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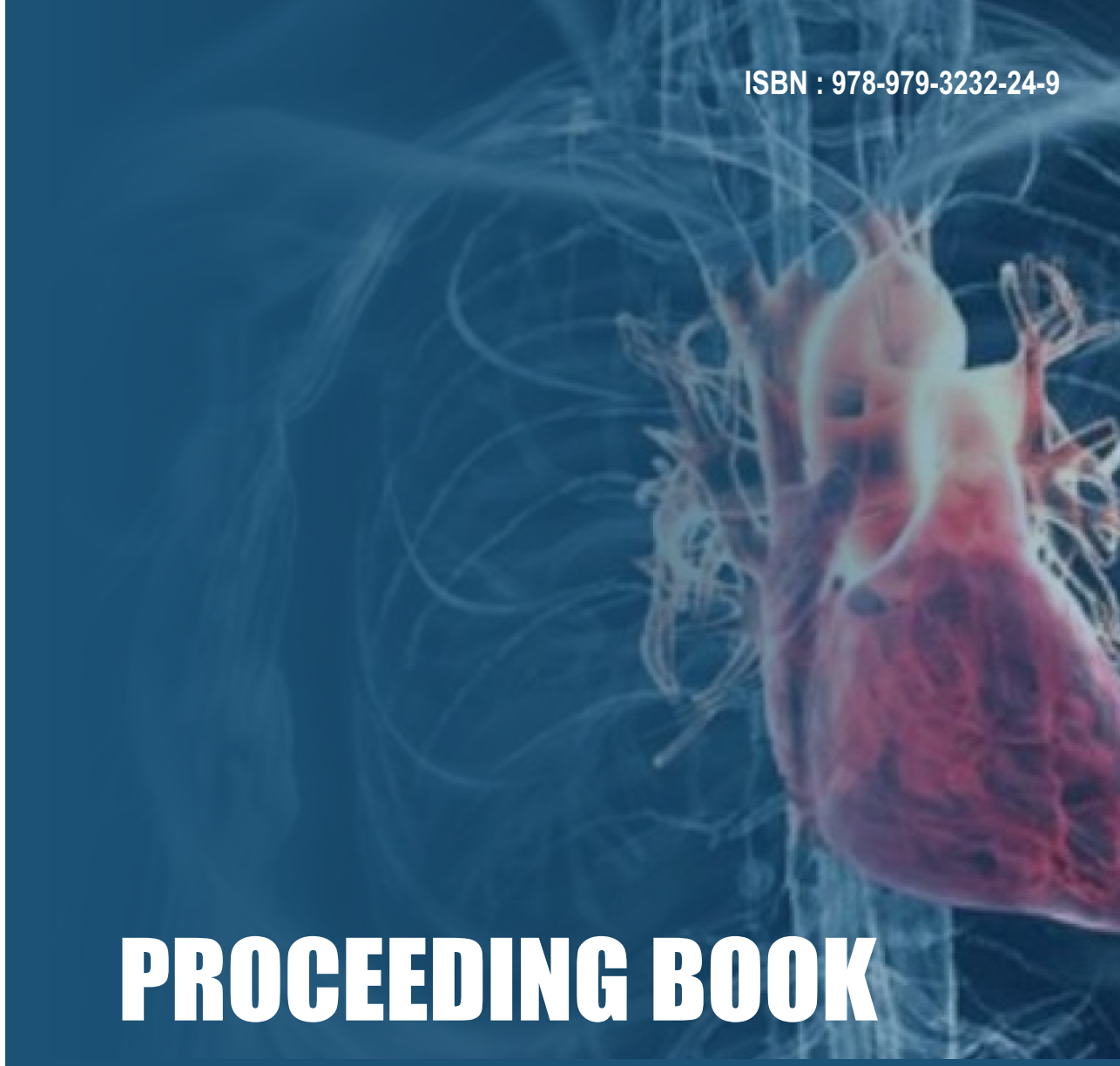

PROCEEDING BOOK

International Conference
CARDIOVASCULAR DISEASES
CVD-IA

**Integrated Approach From Basic,
Clinical Science, Public Health and Bioethics**

Yogyakarta, May 14-17, 2016

Abdul Kahar Muzakkir, Conference Hall
Universitas Islam Indonesia, Yogyakarta
UII Main Campus, Jl. Kaliurang KM 14.5 Sleman Yogyakarta

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RECTOR SPEECH

The Honorable:

- ✓ All the impressive keynote speakers, lecturers, delegations, and participantsof the International Conference on Cardiovascular Diseases.
- ✓ Distinguished guests, ladies and gentlemen.

AssalamualaikumWarahmatullahiWabarakatuh.

First of all, I would like to express my gratitude to you, all the keynote speakers, lecturers, delegations, and participants of the International Conference on Cardiovascular Diseases. Welcome to Universitas Islam Indonesia (UII), the oldest national university in the country.

UII is one of the oldest private universities in Indonesia, established on July 8, 1945 , just 40 days before the proclamation of Indonesian independence. Located in the northern outskirt of Yogyakarta, the heart of Javanese culture, the main campus overlooks the stunning beauty of Merapi volcano, which is a perfect place to study. Currently we have 3 doctoral, 9 master's, 4 professional, 25 undergraduate, and 4 vocational programs covering a wide spectrum of knowledge.

One of these undergraduate programs is Faculty of Medicine (FM) UII, where the interest of new student enrollment continues to increase every year. On this special occasion, FM UII is collaborating with other leading institutions (CUCMS Malaysia, STMU Pakistan, UP Manila, UIS Bangladesh and IMU Turkey) to organize an International Conference on Cardiovascular Diseases with the theme 'Integrated Approach for Cardiovascular Diseases from Basic, Clinical Sciences, Public Health and Bioethics'.

Distinguished guests, ladies, and gentlemen,

Cardiovascular Diseases (CVD), included hypertension, stroke, ischemic heart diseases, and heart failure, plays as the number one cause of death from non-communicable diseases, this disease is caused by impaired function of the heart and blood vessels. In Indonesia, the prevalence of heart disease average of 9.2% and could reach 17.3% at the age of 65 years. The highest prevalence reached 16.9% in Central Sulawesi and the lowest prevalence in Lampung with figures of 3.5%. The cases of heart disease each year in the world is likely to increase as well as the conditions in Indonesia.

Based on these problems, it would be very important to take initiative to reduce the risk of CVD for individuals, families, and people around. The prevention of CVD needs to be done by increasing public awareness to recognize the symptoms and the risk. Through this occasion, we hope this awareness can be spread even more to public and society alike, and not only limited in the medical world.

Distinguished guests, ladies, and gentlemen,

Today, we are conducting an International Conference on Cardiovascular under the theme 'Integrated Approach from Basic, Clinical Science, Public Health, and Bioethics'. This conference is aimed to enhance knowledge with recent advancement and management of CVD, to analyze the integrated approach for handling CVD, and to develop the CVD prevention effectively based on local wisdom culture in line with bioethics.

With so many experts coming from various leading institutions in the world, I do hope this conference will be a good chance to discuss all the problems and preventions of CVD, will be a good place that we can take many advantages from. Last but not least, once again I extend a very warm welcome to everyone visiting our university. Thank you very much, and hope you are having a great time.

Wassalamu'alaikumWr. Wb.

Dr. Ir. Harsoyo, M.Sc.
Rector of Islamic University of Indonesia

DEAN SPEECH

Assalamualaikum warahmatullah wabarakatuh,

Let us praise to the Almighty Allah SWT, for all His Blessings, His Mercy, and His Guidance for holding this International Conference on Cardiovascular Disease : Integrated Approach from Basic, Clinical Science, Public Health and Bioethics. And may peace be upon the best role model for all human being, Muhammad SAW.

Faculty of Medicine, Universitas Islam Indonesia (FK UII) as higher education institutions have an obligation to carry out the pillars of education, research, society dedication and dakwah. We realize that the pillars of research in higher education should be perceived beneficially either by academics, society and country. Therefore the effort to provide information and development of science in cardiovascular field should is expected to be more useful for health care services. Cardiovascular disease is the leading cause of mortality in many countries. Acute Myocardial Infarction (AMI) is the leading cause of mortality by heart disease and the incidence level is increasing from year to year. AMI diagnosis must be done quickly and accurately because of high mortality in the first hour and early intervention is very helpful.

On this international seminar, we expect to develop communication, cooperation and synergy of all speakers and participants from domestic and abroad. We are ready to work for it, in the form of a research collaboration and other scientific conference.

Before ending my speech, we say welcome to all the attendants from other countries as well as from all over Indonesia both as participants and speakers. We extend our thanks to the speakers who are willing to share knowledge, experience and build silaturahmi at this International Seminar on CVD. May we all receive additional knowledge, and continuously

improve our good deeds in the development of science in our respective institutions and hopefully it will be useful for society. I also would like to ask for all participant's apology for any possible lack or mistake found in this event. Let's begin the seminar and see you on the next events of FK UII.

Wassalamua'alaikum warahmatullah wabarakatuh

dr. Linda Rosita, MKes, Sp. PK
Dean of Faculty of Medicine, Islamic University of Indonesia

MESSAGE FROM CONFERENCE'S CHAIR

Dear conference attendees,

On behalf on the organizers at The International Conference in Cardiovascular: Integrative aspect from basic science, it is my great pleasure welcome you to Yogyakarta, Indonesia and I believe it will be a wonderful two-day conference.

This conference is an effort to share many interesting topic, consisting of updates on preclinical studies, diagnosis and management of cardiovascular disease. We hope that through this conference and workshop , we can bring the best experience and expertise into the improvement of patient's care in cardiovascular problems. We would like to thank you to our distinguished speaker for their contribution in the realization of this event.

We hope all the participants will enjoy this conference and benefit from the knowledge and experience shared in this meeting.

For the last, I would be remiss if I did not mention that I hope you will get to take time to enjoy our social event in the end of conference and to experience the friendliness, hospitality and excellent Yogya's cuisine offered. Ramayana Ballet, Ullen Sentanu, and sightseeing in Keraton and Malioboro will be unforgettable experience.

Regards,

dr. Putrya Hawa, M.Biomed
Chief of Committe
CVD-IA Conference 2016

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BATTLING CARDIOVASCULAR DISEASE WITH NUCLEAR MEDICINE SHORT TITLE: NUCLEAR MEDICINE IN CARDIOLOGY

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ABSTRACT

With the advances of prevention and treatment strategies, we have encountered the reduction of mortality in cardiovascular disease (CVD) during these 2 decades. In contrast, the percentage of death rate in increases in aging population. There are still regional differences of CVD death ratio among the territories or countries around the world. These increasing aging population and regional differences are next challenges to overcome for the cardiology societies. Among several heart disease, coronary artery disease (CAD) continues to be a leading cause of death in industrialized society and in developing countries as well. Diagnostic techniques for evaluating myocardial perfusion play an important role in the identification of CAD and determination of their prognosis.

Nuclear medicine has played an important role for diagnosis of CAD since early 1970. Myocardial perfusion single photon emission tomography (SPECT) has been applied for detection of extent and severity of myocardial ischemia. The risk assessment using myocardial perfusion SPECT is now well established based on large number of well-designed studies. Myocardial perfusion imaging (MPI) greatly contributes the clinical decision making and patient's care.

Positron emission tomography (PET) MPI is state of the art diagnostic imaging technique. PET has high diagnostic accuracy and can quantitatively estimate regional myocardial blood flow (MBF) in patients with CAD. PET MBF estimation has additional diagnostic value and prognostic value over standard visual assessment.

In the current situation, another challenge of cardiology is increasing the number of heart failure (HF) patients. Sufficient revascularization may improve exercise capacity and outcome in patients with ischemic heart disease with reduced LV dysfunction. PET glucose metabolism imaging, ^{18}F fluorodeoxyglucose (FDG), can identify viable myocardium suitable for revascularization. Cardiac sympathetic nervous imaging has been applied for predicting cardiac events and life threatening arrhythmic events in patients with HF.

Recent technological advances, such as PET and molecular imaging have contributed to enhance diagnostic accuracy. These new developments would further raise an importance of nuclear cardiology imaging for myocardial ischemia detection, viable myocardium detection, and hence patient's care.

Key Words: Blood flow; Myocardial ischemia; Prognosis; Tomography

Introduction

Recent development of prevention and therapeutic approaches have contributed greater reduction of mortality related to heart disease (1). Although the total numbers of heart disease related mortality has been declined, the percentage of death in elderly population has increased. With increasing aging population, this is the new challenge for cardiologists (2). In addition, there are regional differences in the decline of heart disease mortality and this should another challenge has to be overcome (1).

Nuclear medicine has played an important role in the diagnosis of coronary artery disease (CAD) since early 1970 (3). With the good diagnostic accuracy, single photon emission computed tomography (SPECT) myocardial perfusion imaging (MPI) have been widely applied for detecting myocardial ischemia and cardiovascular event risk prediction in patients with suspected or known CAD (4,5). Nuclear medicine does not require contrast agents and using technetium labeled radioisotopes or positron emission tomography (PET) tracers causes limited .radiation exposure (6). Therefore, most patients are able to have this imaging test even if they have other disease, such as renal dysfunction. New nuclear medicine imaging technique including positron emission tomography (PET) and molecular imaging, have also provide crucial diagnostic information in patients with CAD and heart failure (HF).

Detecting Myocardial Ischemia using Myocardial Perfusion Imaging in Coronary Artery Disease Patient

In stable CAD patients, percutaneous coronary intervention (PCI) improves angina symptoms but does not improve the survival in the general population. However, PCI provides a survival benefit in patients with significant ischemia reduction after PCI (7). Thus, the physiological assessment of coronary arterial stenosis severity is a more critical component in the

management of patients with CAD than are morphological approaches.

The most important application of nuclear cardiology imaging should be MPI for detecting myocardial ischemia (3,8,9). For stress and rest MPI, thallium-201 (^{201}Tl) and Tc-99m (^{99m}Tc) labeled tracers such as sestamibi and tetrofosmin have been applied for detecting myocardial ischemia. Because of longer physical short life, ^{201}Tl has higher radiation exposure. American Society of Nuclear Cardiology (ASNC) issued a statement for reduced radiation exposure less than 9 mSv for diagnostic imaging (10). In this regard, ^{99m}Tc labeled MPI has replaced ^{201}Tl but we still need to encourage our colleague to switch from ^{201}Tl to ^{99m}Tc (11). Stress and rest imaging is applied for detecting myocardial ischemia. Exercise stress test can obtain exercise capacity and exercise induced ischemia. Therefore, exercise stress protocol is preferable stress approach. However, recently numbers of patients who cannot exercise or have contra indications for exercise stress have been increased not only aging population but also younger population (12). Pharmacological using vasodilator agents can be applied for stress MPI in this population. Pharmacological stress is basically safe and almost no contraindication. In this regard, there is almost no contraindication for MPI using nuclear imaging (3). This is major advantage of nuclear cardiology imaging. Pooled data showed high sensitivity (87%; range from 71% to 97%) and relatively better specificity (71%; range from 36% to 100%) in exercise stress MPI (8). Pharmacological stress MPI also shows similar diagnostic accuracy compared to stress MPI. Systematic image interpretations can also contribute to predict cardiac events. Summed stress score (SSS) has been most widely used for the risk assessment and increasing SSS is associated with higher cardiac events (5).

Electrocardiography (ECG) gated data acquisition provides additional cardiac functional information over myocardial perfusion. Increasing end systolic volume has incremental prognostic value over MPI (13).

Positron Emission Tomography Myocardial Perfusion Imaging

Positron emission tomography (PET) represents an advanced nuclear imaging technology. PET has high spatial resolution among nuclear techniques and its high temporal resolution. Positively charged positrons are emitted from the nuclei of unstable isotopes during

radioactive decay and travel a short distance in tissue before interacting with electrons. This interaction results in the annihilation of the electron-positron pair. Then, two 511-keV gamma rays are emitted in opposite directions named coincidence (14). This coincidence detection contributes the accurate measurement of the concentration of a radiolabeled molecule in a given tissue. Current PET/CT scanners performed CT based tissue attenuation corrections. This improved the image quality.

The applications of PET/CT in cardiology have been MPI and myocardial viability assessments (15). In fact, FDA has approved Rubidiu-82 (^{82}Rb), ^{13}N ammonia as MPI, and ^{18}F fluorodeoxyglucose (FDG) as viability imaging (15). Japanese Ministry of Health, Labor and Welfare (JMHLW) have approved ^{13}N ammonia, and ^{18}F FDG (11).

^{82}Rb has short physical half-life, 76 seconds (Table 1). After 5 decay, the effect of initially given radiopharmaceutical disappears and second imaging acquisition can be performed. The short physical half-life makes ^{82}Rb suitable for repeated and sequential perfusion studies, which usually require 10-minute intervals (16). ^{82}Rb is generator produced tracer and does not require on-cite cyclotron. Thus, ^{82}Rb is widely available in the US. ^{82}Rb has high image quality. PET MPI showed better sensitivity (91% vs. 82%) and specificity compared to SPECT MPI (90% vs. 72%) (17). PET MPI has advantage for the CVD risk assessment especially in obese patients and patients who have inconclusive SPECT MPI (18). In fact, ACC/AHA guidelines addressed that in case of equivocal or inconclusive stress diagnostic tests is class 1 indication for PET MBI (8).

Myocardial blood flow (MBF) quantification is another advantage of PET MPI (19). The limitation of relative perfusion imaging is underestimation of myocardial ischemia in the multi-vessel CAD. In this regard, quantitative MBF provides accurate diagnostic information over relative perfusion imaging (19). In addition, even in visually normal stress images, patients with reduced coronary flow reserve (CFR) had higher cardiovascular events (20).

Assessment of Viable Myocardium

Improvement of the acute coronary syndrome (ACS) treatments have increased the number of survivors after ACS. This increase the patients with heart failure (HF) due to the CAD. Among the several treatments option for HF, the best treatment contributes improving the survival and functional capacity should be coronary revascularization (21). In this regard, myocardial viability assessment is quite important for deciding treatment strategy. Although stress and rest MPI is usually applied for detecting ischemic and viable myocardium, MPI often underestimates viable myocardium (22). Dysfunctional and viable myocardium changes myocardial metabolism from fatty acid metabolism to glucose metabolism. Therefore, ^{18}F FDG PET can detect viable myocardium. The typical dysfunctional but viable myocardium shows perfusion and glucose metabolism mismatch pattern (23). Among several cardiovascular imaging techniques, ^{18}F FDG PET has highest sensitivity for detecting viable myocardium (22). In terms of evidence point of view, large scar detecting MPI and ^{18}F FDG PET is associated with poor survival (24).

Risk Assessment in Heart Failure

The cardiac sympathetic nervous function plays an important role in regulating cardiac function (25). The sympathetic nervous function is divided into two parts such as sympathetic nervous system (SNS) and parasympathetic nervous system (PNS). The activation of the SNS play key roles in the pathogenesis of HF. Dysfunction of the SNS is also associated with the development of life threatening ventricular tachyarrhythmia.

Iodine-123-metaiodobenzyl-guanidine(^{123}I-MIBG) is an analogue of the false neurotransmitter guanidine and accumulates in adrenergic nerve terminals via the uptake-1 mechanism. Reduced myocardial ^{123}I-MIBG uptake is linked to the occurrence of ventricular tachyarrhythmia and is an independent predictor of outcome in HF (26).

Assessment of New Heart Failure Treatments using PET

Many new therapeutic approaches have been developed for HF. HF is characterized by an energy depleted state. Elevated wall stress from increased afterload increases metabolic demand at the expense of forward kinetic work. Increased metabolic demand [myocardial oxygen consumption

(MVO_2)] associated with impaired ventricular function is associated with a reduced myocardial efficiency and correlates with a poor prognosis (27). Treatments reduced excessive MVO_2 improve HF patient's survival. Therefore, MVO_2 measurements play important roles for evaluating new HF treatments (28).

^{11}C acetate PET provides a non-invasive technique for measuring myocardial oxidative metabolism which is closely correlated with TCA cycle flux and myocardial oxygen consumption. Combined with an assessment of cardiac function (such as echocardiography or MRI), ^{11}C acetate PET can be applied as a noninvasive approach to study myocardial energetics and efficiency (29). ^{11}C acetate PET has revealed therapeutic effects of important newly developed HF treatments including beta blockers, continuous positive airway pressure therapy in patients with sleep apnea, cardiac resynchronization therapy, and surgical ventricular reconstruction (29-32).

Conclusions

Nuclear cardiology imaging has excellent diagnostic accuracy for myocardial ischemia and viable myocardium in patients with CAD. Metabolic imaging has contributed to evaluate new HF treatments. With an aging population, percentage of CVD are increasing. In this regard, the non-invasive approach of nuclear medicine can contribute these patients care. Therefore, nuclear cardiology imaging should continue to the main stream of non-invasive cardiac imaging in order to enhance patient's care.

Conflicts of Interest

None

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Table 1. Common Cardiovascular PET and SPECT Tracers

Radioisotope	Pharmaceutical	Half-life	PET or SPECT	Indications
^{82}Rb	^{82}Rubidium	76 sec	PET	Myocardial perfusion imaging
^{15}O	^{15}O water	2 min	PET	Myocardial perfusion imaging
^{13}N	^{13}N ammonia	10 min	PET	Myocardial perfusion imaging
^{11}C	^{11}C acetate	20 min	PET	Myocardial oxidative metabolism
	^{11}C hydroxyephedrine		PET	Cardiac sympathetic nervous function
^{18}F	^{18}F fluorodeoxyglucose	110分	PET	Myocardial glucose metabolism
^{99m}Tc	^{99m}Tc tetrofosmin ^{99m}Tc sestamibi	6 h	SPECT	Myocardial perfusion imaging
^{123}I	^{123}I MIBG	8 h	SPECT	Cardiac sympathetic nervous function
^{201}Tl	^{201} Tl	73 h	SPECT	Myocardial perfusion imaging

CURRENT EVIDENCE ON CAROTID ENDARTERECTOMY (CEA) IN ISCHEMIC STROKE

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LONG ABSTRACT

Stroke is the third global leading cause of death after cancer and cardiac disease and also the number one cause of long life disability with narrow therapeutic windows for pharmacological reperfusion therapy. Futhermore, stroke is now first leading cause of death in Indonesia. According to recent studies, one in every 6 person is going to have stroke in their lifetime. The improvement of treating acute phase is accompanied by the fact that successful recanalization and subsequent reperfusion is not necessarily followed by brain tissue reperfusion. Recently, several mechanical recanalization methods were introduced in addition to thrombolytic therapy. Nevertheless, the evidence supporting these methods is still limited and further studies are needed to establish the role of such methods in treatment of stroke. Given the aforementioned facts, it seems preventive measure will be more favorable to provide better outcomes. One of the commonest underlying diseases of ischemic stroke is carotid stenosis. Therefore, it is important to correct the stenosis to prevent stroke. Two approaches are availabel for correcting stenosis in carotid artery: carotid endarterectomy (CEA) and carotid artery stenting (CAS).

Prompt evaluation and triage of patients with symptomatic carotid artery stenosis are essential to minimize the risk of early recurrent cerebrovascular events. Prospective studies have shown that the risk of ipsilateral stroke is high within the first 90 days, and especially within the first month, after TIA. Urgent initiation of treatment can reduce the risk by up to 80%. A recently developed scoring system ABCD score (age, blood pressure, clinical features, duration of TIA, and diabetes mellitus) could give a clue in estimating the short-term risk of ipsilateral stroke after TIA, as well as guiding the decision of intervention.

Based on The European Carotid Surgery Trial (ECST) and The North American Symptomatic Carotid Endarterectomy Trial (NASCET) in early 2000s, the indication of CEA is established for 70-90% stenosis. Then, based on VA Clinical Studies Program 309, CEA indication also established for moderate (>50%) stenosis. Currently, CEA also proven appropriate for some asymptomatic

patients. A very large study (n = 1.719) published in 2012, named ACAS study which try to develop risk prediction model or scoring system for determining appropriateness of CEA in ACAS patient based on the 5-year survival after CEA. A similar study (n=4.114) published in 2014, also to develop risk prediction model or scoring system, which classified major risk factors and minor risk factors for factors of 5-year survival after asymptomatic CEA.

Since the introduction of carotid artery stenting (CAS) as an alternative method for CEA in CAVATAS study, there has been increasing tendency to replace CEA with CAS. Recent larger studies like CREST and ICSS (Lancet, February 7th 2015) found out that, though CAS and CEA share similar primary outcome (30-day rate of stroke, death, MI, and 4-year ipsilateral stroke), CAS was associated with higher periprocedural stroke, and higher 4-year rate of stroke/death, which is unfavorable due to higher level of disability. Moreover, CEA also showed superiority in elderly patients. Currently, a study published in 2015 found that postoperative stroke or in-hospital death was more frequent after CAS (4.0% versus 1.5%; $p<0.001$). The more number of stroke in CAS can be explained if by ultrasonographic examination, which done by Mitsuoka, et al. They found that ultrasonographic character of the plaques in CEA cases more severe than the one in CAS cases. They also found that the incidence of brain embolism in the CAS cases was 52.6 % while 0% in the CEA cases (p = 0.00037). Thus, CAS should be treated as an alternative for CEA in very limited group of patients with average or low risk of complications associated with endovascular intervention.

In the era of 'evidence jungle' the interdisciplinary approach seems to become an inevitable option in treating stroke to provide the best possible therapy for patients. The adaptation of interdisciplinary acute stroke center concept was pioneered in Indonesia by Faculty of Medicine Universitas Indonesia and Cipto Mangunkusumo General Hospital in form of establishment of a Carotid Team; which comprises of experts from neurology, vascular surgery, neurosurgery, cardiology, radiology, and clinical pathology field. Furthermore improving public stroke-information may influence the main sociocultural and economical factors and the access to medical and surgical care for many stroke victims. With well-organized stroke-emergency transport services, well-educated neurological and vascular expertise especially for performing medical and surgical will make treatment like Carotid Endarterectomy (CEA) possible and available for patients.

After establishment of interdisciplinary team (the Head of which Prof. Dr. Teguh Ranakusuma, SpS) i.e. from 2009-2014 in RSCM Kencana (Faculty of Medicine, Universitas Indonesia), 34 CEA procedures in all symptomatic patients with 1 until 3 times history of TIA and hypertension previously, can be performed safely. In the end, we believe that we all share a similar hope that what we did contributes to the improvement of stroke care in Indonesia. With well-organized

stroke-emergency transport services, well-educated neurological and vascular expertise especially for performing medical and surgical will make treatment like Carotid Endarterectomy (CEA) possible and available for patients.

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NEW GUIDELINE IN ACUTE CORONARY SYNDROME

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INTRODUCTION

Acute Coronary Syndrome (ACS) management usually started from the presence of chest pain, and would finally be followed by an Electrocardiogram (ECG) examination. Based on the results of ECG, it could be classified into (Roffi, 2016):

1. Individuals with acute chest pain symptom and persistent ST-elevation (>20 minutes) (*ST-elevation Miocard Infarct*/STEMI). This condition is called ACS ST-elevation and reflected the presence of total blockage. Most patients would eventually develop into ST-elevation miocard Infarct. The main goal of the management for this group is immediate reperfusion using fibrinolytic or primary angioplasty.
2. Individuals with acute chest pain without persistent ST-elevation (*Non ST-Elevation Miocard Infarct*/ Non-STEMI). Changes in ECG include transient ST-elevation, persistent or transient ST-depression, T wave inversion, flat T wave or pseudonormalization of T wave, or normal ECG findings.

The definition of acute miocard infarct is the presence of necrosis in miocard tissues, consistent with acute miocardial ischaemia. To determine the presence of miocardial necrosis, a combination criteria is needed through the increase or decrease of cardiac biomarkers levels, in which Troponin has the highest sensitivity (Roffi, 2016).

Unstable angina is defined as miocardial ischaemia during resting or low-impact activity without any miocardial necrosis indications. Compare to ACS Non ST-elevation patients, patients with unstable angina do not require immediate invasive management (Roffi, 2016).

The management of ACS are always evolving in line with new clinical evidence. This article will describe the difference between newest and previous treatment guideline.

Due to our limitation in the field, a development of new strategy is needed in order to provide proper treatment that are compatible with newest *Non Communicable Diseases* (NCD) treatment guideline.

CURRENT MANAGEMENT OF ACUTE CORONARY SYNDROME

The management of ACS is constantly updated to decrease its morbidity and mortality. These changes are based on the existing clinical evidence. Clinical evidences will be use as a reference to determine treatment guideline (O'Gara Patrick T., 2013).

In ACS with persistent ST-elevation (STEMI), the emphasis is reperfusion therapy. Whether pharmacological reperfusion using fibrinolytic or mechanical reperfusion using coronary angioplasty technique. Current treatment guideline from AHA (O'Gara Patrick T., 2013) and ESC (Steg Ph. Gabriel Steg, 2012), both reccomend the same thing. If the diagnosis of ACS with ST-elevation has been established, consider : whether the health facility were able to perform primary angioplasty procedure? If the health facility are able to perform primary angioplasty, consider whether it can be done within 60 minutes (Steg Ph. Gabriel Steg, 2012) or 90 minutes (O'Gara Patrick T., 2013) since the first contact with medical personel until percutaneous coronary intervention.

