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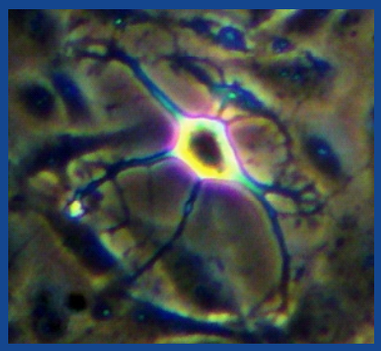

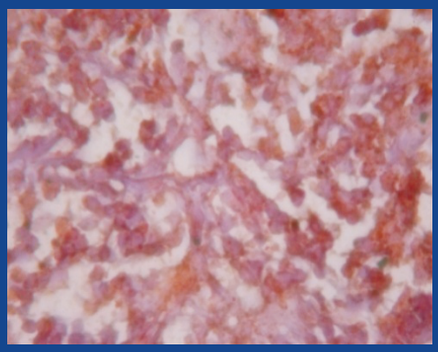

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Research Report

MOLECULAR SURVEILLANCE OF DENGUE VIRUS SEROTYPE USING POLYMERASE CHAIN REACTION IN SURABAYA 2013

Teguh Hari Sucipto[1], Amaliah Labiqah[1], Siti Churrotin[1], Nur Laila Fitriati Ahwanah[1], Kris Cahyo Mulyatno[1], Soegeng Soegijanto[1], Tomohiro Kotaki[2], Masanori Kameoka[2], Eiji Konishi[2]
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ABSTRACT

Dengue is one of the infectious diseases which is endemic in the tropical and sub-tropical country. The disease found in Indonesia Surabaya, 1968. The symptoms of Dengue virus infections are two kinds, first DF (Dengue Fever), second DHF (Dengue Hemorrhagic Fever). This infectious disease transmitted by Aedes aegypti mosquito. Mosquitoes breed in clean water areas. More than 100,000 cases of DF/DHF occurred in Indonesia every year. The purpose of this study were to provide information and the spread of dengue virus types in Surabaya from January 2013 to September 2013. The analysis technique used to determine the type of dengue virus infection was used PCR (Polymerase Chain Reaction). The results obtained 69% DENV-1, 27% DENV-2 isolates, 4% isolates DENV-3, and 0% DENV-4 isolates.

Key words: *Dengue, DENV, Surabaya 2013, Polymerase Chain Reaction*

ABSTRAK

Dengue merupakan salah satu penyakit infeksi yang endemik di daerah tropis dan sub tropis. Penyakit ini ditemukan di Indonesia pada tahun 1968 tepatnya di kota Surabaya. Gejala infeksi virus Dengue ada 2 macam, yaitu DF (Dengue Fever) dan DHF (Dengue Hemorrhagic Fever). Penyakit infeksi ini ditularkan melalui nyamuk Aedes aegypti, nyamuk ini berkembang biak pada daerah air yang bersih. Lebih dari 100.000 kasus DF/DHF terjadi di Indonesia setiap tahunnya. Tujuan dari penelitian ini adalah untuk memberikan informasi persebaran dan tipe virus dengue yang ada di Surabaya pada periode Januari 2013 sampai dengan September 2013. Teknik analisis yang digunakan untuk menentukan tipe infeksi virus dengue adalah menggunakan PCR (Polymerase Chain Reaction). Hasil isolat yang diperoleh 69% DENV-1, isolat DENV-2 27%, isolat DENV-3 4%, dan isolat DENV-4 0%.

Kata kunci: *Dengue, DENV, Surabaya 2013, Polymerase Chain Reaction*

INTRODUCTION

Dengue fever (DF) is a kind of infectious diseases that is distributed in the tropical and sub-tropical country.[1] This infectious disease is transmitted by *Aedes aegypti* mosquitoes, mosquitoes breed in clean water areas. This infectious diseasehas been found in 18 and 19 centuries ago.[2] Later, in 1953–1954 has been reported that the presence of DHF (Dengue Hemorrhagic Fever) in Manila Philippines. In, 1958 was found in Bangkok Thailand. In the 1960's has been found in Malaysia, Singapore, and Vietnam. This was due to the increasing influence of geographical and the density of *Aedes aegypti*.[3]

Dengue virus infection in human may be subclinical and clinical, with mild symptoms such as fever / flu - like syndrome or Dengue Fever (DF).[4] Dengue Fever are limited and rarely fatal. However, it is becoming high risk if the infection develops into Dengue Hemorrhagic Fever (DHF) or Dengue Shock Syndrome (DSS) can turn into death. Hemorrhagic Fever (DHF) is caused by vascular permeability that is characterized by capillary leakage, thrombocytopenia and hypovolamic shock.[5]

In Indonesia, Dengue Hemorrhagic Fever (DHF) occurred for the first time as an outbreak in Surabaya, in 1968. Dengue fever has spread to all regions of the province with the number of cities infected were increasing. More

than 250,000-500,000 cases of DF / DHF occurred in the world each year.[6] Genetically, there are 4 types of dengue viruses, they are DENV-1, DENV-2, DENV-3 and DENV-4. Until now, has not been found effective antivirus for dengue.

The purpose of this study was to provide information and the spread of dengue virus types in Surabaya from January 2013 to September 2013. The analysis technique used to determine the types of dengue virus infection was used PCR (Polymerase Chain Reaction).

MATERIALS AND METHODS

Population Sample

Epidemiological studies has conducted in Surabaya, East Java. Samplings were conducted at Soerya Children's Hospital and Maternity Sepanjang Sidoarjo accompanied by a certificate of Ethics from LPPM (Institute for Research and Community Service) Airlangga University. 800 serum samples were obtained. The study populations were all patients with Dengue Hemorrhagic Fever (DHF) which met the sample criteria according to WHO (World Health Organization) 2009.

Collection of Samples and the Diagnosis of Dengue

Blood samples were taken from DENV - infected patients diagnosed with IgG and IgM examination. Diagnosis based on WHO 2009 criteria[7] consists of clinical and laboratory criteria were as follows, sudden high fever, with no apparent reason, lasted continuously for 2–7 days, there were manifestations of bleeding, including testing positive tourniqued, petechical, ecchymosis, epistaxis, bleeding gums, and haemasthasis or malena, liver enlargement, shock, marked rapid and weak pulse, and pulse pressure, hypotension, feet and hands cold, moist skin and the patient was restless. Based on laboratory criteria, thrombocytopenia (100.000/mm^3 or less), haemoconcentration, can be seen from the increase in hematocrit of 20% or more, according to age and type of gender, or a decrease in hematocrit of 20% after fluid therapy.

Sampling operational procedures were as follows, the doctor asked the willingness to research subjects to participate. If the doctor was willing to coordinate with the hospital laboratory personnel to take the patient's blood, hospital laboratory personnel take as much as 8–10 cc blood of patients put in the tube on ice, prepared the shipment of samples to researcher at Airlangga University in LPT, then the sample were being examined at LPT Airlangga University.

Processing of Samples

Serum used for viral culture, formed after the CPE (Cytopathic effect) by viral infected examination of molecular biology ranging from the extraction of RNA, RT-PCR examination, and PCR using primers specific to determine the serotype of the virus.

Isolation of Virus

Blood serum taken from patients, inoculated into cell cultures (vero cell). Cells were grown for 2–3 passages, the first time the passage of time was 5–7 days. After positive cells showed CPE or infected, the culture fluid were collected and molecular biological examination can be done.

Extraction of RNA

RNA extraction using Trizol solution, 200 mL liquid culture, then added a solution of 800 mL Trizol, then mixed with pipette for several times and incubated in the mixture and incubated at room temperature. 200 mL chloroform was added to the mixture and allowed to stand at room temperature for 5 minutes, then centrifuged 12,000 rpm for 15 min at 4ºC. 500 mL of supernatant was taken and then added 500 mL 2 - propanol as much into a new Eppendorf tube, vortex and left at room temperature for 10 minutes. Centrifuge was repeated 12,000 rpm for 10 min at 4ºC. Supernatant layer then removed slowly by pipette carefully so that the RNA formed is not fetched. Ethanol 70% in increments of 1 mL was added to the sediment above the vortex and in this phase deposition can be saved or resumed by a centrifuge 12,000 rpm for 10 min at 4ºC. Disposal repeated supernatant layer was then dried with a vacuum pump for 10 minutes. Pellet suspended with DW as much as 10 mL, ready for further examination cDNA synthesis using (Reverse Transcriptase Polymerase Chain Reaction) RT-PCR method and PCR method for DNA synthesis.

Synthesis of DNA by PCR

Reactions for cDNA synthesis using TS primer (type-specific) is TS1 (5´-CGTCTCAGTGATCCGGGGG-3´), TS2 (5´-CGCCACAAGGGCCATGAACAG-3´), TS3 (5´-TAACATCATCATCATGAG ACAGAGC-3´), and TS4 (5´-CTCTGT TGTCTTAAACAAGAGA-3´), and then incubation for 3 phase, phase 1 were incubated at 65ºC for 5 minutes. Phase 2, incubated at 50ºC for 45 minutes and incubated at 85ºC for 5 minutes. Then, phase 3 incubated 37ºC for 20 minutes.

Doing PCR using primers D1 (5´-TCAATATGCTGAAACGCGAGAA-3´) and TS (type-specific) is TS1, TS2, TS3, and TS4 together in a 1.5 mL Eppendorf tube. Principles of PCR consisted of three stages: denaturation of double-stranded DNA, subsequent annealing (annealing) primer on the target DNA, primer extension last (primer elongation) at the presence of DNA polymerase. The results of DNA that occurs was ex-potential accumulation of the specific target DNA, approximately 2^n where n is the number of cycles set in the PCR process. At this stage PCR was performed in 35 cycles, the temperature and time used for denaturation 94°C for 30 s, annealing 55ºC for 60 sec and extension 72ºC for 2 min.

Electrophoresis

Validation was the process of DNA replication can be done with electrophoresis gel using ethidium bromide

which has been given to tracer tape from serotype to be searched.

RESULTS AND DISCUSSION

Virus isolation resulted from the sample collection from Soerya Children's Hospital and Maternity Sepanjang Sidoarjo from January 2013 to September 2013 were 68 virus isolates obtained from total of 800 serum samples.

In January obtained 34 isolates of dengue viruses which consists 31 isolates DENV-1, 1 isolate of DENV-2, and 2 isolates of DENV-3, in January 2013 rainfall was very high so dengue virus isolates obtained higher than other months (February to September) 2013. Due to higher rainfall was a good time for *Aedes Aegypti* mosquitoes to breed. From February to June, 16 isolates obtained (February DENV-3 by 1 isolates, March DENV-1 as 6 isolates, 4 isolates as DENV-1 April, May DENV-1 3 isolates, and June DENV-1 2 isolates) due to rainfall was very low in so the decline of dengue vector breeding. On July, 12 isolates obtained DENV-2, while on Aug. 5 isolates obtained DENV-2, and in September 1 isolates obtained DENV-2.

In figure 2 it can be seen that every month in 2013 (from January to September) there were cases of dengue and there are many different types of dengue virus every month. Dengue Virus Type 1 (DENV-1), from January to September 2013 still dominate as before. Based on research conducted by Yamanaka (2011) DENV-1 type of dengue virus was dominated since 2009 to 2010.[8] As well as in 2011 and 2012, has done research that DENV-1 obtained 90.3% and 93.2% but this result was still in the process of publication. This caused by a secondary infection that occurs when a carrier mechanism and progression of the virus increases immunity. Figure 4 was the result of electrophoresis showed positive samples of fragments coding genes NS1 protein of infected DENV-1 (band at 482 bp).

