of Indonesian Consortium for Biomedical Sciences
(ASMICBS) 2018***

Master Program in Biomedical Sciences

1st Floor, Graha Medika (C) Building, Faculty of Medicine, Universitas Brawijaya

Jl. Veteran Malang, East Java, Indonesia, 65145

3rd Annual Scientific Meeting of Indonesian Consortium for Biomedical Sciences



Pre-Conference Workshop

Will be Held at **Faculty of Medicine, Universitas Brawijaya,**
21st November 2018. We will conduct **3 kinds of workshops :**

1. Bioinformatics , 2. Nanomedicine, 3. Isolation-purification of natural resources and Flowcytometry.

Conference Schedule

Venue : Savana Hotel (Jl. Letjen Sutoyo No. 79 Malang)

Important Date :

1. **Abstract submission, 26th October 2018**
2. **Conference : 22nd -23rd November 2018**

Registration : <http://fk.ub.ac.id/asmicbs2018/en/registration/>

1

CONTACT PERSON

Husnul : 08113646739
Yayan : 081334756682:
asmicbs2018@gmail.com

2

CONFERENCE FEE

Early bird : 600K IDR
After 1 November 2018
: 800K IDR

3

WORKSHOP

WS I or II : 1000K IDR
WS III : 1500 IDR
Before 10th November
2018



Dr. dr. Ina Rosalina,
SpA(K), MKes,
MH.Kes

Director of
Traditional Health
Services



Prof. Mikio Nishizawa, MD.
PhD

Dept. of Biomedical
Sciences,
Ritsumeikan
University, Japan



Sharida Fakurazi, PhD

Faculty of Medicine
and Health
Sciences, University
Putra Malaysia

Conference Schedule, Day I, 22nd November 2018

Time	Agenda	PIC
07.30 - 08.00	Registration	Committee
08.00 - 08.30	Speech and Opening Ceremony	
	Dean Faculty of Medicine UB	Dr. dr. Sri Andarini, MKes
	Chair Person of KIBI	Dr. rer. physiol. dr. Septelia Inawati Wanandi
	Rector of UB	Prof. Dr. Ir. Nuhfil Hanani, MS
08.30 - 09.00	Keynote Speech : Herbal Medicine in Indonesia	Dr. dr. Ina Rosalina D, Sp.A(K) (Director of Traditional Health Services, Ministry of Health Republic of Indonesia)
09.00 - 09.15	Coffee break	Committee
	Session 1	Moderator : Prof. Dr. dr. Loeki Enggar Fitri, MKes, SpParK
09.15 - 09.45	The Role of Natural Resources on Cytokines production for Drugs Development	Prof. Mikio Nishizawa, PhD (Ritsumeikan University, Japan)
09.45 - 10.15	Nano-medicine in Biomedical Sciences	Sharida Fakurazi, PhD (University Putra Malaysia)
10.15 - 10.45	Discussion	
	Session II	Moderator : Prof. Dr. dr. Soemarno, DMM., SpMK(K)
10.45 - 11.15	Bioinformatics for Screening and Prediction Pathways and Diagnosis of Diseases	Setia Permana, SSI, PhD (Polytechnic of Statistics - STIS Jakarta)
11.15 - 11.45	Herbal for SLE	Prof. Dr. dr. Kusworini, MKes (Universitas Brawijaya)
11.45 - 12.15	Discussion	
12.15 - 13.15	Break	Committee
13.14 - 16.00	Oral and Poster Presentation	Committee

Conference Schedule, Day II, 23rd November 2018

Time	Agenda	PIC
07.30 - 08.00	Registration	Committee
	Session I	Moderator : Dr. Drs. Swi Winarsih, Apt. MS
08.00 - 08.30	Design and Characteristics of Nano-medicine	Dr. rer.nat. Fredy Haryanto, SSI, MSi (Institut Teknologi Bandung)
08.30 - 09.00	The Formulation of Herbal Medicine for Cancer	Prof. Dr. Djoko Agus Purwanto, Apt, MSi (Universitas Airlangga)
09.00 - 09.20	Discussion	
09.20 - 09.30	Coffee break	Committee
	Session II	Moderator : Dr. rer.nat. Tri Yudhani MR, MSc
09.30 - 10.00	Herbal for Malaria	Prof. Dr. dr. Loeki EF., MKes, SpParK (Universitas Brawijaya)
10.00 - 10.30	Physalis for Hypertension	Dr. drg. Nur Permatasari, MS (Universitas Brawijaya)
10.30 - 11.00	Herbal for Cardiovasculer	dr. M. Saifur Rohman, PhD, SpJP, FICA (Universitas Brawijaya)
11.00 - 11.30	Discussion	
11.30 - 12.30	Break	Comittee
12.30 - 15.00	Oral Presentation	
15.00 - 15.20	Closing Ceremony	Committe

Rakernas KIBI : 23rd November 2018 at Hotel Savana

Supported by :



BUCHI Corporation