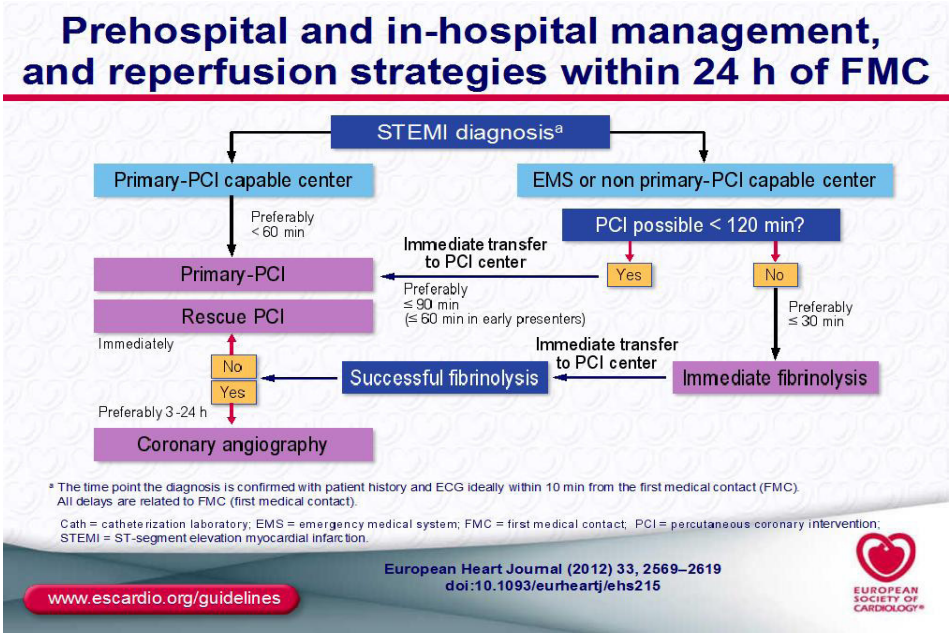

Figure 1. STEMI management algorithm based on ESC (Steg, 2012)

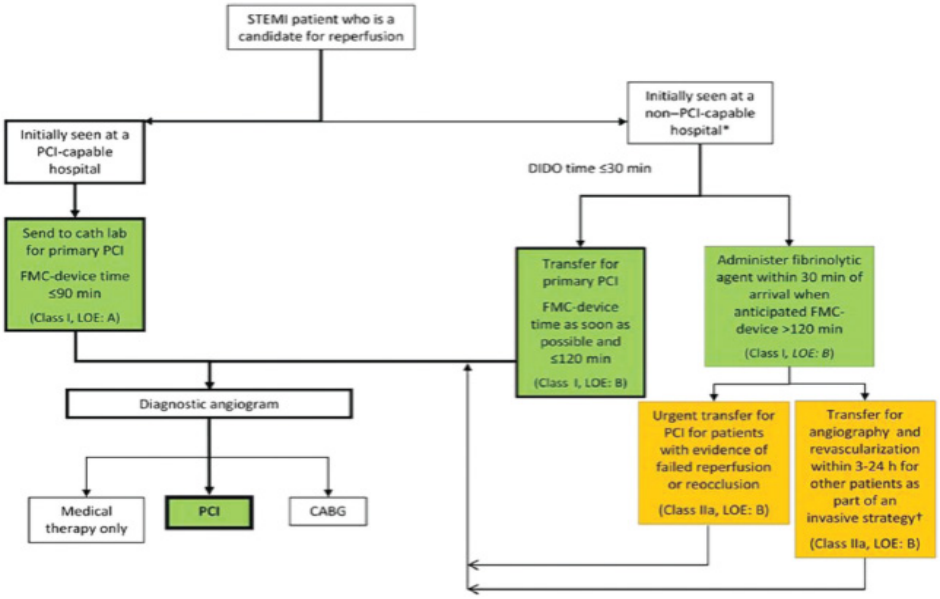

Figure 2. STEMI management algorithm based on AHA (O'Gara, 2013)

If the health facility are not able to perform primary percutaneous coronary intervention (primary PCI), patients should be transfered to another health facility with the ability to perform primary PCI within 120 minutes since the first contact with medical personel until percutaneous coronary intervention. If both options are not possible, then reperfusion should be done using fibrinolytic. After fibrinolytic reperfusion, it is recomended that patients are transfered for invasive procedure. If the result of fibrinolytic reperfusion is not as expected, invasive procedure should be done immediately. While if the result of fibrinolytic reperfusion is as expected, invasive procedure can be planned within 3 to 24 hours (O'Gara, 2013;Steg, 2012).

In the group without persistent ST-elevation (NonSTEMI), there are a few differences between treatment guideline in 2013 and 2015 (Roffi, 2016). Risk criteria is needed in this group to determine treatment plan—whether or not invasive procedure is needed and when it should be done.

In 2013 guideline :

- *Invasive strategy* is needed within < 72 hours with I-A indication in patents with:
    - One high risk criteria
    - Recurrent angina symptoms
- *Early invasive strategy* (<24 hours) with I-A indication in patients with :
    - GRACE score more than 140 or at least one *primary high-risk* criteria
- *Urgent coronary angiography* (<2 hours) with I-C indication in patients with :
    - *Very high risk* ischemia criteria (recurrent angina, related with cardiac failure, impending ventricular arythmia, or unstable hemodynamic.

In 2015 ESC Guideline, there are a few changes. Risk classification are changed into 4 class. From this risk classification, treatment plan can be determined. Risk classification as listed below (Roffi, 2016).

Risk classification for invasive strategy guideline (Roffi, 2016).

Very-high-risk criteria

- *Haemodynamic instability or cardiogenic shock*
- *Recurrent or ongoing chest pain refractory to medical treatment*

- *Life-threatening arrhythmias or cardiac arrest*
- *Mechanical complications of MI*
- *Acute heart failure*
- *Recurrent dynamic ST-T wave changes, particularly with intermittent ST-elevation*

High-risk criteria

- *Rise or fall in cardiac troponin compatible with MI*
- *Dynamic ST- or T-wave changes (symptomatic or silent)*
- *GRACE score >140*

Intermediate-risk criteria

- *Diabetes mellitus*
- *Renal Insufficiency (eGFR<60 mL/min/1.73m2)*
- *LVEF <40% or congestive heart failure*
- *Early post-infarction angina*
- *Prior PCI*
- *Prior CABG*
- *GRACE risk score >109 and <140*

Low-risk criteria

- *Any characteristics not mentioned above*

ACS non persistent ST-elevation patients with very high risk criteria would need faster *immediate invasive strategy* (<2 hours after hospitalization). In this group, management is similar with STEMI group. Patients with high risk criteria would need invasive procedure within 24 hours. While patients with intermediate risk criteria would need invasive procedure within 72 hours. This management algorithm can be seen in Fig.2 from Guideline NSTEMI ESC-2015 (Roffi, 2016).

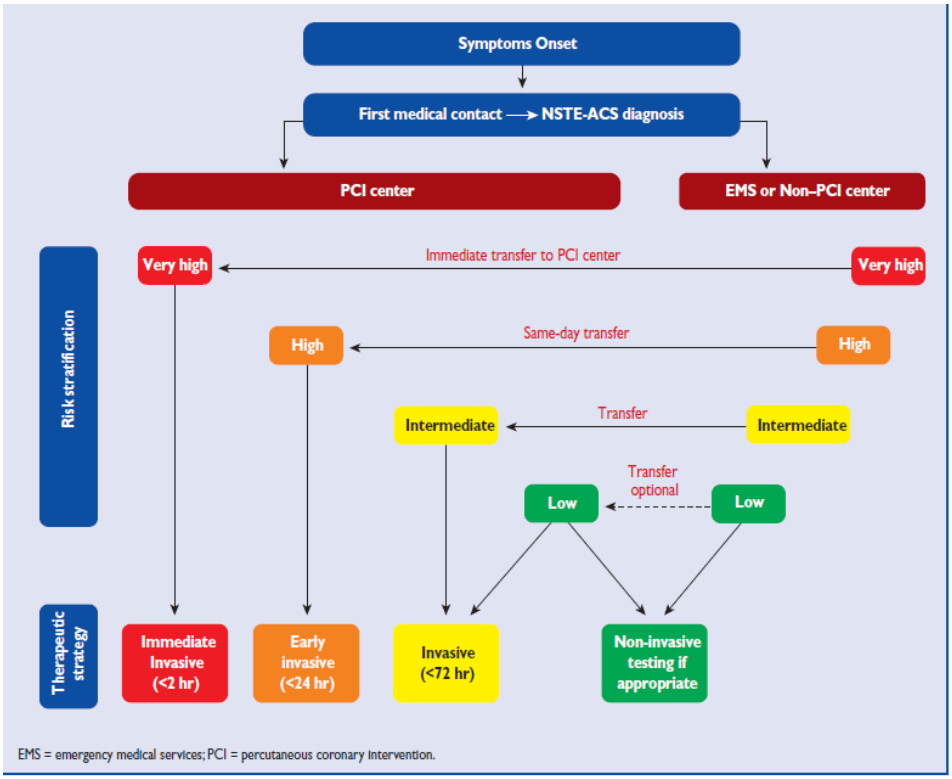

Figure 3. NSTEMI management algorithm based on ESC (Roffi, 2016).

CONCLUSSION

The management of ACS with persistent ST-elevation (STEMI) is reperfusion. Reperfusion modalities include fibrinolytic and primary percutaneous coronary intervention (Primary PCI). The choice between fibrinolytic or Primary PCI are based on the presence of Cardiac chateterization laboratorium and should consider the time frame of reperfusion in order to limit the scale of miocard infarct.

In ACS without persistent ST-elevation (Non-STEMI), percutaneous coronary intervention is also being considered by determining the risk and clinical conditions of the patients. Whether PCI should be done within < 2 hours, <24 hours or within <72 hours.

Due to resource limitations, a breakthrough strategy needs to be considered in order to implement current Acute Coronary Syndrome treatment guideline.

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A PRELIMINARY STUDY ON PLATELET REACTIVITY IN NORMOTENSIVE SUBJECTS WITH A FAMILY HISTORY OF HYPERTENSION

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ABSTRACT

Backgrounds: Platelets adhesion, activation, and aggregation play an important role in cardiovascular pathogenesis. These events emerge as consequences of the endothelial dysfunction and activation. Hypertension is known as a risk factor for cardiovascular disease.

Aim: To know whether endothelial dysfunction and activation have been occurred in prehypertension subjects.

Methods: We did a quasi experimental, ex vivo, human study, by examining platelet aggregation using the turbidimetric method in 10 undergraduate students with a family history of hypertension and 10 undergraduate students without a family history of hypertension.

Results: The two groups did not differ significantly in gender proportion, age, body mass index, resting blood pressure, and platelets counting. The two groups did not differ significantly in the percentage of maximal platelet aggregation to 2, 5, and 10 μM adenosine 5'-diphosphate (ADP). These results showed that normotensive subjects with family history of hypertension do not have exaggerated platelets reactivity as compared to their counterparts.

Conclusion: Under resting condition endothelial dysfunction may not manifest yet in the form of increased platelets aggregation in normotensive subjects with a family history of hypertension.

Relevance for patients: Normotensive subjects with a family history of hypertension can be considered as prehypertension subjects. To prevent the development of hypertension in these subjects and its collateral events, we need to explore the structural and functional changes in their cardiovascular system.

Key words: platelets reactivity, prehypertension, normotensive subjects, a family history of hypertension, endothelial dysfunction.

INTRODUCTION

Primary or "essential" hypertension accounts for 90-95% of hypertension worldwide. Its etiology is unknown; however, it results typically from an **increase in systemic vascular resistance** rather than exaggerated cardiac output [1-3].

Physiologically, the endothelium functions as an inhibitory mode to the vascular tone since it releases endothelium-derived relaxing factors, such as nitric oxide (NO) and prostacyclin (PGI_2), which influence the vascular smooth muscle cells. NO and PGI_2 also act as thromboregulators because they inhibit platelets activation and aggregation. Thus, in normal condition endothelium has the capacity as a vasodilator and **antithrombotic** [4-5].

However, endothelium may undergo several phenotypic modulations that lead to the impairment of mechanisms that maintain homeostasis in healthy circulation. The condition commonly named as "**endothelial dysfunction**" is associated with a proinflammatory phenotype, increased oxidative stress, and abnormal modulation of vasoactive pathways, which may lead to several manifestations, including impaired endothelium-dependent vasodilation and thrombotic-prone [6].

Essential hypertension is likely to aggregate in the family. Normotensive subjects having a blood relative, such as a mother, father, sister, or brother, who has high blood pressure before the age of 60 have two to three times the risk of developing hypertension in the later life [7-8].

MATERIALS AND METHODS

Study design

If normotensives subjects with a family history of hypertension already have endothelial dysfunction, thus they will have exaggerated platelets aggregation. To test this hypothesis we did a quasi-experimental, ex vivo, human study. In this study, we compared platelet aggregation in 10 normotensive subjects with a family history of hypertension and other 10 normotensive subjects without a family history of hypertension.

Study subjects

We collected 20 study subjects from undergraduate students of Faculty of Medicine and Health Sciences, Universitas Muhammadiyah Yogyakarta, by a purposive sampling, with equal gender proportion. Normotensive subject was defined as having blood

pressure ≤ 140/80 mm Hg measured in sitting position. A family history of hypertension was defined as having mother, father, or both with high blood pressure (systolic blood pressure {SBP} ≥ 140 mm Hg or and diastolic blood pressure {DBP} ≥ 90 mm Hg) based on a self-reported questionnaire. We collected and measured characteristics data of study subjects, such as age, body mass index (BMI), SBP, DBP, heart rate (HR), and platelets count. BMI was calculated from the formulae: (body weight in kilograms)^2/height in meters. SBP and DBP were measured using a non-invasive, oscillometric method, automatic vital sign monitor device TM-2551 P (A & D Co. Ltd., Tokyo, Japan) from the brachial artery of the subjects' dominant hand. Heart rate was counted manually using a radial pulse of the subjects' dominant hand. All measurements were done two times, then be averaged, and blindly.

Platelets aggregation test

Whole blood was taken from subject's arm and collected in tubes containing sodium heparin. At least 2 hours before blood withdrawal, the subjects were refrained from eating, exercising, and drinking coffee. One week before blood withdrawal, the subjects were not taking any drugs. The percentage of maximal platelets aggregation in response to adenosine 5'-diphosphate (ADP) as the platelets agonist was measured using the turbidimetric method (Helena Lab.).

Statistical analysis

Subjects' characteristics as a whole sample were presented as minimum, maximum, and mean ± standard deviation (SD). Except for gender proportion, group differences in subjects' characteristics were presented as mean ± SD, and were analyzed using independent t-test. The group differences in the percentage of maximal platelets aggregation was analyzed using independent t-test. For further analysis, the results of maximal platelets aggregation were categorized into three levels, namely hypo aggregation, normal aggregation, and hyper aggregation. To analyze the group difference in the level of maximal platelets aggregation we used chi-square. p value < 0.05 was considered significantly difference. Statistical analysis was done using SPPS 15.0 for Windows.

RESULTS

Subjects' characteristics

The subjects of this study have the following characteristics: late adolescent to young adult

age, underweight to normal body mass index, normal blood pressure, normal heart rate to slight tachycardia, and normal platelets counting (Table 1).

Table 1. Subjects' characteristics (n = 20)

Characteristics	Minimum	Maximum	Mean ± SD
Age (years)	18	23	20,05 ± 1,32
Body mass index (kg/m^2)	17,8	28,1	21,04 ± 2,75
Systolic blood pressure (mmHg)	94	126	112,6 ± 8,52
Diastolic blood pressure (mmHg)	54	83	71,3 ± 6,4
Heart rate (times/minute)	58	108	85,1 ± 11,63
Platelets count (10^3/mm^3)	244	451	318,1 ± 52,25

Note: SD = standard deviation

Normotensives subjects with a family history of hypertension did not differ significantly regarding gender proportion, age, body mass index, SBP, DBP, HR, and platelets counting (Table 2).

Table 2. Group comparison of subjects' characteristics

Characteristics	Normotensive without a FHoH (n = 10)	Normotensive with a FHoH (n = 10)	*p* value
Gender proportion (F:M)	1:1	1:1	-
Age (years)	20,3 ± 1,42	19,8 ± 1,23	0.41
Body mass index (kg/m^2)	21,72 ± 1,23	20,36 ± 2,39	0.28
Systolic blood pressure (mmHg)	112 ± 9,49	113,2 ± 7,91	0.76
Diastolic blood pressure (mmHg)	71 ± 5,05	71,6 ± 7,81	0.84
Heart rate (times/minute)	87,7 ± 7,21	82,5 ± 14,79	0.33
Platelets count (10^3/mm^3)	326,5 ± 47,92	309,7 ± 57,55	0.49

Note: FHoH = family history of hypertension. F = female. M = male. Data are summarized as a mean ± standard deviation, except for gender proportion.

Platelets aggregation test

The results of maximal platelets aggregation in response to different doses of ADP (2 µM, 5 µM, 10 µM) between normotensives subjects with a family history of hypertension and those without a family history of hypertension did not differ significantly (Table 3).

Table 3. Group comparison of maximal platelets aggregation

Agonist	Normotensive without a FHoH (n = 10)	Normotensive with a FHoH (n = 10)	p value
ADP 2 µM	31 ± 27,56	31,14 ± 14,74	0.99
ADP 5 µM	64,76 ± 24	65,43 ± 21,97	0.95
ADP 10 µM	81,4 ± 15,47	83 ± 10,7	0.79

Note: FHoH = family history of hypertension; ADP = adenosine 5'-diphosphate. Maximal platelets aggregation was measured as a percentage (%). Data are summarized as a mean ± standard deviation.

As the results of maximal platelets aggregation were categorized into hypo-, normal-, and hyper aggregation, the proportion of subjects with hypo aggregation, normal aggregation, and hyper aggregation did not differ significantly between normotensives subjects with a family history of hypertension and their counterparts. However, we can see a trend of increasing platelets aggregation in normotensive subjects without a family history of hypertension as compared to normotensive subjects with a family history of hypertension (Table 4).

Table 4. Group comparison of the level of maximal platelet aggregation

Agonist	Classification	Normotensive without a FHoH (n = 10)	Normotensive with a FHoH (n = 10)	*P value*
ADP 2 µM	Hypo aggregation	3	2	0.23
	Normal aggregation	5	8	
	Hyper aggregation	2	0	

Agonist	Classification	Normotensive without a FHoH (n = 10)	Normotensive with a FHoH (n = 10)	*P value*
ADP 5 μM	Hypo aggregation	2	1	0.36
	Normal aggregation	2	5	
	Hyper aggregation	6	4	
ADP 10 μM	Hypo aggregation	1	1	0.87
	Normal aggregation	2	3	
	Hyper aggregation	7	6	

Note: FHoH = family history of hypertension; ADP = adenosine 5'-diphosphate

DISCUSSION

Normotensive subjects with family history of hypertension may have preclinical cardiovascular disease state, in which the cardiovascular system undergoes structural and functional changes. This condition, without any further intervention, can lead to cardiovascular diseases state [9]. Previous studies showed that ED, one of the initiators of the development of hypertension, already occur in children and adolescents having risks of cardiovascular diseases, such as having a family history of hypertension [10]. Li et al. [11] measured endothelium-dependent vasodilatation by the percentage of brachial artery diameter in response to reactive hyperemia. They found that the brachial artery response to reactive hyperemia was significantly reduced in late young adults (age 44.5 ± 11.2 years) normotensive subjects with a family history of hypertension as compared to the counterparts without a family history of hypertension. Teixeira et al. [12] also confirmed that young adult normotensive subjects with a family history of hypertension were associated with ED.

This study showed that late adolescence-early young adult normotensive subjects with a family history of hypertension have no heightened platelets aggregation in response to different doses of ADP as compared to their counterparts without a family history of hypertension (Table 3). The results of our study are not in line with Nara et al. [13] who reported that platelet aggregation induced by ADP was significantly higher in men with a family history of essential hypertension as compared to men without such a family history of essential

hypertension taking low cholesterol and low salt diet. However, Akbar [14] reported that platelets from spontaneously hypertensive rats (SHR) showed greater platelets aggregation in response to thrombin and prostaglandin E_1 (PGE_1) but lesser platelets aggregation in response to ADP as compared to Wistar-Kyoto (WKY) rats. Akbar's study [14] supports our results regarding the platelets response to ADP. Akbar [14] showed that platelets aggregation to ADP is greater in healthy animal (WKY rats) as compared to hypertensive animal model (SHR rats). In our study an increasing trend in platelets aggregation in normotensive subjects without a family history of hypertension was found as compared to normotensive subjects with a family history of hypertension (Table 4).

Akbar [14] did not explain the causes of the different responses of platelets of the animal model of hypertension (SHR) to different agonists. ADP, thrombin, and PGE_1 activate platelets through the same pathway, namely G protein-coupled receptors (GPCR) [15]. Thus, the different responses to different agonists in Akbar's study may be caused by, in our opinion, the differences in the intracellular biochemical signaling after GPCR activation. It also explains why in our study there was a trend of heightened platelets aggregation in normotensive subjects without family history of hypertension as compared to normotensive subjects with family history of hypertension.

Our study, however, did not measure ED. Our study based on the assumption that ED occurs in normotensive subjects with family history of hypertension [10-12]. Moreover, Schlaich et al. [16] found that L-arginine transport in the endothelial cells was substantially reduced in normotensive subjects with a family history of hypertension compared to normotensive subjects without a family history of hypertension. L-arginine is needed in NO synthesizing in endothelial cells. The disturbance in L-arginine transport in normotensive subjects with a family history of hypertension can lead to the decrease of NO release from endothelial cells, then, in turn, it fails to inhibit platelets activation and aggregation. The ED that occurs early in this prehypertension model is associated with praecox atherosclerosis, as reported by Solini et al. [17]. They found that young adult (age 25.2 ± 2.4 years) normotensive subjects with a positive family history of hypertension is associated with an initial increase in markers of inflammation (such as adhesion molecule P-selectin) and plaque

instability (such as protease matrix metalloproteinase {MMP}-9). The development and progress of atherosclerosis due to ED may give risk to atherothrombosis in later life. The greater risk to develop thrombosis than to bleed in hypertensive patients is known as "the thrombotic paradox" [18].

CONCLUSION

Because the exaggerated platelets reactivity is not found, endothelial dysfunction may not occur yet in our human model of prehypertension, i.e. normotensive subjects with family history of hypertension.

However, in our opinion, the temporary conclusion inducted by this preliminary study needs further investigation. For example, platelets aggregation in normotensives subjects with a family history of hypertension can be measured after endothelial cells are activated or stimulated. Moreover, different platelets agonists can be used, such as ADP, thrombin, thromboxane, and epinephrine, which act through G protein-coupled receptors (GPCR), and collagen, which acts through tyrosine kinase [15, 19].

CONFLICT OF INTEREST

The authors have no conflict to disclosure.

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Fruit and Vegetable Consumptions Simulation of Type 2 Diabetes Mellitus Prevention Using Dynamic Model Approach In South Sulawesi, Indonesia

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ABSTRACT

The study aimed to assess fruits and vegetables consumption portion for reducing the rate increase of type 2 diabetes mellitus (DM) incidence in the next 10 years. The study design was a cross sectional study using the data from Basic Health Research (Riskesdas 2013). The subjects were themembers of the household aged ≥45 years. The research variables are the number of type 2 DM, the obesity prevalence, the consumption of sweet, salty and fatty foods as well as fruits and vegetables consumption and physical activity. Powersim was used as dynamic systems analysis . The results showed that without any controls of diabetes, the incidence of type 2 DM estimated to increase 3,53 times from 692 (2013) to 2243 people (2022) in the next 10 years. Consumption of one serving of fruit and vegetables are estimated to prevent the incidence of type 2 DM for 7.62%, two servings of fruit and one serving of vegetables prevent 16.22%, two servings of fruit and vegetables prevent 21.89%, three servings of fruit and two servings of vegetables prevent 25.50%, and three servings of fruit and vegetables prevent 28.31%. Increased portion of fruits and vegetables consumption every day have a greater impact on preventing the incidence type 2 DM.

Keywords: Fruit and Vegetable, Dynamic Model, Type 2 DM

INTRODUCTION

Model is a process of imitating an object / a system that contains important information with the purpose of forecasting and policy design. In contrast to the static model, dynamic model emphasizes the process of change from one condition to another (Ahmad, 2012).

Estimating the incidence and impact of type 2 diabetes mellitus (DM type 2) in the future is an important aspect of public health planning, this is due to the prevalence of type 2 diabetes is increasing

from year to year(Rosella, 2009). DM Type 2 has become one of the most common worldwide non-communicable diseases (NCDs) and also a major cause of death in most developed s and developing countries (Richard Sicree JS, 2011).

The data of World Health Organization (WHO) showed that 171 million people suffered from diabetes in 2000 and estimated to be 360 million in 2030. In Indonesia, the prevalence of DM was 8.426 million in 2000 and was projected to reach 21.257 million in 2030 with an increaseing of three-fold in 30 years. Indonesia also known as the two most populous suffering from diabetes after India. This suggests that diabetes is a serious public health problem (National Health Department, 2008).

In 2013, basic health survey (Riskesdas) showed that the tendency of DM prevalence in Indonesia increased by 1% from 1.1% in 2007 to 2.1% in 2013. The proportion of diabetes at age 15 years is 6.9%. In South Sulawesi the increased prevalence even higher, from 0.8% in 2007 to 3.4% in 2013. This province is one of the provinces with the highest prevalence of DM in Indonesia (Litbang-Kementerian-Kesehatan, 2013).

A healthy diet and an active lifestyle can significantly reduce the risk of Type 2 diabetes even with a family history of diabetes (Sharaf, 2010). Futhermore, dietary pattern characterized by high intake of fruits and vegetables, whole grains, low fat dairy products, and low glycemic load inversely associated with risk of type 2 diabetes mellitus (Heidemann, 2005).. Consumption of vegetables and fruits in Indonesian population is still low at 57.1 grams per person per day and 33.5 grams per person per day respectively (Siswanto, 2014).

To date, the current research in the prediction of non-communicable diseases have only found for dyslipidemia in patients with hypertension (Ompusunggu, 2010), Type 2 diabetes in urban society Indonesia (Irawan, 2010), and a predictor of risk of diabetes mellitus in Taiwan and Thailand (Chien K CT, 2009) (Aekplakorn W BP, 2006) using a static model approach. No research found to includefruit and vegetable consumption variables in the model .

The setting portion of fruit and vegetable consumption on groups of patients and non patients of type 2 diabetes mellitus are allowed to do. However, the facts that it is costly and impractical are present,

Thus, modeling is required to make an ideal representation of a real system to explain the behavior of the system.

This study aimed to estimate the incidence of type 2 diabetes with a focus on setting the portion of fruit and vegetable consumption using dynamic modeling approach in order to obtain the size of servings of fruits and vegetables that are appropriate in reducing the rate of incidence of type 2 diabetes mellitus.

MATERIAL & METHOD

The study was an observational analytic study with a cross sectional design using data from the Basic Health Research Survey (Riskesdas) of South Sulawesi in 2013. The subjects are members of the household age ≥45 years. High prevalence of type 2 diabetes were mostly found and generally occurs in this age (Irawan, 2010). In total 12,649 subjects were included.

Data processing and analysis were devided into several stages. (1) Causal Loop Diagram type 2 diabetes (Figure 1). (2) Flow Chart incident type 2 diabetes mellitus Model (Figure 2) that were made in powersim. (3) Calculating the mean value of each variable based on the group with type 2 diabetes and non-type 2 diabetes group obtained from Basic Health Survey Data in 2013 (Table 1). (4) Analysis of DM and non-DM incidences data using powersim sofware.

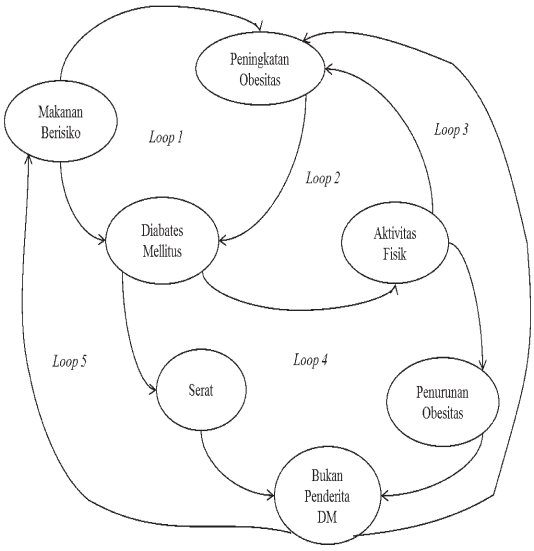


Figure 1. Causal Loop DM Type 2 (Modified from Seoul, 2014)

Figure 1 shows the risk of obesity and DM are higher for higher consumption of sweet, salty and fatty food (loop 1) as well as lower physical activity (loop 2) The number of obesity will also reduce while the number of non-DM increases by increasing physical activity and fiber intake (fruits and vegetables) (loop 4). Non-DM population potensially suffering from DM if risky food consumption as well as obesity are also increasing (Loop 3 and 5)

Figure 1 then translated in to the form of Dynamic Model Flow Chart DM incidence in Powersim program.