Dengue virus type 2 (DENV-2), figure 3 was the result of electrophoresis showed positive samples of fragments coding genes NS1 protein of infected DENV-2 (band at 119 bp) using agarose gel 1.5% and DNA ladder for marker, in July, August, and September dominates like in 2007, specifically in April, 53 isolates were obtained. From June 2008 to April 2009 there were 68% infections due to DENV-2.[8] Dengue virus type 2 (DENV-2) appeared back in July, August, and September 2013 did not find the results

Table 1. Result of Band Electrophoresis (bp)

No.	Type of Dengue Virus	bp
1	DENV-1	482
2	DENV-2	119
3	DENV-3	290
4	DENV-4	392

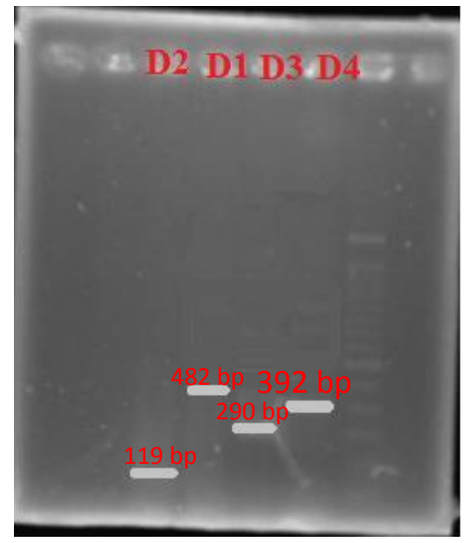

Figure 1. Serotype DENV

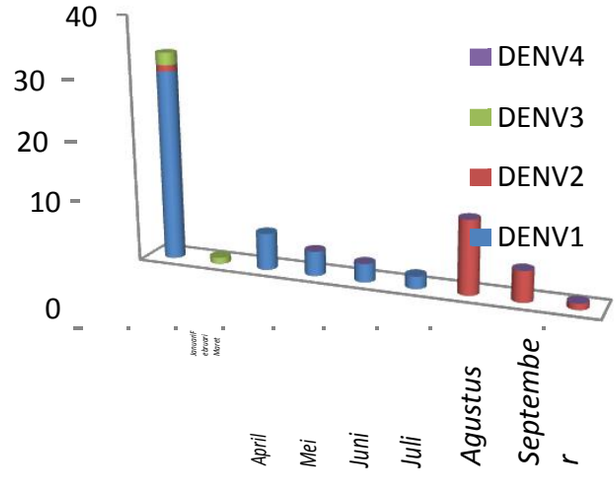

Figure 2. Diagram of Dengue Virus Isolates in Surabaya 2013

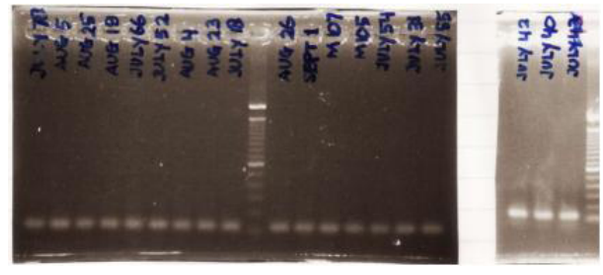

Figure 3. The results of electrophoresis of DNA fragments coding genes NS1 protein of dengue virus serotype 2 (DENV-2) at position 119 bp.

Figure 4. The results of electrophoresis of DNA fragments coding genes NS1 protein of dengue virus serotype 1 (DENV-1) at position 482 bp

of dominance (the previous month). Was this indicate a change of dominance back of dengue virus type 1 to type 2 as it happened in 2008 to 2010?

Dengue Virus Type 3 (DENV-3), for so long has been conducted research at the Institute of Tropical Disease, Airlangga University from 2007 to 2012 has never gained DENV 3 isolates. However, in January 2013 gained 2 isolates DENV-3 and February 2013 1 isolates obtained DENV-3, it has been published by Kotaki (2013).[9] In 2008, Ong et al., has published the same thing, namely the Jakarta DENV-3 of samples collection.10 At the time of the 69 samples obtained 15 isolates, these isolates comprised 10 isolates DENV-3, two isolates DENV-2, two isolates DENV-4, and 1 isolates DENV-1. Based on these publications it can be seen that actually has been dominated DENV-3 infection. Thus, it was possible that this DENV-3 cycle will return again and especially in Surabaya.

Dengue Virus Type 4 (DENV-4), no DENV-4 isolates obtained from January to September 2013. The possibility of this was due to declining circulation and spreading viral evolution.

CONCLUSION

DENV 1 dominance in Surabaya (from January 2013 to September 2013) was still ranked first, followed by DENV 2, DENV 3 gained 2 isolates in January 2013 and 1 isolate of DENV 3 in February. Not obtained isolat DENV 4.

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Research Report

THE PHOTODYNAMIC EFFECT OF LED-MAGNETIC EXPOSURE TO PHOTOINACTIVATION OF AEROBIC PHOTOSYNTETIC BACTERIA

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ABSTRACT

All photosynthetic bacteria have a major pigment of bacteriochlorophyl and accessor pigment e.g. the carotenoids, which both have an important role in photosynthesis process. This study aim to explore the exogenous organic photosensitizer from photosyntetic bacteria for photodynamic therapy application. This study is an experimental research aiming to test the potential illumination of LED with wavelength 409, 430, 528 and 629 nm, and power optimization and time exposure LED-magnetic for optimum photo activation Rhodococcus growth. The reseach design use a factorial completely randomized design with factor of power and exposure time. The number of bacterial colonies grown measure using of Total Plate Count (TPC) methods. The result of anova test shows that irradiation treatment with LED 409 nm, 430 nm, 528 nm and 629 nm significantly affects on bacterial colony growth. LED 409 nm exposure has the greatest potential to boost the growth of bacterial colonies by 77%. LED exposure and the addition of 1.8 mT magnetic field increases bacterial colony growth by 98%. Results of optimization of LED and magnetic fields show power 46 mW and a 40 minute (energy dose 110 J/cm^2) optimum growth of bacterial colonies increase by 184%. So LED and magnetic illumination has potentially increased the viability of an aerob photosyntetic bacteria colonies.

Key words: *photosynthetic bacteria, optimum energy dose, LED-magnetic, Rhodococcus*

ABSTRAK

Semua bakteri fotosintetik memiliki pigmen mayor yaitu bakterioklorofil dan pigemen asesoris seperti karotenoid, yang memiliki peran penting dalam proses fotosintesis. Penelitian ini bertujuan untuk mengeksplorasi eksogen fotosensitiser organic dari bakteri fotosintetik untuk aplikasi terapi fotodinamik. Penelitian ini merupakan penelitian eksperimental bertujuan untuk uji potensi iluminasi LED dengan panjang gelombang 409, 430, 528 dan 629 nm, dan optimasi daya dan lama waktu pemaparan LED-magnet fotoaktivasi pertumbuhan Rhodococcus. Desain penelitian ini menggunakan desain acak lengkap pola faktorial dengan faktor daya dan waktu pemaparan. Jumlah koloni bakteri yang tumbuh dihitung dengan menggunakan metode TPC. Hasil uji anova menunjukkan bahwa perlakuan penyinaran dengan LED 409, 430, 528 dan 629 nm berpengaruh signifikan terhadap pertumbuhan bakteri. Pemaparan LED 409 nm berpotensi terbesar untuk meningkatkan koloni bakteri 77% . Pemaparan LED-magnet meningkatkan pertumbuhan koloni bakteri 98%. Hasil optimasi LED-magnet menunjukkan daya 46 mW dan waktu 40 menit (dosis energi 110 J/cm^2) optimum meningkatkan pertumbuhan bakteri sebesar 184%. Jadi iluminasi LED dan magnet meningkatkan viabilitas koloni bakteri fotosintetik aerob.

Kata kunci: *bakteri fotosintetik, dosis energy optimum, LED-magnet, Rhodococcus*

INTRODUCTION

All photosynthetic bacteria have photosynthetic pigments that are sensitive to light (photosensitizer). The main pigment in photosynthetic bacteria is bacteriochlorophyl and accessory pigments which one carotenoid, which both have an important role in photosynthesis process. Bacteriochlorophyl have major role as a light harvesting which packaged in the form of first light harvesting (LH1) and second light harvesting (LH2) and as a charge separation in the form of the reaction center (RC)[1]. Carotenoid also have major role as same as bacteriochlorophyl that is as an accessory light-harvesting pigment and as a triplet quencher to provide protection again photooxidative damage[2]. That is the difference between absorbance spectrum when photophysical process.

Exposure light will be absorbed by the photosensitizer molecules in photosynthesis bacterial, light photon energy absorbance will excite the photosensitizer molecules in the singlet excitation and triplet. Excitation of photosensitizer molecules occurs only if of light photons spectrum correspond to the photosensitizer absorption spectrum. Subsequent excitation energy is transferred and converted into electrochemical potential energy in the form of transmembrane charge separation as well as the synthesis of adenosine triphosphate (ATP)[3] for the activation of photosynthetic bacteria. Amount of energy converted to ATP (photoactivation) depends on the number of photosensitizer molecules and the number of photons of light absorbed.

One of the photosynthetic pigment producing bacteria is *Rhodococcus*. The *Rhodococcus* include Eubacteria subkingdom members, a group of bacteria autotrophs (able to make their own food from inorganic substances), have chlorophyl and is able to photosynthesize like plants. *Rhodococcus* have chlorophyl pigment (green), carotenoids (orange) and pigment phicobilin consisting of phycocyanin (blue) and phicoeritin (red). Combine of these pigments produce the color to be turquoise. Cell wall contain peptides, hemicellulose and cellulose, and have a slimy membrane. These bacteria use two photosystems to split water and produce oxygen as a byproduct. These bacteria like to live in fresh water, but there are some that live in the sea.[4]

This study is an experimental research laboratory, aimed to determine the potential irradiation of LED Purple 409, LED Blue 430 nm, LED Green 528 nm and LED red 629 nm and 1.8 mT magnetic field for Rhodococcus growth activation as well as effectivel dose energy optimization of LED-magnetic for photoactivation. Giving of 1.8 mT magnetic field from a bar magnet aims to increase the biosynthesis of photosensitizer molecules, thereby increasing the amount of light-absorbing molecules.

MATERIAL

The Sample *Rhodococcus* bacteria were isolated from water river Mas, Surabaya. The Bacteria are grown on the photosynthetic media (PMS).

Irradiation Equipment

Irradiation equipment is a source LED light instrument, microcontroller, cervo motors, temperature sensor and LCD display. The Source LED light is used i.e. 409nm Purple, 430 nm Blue, 528 nm Green and 629 nm Red for exposure the in vitro bacterial. The type AVR 8535 microcontroller is used for setting an exposure time and power LED. The Paralax Continous servo motors is used for rotating bacterial petri dish on holder for flatten irradiation. The temperature sensor type LM35 is used for controlling the room temperature remains constant. The LCD display is used for showing given the pulse width modulation of irradiation and equipped by the timer running according to the length of time a given input, and the room temperature is detected by the temperature sensor. Before the LED instrument used for irradiation, temperature and time exposure calibration were done before.

METHODS

This study are prepared using a completely randomized design (RAL = Fully Randomized Design) factorial[5], which consists of two factors; A factor (irradiation power) with 4 levels (i.e. 17 mW, 28 mW, 34 mW and 46 mW) and B factor (exposure time) with 4 levels (i.e. 20 min, 30 min, 40 min and 50 min). Each treatment is accompanied by the control group using 3 times replication.

The Bacterial Growth

The Bacteria are grown on sterile PMS medium (photosynthetic medium) for 48 hours at 37°C temperature on the shaker incubator until the absorbance values obtained (OD) solution in 0.15 at 600 nm wavelength. The two ml of each bacterial sample is put in a sterile plastic dish diameter 3.5 cm and ready for irradiated.

Irradiation Bacteria by LED

Petri dishes containing bacteria are placed on the holder in the acrylic box above platform servo motors. The distance between the LED and the cup is permanently made in 2 cm. The source LED light are performed at various power and exposure time. Subsequently, the bacteria in the treatment group and the control are grown in PMS *agar* medium.

Counting the number of bacterial colonies

Samples are taken out from the incubator and counted the number of bacterial colonies growing by Total Plate count method using a Quebec Colony Counter. Next step, calculating the percentage would decrease the number of bacterial colonies that grow on each treatment using the equation:

| (ý Treatment colonies – ý control colonies)/ ý control colonies | × 100%

Statistical Analysis

Analysis of research data use SPSS statistical analysis (Statistical Package For Social Science) 13.0 for windows, i.e. factorial ANOVA for determining the effect of each factor and the interaction between factors. To show couple of different treatment groups, the analysis use multiple comparison test, on SPSS using Multiple Comparison Post Hoc5.

RESULTS AND DISCUSSION

The LED instrument has a temperature gauge and a time duration of irradiation designed with precision calibrator. Data of temperatur and time exposure calibration were showed in Table 1 and 2.

In LED instrument performance measurement, the time duration and temperature of LED exposure was set up by calibrator ENKO Sport Digital Stopwatch Timer and calibrators *Atech Thermo* L87AD (Figure 1). The regression graph of time duration and temperature LED instrument yield $R^2 = 1$ and $R^2 = 0.9995$ that means the LED instrument has a good performance.

Table 3 and 4 showed the percentage of bacterial colonies growth after LED exposure and with 1.8 mT magnetic field exposure.

Test results of the potential LED irradiation and 1.8 mT magnetic field exposure can be summarized in Table 4. The test results indicate that in LED exposure has potentially increase the number of *Rhodococcus* growth colonies at 77% and increase to 98% by the addition of 1.8 mT magnetic field exposure.

In Figure 3 and Figure 4 show that potentialy percentage reduction in the number of *Rhodococcus* colonies in 469 nm, 541 nm, 626 nm and 409 nm of LED exposure and with 1.8 mT magnetic field exposure.

Table 4 showed the percentage of bacterial colonies growth after LED exposure and with 1.8 mT magnetic field exposure in varying intensity and time exposure.

Figure 5 shows a graph of the percentage growth of *Rhodococcus* colony on a variety of power and duration time in LED irradiation and 1.8 mT magnetic field exposure. Exposure to 409 nm violet LED optimal *Rhodococcus* increase growth by 119% in power 28 mW and 60 minutes duration time exposure (dose 101 J/cm^2). Results of exposure optimization in 409 nm purple LED and 1.8 mT magnetic field shows the percentage of *Rhodococcus* growth by 184% in the power of 46 mW and 40minute duration time exposure (dose 110 J/cm^2). Exposure to

Table 1. Time exposure calibration data of LED instrument

time (s)	Time of LED instrumen (s)										tmean	Time of calibrator(s)
	1	2	3	4	5	6	7	8	9	10		
300	300	300	300	300	300	300	300	300	300	300	300	300
600	600	600	600	600	600	600	600	600	600	600	600	600
900	900	900	900	900	900	900	900	900	900	900	900	900
1200	1200	1200	1200	1200	1200	1200	1200	1200	1200	1200	1200	1200

Table 2. Temperatur calibration data of LED instrument

Temperature (ºC)	Temperature of LED instrumen (ºC)										T mean	ðT	Calibrator temperature
	1	2	3	4	5	6	7	8	9	10			
21	20	20	20	21	21	21	21	21	21	21	20,7	0,48	21
22	22	22	21	21	22	22	22	22	22	22	21,8	0,42	22
23	23	23	22	24	22	23	23	23	23	23	22,9	0,57	23
24	23	23	24	24	24	24	24	24	24	24	23,8	0,67	24
25	25	25	25	25	24	25	25	25	25	25	24,9	0,32	25
26	26	26	26	25	25	26	26	26	26	26	25,8	0,42	26
27	27	27	27	28	27	27	26	27	27	27	27	0,47	27
28	28	28	28	28	27	28	28	28	28	28	27,9	0,32	28
29	29	29	28	28	29	29	29	29	29	30	28,9	0,57	29
30	30	30	30	30	30	30	30	31	30	30	30,1	0,32	30

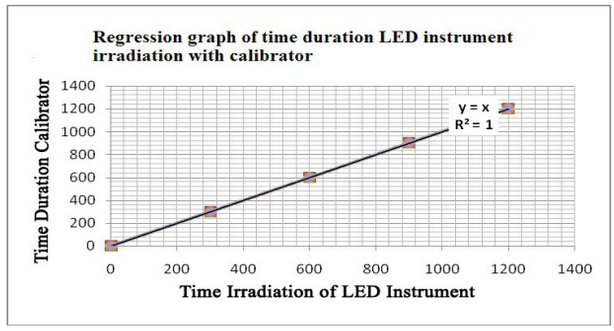

Figure 1. Regression graph of time duration LED instrument irradiation with calibrator.

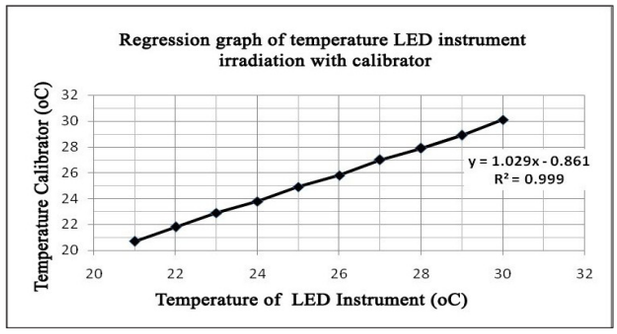

Figure 2. Regression graph of temperature LED instrument irradiation with calibrator.

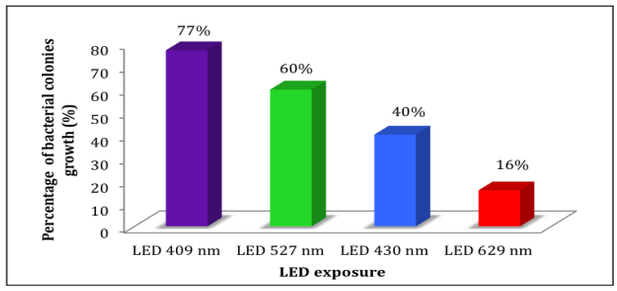

Figure 3. The Graph Percentage of *Rhodococcus* bacteria colonies growth in LED exposure.

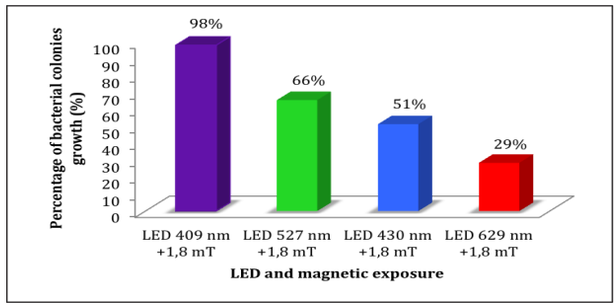

Figure 4. The Graph Percentage of *Rhodococcus* bacteria colonies growth in LED exposure and 1,8 mT magnetic field exposure.

Table 3. Potential of LED irradiation and 1.8 mT magnetic field exposure on the percentage growth of photosynthetic bacteria

		Percentage of Bacterial Colonies Growth (%)
LED exposure	LED 409 nm	77
	LED 527 nm	60
	LED 430 nm	40
	LED 629 nm	16
LED and 1.8 mT magnetic exposure	LED 409 nm	98
	LED 527 nm	66
	LED 430 nm	51
	LED 629 nm	29

430 nm blue LED and 1.8 mT magnetic field *Rhodobacteria* optimal increase by 111% growth in power of 34 mW and 50 minute duration time exposure (dose 102 J/cm^2).

One way ANOVA test results showed that there have influence of LED irradiation treatment 409 nm, 430 nm, 528 nm and 629 nm against *Rhodococcus* colony growth (Table 5).

Statistical test results of exposure energy dose optimization purple LED 409 nm and 1.8 mT magnetic field on *Rhodococcus* data show significance (p) = 0.624 > α (0.05), which means the data i.e. the percentage growth of bacterial colonies on 409 nm purlple LED irradiation normally distributed. The test results showed homogeneity (p) = 0.109 > α (0.05), which means homogeneous data. Factorial ANOVA test results showed that the power factor of 409 nm purple LED irradiation has a significance level (p) = 0.043 < α 0.05, which means that the radiation has an effect on the percentage growth in the number of *Rhodococcus* bacteria colonies. Summary of statistical test results are shown in Table 6 below.

Figure 5 shows a graph of the percentage growth of *Rhodococcus* colony on a variety of power and duration time in LED irradiation and 1.8 mT magnetic field exposure. Exposure to 409 nm violet LED optimal *Rhodococcus* increase growth by 119% in power 28 mW and 60 minutes duration time exposure (dose 101 J/cm^2). Results of exposure optimization in 409 nm purple LED and 1.8 mT magnetic field shows the percentage of *Rhodococcus* growth by 184% in the power of 46 mW and 40 minute duration time exposure (dose 110 J/cm^2). Exposure to 430 nm blue LED and 1.8 mT magnetic field *Rhodobacteria* optimal increase by 111% growth in power of 34 mW and 50 minute duration time exposure (dose 102 J/cm^2).

Table 4. The percentage of bacterial colonies growth after LED 406 nm and magnetic exposure in varying LED time and power exposure

Power time	Percentage of *Rhodococcus* bacteria colonies growth (%)			
	25 mW	46 mW	68 mW	93 mW
20 minutes	102	90	109	111
30 minutes	111	123	98	85
40 minutes	116	148	112	114
50 minutes	126	117	95	81

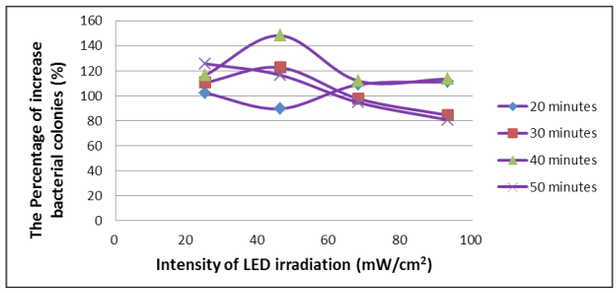

Figure 5. The Graph Percentage of Increase *Rhodococcus* bacteria colonies in 409 nm purple LED irradiation and 1,8 mT magnetic field exposure.

All photosynthetic bacteria have main pigment such as bacteriochlorophyll (BChl) and accessory pigments namely carotenoids. Both of these pigments have an important role in the process of photosynthesis. Bacteriochlorophyll is magnesium porphyrin which has more saturated tetrapyrrole ring. This Porphyrin causes BChl absorbs at a wavelength near-infrared around 620–700 nm[2]. The main role of Bacteriochlorophyll is as a light-harvesting and charge separation. Light-harvesting Bacteriochlorophyll is packaged in first light harvesting (LH1) and second light harvesting (LH2) complex, but charge separation Bacteriochlorophyll is packaged in the form of the reaction center (RC). Reaction center is in the middle of the circle of LH1 and functionally very closely related, so that the unity between LH1 and RC often called RC-LH1 core complex.

Light harvesting is also carried by several major carotenoid pigments that give main pigmentation at visible spectrum area, between 450 to 550 nm.[6] Carotenoids are long chain isoprenoid, usually the amount of carbon is 40. Carotenoids are synthesized from 8 isoprene units (C5), the bonds between the carbon system alternately (double and single). This double bond is able to absorb light.[15] Carotenoids have important roles as light-harvesting especially in environments with limited light conditions and photo-protector bacteriochlorophyll against excessive light. In addition, carotenoids also act to prevent photo oxidation, due to the presence of oxygen in photosynthesis.[11]

The mechanism of energy transfer on photosynthetic bacteria by using inductive resonance and delocation excited. In inductive resonance energy transfer occurred energy transfer. When the bacteria are exposed by appropriate spectrum to the light spectrum, photon absorption then occurs, followed by electron excitation of photosensitizer. The excited electron is unstable and will transfer its energy to another molecule before it fell to the ground state. On excited transfer energy mechanism, the excited electrons move from one molecule to another molecule. This movement is very short and it just occurred on adjacent molecules less than 2 nm at distance.

According Vermeglio and Joliot[12] photosynthesis of bacteria originated from the absorption of light by an antenna system which contains the chromophore such as bacteriochlorophyll and carotenoid polyenes. Singlet excitation energy is quickly transferred between the antenna chromophores, and finally to the reaction center. The role of the reaction center is changing the excitation energy into

Table 5. The result of One Way ANOVA tests for determining the effect of LED irradiation 409 nm, 430 nm, 528 nm and 629 nm

Bacterial Isolates	Group	N	Percentage increase in bacteria colony (%)		T test	
			Rate	SD	Significance	Conclusion
Rhodococcus by LED irradiation	**409 nm LED**	**5**	**76,8**	5,5	p = 0,000	There is a significant difference
	528 nm LED	5	59,8	6,4		
	430 nm LED	5	40,0	5,0		
	629 nm LED	5	15,8	2,7		
	Total	20	45,10	23,8		
Rhodococcus by 1,8 mT magnetic exposure	**409 nm LED**	**5**	**98,2**	12,8	p = 0,000	There is a significant difference
	528 nm LED	5	66,8	12,8		
	430 nm LED	5	51,4	10,1		
	629 nm LED	5	28,4	7,7		
	Total	20	60,95	27,9		

Tabel 6. Statistical test results of exposure energy dose optimization purple 409 nm LED and 1.8 mT magnetic field on *Rhodococcus* colonies

Factor	Group	N	Percentage increase in bacteria colony (%)		Anova	
			Means	SD	significance	Conclusion
Power	46 mW (a)	3	97,4	19,6	p = 0,000	There is a significant difference
	34 mW (a,b)	3	103,3	20,8		
	17 mW (a,b)	3	113,7	22,1		
	28 mW (b)	3	119,2	25,6		
	Total	12	108,4	23,1		

electrochemical energy in the form of a trans membrane charge separation.

The absorption of light is followed by electron transfer from bacteriopheophytin to bacteriochlorophylls, QA quinone, QB quinone and binds with hydrogen to form hydroquinone. Hydroquinone which is produced diffuses to the cytochrome bc complex, which is a trans membrane proton pump.[11] The next step is transferring electrons to cytochrome-c by realizing H+ ions. The energy released is used to transfer protons across the membrane and the resulting energy drives the synthesis of adenosine triphosphate (ATP) from adenosine diphosphate (ADP) and inorganic phosphate (Pi) with catalysis by ATP synthase.