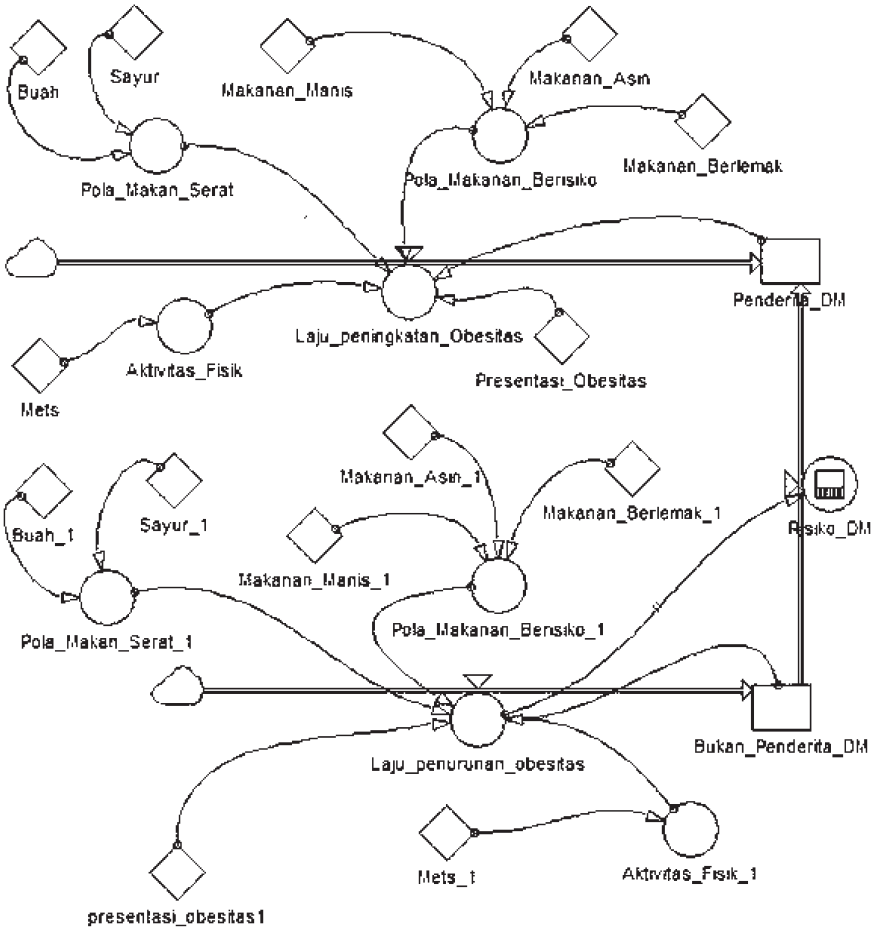


Figure 2 Flowchart Model DM incidence

Model and Structure Validation

The model was built on the basis of type 2 diabetes incident in a causal loop. The causal loop is according to the existing system. The model has been verified by looking to the formulation and test results that running well.

Output validation

Average Marginal Effect (AME), Abstinence Violation Effect (AVE) and Kalman Filter (KF) test to Type 2 diabetes incidence data were used as output validation

Model Simulation I –VI Explanation

Model Simulation is built based on diabetes mellitus and metabolic diseses controlling guidlines in community based. The model simulation is also focusing on the portion setting of fruits and vegetables consumption according to WHO recomendation (National Health Department, 2008) and focus on setting up the

1. Model Simulation 1 estimate the incidence of DM type 2 in the next 10 years (2013-2022) without any controls on the factors affecting the incidence of type 2 diabetes mellitus
2. Model Simulation 2 shows an increased fruits and vegetables servings to be 1 portion each.
3. Model Simulation 3, fruits serving increases into 2 portions and 1 portion vegetables
4. Model Simulation 4, both fruits and vegetables are increasing into 2 portions.
5. Model Simulation 5, increasing servings of fruits into 3 portion and 2 portions for fruits and vegetables, respectively.
6. Model Simulation 6, increasing servings of fruits and vegetables into 3 portions each.

Table 1 Variable Characteristics Based on Model Simulation Dynamic

Variable Characteristics	MS 1	MS 2	MS 3	MS 4	MS 5	MS 6	Non – DM
Fruit Consumption servings / day	0,52	1	2	2	3	3	0,44
Vegetables Consumption servings / day	1,19	1,19	1,19	2	2	3	1,19
Sweet Food/Baverage Consumption	0,49	0,49	0,49	0,49	0,49	0,49	0,67
Salty Food Consumption	0,35	0,35	0,35	0,35	0,35	0,35	0,37
Fatty foods Consumption	0,44	0,44	0,44	0,44	0,44	0,44	0,47
Physical Activity (Mets)	1,95	1,95	1,95	1,95	1,95	1,95	2,09
Basic Level (DM Type 2 Total Incidence) (People)	692	692	692	692	692	692	11957
Percentage of Obesity (BMI) (%)	17,3	17,3	17,3	17,3	17,3	17,3	13,5
Delay (From Non-DM Type 2 diabetes to DM Type 2) (Years) (Barbara, 2000)	4	4	4	4	4	4	-

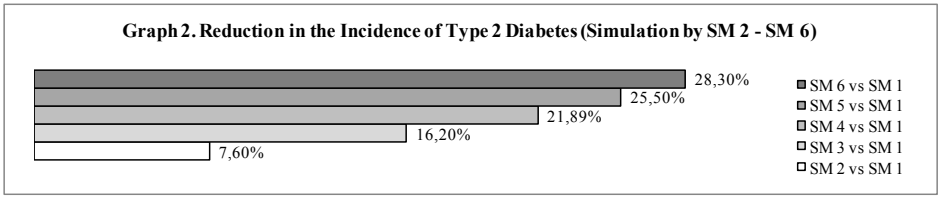
RESULTS

Graph 1 shows the increaseing trend of type 2 diabetes mellitus incidence by 3,53 times in 10 years, from 692 people in 2013 to 2443 people in 2022 without any control over the factors affecting the incidence of DM

Increaseing servings of fruits and vegetables consumption showed a downward trend in the incidence of type 2 diabetes. The more portions of fruits and vegetables consumed, the greater decreaseing of type 2 diabetes incidence shown in the model. Model Simulation 6 shows the greatest decrease of type 2 diabetes incidence compared to other models. 28,3% decreasing incidence found in model simulation 6 compared to model simulation 1 (Graph 2)

DISCUSSION

The study results showed that increasing consumption of fruits and vegetables servings each day can prevent the increased incidence of Type 2 diabetes by 28.3% in the next 10 years.



Fruits and vegetables low consumption from a young age (<45 years) plays an important role in the natural history of Type 2 diabetes diseases characterized by a decrease in insulin sensitivity arising as a result of genetic predisposition and lifestyle (environmental factors) which was initially offset by a decreasinge function of cell β (Hansen, 2002)

Lifestyle factors such as smoking, alcohol consumption, less physical activity, less fiber intake, high fat and calorie intake also initiate high blood glucose levels in the bodies and eventually can be diagnosed as Type 2 Diabetes Mellitus

These study results are consistent with the WHO recomendations of fruits and vegetables consumption of 400 gram per day or at least 5 servings per day that consists of 3 servings of fruit and two servings of vegetables to prevent chronic diseases.

This study is using dynamic modeling simulation of blood glucose levels after complex carbohydrate consumption followed by a delicious dessert. Foods like beans, rice, oats and other cereal grains, fresh fruit, vegetable, bread and pasta, contain combinen sugars and starches. Digestion turns on these complex carbohydrates into glukosa in a more equilibrated

speed the rapid absorption of simple sugars. The rate release of blood of pasta is no as high as that of a candy, it might remain stable at 400 (mg/min) (Pedro Dagoberto, 2013).

The high incidence estimation of type 2 diabetes mellitus in the next 10 years indicates a serious problem. In addition to the drug, redesigning the lifestyle pattern, especially diet and exercises is very important for dibetic and non diabetic patiens.

At the macro level, the high incidence rate of type 2 diabetes mellitus is associated with urbanization and environmental transition, including changes in the work patterns. Heavy works have been shifting into light work (sedentary). The used of computers, mechanics and means of transportations are also rapidly increased. Economic growth and environmental transition led to drastic changes in food production, processing and distribution systems as well as increased accessibility of unhealthy foods (National Health Department, 2008). Changes in diet and decreased physical activity levels mostly resulted on weight gain of men and women around the world (Popkin BM, 2012).

Some researches indicate the positive ability of fiber source food (fruits and vegetables) in reducing the risk of diabetes, improving glucose metabolism and insulin sensitivity in DM patients. Wolfram et al (2011), suggests that increasing the consumption of vegetables, grains, and soluble and insoluble fibers are associated with better glucose metabolism in diabetic and non-diabetic individuals. Improvement on insulin sensitivity and glucose homeostasis are more pronounced in respondents with a plant-based diet compared to respondents with commonly diet (Tungsten T, 2011). Increasing dietary fiber intake was associated with better glycemic control, improved insulin sensitivity and micro-inflammation in patients with type 2 diabetes in Japan. The study encouraged diabetic patients to continue consuming more sources of fiber food in everyday life (Fujii, 2013)

Although the meta-analysis results showed no clearly association betweenfruit and / or vegetables intake with reduced risk on type 2 diabetes mellitus incident. However, increased consumption of green leafy vegetables about one serving per day was associated with a significant decrease risk of type 2 diabetes for about 14%. Futhermore, vegetables consumption may protect individuals from diabetes mellitus development (Fujii, 2013) (Cene, 2011)

Global Action Plan for Prevention and control of Noncommunicable disease 2013 -2033 recommended to increase availability, affordability and consumption of fruit and vegetables, reduce the level of salt/ sodium added to food (prepared or processed), Reduce the content of free and added sugars in food and non-alcoholic beverages. (WHO, Global action plan for the prevention and control of noncommunicable diseases 2013-2020., 2013).

The presence of fast-food restaurants have expanded exponentially in the last few decades. Rapid increasing on the availability of fast food have contributed to unhealthy diets with high calorie content; large portion sizes; and processed meat in large quantities; Very simple carbohydrates consumption; sweet drinks; and unhealthy fats intake. Futhermore, food transition and livestock revolution led to increased production and consumption of beef, pork, , eggs, dairy and poultry products (Ezzati M, 2013).

Health funding disease-related type 2 diabetes estimated to 50% increase from $ 286 billion in 2003 to $ 396 billion in 2025. The increasing prevalence and high cost treatment of type 2 diabetes will lead to heavy economic burden and a major challenge to policy makers on managing this chronic disease in developing countries (Ibrahim, 2010)

The Supports on preventing DM are required. A regulation should ensure that safe fruits and vegetables are available for people. Public facilities should also available to encourage people to be physically active.

Estimates of the incidence of type 2 diabetes mellitus by setting portions of fruit and vegetable consumption using a dynamic model approach provides rapid results. However, the simulation results may not be exactly the same with the real world.The model contains more or less distortion (aberration), and are possible to the GIGO (Garbage In, Garbage Out). Meaning that if we enter the wrong data, theoutput simulation will also get wrong. Thus can be concluded that the results of a simulation depends on the data input (Ahmad, 2012).

In this study, we use computer simulations to overcame the problems. Correcting errors of calculation can be done directly, reviewing, and repetition simulation. Repetition is done primarily in changing various components and variables, such as

parameters andprogram operating conditions.

Be interesting to apply this knowledge in future works to model the behavior of other regulatory mechanisms such as the body temperature control, the heart rate, the water amount contained in the blood and breathing. All of these systems share similar structures like cause-effect cycles with negative feedbacks mechanisms that allow homeostasis and regularizes any changes in the parameters of a healthy person

CONCLUSION

Increased portion of fruits and vegetables consumption every day have a greater impact to prevent the incidence of type 2 DM.

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ANALYSIS OF BIOACTIVE SUBSTANCES IN AVOCADO (*PERSEA AMERICANA* MILL.) LEAVES EXTRACT AND PRECLINICAL TESTING IN LOWERING BLOOD GLUCOSE LEVEL IN MICE (*MUS MUSCULUS*)

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ABSTRACT

Diabetes is the oldest disease in the world, the death rate cause by diabetes mellitus is increasing. This study aimed to know bioactive substances of avocado leaves and effectiveness of concentrate in decreasing the blood glucose level of mices. The study applied experimental method to analyzed bioactive substances in avocado leaves extract qualitatively, whereas the preclinical testing used random group design. This study used 15 male mice as test animal which are induced with etilen diamin tetra asetat (EDTA). The mice are randomly divided into 5 groups (n=5) with different groups. Group I, II, III the mice are given avocado leaves extract each with concentration of 10%, 20% and 40%, group IV is given suspension of Glibenclamide as a positive control (+) and group V as a negative control (-). Data obtained was analysed by using statistical analysis of varians (ANOVA) which is continued with Duncan test. The research result showed that avocado leaves extract positively contains compounds of alkaloid, flavonoid, saponin, tannin and steroid. Preclinic test showed that avocado leaves extract proven can lower the blood glucose level of mice and relatively the most effective concentrate was 10%.

Keyword: alkaloid, avocado leaves, blood glucose level, diabetes mellitus

INTRODUCTION

Type 2 diabetes mellitus (T2DM) now become one of the non-communicable diseases (NCDs) are most common around the world. Type 2 diabetes is also among the major causes of death in most developed countries and developing countries. Complications of type 2 diabetes, such as coronary artery and peripheral vascular disease, stroke, diabetic neuropathy, amputations, kidney failure, impotence, and blindness. These diseases lead to increased disability, reduced life

expectancy and level of health care costs for society at large. Type 2 diabetes has become a public health problem that is most influential in the 21st century (Sicree et al., 2011).

WHO data in 2000 showed that the number of patients with diabetes mellitus in Indonesia ranks sixth, as many as 8.4 million people. Indonesia is expected to rank fourth in the world after India, China, and the United States in 2030. The increase in the number of patients is expected to reach 21.3 million over the next two decades (Wild et al., 2004).

Results of National Basic Health Research of Indonesia in 2013 showed the tendency of T2DM prevalence increased by 1% from 1.1% in 2007 to 2.1% in 2013 (Balitbangkes, 2008 ; 2013). In addition, the proportion of T2DM at the age of ³ 15 years old was 6.9%. If based on the diagnosis and symptoms, the prevalence of T2DM in Central Sulawesi was 3.7%. This showed the Central Sulawesi as the province with the highest prevalence of T2DM on Sulawesi (Balitbangkes, 2008).

Patients with T2DM susceptible to excessive free radical formation. This can caused damage of pancreas and disrupt the function of pancreatic beta cells and insulin resistance, which can aggravate the condition of diabetes (Jeli and Makiyah, 2011). Harmful free radicals requires antioxidants that can coutering them. One is the use of alternative drugs that are relatively inexpensive and usefulness not much different from synthetic drugs.

Traditionally many plants that can lower blood glucose levels, among others avocado leaves. Central Sulawesi Province is rich in natural resources, especially the avocado plants, but the leaves are underused. During this time the water decoction of avocado leaves can be used also as a medication for diabetes mellitus. However, research on the type of phytochemical contents, preclinical testing and clinical, avocado leaves for the prevention of this disease is still relatively limited.

Avocado plant parts, especially the avocado pulp has been reported to lower blood glucose levels significantly. Brai et al. (2014) and Al-Dosari (2011), found that the avocado pulp and seeds contain saponins, alkaloids, flavonoids, and tannins. While the seeds of avocado also reported effectively lower blood glucose levels of mice that had been induced with alloxan (Alhassan et al., 2012; EzEjiofor et al., 2013). Based on that,

allegedly avocado leaves also have antioxidant activity as indicated by the pulp and seeds of avocado.

This study aimed to: (i) identified phytochemical compounds in extract of avocado (*Persea americana* Mill.) leaves extract, (ii) determined the concentration of avocado (*Persea americana* Mill.) leaves extract that effectively lower blood sugar levels in mice (*Mus musculus*).

METHODS

1. Sample Preparation

   Before extraction, avocado leaves are made into powder with the following steps:

   a. Selected parts of leaves that were not too old (fifth leaf from the top).
   b. Leaves cleaned first with water, then be dried.
   c. Avocado leaves areweighed 500 grams and dried for 5 days in the open with good air circulation but not exposed to direct sunlight.
   d. The dried avocado leaves further blended until smooth, then sifted.

Chemicals and Reagents Used

The chemical solution and reagents used were Methanol, Wagner's and Mayer's reagents, distilled water, HCl, $FeCl_3$1%, concentrated H_2SO_4, Na-CMC, Glibenclamide, EDTA (Ethylene Diamine Tetraacetic Acid), lead acetate, ammonia, sulfuric acid, and potassium hydroxide.

2. Screening of Phytochemical

   The screening of phytochemical as follows :

   a. Alkaloids

      0.2 g of the extract was added to 5 ml of 2% hydrochloric acid and heated on boiling water for 10 minutes. It was then allowed to cool and then filtered. To 1 ml of the filtrate in a test tube was tested alkaloids reagent, Wagner's and Mayer's reagent and results are compared with blank. Turbidity or precipitation indicated the presence of alkaloids (Ekeanyanwu et al., 2012).

   b. Flavonoids

      0.1 g of each of the extract was added to a mixture of 10 ml of lead acetate solution (90% w/v) and 20 ml of 50% aqueous ethanol in a 200 ml conical flask. The mixture was placed on boiling water for

2 min, cooled and filtered. Five milliter (5 ml) of dilute ammonia was added to a portion of the aqueous filtrate followed by the addition of concentrated sulphuric acid (1 ml) to 2 ml of potassium hydroxide solution and allowed to mix. Then, into the acid base mixture, a small quantity of aqueous filtrate of the sample was added and observed for colour change (Ekeanyanwu et al., 2012).

c. Saponins

0.1 g of each of the extract was measured into a beaker and 20 ml of distilled water was added, the beaker was heated in a water bath for over 5 min. The mixture was filtered using a filter paper into another beaker to obtain a filtrate. Two milliter (2 ml) of the filtrate was poured in another test tube and 10 ml of distilled water was added, it was shaken vigorously for over a minute. Frothing which persist on warming indicated the presence of saponin (Ekeanyanwu et al., 2012).

d. Tannins

0.2 g of the extract was boiled with 5 ml of 45% ethanol for 5 min. The mixture was filtered hot using a filter paper and filtrate was collected in a beaker. Two milliter (2 ml) of the filtrate was mixed with 10 ml of distilled water and then a drop of iron chloride solution was added. A blue-black or blue-green precipitate indicates the presence of tannins (Ekeanyanwu et al., 2012).

3. Preparation of Solutions and Suspensions

The preparation of the solutions and the suspensions were as follows (Dharmayudha, 2011) :

a. Makingof 1% w/v Na-CMC Colloidal

1% Na-CMC colloidal was prepared by dissolving 1 gram of Na-CMC little by little into 50 ml of hot distilled water while stirred to form a colloid. Making colloids used distilled water made up to 100 ml.

b. Making of Glibenclamide Suspensions

Take 1 tablet of 5 mg Glibenclamide, then crushed in a mortar and then added 1% (w/v) Na-CMC colloidal little by little. After that, that mixture was crushed until homogeneous. The suspension was put in a 100 ml flask were then mixed with 1% (w/v) Na-CMC colloidal by volume to 100 ml.

c. Making of Avocado Leaves Extract in Different Concentrations

Avocado Leaves Extract was made using Dekok Method. Avocado leaves powder weighed 10 grams and then put in a beaker containing about 100 ml of distilled water. Further heated for 15 minutes at a temperature of 90°C. Then the mixture was filtered hot using a flannel. The volume was less than 100 ml, then added warm water through the filter until the residue volume reached 100 ml. The same way done in the manufacture of avocado leaves extract 20% and 40% are made using 20 grams and 40 grams of avocado leaves extract.

4. Animals

Test animals used were mice (*Musmusculus*) health, male, aged 2 to 3 months with weight varying from 25 to 30 grams. Mice are used as much as 15 tails and are divided into five treatment groups. Each group consisted of three mice in which the three mice are placed separately for each treatment.

Mice are fasted for 4 to 5 hours, yet still supplied drinking water *ad libitum*. Their weight was weighed and their fasting blood glucose levels measured on day 0. EDTA was injected once as much as 150 mg/kg of body weight intravenously (Suharmiati, 2003). On day 3, their blood glucose levels re-measured to ensure the levels of EDTA still served as an experimental diabetic. Then they areinduced 10% glucose. During the treatments, the mice were still given feed.

The group is given as follows:

1. Group I (G1): Diabetic mice administered *(Perseaamericana Mill.) Leaf* extract (10%)

2. Group II (G2): Diabetic miceadministered *(Perseaamericana Mill.) Leaf e*xtract (20%)
3. Group III (G3): Diabetic mice administered *(Perseaamericana Mill.) Leaf* extract (40%)
4. Group IV(G4): Diabetic mice administered *Glibenclamide (positive control)*
5. Group V (G5): Diabetic mice*(negative control)*

After being given the treatments, all the mice are put in each cage and given food. Drinks provided *ad libitum*. Blood glucose levels are measured on day 3.

5. Data Collection and Analysis

Data from the measurement of blood glucose levels in mice are then averaged for each group. Then analyzed and evaluated using a randomized block design with analysis of variance (ANOVA) with a 95% confidence interval (F-test). This test was used to determine whether the groups given between a significant differences or not. There were significant differences and the test continued with Duncan test.

RESULTS AND DISCUSSIONS

1. Phytochemical screening

Phytochemical analysis of the extract of *Persea americana* Mill. revealed the presence of flavonoids, tannins, alkaloids, and saponins as shown in Table 1. The phytochemical screening aimed to determine the existence of secondary metabolites, which are expected to act as antihyperglycemia or antidiabetic.

Table 1. The phytochemical composition of the leaves extractof *Persea americana* Mill.

Compounds	**Results***
Flavonoid	+++
Tannin	+++
Alkaloid	++
Saponin	+

*+++ = Present in high amount, ++ = present in moderately high amount, + = present in trace amount

2. Effects of Leaves Extract Avocado (*Persea americana* Mill.) to Decreased Blood Glucose Levels

Effects of avocado's (*Persea americana* Mill.) leaves extract to a decrease of blood glucose levels in mice was shown in Table 1. The data

in Table 1 indicated that the initial glucose concentration in mice was relatively low and there was no significant difference among all groups. Blood glucose concentrations in mice at 48 hours after induction with EDTA increased sharply to 150.67 mg/dl to 161.33 mg/dL or mice that had suffered from diabetes mellitus. Results of homogeneity test of blood glucose levels after all groups of mice induced EDTA, showed no significant difference between groups ($p > 0.05$ or significant value $0.157 > 0.05$).

Perkeni (2011) said that a person is said to suffer from diabetes if they have a fasting blood glucose levels > 126 mg/dl and blood glucose levels at 2 hours after eating (postprandial) > 200 mg/dl. Evacuasiany et al. (2010) also said that the requirement for the state of hyperglycemia in test animals is when the blood glucose levels > 120 mg/dl.

To find out how much decreased in blood glucose levels in mice, they calculated the differences between blood glucose levels after induction with after the treatments. The mean was then calculated as shown in Table 2.

Table 2. Average of initial blood glucose levels of mice, after induction, and after the treatments

Groups	(C0) (mg/dL)*	(C1) (mg/dL)*	(C2) (mg/dL)*	(C3) (mg/dL)*
G1	81.00 ± 3.00^{a}	158.33 ± 10.50^{a}	77.00 ± 23.58^{a}	81.33^{a}
G2	81.33 ± 5.13^{a}	155.33 ± 7.37^{a}	107.33 ± 4.73^{b}	48.00^{b}
G3	74.00 ± 8.54^{a}	150.67 ± 2.52^{a}	106.33 ± 6.66^{b}	44.34^{b}
G4	70.33 ± 13.28^{a}	156.00 ± 4.58^{a}	111.33 ± 4.73^{b}	44.67^{b}
G5	76.67 ± 19.04^{a}	161.33 ± 7.23^{a}	153.33 ± 7.02^{c}	8.00^{c}

*C0 = initial blood glucose concentrations ; C1 = blood glucose concentrations after induction of EDTA ; C2 = blood glucose concentrations after intervention ; C3 = decrease of blood glucose concentrations

^{a,b,c}The numbers with the same letter in the same column showed no significantly different at $\alpha = 0.05$

The data in Table 1 showed after diabetic mice given the leaves extract of avocado with concentrations varied, it appeared that the average blood glucose levels of mice decreased dramatically (G1, G2, G3, and G4), and significantly different from the group who did not used antihyperglycemic medication (Glibenclamide) and avocado leaves extract (G5). The determination of the existence of significant differences between the five groups, conducted by statistical tests using analysis of variance (ANOVA).

Based on the calculation of statistical advanced test (The Real Distance Duncan), with a level of significant 5% ($p < 0.05$), showed that the extract of avocado leaves 10% relative to more effectively reduced blood glucose levels in mice compared extract of avocado leaves 20% and 40% (G2 and G3), as well as Glibenclamide / positive control (G4) and G5 (negative control). The low effectiveness of avocado leaves extract with a higher concentration (G2 and G3) compared to the G1 was often found in the activity of extract of natural materials were multicomponent mixtures. The effects of these components could be mutually synergistic, or antagonistic, so the avocado leaves extract concentrations higher exacerbate insulin-producing tissue damage (Yulinah et al., 2001).

The ability of avocado leaves extract lowered blood glucose levels supposedly influenced by bioactive substances it contained, namely alkaloids, flavonoids, saponins and tannins. Repair damaged of pancreatic cells by EDTA supposedly because of the activity of the active compounds in the extract of avocado leaves were flavonoids and alkaloids. Anti-inflammatory activity of flavonoids and antioxidant activity can prevented and stopped the destruction of pancreatic β cells. While alkaloids played a role in cell regeneration to restore pancreatic β cells that are damaged partially and then increases insulin secretion (Gupta and Neera, 2006; Tende et. al., 2011). Increasing the amount of insulin in the body will increased the amount of blood glucose levels into the cells, resulting in decrease of blood glucose levels.

Flavonoids also had properties as an antioxidant

that can protected damage pancreatic cells from free radicals, while alkaloids lowered blood glucose levels by inhibited glucose absorption in the intestine, increased the transport of glucose in the blood, stimulates the synthesis of glycogen, and inhibited the synthesis of glucose and the enzyme glucose 6-fosfatase, fructose 1,6-bifosfatase, and increased the oxidation of glucose by glucose 6-phosphate dehydrogenase. Glucose 6-phosphatase and fructose 1,6-bifosfatase were enzyme that played a role in gluconeogenesis. Inhibition both of enzymes will reduced the formation of glucose from other substrates apart from carbohydrates (Arjadi and Susatyo, 2010).

Flavonoids also had an activity by inhibited the enzymes involved in the breakdown of carbohydrates into monosaccharides which can be absorbed by the intestine, the α-amylase enzyme and α-glucosidase enzyme (Kurniawati et al., 2010). One flavonoid substance with hypoglycemic effects was quercetin. Hii and Howell (1985) in Arjadi and Susatyo (2010) suggested that quercetin may increased insulin secretion from the islets of Langerhans cells via Ca^{2+} metabolism changed.

Tannins are known to stimulated glucose metabolism and fat, so that the pile of these source of calories in the blood can be avoided. Tannins had antioxidant activity and hypoglycemia; increased glikogenesis. In addition, also served as an astringent tannins or chelated which can be screwed epithelial membrane of the small intestine, reduced the absorption of nutrients thereby blocked the intake of sugar and the rate of increased in blood sugar levels was too high (Monica, 2006).

Synthetic drugs were generally consumed for patients with diabetes mellitus named Glibenclamide. Glibenclamide was sulfonylurea derivatives oral hypoglycemic actively worked to lowered blood sugar levels. Glibenclamide worked by stimulated the secretion of insulin from the pancreas. The active compounds worked in Glibenclamide were alkaloids and flavonoids that helped in the processed of lowered of blood sugar levels and can lowered blood glucose levels

within 3 hours and can sustained for 15 hours (Dharmayudha, 2011). It can be connected to the processed of lowering of blood sugar by used herbal medicine extract of avocado leaves that allowed of the most active compounds to lowered blood sugar were alkaloids and flavonoids, so effectively used as an alternative herbal medicine, especially in patients with diabetes mellitus.

CONCLUSIONS AND RECOMMENDATIONS

Conclusions

The extract of avocado (*Persea americana* Mill.) leaves contained a phytochemical compounds were alkaloids, flavonoids, saponins, and tannins. The extract of avocado (*Persea americana* Mill.) leaves relatively most effectively lower blood glucose levels were at a concentration of 10% (w/v) with $p < 0.05$.

Recommendations

Further research is needed in the form of stage clinical trials for patients with Type 2 Diabetes Mellitus.