Carotenoids are lipids so that this pigment is liposoluble (fat soluble) and soluble in nonpolar solvents.[13] Carotenoid pigments are very efficient in absorbing light at wavelengths (450–550 nm). When the carotenoid molecule is exposed by light, it will be excited to a certain energy level. Opportunities excitation energy levels that occur can be divided into two kinds, namely the singlet and triplet state. The function of carotenoids as photo protector occurs on triplet-triplet (TT) energy transfer and singlet-singlet energy transfer mechanism.[11]

When the light absorption, carotenoid excited to singlet state and immediately transfers the excitation energy to bacteriochlorophyll by using singlet-singlet energy transfer.[11] The process of singlet- singlet (SS) energy transfer is more common than the triplet-triplet (TT) energy transfer.[14] This is due to the energy transfer from carotenoids to bacteriochlorophyll does not take a long time and does not require too much energy to work in the process of photosynthesis so that the cycle of photosynthesis can take place. While the triplet-triplet energy transfer occurs at bacteriochlorophyll have excess energy so that the energy transfer to the carotenoid is happened.

Carotenoids can be excited to a triplet state, i.e. when it receives a transfer of energy from triplet-bacteriochlorophyll via triplet-triplet energy transfer mechanism[7]. The energy transfer happens in order to the carotenoid makes photo protection to bacteriochlorophyll. Photo protector carotenoids function is as a photo protector through suppression mechanism (quenching), either directly or indirectly.

In the directly extinction process, carotenoids receive energy transfer from triplet bacteriochlorophyll directly and disposed the excess energy in the form of heat (energy dissipation). The process of extinction (quenching) is indirectly done by carotenoids in a manner involving singlet oxygen. Singlet oxygen formed naturally from receiving oxygen triplet energy transfer from triplet bacteriochlorophyll. Singlet oxygen is radical. In the process of extinguishing, carotenoid accepts energy transfer from singlet oxygen, so the carotenoid gets the transition to the triplet state. Finally, the triplet carotenoids release the excess energy as heat.

Photophysical process also can be occurred in bacteriochlorophyl. But if bacteriochlorophyl absorb more photon energy, excitation state bacteriochlorophyl can be exchange to triplet state. An excited electron spin singlet S_n can be reversed, leaving the molecule in the excited state triplet T_n, called intersystem crossing[9]. Intersystem crossing probability increases if the lowest singlet of vibrational level experience overlap with one of the higher vibrational levels of the triplet state. The triplet state bacteriochlorophyl will interact with oxygen molecule so that produce reactive oxygen species (ROS). ROS is a toxic molecule so that make cell damage. In order to avoid such that carotenoids neutralize it with excitation singlet oxygen transfer energy to carotenoid so that carotenoid is at high vibrational levels of the triplet excited state and back to the ground state through transfer heat in environment, the mechanism is called triplet-triplet energy transfer. This energy is used to prevent photooxidation and photoprotection.[1]

Exposure to magnetic fields significantly influence the growth of bacterial colonies. Giving magnetic field causes stress on bacterial cells and activates genes ALA dehydratase (ALAD).[4] ALAD is a key enzyme of porphyrin biosynthesis.[10] Thus giving the magnetic field increases the synthesis of porphyrin photosensitizer in pigment-producing bacteria. Exposure to light will be absorbed by the photosensitizer molecules in bacterial photosynthesis, light energy is absorbed photon will excite the photosensitizer molecules. Further excitation energy is converted into electrochemical potential energy in the form of transmembrane charge separation and synthesis of adenosine triphosphate (ATP).[3]

According to the result, wavelength light having increase of the number colonies is 409 nm purple LED which it is accordance with maximum Soret absorbance. Photophysical process initiate the photochemical process.[8] Almost photophysical mechanism occurs in carotenoid. Photon light absorption by carotenoids will excite the molecule from the singlet ground electronic vibrational levels S_0 to one of the vibrational levels of the electronic excitation. Excitation of the molecule to the higher energy state is likely to return to the ground state, either through

chemical reactions or changes to the heat released into the environment in the process of internal conversion or vibrational relaxation. In the singlet excited state, the carotenoids transfer energy to Bacteriochlorophyls via singlet-singlet energy transfer.[6]

CONCLUSIONS

As briefly described in this article, the research results indicate that in 409 nm purple LED irradiation has potentially increased the number of *Rhodococcus* growth colonies at 77% and increase to 98% by the addition of 1.8 mT magnetic field exposure. Results of exposure optimization in 409 nm purple LED and 1.8 mT magnetic field shows the percentage of *Rhodococcus* growth by 184% in the power of 46 mW and 40 minute duration time exposure (dose 110 J/cm^2).

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Research Report

THE UTILIZATION OF *ACHATINA FULICA* MUCUS IN ALGINATE MEMBRANE AS WOUND HEALING ACCELERATOR AND ANTI-INFECTION MATERIAL

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ABSTRACT

Wound should be covered with bandage that is called wound dressing. Most people use synthetic materials such as gauze dressing. Gauze has high absorption of NaCl, which is often used to cleanse the wound. However, discomfort and pain arise since the gauze becomes sticky on the wound. Therefore, we need other alternatives instead of gauze to cover wound. One such alternative is the alginate membrane. This study used alginate membrane with mixture of mucous of the snail Achatina fulica, which contain proteins such as proline, serine asparagine, glycosaminoglycan, hydroxylysine, trionin and so forth, to activate the growth factor. Alginate powder and carboxymethl cellulose (CMC) was dissolved in distilled water mixed with mucus of the snail Achatina fulica in four variations (4:0; 4:1, 4:2, 4:3) through a magnetic stirrer, and casted on a baking sheet covered with sterile gauze. High Performance Liquid Chromatography (HPLC) test showed that the glycosaminoglycan content was found on the mucous of Achatina fulica. This was indicated by the appearance of peak at 325–350 second. The most optimum alginate and mucus composition was in ratio of 4:2. This ratio resulted in a wound dressing that was still able to absorb the exudate and optimally accelerated wound healing.

Key words: *alginate, Achatina fulica, wound healing accelerator*

ABSTRAK

Luka seharusnya ditutup dengan perban yang dinamakan dengan penutup luka. Banyak orang menggunakan material sintetik seperti penutup kasa. Kasa memiliki daya serap NaCl, yang biasa digunakan untuk membersihkan luka. Bagaimanapun, ketidaknyamanan dan rasa sakit timbul ketika kasa menjadi lengket pada luka. Oleh karena itu, kita membutuhkan alternative kasa untuk menutup luka. Salah satunya ialah membran alginat. Pada penelitian ini menggunakan membran alginat yang dicampur dengan lender bekicot Achatina fulica,yang mengandung protein seperti proline, serine asparagines, glycosaminoglycan, hydroxylysine, trionin, dan sebagainya untuk mengaktifkan growth factor. Bubuk alginat dan carboxymethl cellulose (CMC) dilararutkan pada air suling dan dicampur dengan lender bekicot Achatina fulica dengan empat variasi komposisi (4:0; 4:1, 4:2, 4:3) menggunakan magnetic stirrer dan diletakkan pada tempat oven dengan dilapisi kasa steril. Uji High Performance Liquid Chromatography (HPLC) menunjukkan bahwa kandungan glycosaminoglycan ditemukan pada lendir bekicot Achatina fulica. Hal diindikasikan dengan munculnya puncak pada detik 325-350. Alginat yang paling optimum dan dan lendir pada komposisi 4:2. Komposisi ini menghasilkan penutup luka yang tetap bisa menyerap nanah dan optimal dalam mempercepat penyembuhan luka.

Kata kunci*: alginate, Achatina fulica, percepatan penyembuhan luka, , glycosaminoglycan, carboxymethl cellulose*

INTRODUCTION

Traffic accident is one of the causes of high mortality in Indonesia. Based on data from the Jakarta Police Department, the number of accidents during 2010 reached 8059 cases, from which the number of people killed was as many as 1,032, seriously injured 3,429 people, and minor injuries 5,679 people. Data toward the end of 2011 stated, 8,468 victims of accidents in and around Jakarta, as many as 11.04% died. A total of 2,241 people were seriously injured and those with slight injury were as many as 5,292 people (62.49%).[1] Each accident victim requires treatment to his injuries. By default of wound management, the wound should be covered with a membrane or a bandage called the wound dressing. Usually people use a synthetic form of gauze dressing as wound dressings. The gauze is keeping the wound from the surrounding trauma. However, the gauze quickly absorbs NaCl which is previously used to wash wounds. This raises the patient's discomfort and worry if infection may occur due to sticky gauze on the wound.[2]

There are some research that conducted an experimental study comparing the use of conserved amnion with synthetic wound dressing material coated with gauze to cover circumcision wound in 16 children. From the results, it can be concluded that the use of conserved amnion as a circumcision wound closure is more effective in reducing pain when removing dressings circumcision and can reduce the risk of infection in treating circumcision wound, compared to the use of gauze coated synthetic materials on the outside. Besides gauze, wound dressings as amniotic membrane and membrane alginate have been widely used today.

Amnion has great benefits as a wound cover because it contains growth factors that help natural process of cell proliferation. However, not all pregnant women and their families allow to donate the placental membranes to take the amnion, so that the source of the amnion becomes very limited.[3] Therefore, we need another alternative as a substitute for the amnion to function as wound closure. One alternative is the alginate membrane. Currently, the widely used wound closure is pad or wick-shaped alginate. Imported foam products are typically that of hydrogel material. Based on studies, it is known that the foam or sponge can be made as alginate material that have a high absorption of wound containing liquid such as exudate. To accelerate wound healing, the membranes are usually given with medication or substance that could cause more active growth factor in human skin. The main element that can activate the growth factors are proteins such as proline, serine asparagine, hydroxylysine, trionin and so forth.[4] The material contained in one species *Achatina fulica* snail mucus.

During this time, snails are only be used as a food ingredient in the form of chips or sate. Moreover, villagers apparently use the mucus from these molluscs as toothache medicine. It is less hygienic. Therefore, we need the development of research on snail mucus to be used as a wound healer.[5] The combination of alginate, carboxymethyl cellulose (CMC) as a thickener, and snail mucus, is a solution to the needs of eco-friendly wound closure membrane and accelerate wound healing that is expected to reduce the incidence of wound infection and treatment costs.

MATERIALS

Materials used in this study were *Achatina fulica* snail mucus, sodium alginate with brand Sigma Aldrich 71238, and carboxymethyl cellulose (CMC) of Brataco technical types.

METHODS

Early Preparation

The first stage was the process of snail mucus snail mucus removal by preparing some snails from which the mucus would to be removed. Then, part of the shell was washed with water until clean. After that, the tip of its taper-shaped shell was cut about 0.5 cm.

Samples Preparation

Alginate membranes – snail mucus – carboxymethyl cellulose (CMC) was made from mixing powdered sodium alginate and CMC that had been diluted with a solvent, such as distilled water, and snail mucus. Each solution was mixed and stirred with a magnetic stirrer in order to be homogeneous. This solution was then casted on a round baking pan that has been coated with sterile gauze, and then stored in a deep freezer with a temperature range of –80°C to –100°C for ± 24 hours. After 24 hours in the freezer, the samples were removed and immediately lyophilized for 72 hours at a temperature of about –105°C and pressures in miliTorr. There were four variations of composition in this study using a total weight of 1% (Table 1).

Table 1. Composition Variations

Sampel	Alginate: Achatina fulica Mucus	Alginate (gr)	Distilled water solvent (gr)	Achatina fulica mucus (gr)
A	4 : 0	1.4	140	0
B	4 : 1	1.1	110	2.8
C	4 : 2	0.92	92	4.6
D	4 : 3	0.80	80	6.0

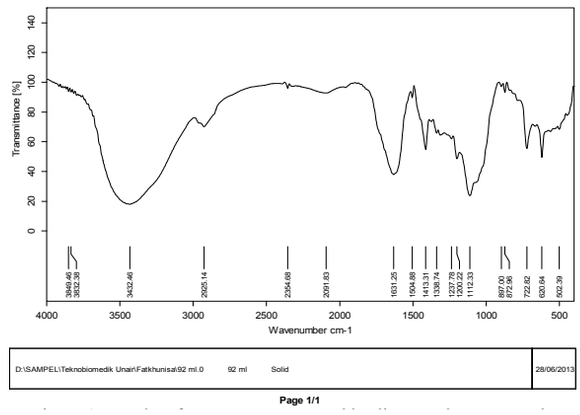

Figure 1. Results of FTIR Test on wound healing accelerator sample.

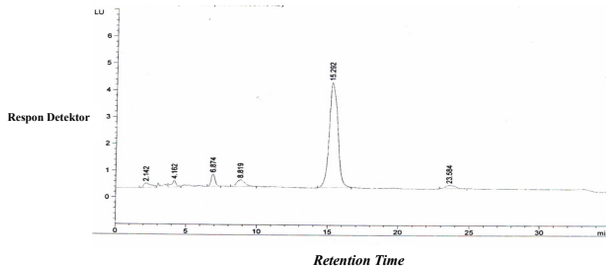

Figure 2. Spectrum of HPLC test results on *Achatina fulica* snail mucus.

Test Taxonomy Biological Characterization

Test taxonomy is the earliest process prior to the study as it aimed to get *Achatina fulica* snail species, according to the characteristics of the animal molluscs, based on accountable literature references.

Characterization of Fourier Transform Infra Red (FTIR)

Fourier Transform Infra Red (FTIR) test was used to determine the peak characteristics of functional groups described as transmittance curve (%) against wave number (cm^{-1}) on the material that has been made.

Characterization of High Performance Liquid Chromatography (HPLC)

To find out how much glycosaminoglican levels, one of the important growth factors activating proteins contained in the snail mucus wound closure as the main ingredient in accelerating wound healing.

Characterization of Anatomic Histopathology (AHP)

Wound healing and histology test were in vivo tests by observing the development of wound healing in mice (*Mus musculus*) macroscopically by the intensity of wound color, wound fluid, and wound type.[10]

Characterization of Antibacterial Test

This test used a disk diffusion method with *Staphylococcus aureus* and *E. coli* bacterial culture, whose inhibitory zone were analyzed. The qualitative disk diffusion test here used the colony units forming of 10^5 *E. coli* cultured on nutrient agar agent.

RESULTS AND DISCUSSION

Snails used in this research belonged to the phylum *Mollusca*, Classis *Gastropoda*, Order *Stylommatophora*, Familia *Achatinidae*, Genus *Achatina*, and species *Achatina fulica*. Snail shell length was 5.3 cm, it has smooth surface without ornament and has a color pattern of longitudinal stripes, alternating dark brown and yellowish white, the color lines are uneven edge, the dark brown part is wider than the white part. The shell is circular cone-shaped, the bottom coil is much larger than the other coil, and the concave coil has very clear boundaries. The shell appears strong and bold, but has very thin edge of the aperture, and the aperture is without cover.[6,7]

The result of Fourier Transform Infra Red (FTIR) test revealed a peak in the wave number 3750–3000 (showing O-H bond in alginate), 1900–1650 (showing C=O bond on the alginate), 1250–1050 (showing C-O bonds on the alginate) in accordance with existing references.[8]

High Performance Liquid Chromatography (HPLC) test showed that glycosaminoglycan content was found on the mucous of *Achatina fulica*. This finding is consistent with the literature, where the appearance of peak was at 165,477 in minutes.[9]

The final characteristic was the result of anatomic histopathology test conducted in mice (*Mus musculus*). The mice were given cut wound, and the healing process was observed through macroscopic test until the proliferative phase (around 13 days). Each sample group consisted of four mice. Parameters observed were the intensity of wound color, wound fluid, and wound type. The formed wounds contained liquid with a reddish color in each animal. The type of wound was open wound. On day 14, it could be seen that the composition of 4:2 was the most optimal composition in healing wounds since the reddish color in this group as a whole has faded, wound fluid in mice 1,3, 4 from a total of 4 mice had been absorbed by the wound healing accelerator membrane, whereas wounds in mice 1,2, and 4 had been completely closed. The next best composition was 4:3, then 4:1, and the last was a 4:0 or a group of negative samples. Through these observations, we observed that when the sample does not contain snail (*Achatina fulica*) mucus, the benefits of wound closing does not work et al.

The Antibacterial test results showed that from various concentrations used in tube dilution method, the concentration of 8% was the minimum dose of bacterial growth inhibition (MIC) and the concentration of 15% was the minimum concentration to kill the bacteria (MBC). Antibacterial test was only performed in a solution of 4:2 ratio since in macroscopic test the ratio had been proved as having most optimum wound healing.

Since the healing is known to be affected by tissue bacteria concentrations higher than 10^5 microorganisms per gram,[11] the use of dressing materials were able to reduce content of these microorganisms in surgical dermal wounds, as performed in this study, might be useful to avoid wound infection and, therefore, favor wound healing.[12] In addition, the release of angiogenesis-associated growth factors, such VEGF (vascular-endothelial growth factor) and PDGF (platelet-derived growth factor), after inflammatory chronification, is a key-step to the development of the granulation tissue during wound healing,[13] which could support our findings regarding to enhanced vascularization.

The composite of alginate, carboxymethyl cellulose (CMC) and *Achatina fulica* mucus can be produced in the early stages as an alternative wound dressings that have the potential of accelerating wound healing, absorbs excess exudate and prevent infection. From the above data it can be concluded that the most optimum composition of alginate and mucus are in 4:2 ratio. This comparison produces wound dressing that is still able to absorb exudate and optimally accelerate wound healing. In conclusion, we have demonstrated that the mucous secretion of *Achatina fulica* presents antibacterial properties. In addition, the use of dressing films based on this mucous secretion improved wound healing model.

CONCLUSION

The physical characteristics of accelerator wound healing membrane pore size are approximately 1,457-2,687 μm. The effect of Achatina fulica mucus as accelerator wound healing has been proved by the fact of faster healing process of wound compared with the group without the mucus.

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Literature Review

HIV AND MALARIA

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ABSTRACT

HIV/AIDS is a global problem involving industrialized and developing country including Indonesia. Malaria has killed millions of human beings almost 3 million people each year, whereas since 1999, nearly 36 million people in the world infected with HIV and 3 million more have died (Kakilaya, 2006). HIV infection increases the risk and aggravate malaria. In Africa in the area of malaria transmission intensities high and low, HIV aggravate malaria and improve case fatality at any age (Eline 2006). HIV is an RNA viruses whose hallmark is the reverse transcriptation of its genomic. Malaria is a protozoan disease transmitted by the bite of infected anopheles mosquito. Infection malaria can stimulate HIV replication and may cause faster progression of HIV disease.

Key words: *HIV, Malaria, infection, RNA, Progression*

ABSTRAK

HIV/AIDS merupakan masalah global yang meliputi industri dan negara berkembang termasuk Indonesia. Malaria telah membunuh jutaan manusia yaitu sekitar 3 juta orang tiap tahunnya, dimulai tahun 1999, 36 juta orang di dunia terinfeksi HIV dan lebih dari 3 juta telah meninggal[1]. Infeksi HIV dapat meningkatkan dan memperburuk malaria. Di Africa yang mana area transmisi malaria intensitasnya tinggi dan rendah, HIV memperburuk malaria dan meningkatkan kasus fatal di beberapa usia[2]. HIV adalah virus RNA yang ditandai dengan reverse transcriptation gennya. Malaria merupakan penyakit yang disebabkan oleh protozoa yang ditransmisikan oleh gigitan nyamuk anopheles yang terinfeksi. Infeksi malaria dapat menstimulasi replikasi HIV dan menyebabkan percepatan progesi penyakit HIV.

***Kata kunci**: HIV, Malaria, infeksi, RNA, perkembangan*

INTRODUCTION

HIV/AIDS is a global problem involving industrialized and developing country including Indonesia. The problem is growing increased rates pain and death. With respect to the decreasing immune from intervention HIV encourage micro growing organism one is a plasmodium malaria.

With technological progress digit HIV medicine supposed decline in pain in fact the pain remains high.

Malaria has been known for centuries, while the HIV in the last 2 decades. Malaria has killed millions of human beings almost 3 million people each year, whereas since 1999, nearly 36 million people in the world infected with HIV and 3 million more have died. Both of these diseases is expected to infect and kill a lot of people in the world because HIV was increased dramatically in countries with malaria are not controlled.[1]

HIV infection increases the risk and aggravate malaria. In Africa in the area of malaria transmission intensities high and low, HIV aggravate malaria and improve case fatality at any age.[2]

Has known that HIV reduce the number of lymphocytes, especially CD 4, so the immune response decline consequently micro an organism grows with fertile, one is malaria. Malaria while interference red blood cells and take glucose so happen deficiency nutrients that can result in immune deficiency. Malaria burdensome travel HIV infection being aids. On the basis of various fyl above necessary knowledge deep, mutual the relatedness of HIV and malaria so as to be done steps to reduce the rate in pain and death.

HIV

HIV aids first known in 1981 when acquired are sufferers by infection has opportunity without predisposing disorder of the immune system (Crowe, 2001; Fauci, 2005).Cause aids this is known as retrovirus divided into 2 groups namely human T lymphotropid viruses (HTLV) I and HTLV-II as "transforming retrovirus and human immunodeficiency viruses (HIV)-1 and

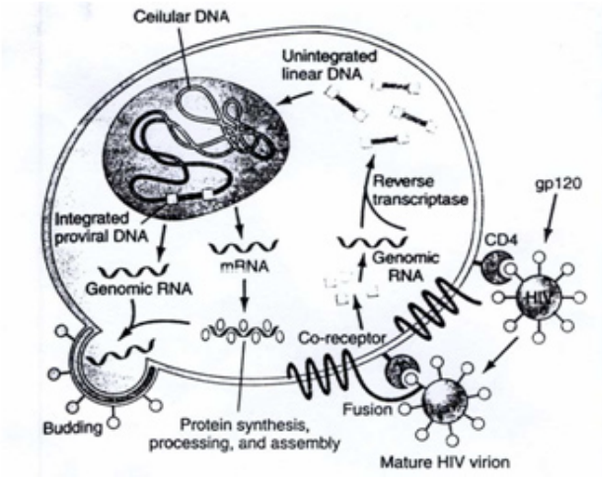


Figure 1. HIV replication cycle.

THE CYCLE OF HIV REPLICATION

HIV is the RNA virus that marked by a transcription inverted namely from RNA to DNA through enzyme reverse transcriptase. Started from the bonds protein gp120 receptor surface host namely molecules CD 4. Molecules CD4 is lymphocytes t that are responsible for helper T cells or induction the immune system. Cd4 expressed also at monocytes/macrophages and cells dendritic/langerhans. After the bond between gp120 with CD 4, gp120 will facilitate bond with co-receptor namely ccr5 and cxcr4. Bond with one or both these receptors will bring virus entering cells. Dendritic cell expressing receptors lectin type c different one of them is called dc-sign on surface that will make proteins gp120 HIV. Cell dendritic will facilitate binding virus T cell with CD 4+. After the ties it forms the surface virus will change, fusion to the cell membrane host would happen through penetration molecules gp 41 to a membrane plasma cells target. Capsule RNA viruses free and went into the cell target. Enzyme resvers transcriptase then catalyze RNA into double-strand DNA. DNA will hold translokation into the cell nucleus host and into the chromosome. Integration HIV this reactivated gene and produce provirus that may be latent or manifest at several levels genes expressions to a virus active. Next several levels activity cell host needed to start a transcription from proviral DNA into RNA or mRNA. MRNA HIV virus will be translation into protein were afterward subjected to modification through glicosilasion, miristilasion, fosforilasion, and division. The viral particles is formed of protein, enzyme, the genome RNA to membrane plasma of the host. Budding of virus happened through a special area in two layers of fat cell membranes host known as lipid rafts which will then be the outer layer of the virus.[3]

PATOFISIOLOGI AND PATOGENESE OF HIV

Sign of HIV is found immune deficiency caused by deficiency of lymphocytes t namely helper t cells or inductor T cells. CD4 that is part of t cells role as receptors of HIV. A co-receptor CD4 required to hold fusion and insert a virus hiv-1 into cells target for CCR5 and cxcr4. Although some mechanism responsible on decline and dysfunction immune T cells CD4+ have been demonstrated in vitro, currently it is not clear mechanism which most responsible on decreasing and impaired function in in vivo. When the number of T cells CD4+ declining the HIV have risk suffer hierograms disease opportunistic, several infections and a neoplasm (Fauci, 2005, O'neil, 2002).