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THE CORRELATION BETWEEN OBESITY AND PHYSICAL ACTIVITY AND HYPERTENSION INCIDENCE OF INPATIENT AT ANUTAPURA PUBLIC HOSPITAL IN PALU

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ABSTRACT

Hypertension is a disease that the blood pressure is more than 130/85mmHg. Hypertension is a chronic medical condition of elevated blood pressure in cardiovascular system hich can caused damage in heart, kidneys, eyes, and brain. The research as aimed to find out the correlation between obesity and physical activity and hypertension incidence of inpatient at Anutapura Public Hospital in Palu. The design of this research was an observational analytical with cross sectional study approach. There were 78 samples, which were retrieved by purposive sampling technique. The result shows that there is no correlation between obesity (p=0.055) and hypertension incidence. there was correlation between physical activity (p=0.000) and hypertension incidence. To prevent from hypertension, ideal physical activity is needed to get healthy body and lower the blood pressure.

Keywords: obesity, physical activity, hypertension

INTRODUCTION

The prevalence of hypertension in the world by 15 to 20%, while in Asia has reached 8 to 18%. Based on a report from the University of Auckland New Zealand showed that more than 80% of hypertension disease occured in developing countries (Asdi, 2008).

Based on the results of the Household Health Survey in 2009, it is known that the prevalence of hypertension in Indonesia of 8.3%, and rising to 21% in 2010. Results ofIndonesia Basic Health Research showed that the national prevalence of hypertension reached 31.7% of the total adult population, while for Palu City's prevalence was 12% (Balitbangkes, 2011).

Hypertension hospitalized patients in Anutapura Palu General Hospital during 2009, there were 409 patients. Although in 2010 had decreased by 264 patients, but in 2011 increased again, reaching

434 patients. Meanwhile, there were 743 outpatients in 2010 and increased to 985 patients in 2011 and for hospitalized patients during 2012 as many as 546 patients at one time put this disease is ranked 4th out of 10 diseases suffered by hospitalized patients and also signified that sufferers increased from the previous year (Anutapura Palu General Hospital Profile, 2012).

Based on preliminary studies, having conducted interviews in some patients with hypertension in Anutapura Palu General Hospital, information was obtained that of the 20 patients, 15 of whom were obese (Anutapura Palu General Hospital Profile, 2012). This is supported by several studies that claimed that the number of obese people continues to increase every year, especially in big cities. Meanwhile, the results of a national survey conducted in 2009 in the capital of all provinces in Indonesia showed that in the age group 18 years old and over, that were overweight category by 8.1% in men and 10.5% in women, while in men with obese by 6.8% and in women by 13.5%.

In addition, based on preliminary studies that out of 20 patients with hypertension in Anutapura Palu General Hospital only 5 patients who regularly perform physical activity (an average of 3 times per week). Some studies related to the relationship between physical activity and the incidence of hypertension that showed that men with physical inactivity, were at risk of suffering from hypertension of 1.39 times, whereas in women by 1.28 times. Furthermore, the results of Kaplan (2008) research noted that men with physical inactivity, were at risk of suffering from hypertension of 1.43 times and women of 1.27 times (Sudikno, 2010).

Therefore, based on these data, the research about the relationship between obesity and physical activity factors with hypertension hospitalized patients in Anutapura Palu General Hospital.

METHODS

Type of research

This type of research used was observational analytic method with cross sectional study approach. This research was conducted inHospitalized Department of Anutapura Palu General Hospital. West Palu Subdistrict, Central Sulawesi, Indonesia in 2013.

Population and Sample

The population was the entire of hospitalized patients in internal medicine care room in Anutapura

Palu General Hospital as many as 6025 patients. The sample was part of the population that represents all existing the hospitalized patients. The sample size will be calculated using the Lemeshow formula:

$$n = \frac{NZ^2p(1-p)}{(N-1)d^2 + Z^2p(1-p)}$$

Informations:

n = Large sample

N= Population size estimation

Z = Normal standard on confidence interval (with α = 5% = 1.96)

d = level of accuracy (value of 0.05)

p = estimation of proportion would be studied estimation

Thus, a large sample of hospitalized patients in internal diseases care room in the studied as many as 78 respondents.

Data Analysis

Data analysis in this study is done by using a computerized program. Univariate analysis is used to illustrate the characteristics of the study subjects by calculating the frequency distribution of some of the factors associated with hypertension in hospitalized patients as characteristic of obesity and physical activity. The bivariate analysis is used to determined the relationship of the independent variables and the dependent variables. Test used Chi square (X^2) with α = 0.05 and confidence interval = 95%.

RESULTS

Table 1. Distribution of Respondents According to Characteristics of Hospitalized Patients in Anutapura PaluGeneral Hospital

Characteristics	n (78)	(%)
Sex		
Men	30	38.5
Women	48	61.5
Age Groups		
< 42 years old	29	37.1
≥ 43 years old	49	62.9
Occupations		
Students	9	11.5
Farmers	7	9.0
Civil Servants	20	25.6
Housewives	23	29.5
Entrepreneurs	19	24.4

Table 1 showed that most respondents were female of 61.5% with age were ≥ 43 years old of 62.9% and generally work as a housewife by 29.5%

Table 2. Overview of Research Variables

Research Variables	n (78)	(%)
Independent		
Obese Status		
Obese	26	33.3
Not obese	52	66.7
Physical Activity		
Less	65	83.3
Sufficient	13	16.7
Dependent		
Incidence of Hypertension		
Hypertension	50	64.1
Not hypertension	28	35.9

Table 2 showed that patients were generally not obese by 66.7% with less physical activity of 83.3%. The incidence of hypertension showed that most patients had hypertension of 64.1%.

Table 3. Bivariate Analysis

Variables	Incidence of Hypertension				Total		p
	Hypertension		Not hypertension				
	n (50)	% (64.1)	n (28)	% (35.9)	N (78)	% (100.0)	
Obese Status							
Obese	21	80.8	5	19.2	26	100.0	0.055
Not obese	29	55.8	23	44.2	52	100.0	
Physical Activity							
Less	48	73.8	17	26.2	65	100.0	0.000
Sufficient	2	15.4	11	84.6	13	100.0	

Table 3 showed that there was no significant relationship between obesity and hypertension, with p = 0.055 (p > 0.05) in which patients who were obese and non-obese have the same percentages of incidence of hypertension was same great. As for physical activity showed that there was significant relationship between physical activity with hypertension with p = 0.000 (p < 0.05), where patients have less physical activity had a high percentage of hypertension incidence was 73.8%.

DISCUSSIONS

A. Relation of Obesity with Incidence of Hypertension

The results showed that there was no correlation between obesity and hypertension, with p = 0.055 (p > 0.05). This was consistent with

the theory of Sulastri (2008) which stated that the mechanism is not yet known for sure that explain more particularly the nutritional status of obesity as a cause of hypertension because not all obese people can develop hypertension.It was also reinforced by research conducted by Wilsgard (2008) in Norwey which indicated that there is no significant relationship between obesity and hypertension which the obesity respondents as many as 60% had blood pressure under control, while respondents who are not obese only 58% of respondents had blood pressure controlled.It was also increasingly supported by research conducted by Sari (2009) in Kendari Community Health Center that showed that p = 0.435 (p> 0.05) with test results ofOdds Ratio (OR) = 2.333 with a lower limit (LL) value = 0.264 and the upper limit (UL) value = 20.659. Which found that 15 respondents (23.5%) who had hypertension with obese and the majority of respondents were 49 respondents (76.5%) who had hypertension with no obese.

There are several things that can lead to obesity, one of which is the consumption habits of unhealthy foods (snacking). Snacking is a habit of eating is done outside meal times (snack), and the food that is consumed in the form of a snack that has delicious tastes, sweet and usually fried. If this habits is not controlled can lead to obesity, because these foods include high calories.

This is proven by the results of studies showed that of 26 obese patients only 3 patients who did not have the consumption habits of unhealthy snack, while 23 others had the consumption habits of unhealthy snack.

Type of snacks that was often they consumed consisted of soft drinks, fried foods, fast food, chocolate, bread, biscuits, wafers, and cake which contains a lot of high sugar and fat and low in fiber. When consumed continuously and not balanced with other healthy foods will be cause an imbalance of calories for energy intake far exceeds the needs of the body. Excessive accumulation of fatty deposits that will occur in the body and have an impact on weight gain. This was according to research of Sheps (2007) stated that obesity is closely associated with a penchant to consume foods that contain high fat.

B. Relation of Physical Activity with Incidence of Hypertension

The results showed that there was a relationship between physical

activity (exercises activities) and the incidence of hypertension with a $p = 0.000$ ($p < 0.05$). This happened because patients were less moving so that the condition of the body did not fit because it could cause blood vessels less relaxation. The pressure on the artery until the narrowing of blood vessels that ultimately lead to hypertension. However, in patients suffering from hypertension were still found 15.4% were enough of physical activity.

Cahyono (2008) suggested that exercise can reduce blood levels of low density lipoprotein (LDL). Through exercises activities, the heart can work more efficiently, pulse frequency is reduced, otherwise the heart's pumping strength is getting stronger, a decrease in body fat and weight and also lowering blood pressure. It was also reinforced by research conducted by Paffenbarger (2009) which suggested that the incidence rate of hypertension in the United States is 20 to 40% lower in those who do exercises activities at least 5 hours per week than those who are less active in exercises.

According to Kaplan (2008), physical activity can help prevent hypertension and blood pressure of people who have been suffering from hypertension, can be reduced by regular exercise. This relationship may involved with insulin resistance due to increased insulin resistance associated with exercises activities less in normotensive people with a family history of hypertension. In one session of exercise or physical activity, the average of lower blood pressure of 5 to 7 mmHg. Its effects can last up to 22 hours after exercise. After 4 to 6 months (long term) can lower blood pressure by 7.4 to 8.5 mmHg.Someone who exercise regularly proven to lower blood pressure to normal levels and reduce the risk of hypertension by 50% than not doing exercises.

CONCLUSION

Physical activity was a variable associated with hypertension in hospitalized patients in Anutapura Palu General Hospital.

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CORRELATION BETWEEN P WAVE DISPERSION, QRS DURATION & QT DISPERSION IN HOSPITAL EVENTS IN CASES OF ACUTE CORONARY SYNDROME

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Background: The acute coronary syndromes encompass a spectrum of unstable angina to transmural myocardial infarction. The definition of acute coronary syndrome depends on the specific characteristics of each element of the triad of clinical presentation, electrocardiographic changes and biochemical cardiac markers.

Objectives: To assess Correlation between P wave dispersion, QRS duration & QT dispersion in hospital events in patients with acute coronary syndrome (ACS).

Methods: This prospective study was conducted on 60 patients with acute coronary syndrome admitted to critical care units of Alexandria University Hospitals from first of January 2012 to end of August 2012. An informed consent was taken from relatives of every patient included in the study. This study was approved by ethical committee of Alexandria Faculty of Medicine.

Result: During the observation up to 5 days, 1 patient (1.6%) developed AF. There was no significant correlation between P wave dispersion(PD) in the three studied groups& No significant relation between it & the whole complications developed in failed thrombolysis group (p> 0.05). PD alone couldn't predict AF in this study, as the mean was 20.0 milliseconds, which was too small to predict AF alone. There was no significance difference between QRS duration in the three groups (unstable angina, successful thrombolysis and failed thrombolysis respectively with mean value (75.7 ± 14.02, 75.2 ± 10.88, 73.36± 8.81) (p> 0.05) & the relationship between the QRS duration and the development of complications was non-significant in this study. This study showed that there was a significant relation among the three groups & highly significant correlation between QTD duration and prediction of complications. It was clear that ICU length of stay was longer in patients with failed thrombolysis in comparison with the other two groups (p<0.05).

Conclusions: Importance of measurements of QT Dispersion to patients with acute coronary syndrome especially at admission. Failed thrombolysis patients should be under close observation and monitoring till another option is available like coronary intervention.

Keywords: ACS (Acute Coronary Syndrome), QTC (Corrected QT Interval), QTD (QT Dispersion)

INTRODUCTION

The acute coronary syndromes encompass a spectrum of unstable angina to transmural myocardial infarction. The definition of acute coronary syndrome depends on the specific characteristics of each element of the triad of clinical presentation, electrocardiographic changes and biochemical cardiac markers.[1]

In the emergency setting, ECG is the most important ED diagnostic test for angina. It may show changes during symptoms and in response to treatment, confirm a cardiac basis for symptoms. It also may demonstrate preexisting structural or ischemic heart disease (left ventricular hypertrophy, Q waves). A normal ECG or one that remains unchanged from the baseline does not exclude the possibility that chest pain is ischemic in origin. Changes that may be seen during anginal episodes include the following: Transient ST-segment elevations Dynamic T-wave changes - Inversions, normalizations, or hyperacute changes ST depressions - May be junctional, downsloping, or horizontal. In patients with transient ST-segment elevations, consider LV aneurysm, pericarditis, Prinzmetal angina, early repolarization, and Wolff-Parkinson-White syndrome as possible diagnoses. Fixed changes suggest acute myocardial infarction.[1,2,3,4]

Diagnostic sensitivity may be increased by performing right-sided leads (V4 R), posterior leads (V8, V9), and serial recordings

Correlation between ECG Waves & Events

Atrial fibrillation (AF) is a frequent complication of acute myocardial infarction (AMI), with reported incidence of 7% to 18%. The incidence of congestive heart failure, in-hospital mortality, and long-term mortality is higher in AMI patients with AF than in AMI patients without AF. P wave duration on signal-averaged ECG (PWD) and P wave dispersion on standard ECG (Pd) are noninvasive markers of intra-atrial conduction disturbances, which are believed to be the main electrophysiological cause of AF. PWD and Pd both measured in a very early period of AMI are useful in predicting AF.[5]

QRS prolongation with or without bundle branch block (BBB) has been associated with adverse

outcome in myocardial infarction; we examined the relationship between QRS duration and outcome in a broad spectrum of patients with acute coronary syndrome (ACS). In patients presenting with a broad spectrum of suspected ACS, QRS prolongation—particularly in the setting of LBBB—is an independent predictor of in-hospital and 1-year mortality. [6]

For patients with chest pain and non-diagnostic initial ECG, ACS risk is high if QTD and QTcD values are greater than 40 Ms. Therefore, QTD and QTcD can help identify patients with acute coronary syndrome who present with chest pain and a nondiagnostic initial ECG. However, poor operator characteristics of QT dispersion could limit its value as a diagnostic test in the clinical setting.[7]

The aim of this work was to assess the correlation between P wave dispersion, QRS duration & QT dispersion in hospital events in patients with acute coronary syndrome (ACS).

PATIENTS AND METHODS

Patients

This prospective study was conducted on 60 patients with acute coronary syndrome admitted to critical care units of Alexandria University Hospitals from first of January 2012 to end of August 2012.An informed consent was taken from relatives of every patient included in the study. This study was approved by ethical committee of Alexandria faculty of medicine.

Inclusion Criteria:

The study included patients with Acute Coronary Syndrome (ACS) who had two from three of the following criteria:

1. Clinical manifestation (Typical anginal pain usually with nausea and sweating).
2. Electrocardiographic changes (ECG).
3. Myocardial markers (CK, CKmb, Troponin I).

Exclusion Criteria:

Excluded from the study patients with any of the following:

ECG in which Q-T interval could not be assessed:

1. Unclear QT interval in at least seven ECG leads.
2. Bigeminal ventricular systole, atrial fibrillation, a pacemaker.
3. Current drug use affecting QT interval e.g quinidine, Amiodarone, psychogenic agents (tricyclic antidepressant, tetracyclic agents).

4. Patients on hemodialysis.
5. Post Coronary Artery Bypass Graft (CABG).

The patients included in the study will be classified into three groups:

I) Group 1: (ST segment elevation myocardial infarction with successful thrombolysis):

1. Typical anginal pain.
2. ST segment elevation in two or more contagious leads in ECG.
3. High Cardiac enzymes.
4. Signs of success after thrombolytic therapy (two from three): (1) Resolution of chest pain, (2) Regression of ST elevation by at least 50 % in ECG, (3) Occurrence of reperfusion arrhythmias (e.g. idioventricular rhythm).

II) Group 2: (ST segment elevation myocardial infarction with failed thrombolysis):

1. Typical anginal pain.
2. ST segment elevation in two or more contagious leads in ECG.
3. High Cardiac enzymes.
4. Signs of failure after thrombolytic therapy : (1) Resistant chest pain, (2)No regression of ST elevation by at least 50% in ECG, (3) No Occurrence of reperfusion arrhythmias.

III) Group 3 : (Unstable angina):

1. Typical anginal pain.
2. ECG changes (without ST segment elevation).
3. Negative cardiac enzymes.

METHODS:

1) Full history taking including :

1. Age.
2. Sex.
3. Character of pain.
4. Medical history including: Diabetes Mellitus, Hypertension, Previous heart disease & Drug intake.

2) Clinical Examination including:

1. Pulse rate (beat/min.).
2. Blood Pressure (mmHg).
3. Temperature (c º).
4. Chest & heart examination .

3) Electrocardiography: A 12-lead ECG record was done on admission, before and after thrombolytic (if given) and once daily till discharge.

A paper speed of 25 mm/s and amplification of 10 mm/mV was used for recording 12-lead ECG and the following parameters were calculated:

a- P wave Dispersion (PWD) is the difference between the longest (P max) & the shortest P wave duration (P min) recorded from multiple different surface ECG leads was calculated as (PWD = P max – P min) by milliseconds (ms).

b- QRS duration was measured (ms).

c- QT interval was calculated from the onset of the QRS complex to the point of return of the T wave to the isoelectric line. Three sequential complexes were measured and the mean value was used for QT interval calculation (ms).

The difference between the maximum and minimum QT intervals, occurring in any of the 12 leads, was measured as QTD. QTc max and QTc min will be determined with the Bazett formula (QTc= QT /√RR), and the difference between QTc max and QTc min was calculated as QTcD(9), together with assessment of other ischemic ECG change (T wave changes, S-T segment changes).

4) Cardiac markers [CPK (u/l), CK MB (u/l), Troponin I (ng/ml)] on admission and after six hours.

5) Plain chest X – ray

6) Management:

All patients were managed according to the following Protocol which includes**:**

I) Patients with ST segment elevation myocardial infarction (STEMI) will receive:

1. Oxygen (3-5 l / min).
2. Morphine (4mg I.V. As a pain killer).
3. Nitroglycerine (sub-lingual then intravenous titrated according to response).
4. Antiplatelets (Acetyl salycilic acid 81mg 4 tablets on admission then once daily and clopidogrel 75 mg 4 tablets on admission then once daily).
5. Thrombolytic therapy (streptokinase in a dose of 1.5 million units over one hour).
6. Anticoagulants (low molecular weight

heparin e.g. enoxaparine 1 mg/kg).

II) Patients with unstable angina will receive:

The same treatment except thrombolytic therapy

7) Outcome as regards:

Every patient was followed up for five days (patients who died before fifth day will be excluded due to insufficient data needed for follow up):

1. Duration of stay in ICU:
2. Morbidity: dysrhythmias, heart failure, cardiogenic shock, mechanical ventilation.
3. Mortality:

8) Statistical method:

Records of the studied cases and the results obtained after proposed procedure were statistically analyzed using SPSS 16.0 under Microsoft Windows XP. Continuous data were expressed in the form of mean ± SD. Categorical variables were expressed in the form of number and percent. One way ANOVA were utilized to compare numerical data. Chi-square test was used to compare categorical data as appropriate.

Correlation was done between quantitative and qualitative variables using Spearman correlation test and scatter dot graph.

RESULTS

Table 1. Comparison between the different studied groups according to demographic data

	Unstable angina 20 case		Successful thrombolysis 20 case		Failed thrombolysis 20 case		*p*
	No	%	No	%	No	%	
Sex							
Male	**14**	**70.0**	**19**	**95.0**	**15**	**75.0**	$^{MC}p = 0.124$
Female	**6**	**30.0**	**1**	**5.0**	**5**	**25.0**	
Age (years)							

Min. – Max.	**39.0 – 75.0**	**30.0 – 73.0**	**47.0 – 70.0**	
Mean ± SD	**57.35 ± 9.87**	**57.20 ± 11.37**	**56.30 ± 8.24**	**$^{F}p = 0.937$**
Median	**56.50**	**60.0**	**52.0**	

*: Statistically significant at $p \leq 0.05$

There was no significant difference between the gender of patients in the three groups of patients (p >0.05), total number of males was 48, however in females was 12. There was also no significant difference between the three age groups of patients (p >0.05), mean of age was similar (57.3, 57.2 and 56.3 years) respectively.

Table 2. Comparison between the different studied groups according to medical history

	Unstable angina		**Successful thrombolysis**		**Failed thrombolysis**		***p***
	No	%	No	%	No	%	
DM							
+ve	11	55.0	4	20.0	7	35.0	$^{c2}p = 0.070$
-ve	9	45.0	16	80.0	13	65.0	
$^{c2}p_1$			0.022*		0.204		
$^{FE}p_2$			0.015*				
Hypertension							
+ve	11	55.0	7	35.0	5	25.0	$^{c2}p = 0.139$
-ve	9	45.0	13	65.0	15	75.0	
Heart disease							
+ve	15	75.0	18	90.0	13	65.0	$^{MC}p = 0.201$
-ve	5	25.0	2	10.0	7	35.0	
Smoking							
+ve	4	20.0	15	75.0	9	45.0	$^{c2}p = 0.002^{*}$
-ve	16	80.0	5	25.0	11	55.0	
$^{c2}p_1$			<0.001*		0.091		
$^{c2}p_2$			0.053				
Cardiac enzymes							
+ve	0	0.0	20	100.0	20	100.0	$^{c2}p <0.001^{*}$
-ve	20	100.0	0	0.0	0	0.0	

	Unstable angina		Successful thrombolysis		Failed thrombolysis		*p*	
	No	%	No	%	No	%		
$^{c2}p_1$			<0.001*		<0.001*			
p_2			-					
ECG changes								
Non significant change	8	40.0	0	0.0	0	0.0	M	C
Positive change	12	60.0	20	100.0	20	100.0	<0.001*	
$^{FE}p_1$			0.003*		0.003*			
p_2			-					

This table shows that there was significant difference in risk factors between the studied groups as following: smoking is higher in the groups (Successful thrombolysis and failed thrombolysis) than the unstable angina group. There was also significant ECG changes and Cardiac enzymes were elevated in the (Successful thrombolysis and failed thrombolysis) than the unstable angina group.

Table 3. Comparison between the different studied groups according to P wave dispersion (PWD)

	Unstable angina	Successful thrombolysis	Failed thrombolysis	p
PWD	**(n = 20)**	**(n = 20)**	**(n = 18)**	
Min. – Max.	20.0 – 60.0	20.0 – 60.0	20.0 – 60.0	
Mean ± SD	48.0 ± 13.61	47.0 ± 13.42	43.33 ± 15.72	$^{KW}p = 0.624$
Median	50.0	40.0	40.0	
$^{MW}p_1$		0.787	0.357	
$^{MW}p_2$		0.485		

This table shows that no difference in PWD between the three groups.

Table 4. Comparison between the different studied groups according to QRS Duration

	Unstable angina	Successful thrombolysis	Failed thrombolysis	*p*
QRS				
Min. – Max.	50.0 – 100.0	40.0 – 120.0	50.0 – 90.0	
Mean ± SD	73.50 ± 14.24	72.0 ± 17.95	76.0 ± 10.46	$^{F}p = 0.681$
Median	80.0	70.0	80.0	
$^{Sch}p_1$		0.948	0.863	
$^{Sch}p_2$		0.687		

*: Statistically significant at $p \leq 0.05$

This table shows that there was no difference in duration of QRS between the three groups (p >0.05), QRS mean was ranged from 73.5 MS in patients with unstable angina, and 76 MS in patients with failed thrombolysis.

Table 5. Comparison between the different studied groups according to QT Dispersion (QTD)

	Unstable angina	Successful thrombolysis	Failed thrombolysis	*p*
QTD				
Min. – Max.	39.0 – 129.0	9.0 – 125.0	28.0 – 187.0	$^{F}p = 0.025$
Mean ± SD	83.50 ± 31.11	72.30 ± 31.47	101.0 ± 36.35	
$^{Sch}p_1$		0.566	0.239	
$^{Sch}p_2$		0.025		

*: Statistically significant at $p \leq 0.05$

This table shows that there is significant difference in QTD between the three groups, it was longer in patients with failed thrombolysis therapy (101msec), than in other groups of patients, (72.3msec) in patients with successful thrombolytic therapy, (83.5msec) in patients with unstable angina .

Table 6. Comparison between the different studied groups according to ICU length

	Unstable angina		Successful thrombolysis		Failed thrombolysis		*p*
	No	**%**	**No**	**%**	**No**	**%**	
ICU length							
<3 days	19	95.0	9	45.0	7	35.0	^{c2}p <0.001*
>3 days	1	5.0	11	55.0	13	65.0	
$^{c2}p_1$			0.001*		<0.001*		
$^{c2}p_2$			0.519				
Min. – Max.	1.0 – 4.0		2.0 – 4.0		2.0 – 5.0		
Mean ± SD	2.45 ± 0.76		3.50 ± 0.61		3.60 ± 0.88		^{F}p <0.001*
Median	2.50		4.0		4.0		
$^{Sch}p_1$			<0.001*		<0.001*		
$^{Sch}p_2$			0.917				

*: Statistically significant at $p \leq 0.05$

This table shows that there is a significant difference between the three groups ICU length of stay, patients with failed thrombolysis stayed more than 3 days in ICU, in comparison to the other groups.

Table 7. Comparison between the different studied groups according to complications

	Unstable angina		Successful thrombolysis		Failed thrombolysis		^{MC}p
	No	**%**	**No**	**%**	**No**	**%**	
Complications							
Absent	20	100.0	20	100.0	13	65.0	0.001*
Present	0	0.0	0	0.0	7	35.0	
$^{FE}p_1$			-		0.008*		
$^{FE}p_2$			0.008*				

	Unstable angina		Successful thrombolysis		Failed thrombolysis		^{MC}p
	No	%	No	%	No	%	
Heart failure	0	0.0	0	0.0	2	10.0	
Cardiogenc Shock	0	0.0	0	0.0	2	10.0	
Accelerated junctiontial rythm	0	0.0	0	0.0	1	5.0	-
AF	0	0.0	0	0.0	1	5.0	
Pulsless VT	0	0.0	0	0.0	1	5.0	

This table shows that the presence of listed complications was only in the failed thrombolysis group in comparison to the other two groups.

Table 8. Comparison between the different studied groups according to survival

	Unstable angina		Successful thrombolysis		Failed thrombolysis		^{MC}p
	No	%	No	%	No	%	
Survival							
Survived	20	100.0	20	100.0	16	80.0	0.029*
Non survived	0	0.0	0	0.0	4	20.0	
$^{FE}p_1$			-		0.106		
$^{FE}p_2$			0.106				

It was found that percent of survivors in unstable angina and patients with successful thrombolysis was 100% in comparison with 80% of failed thrmobolysis group , that was highly significant ($p < 0.001$*)

Table 9. Relation between P Wave Dispersion (PWD) , QRS Duration, QT Dispersion (QTD) and Complications in failed thrombolysis group

	Complications		*p*
	Absent	**Present**	
PWD	**(n = 13)**	**(n = 7)**	
Min. – Max.	20.0 – 60.0	20.0 – 40.0	
Mean ± SD	46.15 ± 17.10	36.0 ± 8.94	$^{MW}p = 0.208$
Median	60.0	40.0	
QRS	**(n = 13)**	**(n = 7)**	
Min. – Max.	50.0 – 90.0	80.0 – 80.0	
Mean ± SD	73.85 ± 12.61	80.0 ± 0.0	$^{t}p = 0.104$
Median	80.0	80.0	
QTD	**(n = 13)**	**(n = 7)**	
Min. – Max.	36.0 – 102.0	28.0 – 187.0	
Mean ± SD	83.15 ± 17.47	121.86 ± 51.68	$^{t}p = 0.022$*
Median	82.0	119.0	

This table shows that there is no significant difference between the PWD, QRS duration and the presence of complications in failed thrombolysis group, however there is a significant difference between the QTD and the complications in the same groups.

Table 10. Relation between QTD and complications:

Complications	QTD (Mean ± SD)	Anova	*p* value
▪ No Complications	79.9 ± 28.39		
▪ Heart Failure	190.0 ± 00.0	6.95	<0.001*
▪ Cardiogenic Shock	154 ± 00.0		
▪ AF	92.0 ± 00.0		
▪ JA	28.0 ± 0.00		
▪ Pulpless VT	187.0±0.0		

There was a highly significant difference between QTD in relation to occurrence of each complications, the largest one was in heart failure (190 msec) and the smallest one was (28 msec) in Junctional arrhythmia

Table 11. Relation between survival and complications in failed thrombolysis group

	Complications				*p*
	Absent (n = 13)		Present (n = 7)		
	No	%	No	%	
Survival					
Survived	13	100.0	3	42.9	0.007*
Non survived	0	0.0	4	57.1	

This table shows that non survived patients were 4 from 7 complicated patients (57.1%), their causes of death were (heart failure, cadiogenic shock, AF and pulseless VT), in comparison to the survived patients 3 from 7 complicated patients (42.9%), with a statistical difference <0.05.

Table 12. Relation between ICU length and complications in Failed thrombolysis group

	Complications				*p*
	Absent (n = 13)		Present (n = 7)		
	No	%	No	%	
ICU length					
<3 days	7	53.8	0	0.0	$^{FE}p = 0.044^*$
>3 days	6	46.2	7	100.0	
Min. – Max.	2.0 – 4.0		4.0 – 5.0		
Mean ± SD	3.23 ± 0.83		4.29 ± 0.49		$^{t}p = 0.007^*$
Median	3.0		4.0		

p: *p* value for comparing between the studied group

*: Statistically significant at $p \leq 0.05$

This table shows significant difference between complicated & non complicated patients regarding ICU stay , All complicated patients (100%)stayed more than 3 days in ICU while only 6 from 13 patients (46%) from complicated patients stayed more than 3 day

DISCUSSION

Acute coronary syndrome (ACS) refers to any group of symptoms attributed to obstruction of the coronary arteries. The most common symptom prompting diagnosis of ACS is chest pain, often radiating to the left arm or angle of the jaw, pressure-like in character, and associated with nausea and sweating. Acute coronary syndrome usually occurs as a result of one of three problems: ST elevation myocardial infarction (30%), non ST elevation myocardial infarction (25%), or unstable angina (38%).[8]

These types are named according to the appearance of the electrocardiogram (ECG/EKG) as non-ST segment elevation myocardial infarction (NSTEMI) and ST segment elevation myocardial infarction (STEMI). There can be some variation as to which forms of myocardial infarction (MI) are classified under acute coronary syndrome .[9]

ACS should be distinguished from stable angina, which develops during exertion and resolves at rest. In contrast with stable angina, unstable angina occurs suddenly, often at rest or with minimal exertion, or at lesser degrees of exertion than the individual's previous angina "crescendo angina". New onset angina is also considered unstable angina, since it suggests a new problem in a coronary artery. Though ACS is usually associated with coronary thrombosis, it can also be associated with cocaine use. [10]

Patients presenting with chest pain and nondiagnostic electrocardiograms (ECG) in the emergency department (ED) often pose a challenge to physicians, Atrial fibrillation (AF) is a frequent complication of acute myocardial infarction (AMI), with reported incidence of 7% to 18%. The incidence of congestive heart failure, in-hospital mortality, and long-term mortality is higher in AMI patients with AF than in AMI patients without AF. P wave duration on signal-averaged ECG (PWD) and P wave dispersion on standard ECG (Pd) are noninvasive markers of intra-atrial conduction disturbances, which are believed to be the main electrophysiological cause of AF.[5]

P wave dispersion, detected from the surface ECG, has been

thought to reflect left atrial enlargement and altered conduction . P wave dispersion and P wave maximal duration reflects the activation of atrial muscle and may depend primarily upon the mass of tissue excited, have been used in the assessment of the risk for atrial fibrillation which is characterized by nonhomogeneous and discontinuous atrial conduction .[11,12] P wave dispersion was defined as the difference between the longest and the shortest P wave duration recorded from multiple surfaces ECG leads. The clinical significance of P wave duration has been demonstrated in many clinical conditions, especially in paroxysmal atrial fibrillation.[11,13] P wave dispersion has been showed to be influenced by the autonomic nervous system activation, which induces changes in left atrial size and the velocity of impulse propagation .[14]

At present, no definitive cut-off value has been determined as to the diagnosis of high-risk patients. This study was undertaken to find out the correlation between the P wave dispersion in hospital events in patients with acute coronary syndrome (ACS).

In Table 3 there was no significant relationship between P wave duration in the studied groups ($p > 0.05$). A meta- analysis study, found that Pd, Pmax, and Pmin span a wide range of values in healthy individuals. Seemingly, abnormal values were often reported in healthy adults. The high variability of P-wave parameters in healthy individuals, and overlapping of the results with those reported for patients with increased risk for atrial fibrillation, might suggest that this technique has limited sensitivity and specificity. The variability between studies may stem from methodological issues and, therefore, there is a definite need for methodological standardization of Pd measurements . This study agreed with our study as PWD alone couldn't predict AF. But, this study disagreed with (*Dilaveris et al., 1999)* who said that Prolonged P wave duration and PD have been reported to represent increased risk of atrial fibrillation in patients with acute coronary syndromes.[15]

This study found that only one patient developed AF on the 3rd day in failed thrombolysis group, this patient was male, 66 years, diabetic, smoker, with typical chest pain, had significant ECG changes, P wave was measured on observing him 5 days in the intensive care unit (ICU), as following: PWD1= 20, PWD2= 40, PWD3 = 0 .0 (AF), PWD4= 0.0(AF) and PWD5=

0.0(AF) milliseconds, he died on the 5th day.

Prolonged QRS duration, and the presence of intraventricular conduction abnormalities, usually indicates the presence of changes in the myocardium due to underlying heart disease. Prolonged QRS duration is often associated with depressed ejection fraction or enlarged left ventricular volumes, but several studies have demonstrated that this simple ECG measure provides independent prognostic value, after adjusting for relevant clinical covariates .[16]

Post-infarction patients with prolonged QRS duration have a significantly increased risk of mortality, although data associating QRS prolongation specifically with sudden death is less supportive. In non-ischemic cardiomyopathy, there is no evidence that QRS duration has prognostic significance in predicting mortality or sudden death. Prolonged QRS duration, and especially presence of left bundle branch block, seems to predict a benefit from cardiac resynchronization therapy in both ischemic and non-ischemic cardiomyopathy patients. Therefore, QRS duration and morphology should not only be considered a predictor of death or sudden death in patients after myocardial infarction, and in those suspected of coronary artery disease, but also as a predictor of benefit from cardiac resynchronization therapy in patients with heart failure, whether of an ischemic or non-ischemic origin.[17]

This present study agreed with Bryneo et alas evaluation of the presenting electrocardiogram in Alexandria University Hospitals intensive care patients (n = 60) showed that there was no significance difference between QRS duration in the three groups (unstable angina, successful thrombolysis and failed thrombolysis respectively with mean value (73.7 ± 14.02, 76.2 ± 10.88, 72.36± 8.81) (p> 0.05) Table (4) & the correlation between the QRS duration and the development of complications was non-significant in this study, ^{t}p = 0.104 (Table 9).[17]

How ever Petrina et al found the contrast which discovered that QRS prolongation-particularly in the setting of LBBB-is an independent predictor of in-hospital and 1-year mortality.[16]

The QT interval represents the total duration of ventricular depolarization and repolarization .QT interval is calculated from the onset of the QRS complex to the point of return of the T wave to the isoelectric line. The normal

value of QTc is up to 0.39 second in men and 0.44 seconds in women. The difference in the QT intervals between the derivations from ECG leads as "QT dispersion" (QTD) and noted that it represents the degree of the repolarization heterogeneity. "QTD" is defined as the difference between the maximum and minimum QT intervals, occurring in any of the 12 leads. Prolonged QTD is associated with an increased risk of serious ventricular arrhythmias in patients with long QT syndrome, hypertrophic cardiomyopathy, chronic heart failure or myocardial infarction *(Suzuki et al., 1998).* [18]

Experimental work has shown that increased dispersion of electrical recovery after activation is a key factor in the development of serious and fatal arrhythmias associated with ischemia *(Janse & Wit., 1989).* This agreed with our study, 53 patients had QTD (mean= 79.95) millisecond with no complications, however 7 patients who developed complications, had QTD between 28 and 190 Millisecond respectively, Table (3).

Table 9, showed that there was a highly significant correlation between QTD duration and prediction of complications, $^{t}p = 0.022^{*}$ which is agreed with Pekdemir et al(123) who described the same results in his study which showed that QTD can help identify patients with acute coronary syndrome who present with chest pain and a non-diagnostic initial ECG. However, poor operator characteristics of QT dispersion could limit its value as a diagnostic test in the clinical setting.

Regarding the complications, it had been found that all of them had occurred in patients with failed thrombolysis therapy, in comparison to the other two groups, 7 patients (35%), ($p<0.05$) (Table 7)

In the present study ICU length of stay was longer (>3 days) in patients with failed thrmbolysis (13 patients) in comparison with the other two groups, 11 patients with successful thrmbolysis, and one patient with unstable angina stayed more than 3 days in ICU ($p<0.05$) (Table 6). This is an agreement with Craig et al. who found that lower mortality is associated with shorter lengths of stay. Only part of these associations could be attributed to following best practice guidelines and lower rates of preventable complications.[20]

Soon after resuscitation and intensive care was done to acute coronary syndrome patients, it was claimed that survivors mostly would return to their homes. In the present study it was found that percent of patients with favourable

outcomes in unstable angina and in patients successfully thromboylsed 100% respectively compared with failed thrombolysis patients (80% survived and 20% died) Table (10). This is also an agreement with Craig et al**.** [20]

SUMMARY& CONCLUSION

The acute coronary syndromes encompass a spectrum of unstable angina to transmural myocardial infarction. The definition of acute coronary syndrome depends on the specific characteristics of each element of the triad of clinical presentation, electrocardiographic changes and biochemical cardiac markers.

This prospective study was conducted on 60 patients with acute coronary syndrome admitted to critical care units of Alexandria University Hospitals from first of January 2012 to end of August 2012. An informed consent was taken from relatives of every patient included in the study. This study was approved by ethical committee of Alexandria Faculty of Medicine.

During the observation up to 5 days, 1 patient (1.6%) developed AF. There was no significant correlation between P wave dispersion (PD) in the three studied groups& No significant relation between it & the whole complications developed in failed thrombolysis group (p> 0.05). PD alone couldn't predict AF in this study, as the mean was 20.0 milliseconds, which was too small to predict AF alone.

There was no significance difference between QRS duration in the three groups (unstable angina, successful thrombolysis and failed thrombolysis respectively with mean value (75.7 ± 14.02, 75.2 ± 10.88, 73.36± 8.81) (p> 0.05) & the relationship between the QRS duration and the development of complications was non-significant in this study.

This study showed that there was a significant relation among the three groups & highly significant correlation between QTD duration and prediction of complications.

It was clear that ICU length of stay was longer in patients with failed thrombolysis in comparison with the other two groups (p<0.05).

RECOMMENDATIONS

1. Importance of measurementsof QT Dispersion to patients with acute coronary syndrome especially at admission.
2. Failed thrombolysis patients should be under close observation and monitoring till another option is

available like coronary intervention.

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A JUSTICE PROSPECTIVE OF CARDIOVASCULAR DISEASES IN BANGLADESH

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ABSTRACT

Over the last 15-20 years Cardiovascular disease (CVD) in Bangladesh has been increasing gradually, perhaps of all South Asian countries Bangladesh has the highest incidence and prevalence of CVD, and yet is one of the most forlorn for vascular research. Coronary Artery Disease (CAD) and stroke are already the leading causes of mortality and morbidity in Bangladesh. Therefore, this review article attempted to be an excellent source of information for future study and research on CVD in Bangladesh. Amongst the heart diseases hypertension, rheumatic fever, rheumatic heart diseases, ischemic heart diseases (IHD) and congenital heart diseases are commonly prevailed in Bangladesh. Latest research findings on Cardiovascular diseases executed in Bangladesh revealed prevalence of Hypertension in adult population about 20-25%, Ischemic Heart Disease in adult population about 10%, Rheumatic Heart Disease (RHD) 1.2 per thousand and Congenital Heart Disease 8 per thousand new born baby. Environmental, genetic and several classic factors such as diabetes, blood pressure, lipids and smoking, obesity are involved with CVD in Bangladesh. Soon countrywide survey and clinical research should be carried out extensively to determine the different aspects of CVD in Bangladesh.

Keywords: Cardiovascular Disease (CVD); Coronary Artery Disease (CAD); Hypertension; Ischemic Heart Diseases (IHD) ; Clinical Research.

1. INTRODUCTION

Being heaven ground of the world's heart disease, the total number of Cardiovascular Disease (CVD) patients has been increasing significantly over the last few decades in Bangladesh and now CVD is emerging as a serious health problem in this country. Unless concerted efforts are made and national policy of prevention of risk factors are not undertaken, it is feared that by next 10-15 years the number of patients will increase dramatically. While the development in clinical and interventional cardiology technology has been moving at a haste speed all over the world, on the other hand, the infrastructure

improvement in cardiology both in govt. and private sector has been progressing at a snail's space. Due to limited number of specialists in the field and the high cost of technology, it has become difficult for a country like Bangladesh to transfer modern technology. Besides, every year a significant number of patients go to abroad for cardiovascular treatment. Therefore the total scenario creates a embarrassed stage to face the large number of patients who will be seeking medical help and treatment facilities.[1] It is high time to address the all risk factors associated with CVD in Bangladesh and combat the problem through holistic way.

2. STATISTICAL VIEW OF CVD IN GLOBAL AND BANGLADESH

Cardiovascular disease is the main cause of morbidity and mortality in the world. Based of WHO's data in 2011, it is the first cause of mortality in the world and 60% of cardiac mortality is caused by ischemic heart disease, and at least 17.5 million or equal to 30% of mortality is caused by other cardiovascular disease. It is predicted 23.6 million people would death by cardiovascular disease in 2030. Bangladeshis are having heart attacks at least 10 years earlier than the typical sufferers of the condition in western countries, according to preliminary findings of BRAVE, the largest study of cardiovascular disease ever held in Bangladesh. [2]

Among the first heart attack cases admitted at the National Institute of Cardiovascular Diseases (NICVD), approximately 12% were women, meaning there was a possibility that a majority of the early heart attack symptoms in women may have been ignored at the households, raising a grave likelihood of substantial case fatalities in women. The average age among Bangladeshis to have heart attacks was 52 years, with approximately 40% of all cases occurring in people aged less than 50 years.[2]

Latest research findings on Cardiovascular diseases executed in Bangladesh revealed prevalence of Hypertension in adult population about 20-25%, Ischemic Heart Disease in adult population about 10%, Rheumatic Heart Disease 1.2 per thousand and Congenital Heart Disease 8 per thousand new born baby.[3]

3. MAJOR RISK FACTOR FOR CVD IN BANGLADESH

3.1 Environmental Risk Factor

3.1.1. Smoking

In Bangladesh smoking is considered as no.1 risk factor for ischemic heart disease. Despite showing the health hazards of smoking, the number of smokers is increasing at alarming rate.[1] Recently, a large prospective cohort study involving 11746 participants and 6.6 years of average follow up in Bangladesh has found positive association between long-term arsenic ingestion and increased cardiovascular mortality while the association was even worse among smokers.[4]

3.1.2. Hypertention

In Bangladesh, approximately 20% of adult and 40–65% of elderly people suffer from hypertension; high incidence of metabolic syndrome, and lifestyle related factors like obesity, high salt intake and less physical activity may play important role in the pathophysiology of hypertension.[5]

According to the survey by Bangladesh Society of Medicine in collaboration with Directorate General of Health Services and World Health Organization, prevalence of hypertension is 17.9% in general, 18.5% in men and 17.3% in women. Such a high prevalence of hypertension may contribute to the high prevalence of CAD in Bangladesh.[6]

3.1.3. Diabetes mellitus

Like in all other developed and developing countries, prevalence and incidence of type 2 DM is increasing in Bangladesh. In 2010, the International Diabetes Federation (IDF) estimated that 5.7 million (6.1%) and 6.7 million (7.1%) of people living in Bangladesh are suffering from DM and impaired glucose tolerance (IGT), respectively; by 2030, that number of diabetic population is expected to rise to 11.1 million. This explosion in diabetes prevalence will place Bangladesh among the top seven countries in terms of the number of people living with diabetes in 2030.[7]

3.1.4. Dyslipidemia

Despite considerable disparities in the prevalence of individual components of abnormal lipid profile, it is apparent that dyslipidemia is prevalent among the Bangladeshis in general. Another recent study involving 3201 individuals found rising trend of dyslipidemia in sub-urban population; prevalence of

dyslipidemia was 16.6% in general, 22.2% in males and 15.9% in females. Total cholesterol was high (>240 mg/dl) in 16.9%, LDL-C was high (>160 mg/dl) in 15.7%, HDL-C was low (<40 mg/dl) in 8.8%, and TG was high (>200 mg/dl) in 17.8% and very high (>350 mg/dl) in 2.0%. Women had significantly higher TC and LDL-C in comparison to men after the age of 40 years.[8] Liberal use of saturated fats and trans fats, deep frying, reuse of cooking oil, and overcooking leading to destruction of folates may all contribute to dyslipidemia in this population.[9]

3.1.5. Unhealthy and Imbalanced Food Consumption Style

Nearly two-thirds of the typical daily diet of rural people in Bangladesh consists of rice, some vegetables, a little amount of pulses and small quantities of fish if and when available, milk, milk products and meat are consumed only occasionally and in very small amounts.[10] Though over the years the consumption of rice and wheat has decreased, resulting in an overall decrease in cereal consumption from 59% to 41.33% as a percentage of monthly expenditure on major food items, still the proportion is quite high.[11] This may contribute to hypertriglyceridemia. On the other hand, use of liberal amount of cooking oil, fried vegetables and food preparations, extra salts added during preparation and pickles are important aspects of traditional Bangladeshi cuisine. Soybean oil, palm oil and mustard oil are the main edible oils. Vegetable oil, butter oil and ghee are also used. Deep-frying and reuse of cooking oil, the latter for financial constraint and ignorance, may lead to conversion of cis fat to trans-fat. Food preparation methods result in significant nutrient loss; upto 40% of thiamine and niacin are lost during washing of rice before cooking, boiling rice and then discarding the water results in even more nutrient losses.[12] Chronic vitamin B complex deficiency may be associated with hyperhomocysteinemia. Traditional fast foods including Singara, Samucha, Puri, Piaju, and Paratha all are generally deeply fried. Commercially available packaged bakery and fast foods often do not contain any declaration of their fat content. With the social changes in recent years, Bangladesh is now experiencing a fast food culture, especially in urban areas. These commercial fast foods of western type, as well as, the traditional snacks were found to contain high amounts of cholesterol and saturated fatty acids, mainly derived from animal fats and palm oil.[13,14]

On an average, a Bangladeshi person consumes 126 g of fruit and vegetables daily,[12] which is far below the minimum daily consumption of 400 g of vegetables and fruit recommended by the Food and Agriculture Organization (FAO) and the World Health Organization (WHO).[15] In the Bangladesh NCD Risk Factor Survey 2010, consumption of inadequate fruit and/or vegetables (<5 servings per day) was found in 95.7% people.[16]

3.1.6. Air pollution

In the recent years, air pollution has been suggested to contribute to cardiovascular illness. The overall evidence is consistent with a causal relationship between exposure to particulate matter <2.5 μm in diameter ($PM_{2.5}$) and cardiovascular morbidity and mortality.[17] Air pollution is a significant problem, especially in the urban areas of Bangladesh. A study to evaluate the emissions and air quality in megacities found Dhaka to have the poorest air quality in respect of total suspended particles (TSP), sulfur dioxide (SO_2), and nitrogen dioxide (NO_2) among the megacities, and the pollutant levels were far beyond the WHO standard.

3.1.7. Chronic arsenicosis

Arsenic contamination of groundwater in Bangladesh has been recognized as a massive public health hazard.[18,20] An estimated 57 million people have been chronically exposed to groundwater with arsenic concentrations exceeding the WHO standard[21] and 85 million people at risk from arsenic in drinking water and in food crops.[22] Arsenic in irrigation water has been found to accumulate in soils, from where it is taken up by crops, thereby entering the food chain and contaminating crops, vegetables and fish.[23,24] Although maximum uptake has been observed in some leafy vegetables and spices, arsenic uptake in rice constitutes the greatest source of exposure to the population from food.[24] Chronic arsenic exposure may facilitate systemic inflammation and vascular endothelial dysfunction, which may, in turn, increase the risk of CVD.

3. 2. Genetic Risk Factor

The Coronary Artery Disease (CAD) Genetics Consortium performed a meta-analysis of 4 large genome-wide association studies of CAD, 2 of European ancestry (PROCARDIS and HPS) and 2 of South Asian ancestry (PROMIS and LOLLIPOP) and found 5 new

loci i.e. LIPA on 10q23, PDGFD on 11q22, ADAMTS7-MORF4L1 on 15q25, a gene rich locus on 7q22 and KIAA1462 on 10p11 for CAD, that have similar associations in Europeans and South Asians.[25] Recently, 6 novel genetic loci have been identified in South Asians, which are associated with type 2 diabetes mellitus (DM), a major risk factor for CAD.[26]

4. Present Scenario of cardiovascular care amenities

First integrated cardiovascular care started in this country with the formal establishment of National Institute of Cardiovascular Diseases (NICVD) in 3rdApril 1981. In this institute, for the first time, percutaneous transluminal coronary angioplasty (PTCA) was done by foreign experts in 1987 and by Bangladeshi team in 1995, coronary stenting in 1997. [27-29] First CABG was done in NICVD in 1985. Since then, cardiovascular care facilities have increased steadily, and at present, a good number of institutions in public, as well as, in private sector are rendering cardiovascular care throughout the country, though they are more concentrated in the capital city. Almost all sorts of coronary interventions and bypass surgery are being done at a relatively low cost. In the recent years, primary percutaneous coronary intervention (PCI) is being performed in Government, as well as, in private centers. Facilities for post-graduation in cardiology have increased in recent years. Research works are being done regularly as a prerequisite for post-graduate courses, and also independent of this.

5. Current Research Project

The Bangladesh Risk of Acute Vascular Events (BRAVE) study, is going on to find out the risk factors, conventional and unconventional, to explain the undue predisposition of the people of Bangladesh to CAD and to build the first epidemiological resource in Bangladesh. The study is the joint collaboration of Cambridge University of UK, International Centre for Diarrhoeal Disease Research, Bangladesh (ICDDRB) and NICVD in Dhaka, Bangladesh. The initial results of BRAVE study have identified several environmental contaminants (e.g. arsenic in the blood) and nutritional elements (e.g. zinc deficiency) as important drivers for heart attacks in this population. An ongoing study at BIRDEM using the VerifyNow P2Y12 assay has found clopidogrel resistance in 46.7%, and prasugrel resistance in 7% of non ST-elevation acute

coronary syndrome (ACS) patients undergoing PCI.[30] Such highly prevalent clopidogrel resistance may be related to the recurrence of ACS and occurrence of stent thrombosis and in-stent restenosis after PCI in this population.

6. Future Responsibilities

Data related to different aspects of CAD in Bangladesh are inadequate. Large, preferably, nation-wide epidemiological and clinical studies should be carried out to gain reliable information on this important public health issue. Cardiovascular disease prevention should be integrated with primary health care.

Cardiovascular health promotion should be part of the national media strategy and the health education curriculum. The public health approach should target population-wide lifestyle intervention, screening for high blood pressure, DM and dyslipidemia. Healthy life styles including consumption of heart-healthy diets, avoidance to smoking and smokeless tobacco, moderation of salt intake and increased physical activity, should be promoted. Limitations can be placed on the concentrations of salt, sugar, trans-fats and saturated fats in manufactured food products. Food labeling should also be introduced to facilitate informed choice by consumers. Food adulteration should be dealt with rigorously. Provision of safe, arsenic-free water and food should be ensured. Necessary legislative and administrative steps should be taken to reduce air pollution. Policy change should address urban planning, transport and preservation of environment. Special attention should be given to stop malnutrition and under-nutrition in fetal and neonatal life through nutrition programs. Public awareness should be created to avoid childhood obesity. If indicated by further research, vitamin D deficiency should be avoided by fortification of food. Intensive research, may be in collaboration with international organizations, should be undertaken to explore the still-unidentified risk factors unique to this nation, which are responsible for the high prevalence of CAD in Bangladesh.

7. Conclusion

Due to the massive prevalence of CVD, it has already put a serious stress on the health services resources and a big burden on health service providers in Bangladesh. At the advent of the new millennium, we are really unclear about the real situation. Along with the classical risk factors, genetic make-up and

environmental factors unique to our population may exist. We have no more time to lose. Large-scale, preferably, nation-wide survey and clinical research should be conducted to determine the different aspects of CAD in Bangladesh. The information available thereby, would help to formulate national policy to combat the deadly epidemic more efficiently in future.

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EVIDENCE-BASED INTRACORONARY ADMINISTRATION OF GPIIB/IIIA INHIBITOR FOR REDUCING NO-REFLOW PHENOMENON AND IMPROVING TIMI FLOW FOR MYOCARDIAL SALVAGE DURING PRIMARY PERCUTANEOUS CORONARY INTERVENTION

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Introduction: Primary percutaneous coronary intervention (PCI) is the best available reperfusion strategy for acute ST-segment elevation myocardial infarction (STEMI). Despite optimal coronary artery reperfusion has been done, up to 60% of primary PCI may fail to restore optimal myocardial salvage and reperfusion rate, a failure at the microvascular level known as No-Reflow Phenomenon (NR). NR refers to the inability to reperfuse myocardial tissue or failure to restore normal myocardial blood flow despite removal of coronary obstruction. Strategy may be pharmacological or device based. Among pharmacological based, Glycoprotein IIb/IIIa inhibitors (abciximab or tirofiban) is indicated by European Society of Cardiology (ESC) guidelines for prevention of no-reflow (class of recommendation IIa and level of evidence B)

Objective: We carried out a literature search aimed at identifying all randomized clinical trials of Intracoronary Glycoprotein IIb/IIIa inhibitors (GPI) administration in the setting of primary PCI. The outcomes examined were post-procedure rates of TIMI 3 flow, TIMI myocardial perfusion grade 3, ST segment resolution >= 50%, No-Reflow Phenomenon (NR), distal embolization, and composite endpoint of death. For each outcome, we fit a Bayesian hierarchical meta-analytic model.

Methods: Literature search was done from PubMed, Cochrane, EMBASE, Google Scholar, EBSCO, Springer and Ovid databases without language or time limitation. Randomized controlled trials were enrolled for analyzing if they investigated the treatment of intracoronary administration of Glycoprotein IIb/IIIa inhibitors (GPI) versus standard treatment for NR during primary PCI.

Results: Ten studies with 702 patients were included. Significantly, the treatment of Glycoprotein IIb/IIIa inhibitors was more effective in improving the thrombolysis in myocardial infarction (TIMI) flow (OR 0.24, 95% CI 0.15-0.37,

P<0.00001) and reducing major adverse cardiovascular events (MACE)(OR 0.09, 95% CI 0.05-0.18, P<0.00001). There was a trend to increase the risk of bleeding, but the data of the result did not reach the statistical significance (OR 1.44, 95% CI 0.69-3.00, P=0.32). In a small randomized study of tirofiban versus verapamil + sodium nitroprusside for patients undergoing Primary PCI, there are similar rates of final TIMI flow grade 3 (86% vs. 88%).

Discussion: There are four mechanisms play role in NR: Distal atherotrombosis embolization, ischemic injury, reperfusion injury, and individual susceptibility for microcirculatory injury. Other phenomenon are known to contribute to complexity of NR, including large lipid rupture, leukocyte infiltration, vascular damage, vasoconstriction, activation of inflammatory pathways and cellular edema. Glycoprotein IIb/IIIa inhibitors (GPI) can block the final pathway of platelet aggregation and distal atherotrombosis embolization, which results better outcome in TIMI flow and improving myocardial viability.

Conclusion: Adjunctive intracoronary administration of Glycoprotein IIb/IIIa inhibitors may improve recanalization therapy and reduces the incidence of No-Reflow Phenomenon during Primary PCI. Further studies for clinical endpoints are needed. It suggested that intracoronary administration of Glycoprotein IIb/IIIa inhibitors during Primary PCI should be fully considered the individuation of patients and balanced the efficacy and the potential hazard.