Lysis of the virus and elimination directly caused by cellular immune response and humoral against viral is an important factor which contribute to the decreasing t cells are infected cd4 +. Allegedly that hiv infection cause chronic conditions of activity immune cells that cause elimination cd4 + uninfected. Mechanism is responsible for elimination cd4 + cell autoimmune and are not infected cell death.[4]

Lymphoid tissue constitutes the main hiv replication of the virus is also resulting in place lesions specific occurring imunodefisensi.[4]

TRANSMISSION OF HIV

HIV transmitted by contact heterosexual, homosexual, through blood and blood products, through mother to child is infected intra partum, perinatal or when breastfeeding. For research more than 20 years there is no evidence that HIV can be transmitted through the bite of an insect or propagated as mosquito.[3,5]

Like a capsule virus other all retroviruses inactive, easily into shape and not, is transmitted via air dust or smoke. Infection started having no direct contact through the tissue or body fluid from a source of infection.[6]

CLINICAL SYMPTOMS AND TREATMENT

The clinical and travel this disease correlated with cd4 number of cells. Divided into four degrees primary, namely infection where the virus in blood and proliferation quickly limfonodi, early deficiency immune/early (cell number CD4 > 500/μL), intermediate deficiency immune (CD4: 200–500/μL) and immune deficiency further/advanced (CD4 < 200/μL).[7]

Tabel 1. Clinical Stadium HIV WHO 2004 [3]

	Stadium 1 *asimtomatik*	**Stadium 2** *Mild disease*	**Stadium 3** *Moderate disease*	**Stadium 4** *Severe disease (AIDS)*
Weight	Tetap	BB ↓ 5–10%	BB↓ >10%	**HIV wasting syndrome**
A symptom of infection generally treatment and opportunistic infection according to the guideline and regulations	❖ Only limfa-denopati	❖ Wound around lips (angula cheilitis) ❖ Itchy (seborrhoea/ prurigo) ❖ Infection up track breathing, as sinusitis or otitis.	❖ Sprue (hairy leukoplakia) ❖ > 1 month • Diarrhea • Candidiasis vagina • Fever ❖ Infeksion of heavy bactery(pneumoni, infeksion muscle) ❖ TB lung in last year.	❖ Spure in esofagus ❖ > 1 month • Ulserasi herpes simpl ❖ Limfoma ❖ Kaposi sarcoma ❖ Ca cervix infasif ❖ Pneumocystis pneu ❖ TB ekstrapulmoner ❖ Meningitiscryptococcal ❖ Ensefalopati
ARV therapy	❖ Only if CD4 < 200	❖ Only if CD4 < 200 or limfosit total < 1200/mm3	❖ If CD4 not available start therapy. Start from CD4 < 350	❖ Further evaluation of patient for starting ARV.

ANTIRETROVIRAL THERAPHY OF HIV

Targets antiretroviral therapy (ARV) that inhibits bond with CD4 (in research) enzyme inhibit reverse trancriptase (*zidovudine, didanosine, zalcitabine, lamivudine, stavudine, foscamet*) a non-nucleoside reverse transcriptase inhibitor (*nevirapine*) termination of DNA (zidovudine, sintesa chain didanosine, zalcitabine, stop and budding assembly (viruses, interferon) hinder maturation protein virion core (a protease inhibitor: example saquinavir).[8]

It has proven that combination therapies more effective for viruses and impede progresivitation disease. Therapy for patients without major complications is d4T-3TC-NVP namely stavudine-lamivudine-nevirapine.[9]

MALARIA

Malaria is a disease due to protozoa that are transmitted through the bite of an infected anopheles mosquito. There were four genus infecting humans, a plasmodium namely *P. Tertian, P. Vivax, P. Ovale,* and *P. Malariae*. Infection began when a female an infected anopheles mosquito inoculation plasmodial sporozoites when suck the blood of humans. A microscopic malaria parasite would be taken quickly off the flow of blood to heart entering parenchyma liver and begin a period of asexual reproduction. Through a process of amplification (known as intrahepatic or preerytrocityc schizogony or merogony) a sporozoites can produce 10,000 until > 30,000 & merozoites. Liver swell will issue merozoites moving into the blood stream. Then went into the red blood cells and multiplication 6 until 20 times every 48–72 hours. Parasitic on sat reached–50/µL blood clinic symptom of infection this will seem (White, 2005). On *P. vivax* and *P. ovale* when phase intrahepatik not occurring cleavage immediately but is dormant between 3 weeks until a year or so before reproduction began, called hepaticae hypnozoit this condition allow parasitic adapted to climate change.[11,12] Upon entering blood flow merozoites entrance to erythrocytes and tropozoit be assisted by receptors surface erythrocytes specific during the early phases intra eritrosit notching the ring (ring forms) of four species parasitic looked same under a light microscope. As makin magnify tropozoit specific characteristic each species more real, pigment more visible, parasitic looked irregular or notching ameboid. After 48 hours the life cycle intraeritrosit (72 hours to *P. malariae* parasitic) has consume almost all hemoglobin and meet red blood cell (SDM), called schizont. Occurring splitting the nuclei of multiple and SDM shatterlng make 6–30 merozoites female that can mengivasi SDM new and cycle as above will recurring again. This disease in humans caused by any indirect effect of his invasion SDM, destructived by parasites phase asexual and reaction host. After cycle asexual (*P. Falciparum*) or immediately after out of liver (*P. Vivax. P. Ovale, P. Malariae*) some parasitic form morphology different namely sexual phases (gametosit) can anything to malaria. After sucked by the bite of an infected anopheles mosquito female, gametosit male and female form a zygote in intestines of mosquitoes. Which the zygote mature ookinet be entering and protected in the intestinal wall. Ookcyst formed from cleavage asexual continue to cleave to a sporozoites moving then migrate to hemolimf further to the salivary glands mosquito waiting inoculation into another human.[12]

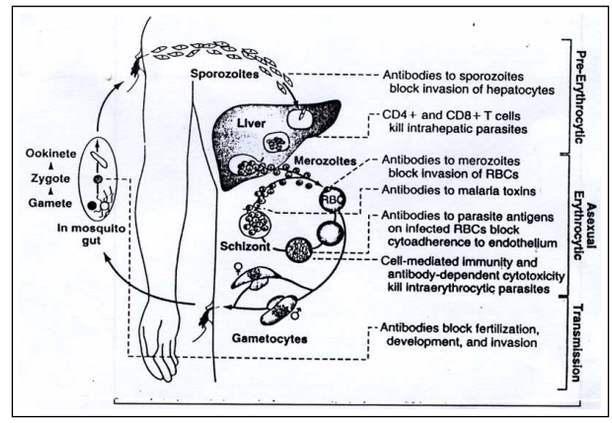
EPIDEMIOLOGY AND TRANSMISSION OF MALARIA

Malaria are found in tropic area. Africa, new guinea, Haiti dominant *P. Falciparum, P. vivax* more numerous in central America and parts of the south north Africa, middle east and zindian subcontinen. *P. Ovale* rare outside west african while *P. Malariae* found in many place.[12]

Malaria transmitted by several species mosquito anopheles. Transmission this is not happening in temperature under 160 c, or over 330 c and height of more than 2000 m. condition optimum is on moisture high with temperature between 20 and 300 c (White, 1996). An area that many obtained the gnat is marshes that deals with the high rate occurrence malaria. Stagnant water into one that support mosquito reproduction.[11]

Malaria can also occurring by sporadic in the non endemic. In example because of malaria latent relapse few months after traveling from the endemic. Sufferers as is generally not getting therapy complete or got kemoprofilaksis inadequate.[11]

Malaria also can be transmitted over blood transfusion, syringe or transplant organs. The incubation generally short having no stadium preeritrositik.[12]



Picture 2. The cycle of the transmission of malaria from the mosquito to humans.

Table 2. A manifestation of malaria clinics difficult because *P. falciparum*[3]

Signs	*Manifestations*
Major	
Unarousable coma / cerebral malaria	Failure to localize or respond appropriately to noxious stimuli; coma persisting for > 30 min after generalized convulsion
Acidemia/acidosis	Arterial pH < 7.25 or plasma bicarbonate level of < 15 mmol/L; venous lactate level of > 5 mmol/L manifests as labored deep breathing, often termed “respiratory distress”
Severe normochronic, normocytic anemia	Hematocrit of < 15% or hemoglobin level of < 50 g/L (< 5 g/dL) with parasitema of > 100,000/μL
Renal failure	Urine output (24h) of < 400 mL in adults of < 12 mL/kg in children; no improvement with rehydration; serum creatinine level of > 265 μmol/L (> 3.0 mg/dL)
Pulmonary edema / adult respiratory distress syndrome	Noncardiogenic pulmonary edema, often aggravated by overhydration
Hypoglycemia	Plasma glucose level of < 2.2 mmol/L (< 40 mg/dL)
Hypotension/shock	Systolic blood pressure of < 50 mmHg in children 1–5 years or < 80 mmHg in adults; core/skin temperature difference of > 10ºC
Bleeding / disseminated intravascular coagulation	Significant bleeding and hemorrhage from the gums, nose, and gastrointestinal tract and/or evidence of disseminated intravascular coagulation
Convulsions	More than two generalized seizures in 24 h
Hemoglobinuriaa	Macroscopic black, brown, or red urine; not associated with effects of oxidant drugs and red blood cell enzyme defects (such as G6PD deficiency)
Other	
Impaired consciousness	Obtunded but arousable
Extreme weakness	Prostration; inability to sit unaidedb
Hyperparasitemia	Parasitemia level of > 5% in nonimmune patients (> 20% in any patient)
Jaundice	Serum bilirubin level of > 50 mmol/L (> 3.0 mg/dL) if combined with other evidence of vital-organ disfunction

SYMPTOMS CLINIC

Malaria is one of the causes of fever in tropical countries. Early symptoms of malaria are non-specific, such as discomfort, headaches, tired, uneasy feeling in the stomach, muscle pain with heat is a symptom similar to some viral diseases. In some circumstances a very head aches, chest pain, abdominal pain, artralgia, myalgia or diarrhea may be due to other illnesses allegedly although headaches on malaria might be heavier. Nausea, vomiting and ortostatik hypotension often occurs as well. The classic symptoms of malaria, such as high heat, chills and stiffness that occurs on a regular basis appropriate intervals, relatively rare and is thought to be caused by *P. vivax* or *P. ovale*. Heat the irreguler on *P. falciparum* in patients with the declined immune and children often achieve above 40° C flutter, and sometimes accompanied by delirium. Seizures can also be caused by *P. falciparum* and is signified the presence of cerebral malaria.[12]

Found little physical examination teratology on patients without complication, malaria is hot malaise, anæmia light and in some cases spleen being palpable. Anemia more often resulted in children living in the transmission stable place that is resistant to partially parasitic chloroquin or other drugs. Enlarged spleen often resulted in patients in the endemic indicating the presence of infection repeated. Enlargement of the liver mild also found chiefly on small children. Mild jaundice associated with adults caused by *P. Tertian* and generally recovered after 1 to 3 weeks.[12]

MANAGEMENT

When a sufferers of an area that get malaria having symptoms heat, hapusan blood with drops thick and thin must be done to diagnose and determine species parasite that infects. Repet of hapusan blood to do at least every 12 to with 24 hours for two days, if hapusan first negative.As an alternative detection antigen or stick test assignment. Some medicines can be used orally and it depends on the sensitivity parasite that infects. Chloroquin is therapy options for malaria a tame *P. vivax*, namely *P. ovale*,

Table 3. Medicine which recomendation for antimalaria[12]

Drug	*Uncomplicated Malaria (Oral)*	*Severe Malaria[a] (Parenteral)*
Chloroquine[b]	10 mg of base/kg followed by 10 mg/kg at 24 h and 5 mg/kg at 48 h or by 5 m/kg at 12, 24, and 36 h (total dose, 25 mg/kg); for *P. vivax* or *P. ovale*, primaquine (0.25 mg of base/kg per day for 14 days[d]) added for radical cure	10 mg of base/kg by constant-rate infusion over 8 h followed by 15 mg/kg over 24 h *or* by 3.5 mg of base/kg by IM or SC injection every 6 h (total dose, 25 mg/kg)[c]
Amodiaquine[b]	15 mg of base/kg followed by 10 mg/kg per day at 24 and 48 h (total dose, 35 mg/kg)	–
Sulfadoxine/ pyrimethamine[b]	25/1.25 mg/kg, single oral dose (3 tablets for adults)	–
Mefloquine[b]	15 mg/kg followed 8-12 h later by second dose of 10 mg/kg	–
Quinine	10 mg of salt/kg q8h for 7 days combined with tetracycline[e] (4 mg/kg qid) or doxycycline (3 mg/kg once daily) or clindamycin (10 mg/kg bid) for 7 days	20 mg of salt/kg by IV infusion over 4 hf followed by 10 mg/kg infused over 2–8 h every 8 h
Quinidine gluconate	–	10 mg of base/kg by constant-rate infusion over 1–2 h followed by 0.02 mg/kg per min, with ECG monitoringg
Artesunate	In combination with 25 mg of mefloquine/kg, 12 mg/kg given in divided doses over 3–5 days (e.g., 4 mg/kg for 3 days or 4 mg/kg followed by 2 mg/kg per day for 4 days); if used alone or in combination with clindamycin or doxycycline, give for 7 days (usually 4 mg/kg initially followed by 2 mg/kg daily)	2.4 mg/kg IV or IM stat followed by 1.2 mg/kg at 12 and 24 h and then daily (or 2.4 mg/kg once daily)
Artemether	Same regimen as for artesunate	3.2 mg/kg IM stat followed by 1.6 mg/kg per day
Atovaquone-proguanil (Malarone)	For adults > 40 kg, each dose comprises 4 tablets (each tablet containing atovaquone 250 mg and proguanil 100 mg) taken once daily for 3 days with food	–
Artemether-lumefantrine	For adults š35 kg, each dose comprises 4 tablets (each tablet containing artemether 20 mg and lumefantrine 120 mg) at 0, 8, 24, 36, 48, and 60 h, taken after food	–

P. Malariae. Malaria on heavy anti arrhythmic quinidine gluconate replace quinine as malaria therapy in us. Discharging quinidine must with the monitor tight if there disritmia and to prevent from happening hypotension. Quinine safer than quinidine and has been much used in the world widely during malaria therapy heavy. In some area of chinese medicines derivable from artemesinin (artemether and artesunat) been first choice to malaria heavy.[12]

THE INFLUENCE OF HIV FOR MALARIA

Currently two health problems in africa HIV and malaria. Research on interaction between both still a little. HIV immunity so on could reduce malaria patients symptoms heavier. While malaria would accelerate HIV infection into AIDS (Chandramohan, 1998, Whitworth, 2005).

Immunologist mechanism will protect the infection. It can be achieved through destructive parasitic on phase preeritrositik in liver by of cytotoxic T cells and other mechanism that related. If this mechanism fail, parasitic will continue easily enter into the bloodstream, it will increase so that manifestation clinic was renewed and will overcome by an antibody in blood. T cells playing an important role in the immune response against malaria. CD4 cells will help production of antibodies against malaria and controls parasitemia by producing cytokines. On HIV with total CD4 decline will give effect on ability of body to form an immune response that is effective against malaria. Resulting from infection HIV in malaria will improve incident pain than healing, increase incident symptoms clinic compared infection asimtomatik, malaria and increase the weight compared light.[13]

THE INFLUENCE OF MALARIA FOR HIV

There is a possibility that cellular aktifasi resulting from infection pathogenic microorganisms will increase replication of the virus. Malaria is strong stimulators the immune system. Someone who exposed to malaria would increase the level of serum imunoglobulin, and occurring IgG acceleration of change. B cells activity excess this may be caused by some specific, the response to an antigen variant malaria and stimulation of non specific derivat a toxin or influential mitogen directly into b cells or through active T cells. The evidence active t cells that remains less strong than b cells but the inductions some immune response specific influence possible active helper t cells that recurs.[13]

There is evidence that function of T cells decline during acute phase episode malaria. Response proliferative are squeezed during acute episode of malaria. Malaria infections have an effect against HIV stimulated change T cells and and failure of function of cytotoxic T cells. Malaria infections also can undermine the placenta to make way for the transmission of HIV in utero.[13]

P. falciparum stimulates HIV replication through production of cytokines (IL-6 dan TNF-α) that is activated by lymphocytes. A study in Malawai, Africa shows that plasmatic viral loads HIV sufferers with higher on malaria compared with only HIV.[14]

THERAPY RESPONE AND INTERACTION OF MEDICINE

Therapy anti malarial will be effective on an individual that have had immunity against malaria. Estimated that response therapy will decline in individuals whose immunosuppressive because HIV infection and living on the transmission malaria stable. Recent observations estimate treatment with artemisin, sulfadoksin-pyrimetamine artemether-lumefantrine and less effective in people with with HIV. Interaction between an anti malarial drug and ARV most involving a protease inhibitor (PI) and nonnucleoside reverse transcriptase inhibitors (NNRTI). An anti malarial drug halofantrine, artemether lumefantrine and should not given by those with that off drugs PI (or NNRTI delavirdine) because will increase the risk toxicity. Among who wears NNRTI another (nevirapine or efavirenz) the lumefantrine and artemether concentration low will increase the risk failure therapy. Acquired also potential interaction between quinine and NNRTI or PI. However potential interaction between drugs is still needs far more research.[14]

Chloroquin commonly used as anti malaria caped inhibits the activity of antiviral namely interferon and beta alpha on experiment animals. Found also that chloroquine increase replication of the virus in mice tried.

Chloroquin often worn as chemotherapy malaria. (Kakkilaya, 2005) chloroquin have an effect immunosuppression and only effective in the central America and middle east. Prophylactic proper to malaria resistant chloroquin is mefloquin and doxycyclin (Rulf at, 1997). In one research farmakokinetik obtained mefloquin norvir lower levels in the blood up to 30%.[1]

Many patients HIV allergic to sulfonamide as pirymethamine-sulfadoxine (Fansidar). Granting doxycycline 100 mg/day for two days before traveling, during traveling and four after that have some benefits because besides can give prophylactic on malaria also in diarrhea and some other infections. Prevent mosquito bite, and diagnosis rapid and on complaint heat especially during and after passing to the regions with risk malaria is important for the hiv who travel.[14,15]

Combination therapy ARV has a great potential to lower hiv dealing with malaria. Prophylactic cotrimoxazol recommended for children and adults with HIV in africa and effective to relieve the symptoms clinic malaria.[2]

SUMMARY

HIV is an RNA viruses whose hallmark is the reverse transcriptation of its genomic. The hallmark of HIV disease is a profound immunodeficiency. Primarily, deficiency of the subset T lymphocytes, CD4, which serves the us primaries celullar receptor of HIV.

Malaria is a protozoan disease transmitted by the bite of infected anopheles mosquito. Four species of the genus a plasmodium cause malaria infection of human. These are *P. vivax, P. falciparum, P. ovale* and *P. malariae*. There are two cycles of a plasmodium life, sexual and asexual cycles which happen in human and female anopheles mosquito.

The association between the two infection has important implications. Malaria and HIV-1 are two most common infection in Sub-Saharan Africa, to a lesser extent, in other developing countries.

HIV-related immunosupression may increase malaria rates of infection and malaria clinical disease. Infection malaria can stimulate HIV replication and may cause faster progression of HIV disease. There are some anti malarias and drug interactions between antiretroviral drugs (ARV).

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Literature Review

CLINICAL DESCRIPTION AND DIAGNOSIS OF HIV/AIDS

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ABSTRACT

Infections of HIV/AIDS currently has become very serious problems for the world health. In the country the first case of HIV/AIDS was discovered in Bali in 1987, in its progress has not the meaning but after 1985 HIV transmission increased considerably. The complex problem that the living and the increasing number of cases should indeed, medical practitioners understand more the clinical and how to diagnose infections of HIV/AIDS. A snapshot of the clinical HIV infection/aids can be seen from grievances and a disease that often accompanies it, a complaint which is found at HIV/AIDS sufferers in the form of suds retroviral acute: fever, weight loss, diarrhea chronic, disphagi, limpadenopati, infections in the skin respiratory disorders and nervous breakdown center. While a disease that often been gained by those with HIV / AIDS as candidiasis, tuberculosis, pneumonia bakterialis, toksoplasmosis and pneumonia pneumocystic carinii. Diagnose HIV infection created based on clinical symptoms which includes major symptoms and symptoms of minor, and the result of the examination of the laboratory.

Key words: *HIV, AIDS, transmission, epidemiology, infectious*

ABSTRAK

Infeksi HIV/AIDS saat ini telah menjadi masalah yang serius bagi dunia kesehatan. Di Indonesia kasus HIV/AIDS pertama kali ditemukan di Bali tahun 1987, dalam perkembangannya tidak mengalami perkembangan yang berarti akan tetapi setelah tahun 1985 penyebaran HIV meningkat dengan tajam. Kompleknya permasalahan yang dihadapi ODHA dan semakin meningkatnya jumlah kasus ini perlu kiranya praktisi kesehatan lebih memahami gambaran klinis dan cara mendiagnosis infeksi HIV/AIDS. Gambaran klinis infeksi HIV/AIDS dapat dilihat dari keluhan dan penyakit yang sering menyertainya, keluhan yang sering ditemukan pada penderita HIV/AIDS berupa sindroma retroviral akut: demam, penurunan berat badan, diare kronis, Disphagi, limpadenopati, infeksi pada kulit, gangguan pernapasan dan gangguan saraf pusat. Sedangkan penyakit yang sering didapatkan pada penderita HIV/AIDS seperti kandidiasis, tuberkulosis, pneumonia bakterialis, toksoplasmosis dan pneumonia pneumocystic carinii. Diagnose infeksi HIV dibuat berdasarkan gejala klinis yang meliputi gejala mayor dan gejala minor, serta hasil pemeriksaan laboratorium.

Kata kunci: *HIV, AIDS, transmisi, epidemiologi, infeksi*

INTRODUCTION

HIV/AIDS infection currently has become very serious problems for the world health. AIDS first found in 1981, but identification of the virus that causes aids new found about 1983–1984.[1,2] This virus were given the name of the human immunodeficiency virus (HIV) that can be found the body fluid especially blood, a liquid sperm, vaginal discharge, and water milk mother. AIDS has been scattered more than 150 countries, until December 2000 58 million people estimated to be infected with HIV, 22 million have died, 3 million died the year 2000.Two thirds of the number of HIV found in the countries of Africa, part of Sahara Africa about 70% and in asia pacific more than 20%.16.000 world's people estimated to be infected with HIV in every day.[1,3,4]

In the country the first case of HIV/AIDS was discovered in Bali in 1987, in its progress has not the meaning but after 1985 HIV transmission increased considerably. Since 1999, there are new phenomenon of the spreading of HIV/AIDS cases included HIV infection started looking for a drug injection users or infecting drug users (IDU). In both groups

IDU happening quickly because of the use of hypodermic needles together. The 2000 increase the pandemics are explicitly through sex workers (dept. of health, 2003). In 2002, they are prone to get HIV in the country among the 13 million to 20 million, the people living with HIV/AIDS (ODHA) an estimated 90,000 people until we got 130,0005.

The problems facing OPDHA very complex, includes physic health problem because decrease CD4, because their physical health psychological problems as shock, depression, denial, angry and sad and sorry and also psychological problem as isolated, expelled from the lives and so on.[6]

The complex problem that the living and the increasing number of cases should indeed, medical practitioners understand more the clinical and how to diagnose infections of HIV/AIDS. This will be the following clinical and diagnoses of HIV/AIDS infections.

HIV/AIDS CLINICAL DESCRIPTON

The clinical description HIV/AIDS can be seen from the disease, a disease that is often found and often accompanying.[1,7,8]

THE ROUTE OF HIV/AIDS

Beginnings arise after HIV infection retroviral acute, called suds, this decline in suds is showed CD4 increasing RNA-HIV levels (and viral load). CD4 count tend to decline gradually within a few years with CD4 faster rate of decrease in 1.5–2,5 years before patients fall in a state of AIDS. Viral load going up fast at the start of an infection and then went down to a point. With continued infectious viral load gradually rising. In the late phase diseases will be found cd4 count & it; 200/mm^3, onset, followed an opportunistic infection the emergence of certain cancers weight down quickly and complication of neurological.[5,9,10]

Symptoms from HIV divided into 4 steps:

Acute Infection Stage: no symptoms typical, arising after 6 first week be either fever, taste tired muscle pain and joints, pain ingest, and enlargement of lymph nodes. May also accompanied inflammation of the membranes of the brain (meningitis aseptic she fever, headache, spasms and nerve paralysis the brain.

Asimtomatic Stage: at this stage usually without symptoms and complaint, this stage can last six weeks to months even years after infection.

Simtomatic Stage light to severe: at this stage weight declining not until 10%, thrush that recurs at the mouth, inflammation of the angles of the mouth, can also found bacterial infection of the breath the top but sufferers can doing activities normal. On the stage that further decline weight more 10%, diarrhea that more than 1 months, heat unknown s why over a month, candidiasis oral, oral hairy leukoplakia, pulmonary tuberculosis, and pneumonia bacteria. At this stage of lying in bed more than 12 hours a day during last month.