Keywords: GP IIb/IIIa inhibitors, No-Reflow Phenomenon, Primary PCI, TIMI Flow

INTRODUCTION

Primary percutaneous coronary intervention (PPCI) for ACS STEMI patient is an effective therapy for myocardial salvage by restoring vessel caliber and improving blood flow to ischemic myocardium. At least 3.000 PPCI are performed annually in Indonesia, and it has been estimated that nearly 2 million procedures are performed annually worldwide.[1] The growth in the number of PCIs has occurred in tandem with technological refinements and advances in peri- and postprocedural medication, which have reduced procedural risk, improved success rates and dramatically changed the prognosis of patients with acute coronary syndromes.[2-8]

The no-reflow and slow-reflow phenomena [9] are poorly understood complications of PCI in which reduced coronary flow persists despite the intraprocedural removal of the occlusive lesion from the epicardial coronary artery or arteries. In as many as 2% of patients without acute myocardial

infarction (AMI) who undergo PCI, restoration of normal anterograde myocardial blood flow (myocardial "blush") is unsuccessful due to microcirculatory dysfunction and/or mechanical obstruction; this failure – slow- or no-reflow – occurs in the absence of postprocedural angiographic evidence of an encroaching luminal lesion (*e.g.*, evolving dissection) or evidence of macroscopic distal embolization.[10-12] The no-reflow phenomenon is encountered most frequently among patients undergoing PCI of saphenous vein grafts (SVGs) and in patients with AMI who undergo thrombolysis or mechanical intervention. In these populations, the incidence of slow- or no-reflow may be greater than 30%.[12-14]

The no-reflow phenomenon following PCI is associated with significant cardiac consequences due to myocardial ischemia caused by inadequate distal microvascular flow and poor myocardial perfusion. There is now clinical evidence that no-reflow is a strong predictor of long-term mortality that is independent of and beyond that provided by infarct size. Identifying and treating no-reflow may have important benefits including enhancing delivery of nutrients and cells required for healing and reducing infarct expansion and ventricular remodeling, which ultimately may reduce congestive heart failure and mortality. Despite widely patent epicardial vessel lumens, no-reflow can be associated with angina and ischemic ST-segment changes in these patients.[10] Consequently, angiographic or contrastenhanced echocardiographic demonstration of open epicardial vessels may not be a reliable indicator of successful myocardial reperfusion in AMI patients with no-reflow and residual perfusion defects in the risk area.[15] No-reflow predicts a poor functional recovery and impaired ventricular remodeling, and its persistence – despite attempted treatment – is associated with a high incidence of ongoing or recurrent acute coronary syndrome (ACS) as well as increased short-term mortality.[11,13,15,16]

In the reported experience of one catheterization laboratory, the no-reflow phenomenon was seen in about 2,0%of the total number of cases. The incidence varies with the clinical setting. Patients undergoing primary intervention for acute myocardial infarction show an incidence of 11,5%, compared to 1,5% of patients undergoing elective coronary intervention. Elective intervention performed on saphenous vein grafts carries a risk of 4,0% or higher. The risk of

developing no-reflow depends also on the nature of the intervention since a higher trend is observed with stenting rotational atherectomy, and directional atherectomy than with conventional balloon angioplasty.[17] Because no-reflow phenomenon occurs in different clinical settings, it is possible that different pathophysiological mechanisms operate in each setting. For instance, embolization of plaque material could be the culprit in saphenous vein graft interventions, while ischemia/reperfusion in acute myocardial infarction and activated platelets in unstable angina would play the major role. Different therapeutic strategies may therefore be needed in each situation.

OBJECTIVE

Before we review some studies about Glycoprotein IIb/IIIa inhibitors for improving no-reflow, it is important to keep in mind that no double-blind, randomized trial has been conducted to assess any of these agents. No trial has been conducted to determine the appropriate dosage either. The use of these drugs is mostly based on clinical experience and their administration limited by the resolution of no-reflow or the development of side effects. No formal study has evaluated any combination of these agents. The purpose of the review is to give some appraisals about the effectiveness and safety of Glycoprotein IIb/IIIa inhibitors as preoperative treatment of primary stenting in patients with ACS STEMI to prevent no-reflow phenomenon during PCI.

MATERIALS AND METHODS

1. Identification and eligibility of the relevant studies

   A comprehensive literature search of PubMed and Embase up to February 28, 2016 was performed using the following keywords: (“No-Reflow Phenomenon” or “Microvascular obstruction”) and (“GP IIb/IIIa inhibitors”, “ abciximab (ReoPro)”, “eptifibatide (Integrilin)”, or tirofiban (Aggrastat)”) and (“Primary PCI”, “Primary Stenting”, “Percutaneous Coronary Baloon Angioplasty”, or “TIMI Flow”). The references in retrieved articles were also reviewed for possible inclusion. Only publications written in English with available full-text articles were included in this meta-analysis. Studies were included if they met the following eligibility criteria: (1) RCT or case-control studies focused on the efficacy and safety of GP IIb/IIIa inhibitors

administration before primary PCI began, (2) more than two articles to compare preoperative GP IIb/IIIa inhibitors with placebo or others antiplatelet drugs, (3) available information about the preoperative intervention and postoperative cardiac care after primary PCI, and (4) published as a full paper in English.

The main reasons for the exclusion of studies were the following: (1) not focused on the successful rate (TIMI flow) for primary PCI, (2) did not study the GP IIb/IIIa inhibitors, (3) did not report the relevant no-relow phenomenon data, (4) not published in English, and (5) non-human research. Ultimately, a total of 12 articles including 1.151 cases and 1.740 controls were included in this meta-analysis

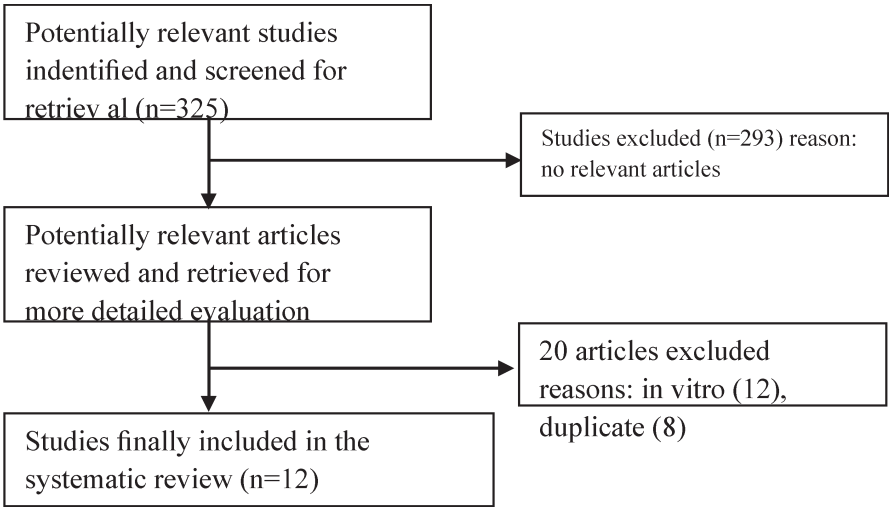

Figure 1. Search flow diagram of this meta-analysis

2. Data extraction

Three investigators (R.A.N, M.J, T.N.O.) independently extracted the data and reached consensus regarding all of the items. The following information was sought from each article: the first author's name, year of publication, country of origin, time door-to-ballooning, type of stenosis and TIMI flow / GRACE / SYNTAX score, numbers of cases and controls, incidence of no-

reflow phenomenon, incidence of MACE. We categorized the different GP IIb/IIIa inhibitors as tirofiban and abciximab.

3. Statistical analysis

The risk of no-reflow phenomenon associated with primary PCI was estimated for each study using the odds ratio (OR) and its 95% confidence interval (95%CI). The between-study heterogeneity was examined with a chi-square-based Q statistic test, and $p \leq 0.05$ was considered as statistically significant. We pooled the results using fixed-effect models when the heterogeneity between studies was absent. Otherwise, a random-effects model was selected. Subsequently, we evaluated the risks of no-reflow phenomenon then evaluated the risks of the MACE and major cardiovascular death between subjects taking GP IIb/IIIa inhibitors vs placebo, while assuming the successfull event for primary PCI were TIMI flow transformation. Funnel plots and Begg's test were utilized to evaluate the publication bias. All analyses were performed using the MedCalc 3000 softwate and SPSS version 17.0 software.

RESULTS

The prevalence of no-reflow occurs in 0.6% to 5% of percutaneous coronary interventions,[21,22] with high incidence in patients undergoing percutaneous coronary intervention for acute myocardial infarction, vein graft interventions and rotational atherectomy.[23] It can be as high as 50% in coronary arteries with high thrombus burden.

In 12 paper, we reviewed an IC bolus dose of Tirofiban (0.25 μg/kg) was administered in the IC group. Microcatheter through coronary guiding catheters was used to administer the IC Tirofiban. When the wire had crossed the occlusion, the drug was administered. In the IV group, two boluses (each 180 μg/kg) of Eptifibatide were administered every 10 minutes. Thrombus aspiration was performed using the Export catheter, if it was necessary. Stenting was performed for all the patients. The intra-aortic balloon pump (IABP) was not used for any patient.

The primary end points of the trial were post-procedural TIMI grade flow / TIMI transformation of the infarct-related artery and no-reflow phenomenon during primary PCI. The secondary end points were major adverse cardiac event (MACE), cardiovascular death, major composite death 1 year,

restenosis, and major bleeding after primary PCI. For the evaluation of the ECG end points, a 12-lead ECG was acquired at the time of presentation and at 90 minutes after primary PCI. Successfull of primary PCI was assessed by comparing the ST-segment elevation in the infarct-related area on the ECG after primary PCI with the ECG at presentation.

"TIMI Grade Flow" is a scoring system from 0–3, referring to the levels of the coronary blood flow as assessed during percutaneous coronary angioplasty:

- TIMI 0 : flow (no perfusion) refers to the absence of any antegrade flow beyond a coronary occlusion.
- TIMI 1 : flow (penetration without perfusion) is a faint antegrade coronary flow beyond the occlusion, with incomplete filling of the distal coronary bed.
- TIMI 2 : flow (partial reperfusion) is a delayed or sluggish antegrade flow with complete filling of the distal territory.
- TIMI 3 : flow (complete perfusion) is a normal flow which fills the distal coronary bed completely.[19]

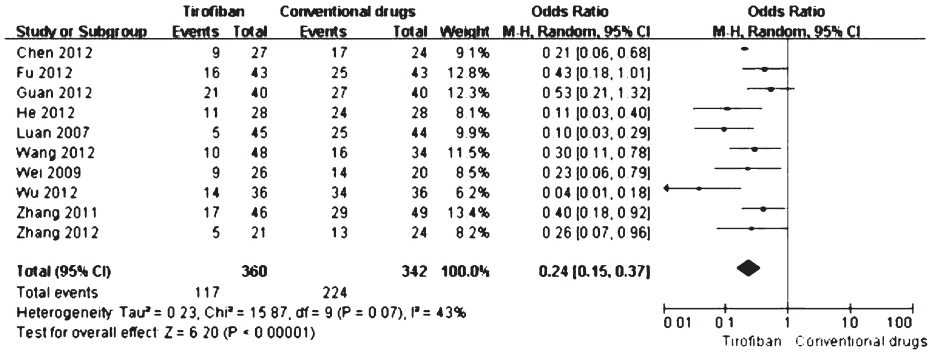

Figure 2. Forest plot of OR for TIMI flow transformation.

There are 10 case controls which research about efficacy of Tirofiban comparing to conventional drugs (standard treatment) during PCI, showing the superiority of Tirofiban for primary outcomes in TIMI Flow Transformation. 10 studies consisting 360 ACS STEMI patients who got Tirofiban before primary PCI shows benefit in transformation of TIMI flow comparing to the 342 ACS STEMI patients who didn't get GPIIb/IIIa inhibitors. Also, the incidence of

no-reflow phenomenon is greater in ACS STEMI patients who took conventional drugs (n=224; 65.5%), comparing to ACS STEMI patients who took Tirofiban (n=117; 32.5%) with OR 0.24 (0.15-0.37). The heterogeneity analysis were not significant.

Secondary outcomes in MACE incidence also shows superiority in Tirofiban groups, comparing to conventional drugs. 6 studies consisting 183 subjects who took Tirofiban vs 176 subjects who took conventional drugs. MACE incidence were greater in subjects who took conventional drugs (n=72; 41%), comparing to subjects who took Tirofiban (n=15; 8.2%). The cardiovascular protection of Tirofiban for MACE were OR 0.12 (0.06-0.23; 95%CI). The heterogeneity analysis for these studies were not significant.

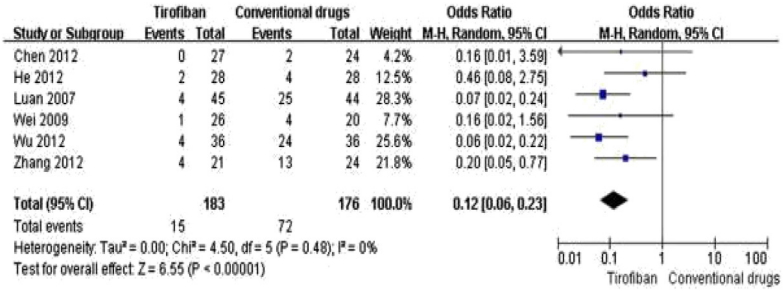

Figure 3. Forest plot of OR for MACE.

Major or minor hemorrhage was determined using the TIMI criteria, including:

A. Major criteria: intracranial hemorrhage or clinical bleeding associated with loss of greater than 5 mg/dl of hemoglobin (or hematocrit decrease by > 15 points or by 10– 15 points with clinical bleeding) and

B. Minor criteria: loss of greater than 3 gm/dl of hemoglobin (or hematocrit decrease by < 10 points) with clinical bleeding or loss of greater than 4 mg/dl of hemoglobin (or hematocrit decrease by 10–15 points) with no clinical bleeding. Clinical bleeding was comprised of large hematoma, gastrointestinal blood loss, and retroperitoneal bleeding.

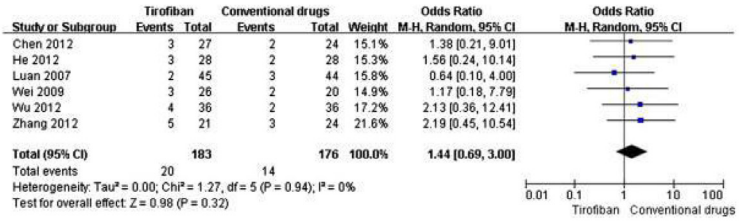

Figure 4. Forest plot of OR for the risk of major bleeding complication.

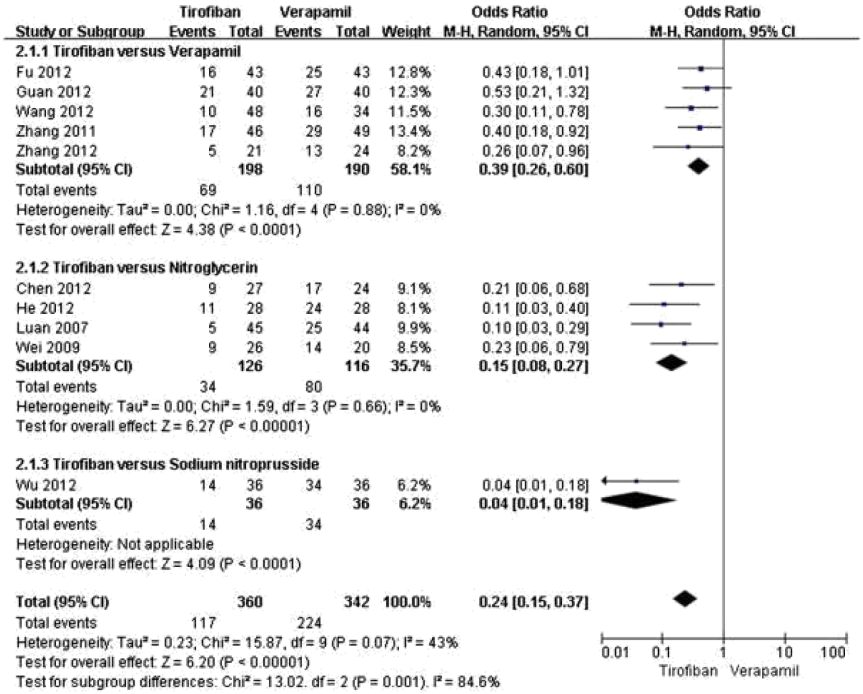

Figure 5. Forest plot of OR for TIMI, compared tirofiban with each kind of conventional drugs.

Due to the absence of normal distribution of the data and the small sample size in each group, the quantitative data are described by median and inter-quartile range and compared by the Mann-Whitney U test. The categorical data are described by frequencies and percentages and analyzed by the chi square test and the Fisher exact test. The AUC was calculated by the trapezoidal rule. Significance

level was determined at a p value < 0.05. For the statistical analyses, the statistical software SPSS version 17.0 for Windows was used.

DISCUSSION

No-reflow describes the persistence of reduced flow and associated myocardial perfusion despite the removal of mechanical epicardial coronary occlusion. The term was first coined by Ames et al in their experimental work on cerebral ischemia.[11] No-reflow is responsible for 40% of the primary percutaneous coronary intervention without complete myocardial reperfusion despite successful reopening of the infarct-related artery. Coronary no-reflow was also described first in an experimental setting[12] but was later noted to occur clinically as well.[13] No-reflow is mainly seen in acute myocardial infarction after catheter based or thrombolytic revascularization.[14] It is also seen during percutaneous intervention on old saphenous vein grafts[15] or on native coronary vessels in the setting of unstable angina.[16] The exact pathophysiologic mechanism behind this phenomenon has not been identified. The epicardial vessel is patent and the flow impairment is the result of pathology in the microcirculation. This microvascular dysfunction usually follows a direct injury. This injury can result from ischemia-reperfusion, distal embolization, etc.

Electron microscopy performed on animal experiments shows plugging of the capillaries with neutrophils, myocyte edema, and endothelial blistering.[22] Capillary resistance to flow is thus increased. Vasospasm, free radical generation and endothelial injury, debris/thrombus embolization, capillary plugging by neutrophils, and myocyte edema with intramural hemorrhage are all potential causes of no-reflow. It has also been postulated that activated platelets secrete potent vasoactive substances that promote distal microvascular constriction, thus impeding flow. In addition, particles from plaques or thrombi that are dislodged and embolized downstream by the revascularization procedure can lead to microvascular spasm. It is important to keep in mind that the responses of the endothelium of the distal microvasculature to these vasoactive substances may not be identical to those of intact endothelial cells. In other words, endothelium dependent vasoactive substances may lead to incomplete or even paradoxical responses once exposed to injured endothelium.

GPIIb/IIIa receptor antagonists have been found to improve procedural outcome and decrease the rate of adverse clinical events in those patients undergoing percutaneous interventions.[18] They also improve patient outcome in the setting of unstable angina or non-ST elevation myocardial infarction, especially when these patients undergo coronary interventions.[19,20] In view of the fact that GPIIb/IIIa antagonists improve percutaneous procedural outcome, one would suspect that they play a favorable role in the prevention of no-reflow, especially that no-reflow is more prone to occur in those situations where the integrin antagonists have proven their benefit. Studies that have evaluated coronary flow after percutaneous intervention have shown that GPIIb/IIIa blockade improves not only epicardial vessel patency, but also microvascular perfusion as well.[21] It appears that GPIIb/IIIa blockade maintains patency of recanalized coronary vessel and may prevent formation and embolization of platelet aggregates into the distal circulation.[22]

As we have discussed earlier, those patients who undergo interventions on saphenous vein grafts are prone to develop No-reflow phenomenon. The effects of GPIIb/IIIa antagonists on saphenous vein graft interventions per se have not been evaluated prospectively. Nevertheless, a review of the effects of Tirofiban on outcome after vein graft procedures did not show a significant reduction in adverse clinical events.[23] This may indirectly imply that Tirofiban does not necessarily improve coronary flow after percutaneous interventions on vein grafts. If this were the case, one would assume that no-reflow in this particular setting is mainly due to mechanisms that do not primarily involve platelet aggregation.

CONCLUSION

No-reflow is usually related to platelet aggregation, distal embolization, spasm of microcirculation, tissue edema, or a combination of many factors. The occurrence of no-reflow has been associated with adverse outcomes, such as periprocedural death, post-procedural Major Adverse Cardiac Event and increase risk of cardiovascular death.

The appointment of the GPIIb/IIIa inhibition before primary PCI contributes to the achievement of full blood flow restoration in the infarct-related coronary artery, and also reduces the risk of intraoperative and postoperative complications. Multiple studies

have shown that Tirofiban inhibits platelet activation that results from stent implantation. If these results are consistent in every trials, GPIIb/IIIa inhibition may be able to play a role in improving blood flow during primary PCI in ACS STEMI patients.

Once No-reflow Phenomenon is established, clinicians have described reversal of flow impairment soon after GPIIb/IIIa administration. The use of GPIIb/IIIa antagonists is indicated in high-risk coronary interventions. They have proven their ability to improve patient procedural and clinical outcome. They are probably not best used as a treatment once no-reflow is established. Rather, they are probably more helpful if used to prevent epicardial vessel occlusion and microvascular dysfunction.

CONFLICT OF INTERESTS

The authors declare that there is no conflict of interests regarding the publication of this paper.

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THE RELATIONSHIP BETWEEN ATHEROSCLEROSIS PLAQUES WITH NUMBER OF MONOCYTE IN BLOOD CELL OF THE WHITE RAT (RATTUS NORVEGICUS STRAIN WISTAR) THAT IS INDUCED BY ATHEROGENIC DIET

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ABSTRACT

Background: Atherosclerosis is signed by the existence of tunica intima vein. The circulation is started with endothelial lesion by low density lipoprotein (LDL), which stimulates process of inflammation therefore monocyte collection can be done with LDL, then macrophage tissue molds foam cell. Observation of lesion size enhancement directly correlate with increased recruitment of monocytes has implication to disease prevention and therapy.

Objective: To understand the relationship between processes of atherosclerosis plaque formation with number of monocyte in the blood of the white rat that is induced by Atherogenic diet.

Methods: The study-an empirical and experimental-uses randomized post test control group design. Data sampling were obtained by measuring the number of monocyte in laboratory rat and observation of laboratory rat's histopathology tunica intima aorta at last time of research, which is divided into two treatments (I = Atherogenic Diet; II = Non-Atherogenic Diet).

Result: Result shows that there are four out of eleven subjects of group treatment I are positively molding Atherosclerosis plaque. In contrast, plaques do not be found in five subjects in treatment II. Data analysis of relation between Atherosclerosis plaque with number of monocyte in laboratory rat that was induced by Atherogenic diet is having $p = 0.431$ ($p>0.05$), CI is 9.05 – 20.05 for treatment in group I; while CI -13.17 – 24.17 treatment in group II. The connection between treatment I and II with Atherogenic plaque molding with $p= 0.032$ ($p<0.05$).

Conclusion: There is no relationship between Atherosclerosis with number of monocyte in blood cell that is induced by Atherogenic. There is relation between treatment (I and II) with Atherosclerosis plaque molding.

Keywords: Atherosclerosis plaque, monocyte number, quantity of monocyte in Atheroclerosis, Atherogenic Diet

INTRODUCTION

Degenerative disease is a non contagious disease, yet chronic and based on World Health Organization (WHO) data, it is primary cause of death. Cardiovascular that is degenerative disease that can be a cause to death, for example atherosclerosis. Furthermore, according to WHO, 37% deathly disease in Indonesia based on gender and age is cardiovascular.[1,2]

In 2020, atherosclerosis is predicted to be first cause of morbidity and mortality in society. The rationale is due to life style shifting, from healthy to unhealthy one; lack of exercise and consume too much food(s), for instance. Around twenty five million people in the United State have at least one complicated atherosclerosis, the cardiovascular.[3,4]

The symptom of Atherosclerosis is forming of atheroma plaque in tunica intima vein. The first process of atheroma plaque forming is endothelin's failure to work because of exposition of low-density lipoprotein (LDL). Therefore, LDL moved into endothelial layers and be oxidized. Oxidized LDL would activate leukocyte receptor, vascular cell adhesion molecule-1 (VCAM-1) and release proinflammatory cytokine.[5,6]

The previous process affects monocyte and lymphocyte absorption. Then, monocyte turns to be macrophage that takes up LDL in tunica intima endothelium and shifts to be foam cell. The foam cell later produces proteolytic enzyme to degrade collagen in fibrosis tissue. Next, the array turns to be thin-atheroma plaque and gradually rupture easily. The rupture will activate thrombosis so thrombus appears.[7,8]

Monocyte is a type of leukocytes in circulation that is able to differentiate itself and then turn to be macrophage and dendritic cell. Monocyte has a pivotal role in inflammation process includes in atherosclerosis molding. One of these types of leukocyte functions to initiate, develop and complicate atherosclerosis lesion. Many related-published researches stated that there were many discussions about monocyte and differential monocyte function in atherosclerosis, and to patients with cardiovascular and healthy subject in atherosclerosis sub-clinics.[9,10]

Observation of lesion measurement is bound up with monocyte absorption, it helps us to do prevention and treatment. Based on the observation, researchers want to know connection between atherosclerosis plaque and number of monocyte in blood of laboratory-rat (*Rattus norvegicus* Stain Wistar) that get atherogenic diet induction.

METHODS

This research is a true experimental, conducted in laboratory with randomized post test control group design. The subject is male-healthy-larboratory-rat (*Rattus Norvegicus* strain Wistar) with weight is around 150-200 gram and age between 2-3 old months. Data sampling taken is by simple random sampling, and as result there are 16 laboratory-rats in each group.

The subjects would be divided into two subgroups: in one side, group I would be undergone atherogenic diet; forage is mixed with 10% pork-oil and this technique is being done in 15 days. In other side, group II gets normal forage with exactly the same period days as group I. Both groups planned to have adaptation process one week in advance.

Termination of subject would be done in 16th week as a last step. Researcher would take blood sampling to create smear, and prepare to dissect heart and aorta from rats. The preparate slides would be counted manually to record number of monocyte, and then move to prepare aorta smear by daubing *Hematoxylin Eosin* (HE), which next would be monitored with 40-times-zoomed microscope, the objective is to see lesion and foam cell.

Available Monocyte cell and histopathology aorta data then are analyzed to understand the causal relationship by using SPSS 21.0 for Windows after being tested to know the spreading with normality test called *Shapiro Wilk*. If the result is normal, then data are analyzed with Independent-test.

RESULT AND DISCUSSION

Result

The research needs an ethical clearance from Medical and Health Research Ethical Committee of Indonesia Islamic University. The research ran for 16 weeks, which first week for adaptation and 15 weeks for experiment. Subject for the research is laboratory rat, *Rattus Norvegicus strain Wistar,* based on eligibility it should be male and healthy rat with weight 150-200 gram and age for 2-3-month-old.

Total laboratory rats are 22 and divided to two subgroups. Group I is 12 rats and are given atherogenic diet in form of forage combined with 10% pork oil and propiltiourasil into drink, while group II is 10 rats as non-atherogenic group and get normal diet. In 15 weeks experiment, there were six rats died and then eliminated from experiment (five rats from group I and a rat from group II).

Table 1 showed that weight characteristic of rat (150 – 200 gram) is appropriate for the experiment. Further, the age also supported the experiment. Number of monocyte examining in laboratory rat, from the result showed in Table 2.

Tabel 1. Characteristics of experimental subject

Group	Subject	Weight	Age
I	1	200 gram	2 month
	2	200 gram	2 month
	3	200 gram	2 month
	4	200 gram	2 month
	5	150 gram	2 month
	6	200 gram	2 month
	7	150 gram	2 month
	8	200 gram	2 month
	9	170 gram	2 month
	10	200 gram	2 month
	11	165 gram	2 month
II	1	200 gram	2 month
	2	200 gram	2 month
	3	180 gram	2 month
	4	150 gram	2 month
	5	200 gram	2 month

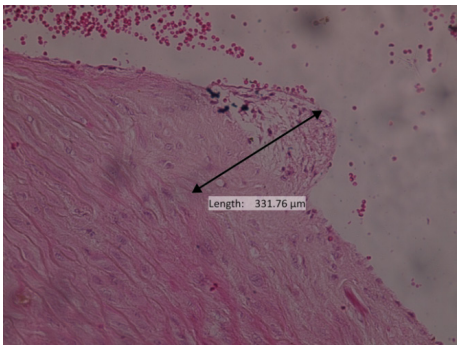
Tabel 2. Hematology examining result

Group	Subject	Monocye (0.00-1.81%)
Experiment I	1	20
	2	12
	3	34
	4	28
	5	28
	6	38
	7	48
	8	18
	9	50
	10	26
	11	36
Experiment II	1	30
	2	28
	3	32
	4	8
	5	18

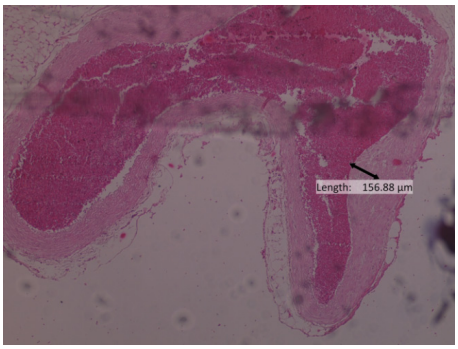
Examination of pathology arcus aorta and its branch that were used to see thickness of atherosclerosis plaque showed only four subjects from group I that was hispathologically change. The success result from experiment in group I by applying atherogenic diet in the forage could be found in subject 5, 7, 8 and 11. The pictures of the results (thickness in half part

of tunica intima aorta and molding of foam cell) could be seen on picture 1, 2, 3 and 4. However, there was no significant pathology change in other subjects. In group II, experiment without addition of atherogenic diet, there was no pathology change in all subjects.

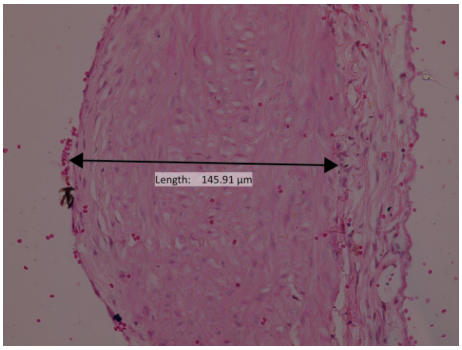
Normality test was done in advance by using *Shapiro-Wilk* to see whether distribution data was normal. Then, researcher did analysis to know relationship between treatment and atherosclerosis plaque molding. The result was $p = 0.000$ ($p<0.05$) meaning distribution was not normal; hence, researcher needed to do nonparametric *Mann-Whitney* test. The result showed in table 3, $p = 0.032$ ($p<0.05$) meaning there was relationship in treatment; atherogenic diet and normal diet with atherosclerosis plaque molding in laboratory rats' blood.



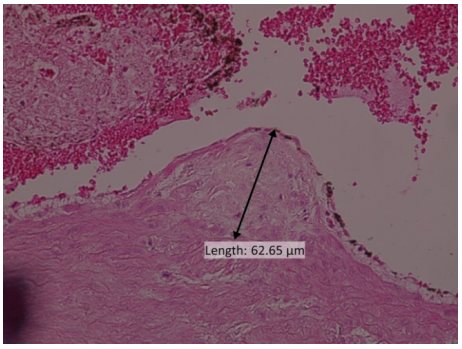
Picture 1. Tunica intima thickening and foam cell in subject 5



Picture 2. Tunica intima thickening and foam cell in subject 7



Picture 3. Tunica intima thickening and foam cell in subject 8



Picture 4. Tunica intima thickening and foam cell in subject 11

Table 3. Relationship between Treatment with Atherosclerosis Plaque

	N (Amount)	p
Plaque forming	4	0.032*
No plaque	12	

Independent T-Test

Significant value p = 0.431 (p>0.05) was got from statistic test by using *Independent t-test* from combination of two groups in Table 4. Result indicated there was no relationship between atherosclerosis plaque molding with number of monocyte in rats' blood.

On the contrary, in group with atherosclerosis plaque there was 95% *Confidence Interval* (CI), -9.05 -23.46; whilst, in group without atherosclerosis plaque, there was CI 95%, -13.17 – 24.17.

Tabel 4. Relationship between Atherosclerosis with Number of Monocyte

	Mean (%)	SD	p	CI (95%)
Plaque molding	32.50	12.68	0.431*	-9.05 – 23.46
No Plaque	27.00	11.48		-13.17 – 24.17

*Uji *Independent T-Test*

Discussion

Atherosclerosis is a deficiency that comprised local fibrolipid in form of plaques that look like lump or thickening, which is called atheroma and could be found in tunica intima and in tunica media. Next, atheroma developed and got complication including calsification, bleeding, ulceration, and thrombosis. Athrosclerosis process was primarily affected by exposition from excessive LDL.[11]

Laboratory animal used was rat (*Rattus Norvegicus strain Wistar)* with criteria: 2 month-old, average weight for group I was 185 gram and group II was 186 gram. Total selected rats were 22 and gradually decreased to 16 rats from both groups in 16th week. Previous researches that used laboratory rats placed the rats in a box (max 3 rats). In contrast, this research grouped five rats in a box. Such treatment could cause dropout in laboratory rats. [12]

Density in rats' box influenced rising stress for rats: competition in food, researcher could not monitor each rat's consumption. Stress increasing of rats caused illness and death as well. Other factor affecting dropout in research was inability to understand rats' health before treatment. In fact, advance check-up, blood checking and the like could help researcher to understand whether rat had infection or other disease. If such finding found, rat would be dropout in the middle of research.

In the research, rats were grouped to two; group I with atherogenic diet (addition 10% pork-oil), normal diet for group II. Result from group I informed positively thickening of tunica intima as a initial atherosclerosis process. This result was parallel with previous research; giving 5% pork oil in forage for eight weeks for laboratory rats could induct significant foam cell forming. Addition of 10% pork oil in forage had objective to get important result. Research was done in 16 weeks and 1 week adaption for laboratory cats, the experiment followed regulation from WHO about using animal for research.[13, 14]

Result indicated 11 subjects in group I that consumed atherogenic diet for 15 weeks, there were 4 subjects positively had thickening tunica intima as mark of atherosclerosis lesion. Obvious difference was seen between group I and II that was no change in group II meant there was no important pathological change. Based on previous theory, atherosclerosis was initiated with failure in endothelium layer on artery periphery, which was followed with change of permeability endothelium cell, addition of LDL, which was oxidized with combination with monocyte that turned to macrophage molded foam cell and in the next step, *cap fibrous* changed to be atherosclerosis. Similarly, in the research, pork-oil has high cholesterol level and affected atherosclerosis lesion in 15 weeks, and thickening of tunica intimia (please see picture 1-4). Analysis result used *Independent t-test* showed correlation of addition of atherogenic diet pork-oil with atherosclerosis forming plaque, $p = 0.032$ ($p<0.05$).[15]

Atherosclerosis process ran through several connected steps. One of indicator from the research was fatty streak and purely inflammation process. On this step, many monocyte derived macrophages and T limfosit emitted to atherosclerosis lesion. The condition would affect

leukocyte from marrow pushed to circulation.[16]

Previous researches, aimed to see relationship between total numbers of leukocyte and thickness of tunica intima-carotid artery showed positive relation, p = 0.000 (significant). The relation has supported a theory stated that leukocyte as marker from inflammation process and atherosclerosis is one of inflammation disease eventhough there was slight difference between previous research with recent research because usage of leukocyte total number. In one side previous research had not seen thoroughly monocyte number as the first cell that had role in inflammation process. In other side, other research had proved important relationship between total number of leukocyte with thickness of plaque on artery layer. Adjusting with this research that did not count total leukocyte but monocyte, neutrophil segment, and lymphocyte.[17, 18, 19]

Hematology examining from rats' blood in week 15 indicated three leukocyte data. The first data is monocyte becomes primary indicator of the research objective. Researcher found number of monocyte correlate with atherosclerosis lesion wide area. Furthermore, the more amount of monocyte shows wider area of atherosclerosis lesion, the more hypercolesterolemia Similar with such research, this research claimed group with forming atherosclerosis plaque, number of monocyte in rats' blood increased more than normal, with mean 36 cell per 100 field of the view.[9]

Previous research had proved that monocyte amount is one sign of atherosclerosis molding with testing result $p = 0.001$ ($p < 0.05$). likewise, some other researches stated monocyte Ly-6C dominated hypercolesterolmia and closely related to piling of macrophage in atherosclerosis plaque. Some researches also showed relationship between amount of monocyte with atherosclerosis plaque. On the other hand, this research with $p = 0.431$ ($p>0.05$) meaning there is no relationship between atherosclerosis plaque with number of monocyte in rats' blood that were given atherogenic diet from manually counting, the number of monocyte in atherosclerosis plaque was higher compared to group with no plaque. Unfortunately, there was important relation after statistic analysis done. [9, 20, 21]

One factor causing the result was treatment for group II, which had not got atherogenic diet, increasing of monocyte cell

significantly occurred even though not in every cell. The result is as influence of treatment in 15 weeks to all rats (with unlimited normal feeding) that caused hyperglycemia. Hyperglycemia could boost protein kinase C in which such protein caused progressive increase of expression transforming growth factor-beta (TGF-β) and then forming structure stiffness and abnormality in vein structure. This process probably could increase monocyte emission to rats' blood.

Other factor causing no relationship in research between atherosclerosis and monocyte amount is found in group II (with no atherogenic diet), all subjects have higher number of monocyte above normal condition. There was no blood check before experiment, while there was possibility leukocyte in some (or more) rats were more than the normal one. Apart from all imperfection of research, a factor causing monocyte or increasing monocyte more than normal number was unwell health condition of rats.

In previous research aimed to see condition causing monocyte, result showed 17 clinical conditions relating to monocytes, chronic virus infection, dengue hemorrhagic fever, malaria, diabetes mellitus, severe pneumonia, severe non-hematopoietic cell, appendicitis, and other chronic infections including fierce virus in bone marrow producing general leukocyte.[23]

Morphologically, atherosclerosis lesion could take place in vein muscles and big and average size of elastic tissues such as aorta, artery popliteal, artery femoral, carotid artery and renal artery. The research did hispathology examining only so that researchers could not control whether there was atherosclerosis plaque in other location besides in arcus aorta.[24]

There was significant increasing of neutrophil segment even though there was connection between number of neutrophil segment and atherosclerosis plaque $p = 0.746$ ($p > 0.05$). All experts who researched about Apo E-/- rat found that hypercholesterolemia inducted neutrophilia in rats' blood, and neutrophilia had contribution in initiating lesion forming of atherosclerosis. While, there has had not profound research of lymphocyte to see relationship with atherosclerosis plaque. In contrast, neutrophilia exists in blood circulation for 4 until 10 hours and could survive for 1-2 days so cannot give indication for chronic inflammation. And for lymphocyte, there has not had deep research

to understand relationship with atherosclerosis plaque. Lymphocyte itself is a type of leukocyte with crucial function in occurrence of chronic infection both viral and bacterial. Some theories stated similar statement regarding chronic response of stress.[25,26]

The limitation of the research is time length, which is 16 weeks or four months, meaning induction process occurred in minimal time to create atherosclerosis so that the result was minimal lesion or initial lesion atherosclerosis. More time spend for research would create other atherosclerosis factors, and probably atherosclerosis lesion would be obvious.

Other incomplete researches caused by small number of data sampling, therefore it is suggested to do with more data sampling. The reason is substitution for data sampling that dropout in the middle of process analysis. Beside, atherogenic diet used pork-oil because there is difficulty in looking for other diet substance with higher cholesterol than pork.

CONCLUSION AND SUGGESTION

Conclusion

1. Result showed no relationship between atherosclerosis plaque with number of monocyte in rats' blood (*Rattus novergicus strain Wistar*) that given atherogenic diet: $p = 0.431$ ($p>0.05$), CI 95% was -9.05 – 20.05 for treatment in group I, and CI 95% -13.17 - 24.17 for group II.
2. There is relationship between treatment (atherogenic and non-atherogenic diet) with atherosclerosis plaque forming with $p= 0.032$ ($p<0.05$).

Suggestion

1. Addition atherogenic diet with formulation under certain period of time could induct atherosclerosis; it needed better formulation, though in order to get clear process of atherosclerosis plaque.
2. It would be better to have pre- and post- experiment in order to see number of monocyte changing in laboratory rats.
3. Lack of other parts beside hearth-muscle and aorta vein to assure atherosclerosis forming in hearth. Next researcher is recommended to do complete check-up for laboratory rat.

4. Suggestion for next research to be done by doing other parameter examination, for examples: cholesterol level, LDL, HDL to acknowledge other factors that possibly give influence to result.

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BETTER ANATOMICAL IMAGING FOR ATHEROSCLEROSIS VIRTUAL RISK ASSESSMENT: SYSTEMATIC REVIEW ABOUT ACCURACY OF ANGIOGRAPHY VS INTRA VASCULAR ULTRA SOUND (IVUS) IN DETECTING SUBCLINICAL CORONARY ARTERY DISEASE

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ABSTRACT

Background: Site, extent, and severity of atherosclerosis can be acquired non-invasively using ultrasound and angiography. Plaque size is an important determinant of IVUS and angiography. We will discuss the current clinical accuracy of IVUS and angiography imaging in the heart.

Objectives: Understanding early detection of atherosclerosis and analyzing the available techinque to image it, focusing on Angiography and IVUS that can detect stenosis.

Methods: Trials were searched in computerized general databases (PUBMED,SCIENCEDIRECT,etc) by checking bibliographies and searching the keywords.Trials were eligible if they were: (1)RCT or controlled clinical trials; and (2)included accuracy and holistic approach for patient. Interventions of patients and outcomes was extracted. Main outcome measure for comparing the accuracy of IVUS and angiography.

Results: Previously published articles showed quantitative coronary angiography (QCA) reference diameter correlated moderately with IVUS. The mean values of minimum lumen diameter (MLD) and reference diameter showed the QCA reference diameter is smaller. The lesion site minimum lumen diameter (MLD) by QCA correlated less well with IVUS.

Conclusion: IVUS is more accurate quantitative predictor of atherosclerosis but more expensive. However, recent advance of the imaging of atherosclerosis have highlighted the inadequacies of it and need better imaging approaches.

Keywords: Atherosclerosis, IVUS, Angiography, Accuracy in Radioimaging

INTRODUCTION

There are several type of techniques for imaging the site and the severity of atherosclerosis both in diagnosis and in guiding future management (Table 1). They can be divided into two groups : invasive and noninvasive techniques.

Table 1. Different types radio-imaging in blood vessels. Source : Davies, John R, BSc, MBBS, et al. Journal of Nuclear Medicine. 2004; 45:1898–1907

Invasive	Non invasive
X-ray Angiography / DSA	B-mode Ultrasound
Intravascular Ultrasound	CTA / MRA
Angioscopy	PET (i.e. PET-FDG)
Intravascular Thermography	SPECT

X-ray Angiography

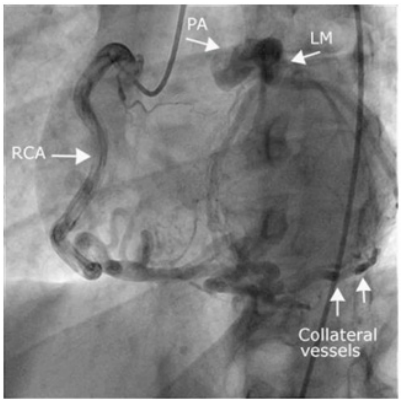

Figure 1. Coronary angiography of the right coronary artery. Source : Kristensen et al. Journal of Cardiothoracic Surgery 2008 3:33 doi:10.1186/1749-8090-3-33

Angiography is a procedure of medical imaging that uses an injection of a liquid contrast (dye) to make the blood vessels easily visible on X-rays. Angiography always using a catheter that able to be entered an artery. Before taking an X-ray, a liquid dye is injected into the blood vessels. When the test is on the arteries of the heart, the carotid artery, or the major arteries coming from the aorta, the catheter is inserted into the groin or most commonly the arm. First of all, a short and thin wire with a rounded tip that carefully inserted to the artery using a needle guide. Sometimes, the using of fluoroscopy (X-ray images) to the spot where the dye is needed. After that, the needle is removed and a vascular sheath inserted to cover the wire. Then, a catheter is inserted along the guide wire. After the catheter has already in the correct position, the wire is pulled out and the dye contrast is inserted by using the catheter. Finally, the artery can be checked on a screen.[1] Until

this moment, X-ray angiography remains the current gold standard imaging technique, although it has considerable limitations.[2-4]

Intravascular Ultrasound Device (IVUS)

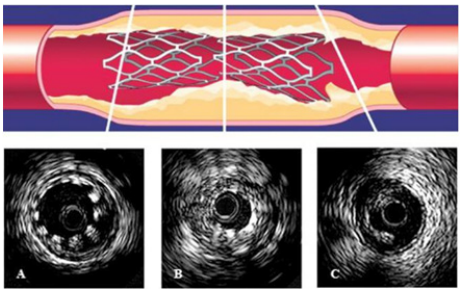

Figure 2. After stent deployment IVUS reveals. A) Malapposition of the stent at the proximal stent edge, B) Malexpansion of the stent in the middle and C) Vessel dissection at the distal stent edge. Source: A Mantziari, A Ziakas, G Stavropoulos, and I H Styliadi. Hippokratia. 2011 Jan-Mar; 15(1): 60–63.

For very long time, the only way of directly imaging the coronary arteries was through angiography, but now, with the recent advance in technology and refinement of intravascular coronary ultrasound (IVUS), it is became possible to thread a tiny ultrasound "camera" into the coronary arteries to give a valuable cross-sectional view of the stenotic lumen,[5] showing the physician where the normal artery wall ends and the plaque begins. Although not a routinely used device in certain situations, IVUS can aid in the selection and sizing of stents and balloons, and can offer assurance that a stent has been properly deployed. This is of increasing importance in the era of drug-eluting stents. Research conducted using IVUS has also shown that one of the causes of restenosis may be inadequate dilatation -- that doctor, concerned with injuring or dissecting the blood vessels itself, have tended to undersize the balloons. Therefore, intravascular ultrasound (IVUS) has become an essential adjunctive imaging modality in which is performed during coronary angiography and allows both precise quantitative measurements and characterization of plaque.[6] Major diagnostic applications of IVUS are to identify occult atherosclerosis in angiographically normal vessels, to evaluate intermediate lesions, and lesions difficult to assess by angiography,[7] to determine the extent of cardiac allograft vasculopathy and to assess the result of percutaneous transluminal coronary angioplasty (PTCA).[8,9]

RESULTS

Recent advance in the CAD diagnostic management should be based on both clinical utilization and cost effectiveness. We were focusing on the clinical

effectiveness of angiography and results supporting previous findings that concluded that the main value of 64-slice Computerized Tomography Angioplasty is to rule out significant CAD. The high NPV that is observed at the patient, artery and segment level establishes angiography as a highly effective alternative for the exclusion of obstructive coronary artery stenosis. Also important, however, that overall diagnostic accuracy varied at the individual artery level, with results being slightly worse for the LAD and CX arteries compared with the RC and LM arteries.[10] It is shown that unlikely that CTA will replace ICA in assessment for revascularisation of patients, particularly as angiography and angioplasty are often performed on the same occasion. However, for those patients who are candidates for stand alone diagnosis with ICA, CTA may be a viable alternative. Furthermore, in under serviced and under resourced health areas, where invasive coronary angiography is not always available, CTA appears a viable alternative.

Table 2. Angiography Finding. Source : Andrea S. Abizaid, MD, et al. Journal of the American College of Cardiology Vol. 34, No. 3, 1999

	Total (n=122)	No Event (n=104)	Any event (n=18)	*p*
Reference (mm)	3.91 ± 0.76	3.98 ± 0.74	3.63 ± 0.81	0.0594
Minimum Lumen Diameter (mm)	2.26 ± 0.82	2.32 ± 0.83	2.00 ± 0.72	0.1086
Diameter stenosis (%)	42 ± 16	42 + 16	42 ± 16	0.4700
Ostial lesion location (%)	21	20	22	0.9000
Left ventricular ejection fraction (%)	48 ± 11	48 ± 12	50 ± 7	0.5800
Any untreated vessel	39 (32)	27 (26)	12 (67)	0.0070
1 untreated vessel	24 (62)	17 (63)	7 (59)	
2 untreated vessels	13 (33)	9 (33)	4 (33)	
3 untreated vessels	2 (5)	1 (4)	1 (8)	
Any treated vessel	63 (52)	48 (46)	15 (83)	0.0140
1 treated vessel	40 (64)	34 (71)	6 (40)	
2 treated vessels	15 (25)	10 (22)	5 (33)	

	Total (n=122)	No Event (n=104)	Any event (n=18)	*p*
3 treated vessels	7 (11)	3 (7)	4 (27)	
Any treated or untreated vessel, n (%)	83 (68)	69 (66)	14 (78)	0.0510

At the patient-level, we find there were 18 studies of meta-analysis by disappearing studies that excluded equivocal test results. Sensitivity in the included studies ranged from 90.9% to 100.0%, with a pooled sensitivity of 98.2% (97.4%–98.8%). Specificity ranged from 45.5% to 100.0%, with a pooled specificity of 81.6% (79.0%–84.0%). The median PPV for the included studies was 90.5% (75.5%–100.0%), with the median NPV 99.0% (83.3%-100.0%). There was little difference between the results of the base case and the alternative analysis.

Table 3. The pooled value (95% CI) for sensitivity and specificity and median (range) for PPV, NPV and overall diagnostic accuracy of angiography.

Table 3. The pooled value (95% CI) for sensitivity and specificity and median (range) for PPV, NPV and overall diagnostic accuracy of angiography.

Study	**Sensitivity Angiography**	**Sensitivity IVUS**	**Forrest Plot Sensitivity**	**Forrest Plot Spesifisity**
Achenbach et al 2008	75	91		
Cademartin et al 2007	95	100		
Ghostline et al 2008	91	98		
Herzog et al 2007	76	100		
Usmann et al 2008	83	93		
Leschka et al 2008a	88	97		
Leschka et al 2008 b	90	95		
Mejboom, Meijs 2008	86	97		
Mejboom et al 2007a	96	100		
Mejboom et al 2007c	87	98		
Mejboom et al 2007b	91	98		
Oncel et al 2007	100	100		
Piers et al 2008	76	100		
Pugliese et al 2008	100	100		
Rixe et al 2009	87	100		
Scheffel et al 2008	94	100		
Shabestri et al 2007	91	83		
Weustink et al 2007	96	95		

At the overall vessel-level, the disappearing studies that excluded equivocal test results, sensitivity ranged from 87.3% to 100.0%, specificity 68.0% to 97.1%, PPV 53.4% to 95.0%, NPV 92.7% to 100.0%, and overall diagnostic accuracy from 73.8% to 98.0%.

In Intravascular ultrasound, plaque composition did not predict events at follow up. Plaque composition was as follows: 42% dominantly hyperechoic, 38% dominantly hypoechoic and 20% a combination of both. None of the plaques was dominantly calcific; the arc of calcium measured 66" ± 107". Patients with events had smaller lesion site lumen CSA (6.8 ± 4.4 vs. 10 ± 5.3 mm^2), maximum lumen diameter (3.07 ± 0.77 vs. 3.85 ± 0.86 mm) and MLD (2.30 ± 0.69 vs. 2.94 ± 0.81 mm); larger lesion site P&M CSA (15.7 + 5.2 vs. 11.9 + 5.9 mm^2) and CSN (70 6 14 vs. 53 ± 18%); larger reference segment CSN (42 ± 13 vs. 35 ± 12%); and larger AS (52 ± 21 vs. 34 ± 20%). For the entire cohort, the event rate was 60% for an IVUS MLD, 2.0 mm, 24% for an MLD 2.0 to 2.5, 16% for an MLD 2.5 to 3.0 mm and 3% for an MLD . 3.0 mm.[11]

Table 4. A paired analysis of angiographic and intravascular ultrasound (IVUS) assessnebts of plaque characteristics in corresponding arterial vessel segments.

Variable	Angiographic Analysis	IVUS Analysis		*p**	*r***
Length of stenosis (mm)	14.3 (±12)	17.3 (±13)		<0.05	0.80
Calcification				<0.05	0.27
None	26%	Calcium 0-5%	51%		
Mild	33%	Calcium 5-15%	42%		
Moderate	28%	Calcium 15-25%	7%		
Severe	12%	Calcium >25%	0%		

p = Significance level of the paired t-test for continuous variables and Chi-squared analysis (or Fisher exact test where appropriate) for proportions.

r = Correlation coefficient for the relationship between angiographic and IVUS measurements.

Source : Arthur ZM, et al. J Vasc Surg. 2010 April; 51(4): 933–939.

Table 3 shown a paired imaging and detection of the length of stenosis, evaluation of concentricity, and estimation of calcification. By angiography, the length of stenosis was 14.3 ± 12

mm, however by IVUS the same stenosis was 17.3 ± 13 mm, a 3.0 mm difference (95% CI: 0.9–5.1mm, $p < 0.05$). When determining the extent of calcification in a plaque, physicians using angiography found higher calcium burden within a lesion. Angiographic interpretation classified 40% of plaques as moderate or severe calcification, whereas IVUS evaluation only yielded 7% of patients to meet criteria for moderate calcification and 0% for severe calcification ($p < 0.05$, 95% CI). In addition, physician using angiography rated 72% of lesions as eccentric and the remaining 28% as concentric. While evaluating concentricity by IVUS, a plaque diameter index was used, where > 0.33 indicated a concentric lesion. By IVUS, only 40% of lesions were eccentric and another is concentric.[12]

DISCUSSION

Contrast angiography has been the standard for evaluating site and extent of coronary artery disease. It uses contrast medium (dye) to visualize the coronary artery lumen in one or more longitudinal planes, measuring a stenosis or reduction in lumen caliber rather than the extent of atherosclerosis. Relative measurements of arterial narrowing (percent diameter stenosis) are the most commonly used angiographic measurements of disease significance and are based on absolute diameters of both the apparently "normal" reference segment and the target lesion.[13] Fundamental in this calculation is the assumption that the normal segment is free of significant atherosclerotic disease. Moreover, the pathologic studies indicate that the extent of coronary atherosclerosis is underestimated by visual analysis of angiographically "normal" coronary artery segments.[14-18] The diagnostic accuracy of angiography in imaging CAD is well established, whereas improvements are still needed for reducing the overestimation of CAD severity and assessment of plaque composition. Angiography is able to be an efficient initial triage tool in patients with stable angina with low-to-intermediate risk in EDs because of its high sensitivity and NPV.[19] In patients with suspected CAD, angiography can be a cost-effective alternative beside exercise ECG for the initial coronary assessment in patients with intermediate pre-test likelihood of suspected CAD. Angiography will undoubtedly provide better quality coronary imaging including structural and functional imaging of myocardium with lower radiation exposure.[20-21] On the other hand,

IVUS has a detailed, higher quality of crosssectional imaging of the coronary arteries in vivo. The normal coronary artery wall (tunica intima, media and adventitia), the major components of the atherosclerotic plaque (lipid, fibrous connective tissue and calcium) and the changes that occur in coronary artery dimensions and anatomy with the pathologic of atherosclerotic disease process can be studied in vivo using intravascular ultrasound that is not achievable using other imaging modalities. Specifically, the presence and composition of atherosclerosis in angiographically normal reference sites can be quantified.[22-24] The major reasons for doctor using angiography or IVUS appear to be the following: 1) Diffuse atherosclerotic involvement affects the DS calculation because of the lack of a normal reference segment, 2) A short coronary artery also makes identification of a normal reference segment difficult, 3) There is compensatory enlargement of the blood vessel as plaque burden increases to preserve lumen size,[25] 4) There may be unique geometric issues in coronary artery disease because the correlation between angiography or IVUS appears to be somewhat better in non-LMCA stenoses[26,27] and 5) There is significant interand intraobserver variability in the angiographic assessment of LMCA disease.[28-30]

CONCLUSION

IVUS seem to be more accurate for diagnosing CAD or atherosclerosis, because the ultrasound camera that inserted in the blood vessels by catheter tends able to found plaque in the blood vessels in vivo. IVUS usually can found and measure the plaque more specifically and accurate rather than angiography. IVUS also tends to quantify and evaluate concentric lesion. The primary disadvantages of IVUS being used routinely in a cardiac catheterization laboratory are its cost expensiveness. At this moment, some limitations of IVUS appear to involve technical aspects and will be solved with future modifications and designs. The present relatively large catheter delivery systems limit intravascular echocardiographic access to distal segments of coronary arteries, including marginal and diagonal branches, which are frequently sites of interventional procedures. Reliable detection of thrombus (fresh and organized) and discrimination of thrombus superimposed upon plaque from soft, lipid-laden plaques also are limitations of the present imaging resolution and processing. The vagaries of coronary artery

poses problems with catheter-vessel coaxial alignment, which can lead to overestimation or underestimation of disease and wall thickness by ultrasonic images. As with any new device or drug, initial equipment costs are high, which may deter early widespread use of the intravascular echocardiograph. However, the potential and real benefits of intravascular ultrasound far outweigh these present limitations. IVUS continues to improve and some manufacturers have proposed building IVUS technology into angioplasty and stent balloon catheters, a potential major advance, but limited by complexity, cost and increased bulk of the catheters. The usage of IVUS at this time has contributed to the optimization of invasive diagnosis and intervertional treatment of coronary lesions by means of evaluating borderline lesions, stenting placement and stent restenosis.

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TOTAL FLAVONOID CONTENT OF EXTRACT AND FRACTIONS FROM *ANDROGRAPHIS PANICULATA* HERBS AND ITS THIN LAYER CHROMATOGRAPHIC PROFILE

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ABSTRACT

Flavonoid is one of the secondary metabolites from plants that have important biological and pharmacological activities. This study was conducted to determine total flavonoid content from *Andrographis paniculata* leaves using aluminium chloride colorimetric assay. The total flavonoid content of water extract from *A.paniculata*, and their fractions of chloroform, ethyl acetate and residue were 3,7%; 5,22%; 9,26%; and 1,80% respectively expressed as quercetin equivalents. The thin layer chromatography profile showed the presence of terpenoids and flavonoids.

Keywords: total flavonoid, *Andrographis paniculata*, colorimetry, thin layer chromatography

INTRODUCTION

Medicinal plants are used as alternative therapeutic tolls to promote health, to prevent, and to cure many diseases (Kamboj, 2000). A plant has a therapeutic activity because of the active compounds contained in the plant. Active compounds from the plants have potential use in medicine (Pal & Shukla, 2003). Medicinal plants have many pharmacologically active compounds such as flavonoids, alkaloids, tannin, steroids, etc (Sharma et al., 2008).