Aids stage: at this stage sufferers was attacked by one or several kinds an opportunistic infection, e.g. pneumocystis carinii, toxoplasmosis the brain, diarrhea due to kriptosporidiosis, viral disease sitomegalo, a viral infection herpes, candidiasis esophagus, trachea, the bronchi or lungs and fungal infection other histoplasmosis, e.g. koksidiodomikosis. Can also found some cancers; e.g. cancer lymph nodes and sarcoma.[5,9,10]

SYMPTOMPS RELATED ON HIV/AIDS

1. **Fever:** rushes often found in people with HIV, CD4 the number could help in evaluating and distinguish a likely cause its fever. On early disease (CD4 > 500) cause fever can occur because of tuberculosis, pneumonia or an acute infection of HIV her. Midstage disease (CD4 200–500) can occur because the spread tubrkulosis or pneumonia. In adults with sexual activity active could by sexual because transmitted disease or infection anorektal. Late disease (CD4 75–200) fever can occur because infection opurtunistic like pneumocystis carinii or malignansi. Another causes can because the spread of tuberculosis, nonthypoid bacteriemia, salmonella bartonellosis, and fungal infection histoplasmosis, and cryptococcosis. Advanced disease (CD4 < 75) all diseases in late disease can occur in this stage and can be found mycobacterium avium complex and infection cytomegalo virus1,[10,12]
2. **Diarrhea chronicle:** diarrhea can be caused for infection, fierceness or HIV his own. Infection can be caused by clostridium: defficile, salmonella, campylobacter, shigella, entamoeba histolytica; giardia lambia, isosporabelli, enterovirus, and strangyloides stecoralis. In late disease besides cause above may also caused by cryptosporidium parvum microsporidium and directorate. On advanced disease can occur because mycobacterium avium complex and citomegalovirus1,[10,12]
3. **Dysphagia:** dysphagi often accompanied by odynophagia and can be developed into a esophagitis. In midstage disease can occur sprue and esophageal disconfort. On late disease can occur infection mucous esophagus kandida accompanied with a lesion in the mouth. On advanced disease equal to late disease but often found infection citomegalo viruses and ulcer aphthous.[1,12]
4. **Respiratory disorder:** can happen because bacterial infection of pyogenic, mikobacterium, fungi, parasitic, virus and ferocity of lymphoma or sarcoma sarcoma. This complaint be either shortness or cough[12]. In late disease can occur because pneumocystis carinii, fungal infection coccidiodes immitis, cryptococus neoformis

or histoplasma capsulatum, can also occurred sarcoma sarcoma. On advanced disease besides cause above can be found again pseudomonas aeuruginosa and aspergilus species especially on the circumstances of netropenia or in hospital.[1]

5. **Skin Infection:** infection in the skin can be varied to suit imunosupresinya degrees. On early disease be either rash, lesions because sexual transmited disease, folliculitis, impetigo, ecthyma and sellulitis. On midstage disease can occur mucocutaneous candidiasis, oral hairy leukoplekia, shingles, psoriasis, dermatitis seborreic, and dermatitis atopy. In late disease skin infection that occurred previously become more chronic and refractory to therapy, can also occurred infection opurtunistic (cryptococcolis or histoplasmosis) and can occur lesion on skin. On advanced disease lesion on the skin not typical so it takes biopsy to its diagnoses, can occur bacillary angiomatosis and moluscum contangiosum.[1]
6. **Central Nerve Disorder:** central nervous breakdown can include the status change, mental and pain the changing status the kognitive mental disorder; impairment of consciousness, delirium and it is psychosis. In early may occur aseptic meningitis disease happens because of its own. On disease midstage aseptic meningitis may become more frequent and chronic meningitis. In late disease can occur cryptococcal meningitis, toxoplasma encepalitis and AIDS dementia complex. On advanced disease the disease may occur in late on this phase and often accompanied prymary CNS lympoma.[1]
7. **Limphadenopathy:** caused by bacterial infection, syphilis; mikobakterium, a virus or fungus, may also caused skin disorder wide as dermatitis seborroik, and pioderma, when swollen lymph nodes happened two locations excess of 1 centimeter, and lasted more than three months called persistent generaled limphadenopathy (PGL). PGL arising during over 50 percent living, is symmetrical, no pain, often in glands behind ears and epitrochlear.[12]
8. **Weight loss:** weight loss is a complaint often obtained in people with HIV, weight loss in line with the progesifitas disease spread. When weight loss more than 10% accompanied diarrhœa chronic over a month or fever over a month not caused another disease called HIV wasting syndrome.[5,13]

HIV/AIDS CLINICAL MANIFESTATION

1. **Candidiasis:** fungal infection kandida this could be this infection at the folding moist, paronychia, angles of the mouth, balanitis, and onychomikosis. Symptoms clinical usually more weight if there infection of the oral mucous, pharinx, and genital. An opportunistic infection by kandida usually more easy to when there is infection bacterium or virus other staphylococcal, like streptococcus, mikobakterium avium complex, cytomegalo herpes viruses and simplek or abrasion the skin and mucosa which are port' entry for kandida to get in circulation and next an undesired effect pathological on an organ local for example in eyes occurring retinitis and endopthalmitis. Candidiasis can cause malnutrition on living due to the lurch swallow (disfagi) and pain ingest (odinofagi).[6]
2. **Tuberculosis:** infection by mycobacterium tuberkolose occurs more frequently in HIV/AIDS sufferers compared with the general population, infection this could happen on all stadium HIV infection and usually occurs in CD4 about 400/ml^3. In an advanced state of the risk of infection mikobakterium tuberkolose by those with HIV 8–10% per year. Marriott, (Smith, 1997; Merati, 2004). Of this number is far higher in a developing country like Indonesia where tuberkolosis is still in endemi (Merati, 2004). Tuberkolosis can be a manifestation of the beginning of HIV so that patients who terdiagnose tuberkolosis should be thought to do with HIV infection especially to a group of high risk are infected with HIV, manifestasinya can include infection of the pulmonary (pulmonary tuberculosis) or infection outside/extra pulmonary stenosis (Smith, 1997; Merati, 2004). TB extra stenosis occurs more frequently in HIV to 70% in the general population, TB extra stenosis this may include: limpadenitis TB, the genital tract infections, urinary, the nerve center and spinal cord. The diagnoses built upon: disease history, the risk of HIV. Photographs thorax hilum, which looks gland enlargement lung, infiltrate at the apex effusion of the pleura, cavity pulmonary or tuberculosis a billion.[6]
3. **Pneumonia bacterialis:** at the HIV pneumonia with bacteria pyogenic occur more often than the general population, but germs the cause same as: streptococci pneumonia, hemopilus influenza, and brahamella catarrhalis. Can also occurred infection with staphylokokus aureus, and gram-negative bacteria. Symptoms may include high heat a sudden, asphyxiate, chest pain, and coughing productive with sputum being purulent. Can also occurred lung infection chronicle suppurative and sinusitis (Smith, 1997). Pneumonia bacterialis often occurs in cd4 & it; 250/ml^3, while suppurative infections happens when cd4 & it; 100/ml^3.[12,14]
4. **Toxoplasmosis:** infection by toxoplasma gondii is an opportunistic infection that often occurs in HIV infection. Common symptom infection toksoplasmosis be either high fever headache and vomiting vomiting, may also form of symptoms ensepalitis neurological or focal plane, as headache, spasm, impaired function cognitive and impairment of consciousness. In disorders more difuse can occur symptoms sudden accompanied fever and spasms or existing bleeding intra cerebral, disorientation, mental disturbance and comma. In the eye can happen retino choroiditis while in mielopathia can occur weakness ektremitas with impaired sensory

and disorders spinter. Diagnose can be made by complaint above accompanied ct a brain scan shown any lesions multiple ring-shaped, the picture is that clearer by contrast or with MRI, lesions lie in cortico-medullary junction or in basal ganglia. Serology tests can help where obtained immunoglobulin G (IgG) a specific for toksoplasma.[6]

5. Pneumonia pneumocystic carinii: pneumonia pneumocystic carinii (ppc) was opportunistic infection frequently found on the HIV (Smith AI, Morris A 2002), in america rate occurrence 70–80% on all the HIV who do not get propilaksis (Zimmerman, 1994). PPC often arise if CD4 < 200/ml^3, symptoms can light to severe form of: dry cough, asphyxiate progressive start from tightness while working until shortness at rest, fever and sweating.[11,15] Photograph roentgen thorax PPC on light, maybe normal, or a little hormonal perihilar, on PPC are occurring abnormality difuse interstitial bilateral shadowing, and at PPC heavy no abnormality that extensive form of bilateral interstitial alveolar and marking. To examination gas blood, PPC light show pao2 normal, and saturation oxygen declining while working, PPC being PaO2 between 60–80 mmHg, PPC and heavy PaO2 & it; 60 mmHg.[11] The diagnosis made based on to be above accompanied microscopic examination to identify pneumocystis of sputum, preparation broncoalveolar fluid or lung tissue and PCR.[11,15]

HIV/AIDS DIAGNOSIS

The diagnosis of HIV infection/aids can be made based on clinical classification organization or CDC (see appendix 1 and 2) (Levy, 1993). In Indonesia diagnose aids for the purposes of epidemiology surveillance made if showing HIV testing positive and lack of symptoms was obtained 2 major and one minor symptoms.[7]

Symptoms Major

1. The weigh decrease more than 10% in 1 month
2. Chronic diarrhea for a month
3. Fever more than a month
4. Impairment of consciousness and neurological disorders
5. Dementia/HIV encefalopati

Symptoms Minor

1. Cough more than a month
2. Dermatitis generalisata
3. Herpes zoster multisegmental and or repeated
4. Kandidiasis oro-faringial
5. Simplek chronic progressive herpes
6. Limfadenopati generalisata
7. Fungal infection of recurring at female genitals
8. Retinitis cytomegalovirus

When acquired one mark/symptoms down here, reported as AIDS cases, without examination laboratory:

1. Sarkoma Kaposi
2. Repeated pneumonia and Life-threatening

HIV EXAMINATION

To detect a person suffering from HIV, the test can be done directly on the HIV virus or indirectly by way of finding an antibody. If someone found antibodies against HIV infected with HIV (Fauci, 2003; Dept. of health, 2004). Inspection strategy for diagnostic lab can be seen in annex.[3]

HIV Serology Examination

Examination first antibodies against HIV can be used rapid a test to tapis test, when acquired positive results done reexamination by using test that having the basic principle different and or using preparasi antigens different of the tests first, usually used enzym-linked immunosorbent assay (ELISA). When available means could pretty conducted trial confirmation to western blot (WB), indirect immunofluorescence assays (IFA), or with radioimmunoprecipitation assay (RIPA) (Depkes, 2003; Crowe, 2004) other checks that can be used to detect antibodies tyerhadap hiv can be used material of saliva (OraSure) and urine (Calypte HIV-1 Urine ELISA).[1]

HIV VIRUS

HIV virus in the body could be detected by a polymerase chain reaction (PCR) technology. This technique was done if the serology test several times not conclusive; in order to make sure there is someone at phase of a window (a window period) to be knowing HIV infection on the baby, and to the interest in certain research. PCR this method includes DNA-PCR can, PCR, RNA (b) DNA assay and p24 antigen joined the.[2]

HIV RISK FACTORS

HIV epidemiological risk factors infection covering (Depkes, 2001):

1. Behavior risky (now or past)
   - sexual intercourse goggle- sexual partners high risk without use condoms
   - narcotics addict syringe
   - sexual intercourse unsecured
     - having many sexual partners
     - sexual partners known patient HIV/AIDS
     - sexual partners from villages in prevalence HIV aids that a high
     - homosexual

2. Workers and customers entertainment as: massage parlor, discotheque, karaoke or prostitution veiled
3. Have the acts of sexually transmitted infection (IMS)
4. The acts of received transfusions blood recurring
5. The acts of wound leather, tattoo, piercing, sirkumsisi or with an instrument not sterile.

SUMMARY

HIV infection has become a serious breakdown in health, the disease have an impact of crimes against victims, their families and surroundings so we needed getting the prompt, therefore understanding to picture clinical and manner diagnoses should be perceptible by practition health.

A snapshot of the clinical HIV infection/AIDS can be seen from grievances and a disease that often accompanies it, a complaint which is found at HIV/AIDS sufferers in the form of suds retroviral acute: fever, weight loss, diarrhea chronic, disphagi, limpadenopati, infections in the skin respiratory disorders and nervous breakdown center. While a disease that often been gained by those with HIV/AIDS as candidiasis, tuberculosis, pneumonia bakterialis, toxoplasmosis and pneumonia pneumocystic carinii. Diagnose HIV infection created based on clinical symptoms which includes major symptoms and symptoms of minor, and the result of the examination of the laboratory.

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Notes to authors

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- **Perspectives**. Articles should be under 3,500 words and should include references, not to exceed 40. Use of subheadings in the main body of the text is recommended. Photographs and illustrations are encouraged. Provide a short abstract (150 words), a one-sentence summary of the conclusions, and a brief biographical sketch**.** Articles in this section should provide insightful analysis and commentary about new and reemerging infectious diseases and related issues. Perspectives may also address factors know to influence the emergence of diseases, including microbal adaptation and change, human demographics and behavior, technology and industry, economic development and land use, international travel and commerce, and the breakdown of public health measures. If detailed methods are included, a separate section on experimental procedures should immediatelly follow the body of the text.
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- **Dispatches**. Articles should no more than 1,200 words and need not be devided into sections. If subheadings are used, they should be general, e, g., "The study" and "Conclusions." Provide a brief abstract (50 words); references (not to exceed 15); figures or illustrations (not to exceed 2). Dispatches are updates on infectious disease trends and research. The articles inlude descriptions of new methods for detecting, characterizing, or subtyping emerging or reemerging pathogens. Developments in antimicrobial drugs, vaccines, or infectious disease prevention or elimination program are appropriate. Case reports are also welcome.
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