Andrographis paniculata is used as traditional herbal medicine for diabetes mellitus, skin diseases, ear infections, and colds.(Departemen Kesehatan Republik Indonesia, 2000). The previous study mentioned that the therapeutic activity of a plant depends on the compounds. The content of the main compounds in extracts of *A. paniculata* herbs were group of andrographolide and flavonoids (Chao & Lin, 2010). Andrographolide has been widely studied as anti-diabetes

mellitus, hypoglycemic and anti-atherosclerosis. Flavonoids are compounds in plants that ~~is~~ are efficacious and potential for therapy. Previous research has found *A. paniculata* contains flavonoids. It is necessary to develop methods of standardization of traditional medicine preparations, one of which is determination levels of compound, which is flavonoid.

Determination of total flavonoid content from *A. paniculata* herbs was done using aluminium chloride colorimetric assay expressed as quercetin equivalents. Quercetin, the most abundant dietary flavonol, is a potentially for therapy and found in most of plants (Pal et al., 2009). Therefore, the objective of our present study is to determine the total flavonoid content of *A. paniculata* using aluminium chloride colorimetric method in extract and fractions. In this study, quercetin was used as a standard.

MATERIALS AND METHODS

Plant material and chemicals

The herbs of *Andrographis paniculata* were obtained from Kulon Progo, Yogyakarta, Indonesia. Plant materials were identified taxonomically by expert taxonomist at the Biology Pharmacy Laboratory, Faculty of Pharmacy, Gadjah Mada University. Potassium acetate, chloroform, ethyl acetate, aluminium chloride, n-buthanol, acetic acid, and quercetin were pro analysis (E-merck).

Preparation of plant samples

The herb of *A. paniculata* were cleaned and cut into small pieces. The herbs was dried and refined with grinder. Dried samples (500 mg) were extracted with aquadest using decoction method, during 30 minutes to have a decoction. After filtration, the supernatant was separated and evaporated to obtain viscous extract. The viscous extract was separated with liquid partitions using chloroform and ethyl acetate. The solvent from each fraction were evaporated to obtain chloroform fraction, ethyl acetate fraction and residue. The rendements were calculated.

Identification

Extract and fractions were analyzed by thin layer chromatography with cellulose as stationary phase and n-buthanol-acetic acid-water (4:1:5, upper layer) as mobile phases. The resulting chromatogram was sprayed by citroborate reagents. Chromatogram was detected in visible light and in UV366. Flavonoid groups would show a specific fluorence..

Determination of total flavonoid content

Determination of total flavonoids UV-Vis spectrophotometry based method performed according to modification of Chang et al. (2002) by measuring the absorbance at the maximum wavelength using quercetin as standard (Kumar et al., 2008).

Measurements of wave length made after $AlCl_3$ reagent and potassium acetate were added to a solution of quercetin in methanol (10 µg/mL).

Operating time was determined by measuring the absorption of quercetin solution with a concentration of 10 ug/mL at the wavelength of maximum every five minutes, in the minutes to 0, 5, 10, 15, 20, 25, 30, 35.40 and 45 to obtain a steady uptake value ,

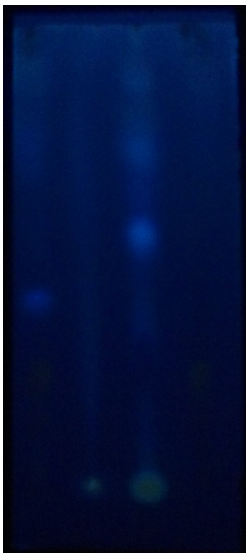
Standard solution of quercetin was created 1.000 ɯµg/mL by dissolving 10.0 mg of quercetin in 1 mL methanol and made a series of volume 10, 20, 30, 40 and 50 ɯµL. 1,990 mL of methanol; 0.1 mL of 10% w/v $AlCl_3$ solution; 0.1 mL of 1 M potassium acetate and 2.8 mL of distilled water were added to every quercetin solution. After allowed to stand 30 minutes at room temperature, absorbance was measured at 415 nm (maximum wave length). (Kumar et al., 2008).

10 mg water extract of *A. paniculata* were weighed and dissolved in methanol to 1 mL then diluted in such a way to obtain a concentration of 400 µg/mL. Taken 0.5 mL of the extract and then added 1.5 mL of methanol; 0.1 mL of 10% of $AlCl_3$ solution; 0.1 mL of 1 M potassium acetate and 2.8 mL of distilled water. After allowed to stand 30 minutes at room temperature, absorbance was measured at maximum wave length.

RESULT AND DISCUSSION

A. paniculata water extract obtained from decoction method (rendement ... %). Water extract was partitioned by liquid-liquid method with chloroform and ethyl acetate. In general, flavonoids are relatively polar compound. The polarity of a flavonoid depends on variety of substituents. Flavonoids in the form of glycosides (bound sugar) are easily soluble in water and diluted ethanol.

Analysis by TLC method performed to water extract, chloroform fraction, ethyl acetate fraction and residue. The stationary phase used was cellulose. The best separation was obtained by using n-buthanol-acetic acid-water (4:1:5, upper phase) as mobile phase. Detection results of TLC under UV366 nm can be seen in Figure 1.



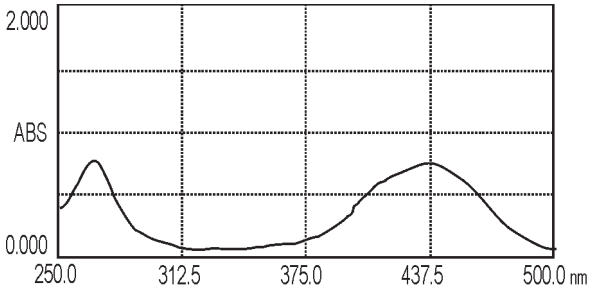
A B C D

Figure 1. TLC Identification of flavonoid of *A. paniculata* on cellulose using n-buthanol-acetic acid-water (4:1:5, upper phase). (A) Water extract, (B) Chloroform fraction, (C) Ethyl acetate fraction, (D) Residue

Identification of flavonoid using thin layer chromatography (TLC) under UV366 nm showed yellow and blue spots on water extract blue spots on chloroform fraction and blue and yellow spots on ethyl acetate fractions. In the water extract (A) obtained one yellow at Rf 0.44 and blue spot at Rf 0.00. The chloroform extract (B) had blue spot in Rf 0.00 and some other blue spots at different Rf values. The ethyl acetate fraction (C) showed one blue spot at Rf 0.00 and four yellow spots at Rf 0.38; 0.55; 0.733; and 0.88. The residue did not contain flavonoids. From the TLC results showed that the water extract and ethyl acetate fraction contained polar and non polar flavonoids whereas chloroform fraction contained non polar flavonoids. Flavonoids which fluoresce blue are nonpolar, which may have methyl or methoxyl group. Flavonoids with yellow fluorescences are more polar, which may bounded with sugar to form glycosides.

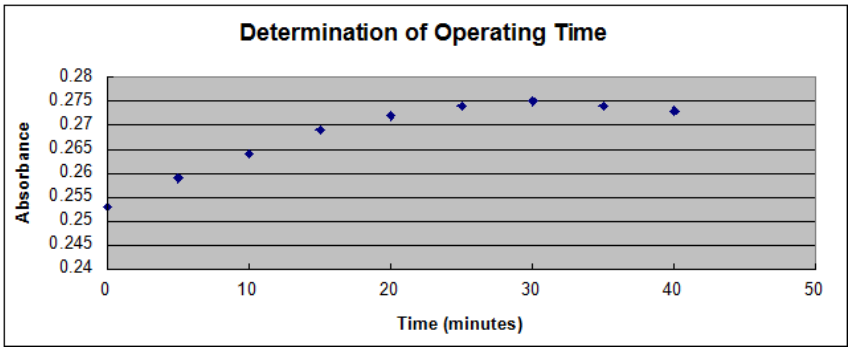
Determination of total flavonoids from extract of *A. paniculata* by colorimetric measurement was based on the formation of color after addition of $AlCl_3$. The principle of determination of flavonoids with $AlCl_3$ is the formation of a complex between $AlCl_3$ with a carbonyl group at C-4 atom and also with ortho-hydroxyl group at C3' and C4' especially for flavones and flavonols. So this method can be used to determine the total flavonoid class of flavones and flavonols.

The calibration curve was done for quercetin as a standard compound because quercetin had group at C-4 and ortho-hydroxyl group at C3'and C4'. The maximum wavelength from quercetine that added $AlCl_3$ was 435,5 nm (the sinamoil group).



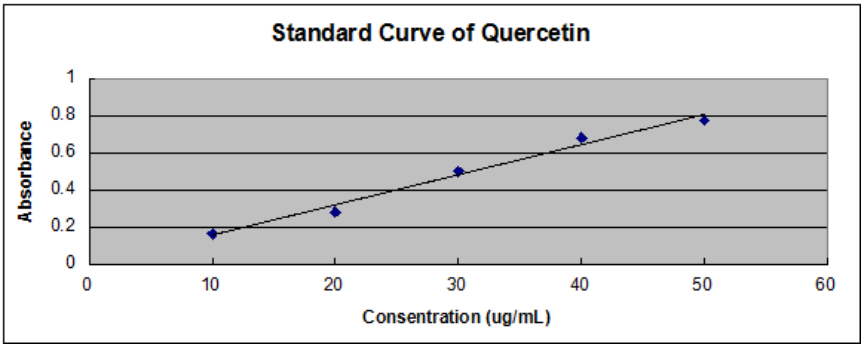
Picture 2. The maximum wave length of quercetin after addition of $AlCl_3$

Operating time obtaining the absorption of quercetin after addition of added $AlCl_3$ reached stable until 30th minutes. The measurement will be held in 30th minutes after the addition of reagents.



Picture 3. The operating time of measurement of quercetin absorption

Determination of standard curve of quercetin.



Picture 4. Standard curve of quercetin

Analysis of the quercetin standard curve obtained by the equation linear regression Y=0.0162x – 0.0040 and the value of the correlation coefficient (R^2) = 0,9843.

Table 1. The total flavonoid content of extract and fractions of *A. paniculata*

No	Extract or Fractions	Total Flavonoid (%)
1	Water extract	3.70
2	Chloroform fraction	5.22
3	Ethyl acetate fraction	9.26
4	Residue	1.80

The results of the analysis of the total flavonoid compounds calculated as quercetin equivalent showed the highes level of total flavonoid was found in ethyl acetate fraction (9,26%). This was the same as shown in the results of the TLC. Ethyl acetate fraction was rich of flavonoids. Chloroform fraction had higher levels of total flavonoids 5.22% than water extract (3.70%). The lowest total flavonoids contained in the residue at 1.80%, and the TLC result showed no flavonoid spot in the residue.

One way ANAVA test performed to determine differences in levels of total flavonoids relative to the quercetin contained in extract and other fractions. Analysis of variance test results showed that the total flavonoid content in extract and fractions of *A. Ppaniculata* were significantly differences.

Flavonoids contained in *A. paniculata,* have many of which were semi-polar, well dissolved in ethyl acetate and had the highest level compared to the others fractions.

In addition, there may be other compounds that participate measured at a wavelength equal to the total flavonoid levels are quite large.

Assay of flavonoid using colorimetric methods was performed with addition of $AlCl_3$ (Chang et al., 2002). The weakness of this method is unable to detect all kinds of flavonoids and could not be used in the example with a complex matrix (Chang et al., 2002). This method could not be used to detect all the flavonoids contained in extracts or fractions. This method can only detect the presence of flavones and

flavonols. *A. paniculata* has more than 10 kinds of flavonoids which generally include 5-hydroxy-7,8-dimetoksiflavon, 5-hydroxy-7,8,2', 3'-tetrametoksiflavon, 5-hydroxy-7,8,2'-trimetoksiflavon 7-metilwogonin and 2'-O-methyl ether (Chao and Lin, 2010). Flavonoids in the *A. paniculata* contains flavones and flavonols, so the assay method was suitable in this analysis.

CONCLUSION

Total flavonoid content in the *A. paniculata* in ethyl acetate fraction, chloroform fraction, water extracts and the residue were 9.26%, 5.22%, 3.70% and 1.80% respectively The highest total flavonoid content was ethyl acetate fraction and it was showed also by TLC analysis.

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THE SUITABILITY OF ORAL ANTI-DIABETIC PATIENTS WITH CHRONIC KIDNEY DISEASE COMPLICATIONS IN PKU MUHAMMADIYAH HOSPITAL YOGYAKARTA

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ABSTRACT

Diabetes mellitus is a metabolic disorder which increasing prevalence every year and diabetes mellitus is the biggest causes for chronic kidney diseases (CKD). Oral antidiabetic (OAD) especially for chronic kidney disease should need more attention because of its decrease renal function. This study aimed to evaluate the suitability of oral antidiabetic selection for chronic kidney disease patient in PKU Muhammadiyah Yogyakarta hospital.This is a cross-sectional study in patients with chronic kidney disease by reviewing medical records from January to December 2014. *National Kidney Foundation-Kidney Disease Outcomes Quality Initiative* (NKF-KDOQI) used as the literature to evaluate the suitability of oral antidiabetic selection.

The number of patients were 32, with the characteristic patients: 17 men (53.1%) and 15 women (46.9%); 5 elderly (15.6%) and 27 non elderly (84.4%); routine hemodialysis 2 patients (6.2%) and non hemodialysis 30 patients (93.8%). The drug profile of antidiabetic were 18 patients (56.3%) use metformin, pioglitazone 4 patients(12.5%), metformin-glimepirid 6 (18.7%) patients, and metformin-acarbose 4 patients (12.5%).The suitability selection for oral antidiabetic showed that 28 patients (87.5%) suitable and 4 patients (12.5%) not. It is conclude that the oral antidiabetic suitability elections were largely in accordance with guidelines NKF-KDOQI.

Keyword: Suitability, OAD, CKD

INTRODUCTION

The incidence and prevalence of diabetes mellitus have grown significantly throughout the world, due primarily to the increase in type 2 diabetes. This overall increase in the number of people with diabetes has had a major impact

on development of diabetic kidney disease. This condition is the leading cause of end-stage renal disease accounting for approximately 50% of cases in the developed world (ADA, 2014).

Diabetic patients with renal impairment are clinically complex and vulnerable to drug-drug interactions and adverse events associated with medication (Johnson et.al. 2008). Sulfonylurea long acting is the leading cause of hypoglicemia in diabetic kidney disease. Biguanide has potential to cause lactic acidosis and nephrotoxic. Glitazone can worsening sodium retention in kidney disease patients (Arroyo et.al. 2011)

The National Kidney Foundation (NKF) published clinical practice guidelines on the management of diabetes in patients with CKD. A recent study found that OAD treatment not concordant with NKF guideline recommendations led to worse clinical and economic outcomes (Chen et.al 2011).

This study aimed to evaluate the suitability of oral antidiabetic selection for chronic kidney disease patient in PKU Muhammadiyah Yogyakarta hospital based on NKF-KDOQI guideline.

METHODS

This is cross sectional study with retrospective database. Subject of study were patients diabetic kidney disease in PKU Muhammadiyah Hospital in Yogyakarta period 2014. Inclusion criteria were diabetic kidney disease patients and use oral anti diabetic. Exclusion criteria was patients use insulin for control glicemia. To evaluate the suitability of OAD be used NKF-KDOQI Clinical Practice Guidelines for Chronic Kidney Disease: Evaluation, Classification, and Stratification.

RESULTS & DISCUSSION

Characteristic respondens

Table 1 showed sociodemographic and characteristif of respondens in this study. The number of male were higher than female respondens. Gender suggested as predictor non-modifiable inisiation and progression chronic kidney disease. A greater incidence of end stage CKD has been reported in men. Female gender has been associated with a slower progression of CKD and better kidney conservation and patient health outcomes (Brown et.al. 2010). The factors involved in this gender disparity may include diet, kidney and glomerular size, differences in glomerular

hemodynamics, and the direct effects of sex hormones (Silbiger and Neugarten, 2008).

Based on Kidney Early Evaluation Program and National Health and Nutrition Examination Survey data biggest prevalence age of chronic kidney disease in USA and other country were more than 65 years old. Based on recent data on the average age who has dialysis were 65 years old (Stevens et.al 2010).

Table 1. Sosiodemographic and characteristic respondens

No	Category	N	(%)
1	Gender		
	Male	17	53,1
	Female	15	46,9
2	Age		
	< 60	27	84,4
	>60	5	15,6
3	Dialysis		
	Non dialysis	30	93,7
	Dyalisis	2	6,3
4	Stage CKD		
	CKD stage <3	9	23,1
	CKD stage 3 – 5	23	71,9
	Total	**32**	**100**

Responden's Medication profile

Table 2 show that metformin dominanly used by 18 respondens (56.25%). Metformin can be used for chronic kidney disease stage 1, 2 and 3 but avoid for stage 4 and 5 (Lipska et.al 2011). The FDA recommends that metformin should not be used with serum creatinine ≥ 1.5 mg/dl in men and ≥ 1.4 mg/dl in women or with decreased creatinine clearance in people over age 80. Because metformin is renally cleared, this recommendation is in place to reduce the risk of lactic acidosis in individuals with even modest renal impairment (Hahr and Molitch, 2015).

Table 2. Characteristic of Medication

Medication class	Oral antidiabetic	Dosage (mg/day)	N	(%)
Biguanide	Metformin	1500	12	37,50
		1700	6	18,75
Tiazolidindione	Pioglitazone	15	1	3,12
		30	3	9,37
Biguanide and Sulfonilurea	Metformin and Glimepiride	1500 + 1	5	15,62

Medication class	Oral antidiabetic	Dosage (mg/day)	N	(%)
		1500 + 2	1	3,12
Biguanide and Inhibitor Alpha-glukosidase	Metformin and Acarbose	1500 + 150	2	6,25
		1500 + 300	2	6,25
Total			**32**	**100**

Combination metformin and glimepirid used by 6 respondens (18.75%). Sulfonylureas and their metabolites are renally cleared, leading to an increased risk of hypoglycemia as GFR declines. Hypoglycemia is greatly increased with glimepiride with GFR <60 ml/min/1.73 m2 (Hahr and Molitch, 2015). This combination should be monitoring cause its potential hypoglicemia (Lacy et.al. 2010; Roger and Flora, 2008). Pioglitazone used by 4 respondens (12.5%). They are metabolized by the liver and can be used in CKD. However, fluid retention is a major limiting side effect and they should not be used in advanced heart failure (Hahr and Molitch, 2015).

Evaluation of suitability anti diabetic oral

Table 3 show that suitability oral anti diabetic largely in accordance with guideline NKF-KDOQI. Largely respondens 28 patients (87.5%) in accordance and 4 patients (12.5%) not accordance with NKF-KDOQI. The use of oral anti- diabetic kidney failure stage 1-3 is still in accordance with the guideline. In this study, the majority of respondents had kidney failure stage 1-3 so that the results of evaluation of the use of oral anti-diabetic largely appropriate . The use of oral anti- diabetic chronic renal failure stage 4-5 need special attention, especially potential hypoglycemic effects of medication due to decreased kidney function.

Table 3. Characteristic Medication and Clinical Guideline

Oral antidiabetic	Stage CKD	GFR (ml/min/1.73m^2)	NKF Guideline	
			Concordant n=28 (87,50%) N (%)	Non concordant n=4 (12,50%) N (%)
Metformin	2	60 – 89	6 (18,75)	-
	3	30 – 59	12 (37,50)	-
Pioglitazon	5	< 15	4 (12,50)	-
Metformin dan Glimepirid	2	60 – 89	3 (9,37)	-
	3	30 – 59	3 (9.87)	-
Metformin dan Acarbose	4	15 – 29	-	2 (6,25)
	5	< 15	-	2 (6,25)

CKD (chronic kidney disease); GFR (glomerular filtration rate); NKF-KDOQI (*National Kidney Foundation-Kidney Disease Outcomes Quality Initiative)*

The use of oral antidiabetic not in accordance with the guidelines, NKF - KDOQI occurred in 4 patients (12.5 %) . This is due to the use of metformin is contraindicated in patients with a GFR < 30ml / min / 1,73m^2. Use of acarbose are also advised to be avoided at GFR < 30ml / min / 1,73m^2.

Sulfonylureas and their metabolites are renally cleared, leading to an increased risk of hypoglycemia as GFR declines. Glipizide recommended as sulfonylurea with minimal potential hypoglicemia effect. Glimepiride in chronic kidney disease stage 3, 4 and 5 need adjusment dose maximal 1mg/day dan should be avoid in dialysis patients (NKF-KDOQI).

Pioglitazone were metabolized by the liver and can be used in CKD. However, fluid retention is a major limiting side effect and they should not be used in advanced heart failure. This also makes their use in CKD, particularly patients on dialysis, limiting. They have been linked with increased fracture rates and bone loss, thus use in patients with underlying bone disease (such as renal osteodystrophy) needs to be considered. No dose adjustment is indicated with either in CKD (NKF-KDOQI; Hahr and Molitch, 2015).

CONCLUSION

This study conclude that the oral antidiabetic suitability elections were largely in accordance with guidelines NKF-KDOQI.

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PRELIMINARY STUDY : (ALERT) BASIC LIFE SUPPORT GROUP ESTABLISHMENT OF POLICE FOR OUT OF HOSPITAL CARDIAC ARREST TREATMENT IN YOGYAKARTA

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ABSTRACT

Objective: The purpose of this present study was to systematically review the existing literature about training of basic life support for Out of Hospital Cardiac Arrest Treatment. Materials and methods: 10 studies were identified through the Resuscitation journal, science direct, simulation in health care, and American Heart Association. We conducted a comprehensive search for eligible studies as using keyword basic life support, cardiac arrest, out of hospital cardiac arrest, traffic accident, trauma, and police officer.

Result: Preliminary study done in ten different articles showed that basic life support simulation and training have positive effect on respondents. Five of studies showed that basic life support training and simulation increase respondent's skill and performance, four studies stated the increasing of self efficacy and confident, and one of the study showed that respondents still have a good memory on basic life support training after several months.

Conclusion : Based on the reviews mentioned above, simulation based training plays role in increasing respondents' skills and performance in conducting basic life support. Some journals also state that basic life support increases survival rate of out of hospital cardiac arrest (OHCA).

Keywords : Cardiac arrest, basic life support, out of hospital cardiac arrest, police.

INTRODUCTION

Traffic accidents resulting casualties over 1.2 million people each year and have a major impact on public health and development sectors. More than 3,400 people died on the road every day and ten millions of people were injured and disabled each year. Data from

the World Health Organization (WHO) shows that Indonesia is ranked fifth in the case of traffic accidents. According to data from the Global Status Report on Road Safety released by WHO, Indonesia has increased the number of traffic accidents by 80%.[1]

The number of victims of traffic accidents that lead to emergency conditions needed help quickly at the scene to prevent high victim's mortality. Treatment given at the scene is part of the Prehospital care. Prehospital care is done to victims before transporting victims to the hospital. Proper prehospital treatment can reduce the risk of death from trauma.[2] Treatment in this case is providing basic life support. Basic life support or BLS is the basic course of action given to a person with several conditions. Those are no movement, unconscious, and no breathing. The administration of basic life support in case of accident with cardiac arrest done by bystander significantly can increase up to 3 times the chance of survival, but only 32% of cardiac arrest victims get help from bystander.[1] It stated that we should increase the number of bystander-trained basic life support to cope with the amount of emergency cases every day. In most cases, the first helper that arrives in traffic accident scene is police officer, followed by ambulance from hospital. The response time of ambulance to come to the traffic accident scene is often slower than police officer, which makes the administration of first aid is delayed. Therefore, police officer have important role in providing first aid to the victims of accident if there is limited medical personnel. Mentioned in the regulations of the police chief of the Republic of Indonesia Number 23 Year 2010 on Organizational Structure and Work at the level of Police and the Sector Police stated that the traffic police are officers responsible for maintaining road safety and minimalize accident victim. The working procedures of the police set up a task related to the responsibility of the police on road safety.[3] It shows that the skills to provide basic life support to accident victims becomes important to be known by the police officer as an effort to carry out the task that has been entrusted to the police officer including the knowledge of basic life support until the ability to rescue the accident victims whose suffered cardiac arrest and no breathing condition.[4] Increasing the number of people-trained basic life support in social environment will give victims of cardiac arrest a greater chance of survival due to the administration of bystander

basic life support before the assistance of medical personnel or even the hospital came. Based on the background mentioned above, the authors interested in taking a big step to form a basic life support standby group in Yogyakarta police force that has function to provide basic life support to the victims of traffic accidents.

METHOD

This preliminary study uses literature reviews which have the validity and reliability that are accountable by combining notions from literature studies. The data have also been analyzed with descriptive argumentative analytical method in order to be transformed into a new notion. We summarize them briefly below:

Study Eligibility

We define technology-enhanced simulation as an educational tool or device with which the learner physically interacts to mimic an aspect of clinical care for the purpose of teaching or assessment.[7] We included studies published in any language that investigated the use of technology-enhanced simulation to teach police learners in comparison with another instructional modality, using outcomes of reaction (satisfaction), learning (knowledge or skills in a test setting), and behaviors (in practice).

Study identification

We conducted a comprehensive search for eligible studies as previously described using search terms for the intervention (e.g, simulator, simulation, manikin, basic life support), topic (e.g, cardiac arrest, out of hospital cardiac arrest, traffict accident, trauma), and learner (police, trafict police).[7] This search was supplemented by adding the entire reference lists from several published reviews.

Study Selection

Working independently and in duplicate, we screened all titles and abstracts for inclusion. In the event of disagreement or insufficient information in the abstract, we reviewed the full text of potential articles independently and in duplicate and resolved conflicts by consensus. Chance-adjusted interrater agreement for study inclusion, determined using intraclass correlation coefficient (ICC), was 0.69.[6]

Data Extraction

Using a detailed data abstraction form, we abstracted data independently and in duplicate

from each article, resolving conflicts. We abstracted information on the clinical topic, training location, study design, method of group assignment, outcomes, and methodological quality. We recorded data separately for learning

Outcomes of reaction (satisfaction), learning (knowledge and skills, with skills further classified as time, process, and product measures), behaviors with patients (time and process), and results (patient effects). We also

RESULT

abstracted information on monetary and time costs associated with training development and maintenance.[5]

No	Author	Title	Result
1.	Meissner, T. M., et al. (2012)	Basic life support skills of high school students before and after cardiopulmonary resuscitation training: a longitudinal investigation	Before the training, 29.5% of students performed chest compressions as compared to 99.2% post-training (P <0.05). At the four-month follow-up, 99% of students still performed correct chest compressions. The overall improvement, assessed by the BLS performance score, was also statistically significant (median of 4 and 10 pre and post-training, respectively, P < 0.05). After the training, 99.2% stated that they felt confident about performing CPR, as compared to 26.9% (P < 0.05) before the training.[8]
2.	Mundell, W. C., et al. (2013)	Simulation technology for resuscitation training: A systematic review and meta-analysis	Meta-analysis of 114 studies indicates that simulation-based resuscitation training is highly effective when comparedwithno-intervention. Evidence from 21 studies further suggests that simulation-based training may be slightly more effective than non-simulation interventions for process skills, but not for knowledge or procedural speed.[9]

No	Author	Title	Result
3.	Toubasi, S., et al. (2015)	Impact of simulation training on ordanian nurses' performance of basic life support skills: A pilot tudy	BLS simulation training sessions are associated with significant improvement in skills and performance amog Jordanian nurses. A refreshment BLS training session for nurses is highly recommended in actual CPR scenarios.[10]
4.	Cook, D. A., et al. (2012)	Comparative Effectiveness of Technology-Enhanced Simulation Versus Other Instructional Methods	In comparison with other instruction, technology-enhanced simulation is associated with small to moderate positive effects, which are satisfaction outcomes, knowledge, process measure of skills, and product measure of skills. [11]
5.	Roh, Y. S., et al. (2013)	The effects of simulation-based resuscitation training on nurses' self-efficacy and satisfaction	Self-efficacy after simulation based training was not different between computer-based and mannequin-based simulations. But learning through computer-based simulation resulted in higher satisfaction in implementing decision making and nursing skills compared to mannequin-based simulation. [12]
6.	Petrić, J., et al. (2013)	Students'and parents' attitudes toward basic life support training in primary schools	The study showed that in Croatia both students in their last two years of primary school and their parents had a positive attitude toward BLS training in primary schools. Implementing compulsory BLS training in Croatia's primary schools could help increase students' confidence, quell their fears toward applying BLS, and possibly even increase the survival of bystander-witnessed cardiac arrests. 13
7.	Li, Q., et al. (2011)	Pre-training evaluation and feedback improve medical students' skills in basic life support	In undergraduate medical students without previous BLS training, pre-training evaluation and feedback improve their performance in followed BLS training. [14]

No	Author	Title	Result
8.	Adelborg, K., et al. (2011)	Benefits and shortcomings of mandatory first aid and basic life support courses for learner drivers	Upon completion of first aid and basic life support courses, 95% or more of the respondents knew how to prioritize treatment of several casualties, knew how to relieve a foreign body airway obstruction, and knew the recommended compression-ventilation ratio during CPR. Despite significant improvements after the course only 64% knew how to diagnose cardiac arrest, 44% knew when to activate an automatic external defibrillator and 23% were aware of when to activate the emergency medical services. Respondents significantly increased their self-confidence in own skills after the course. [15]
9.	Aaberg, A. M. R., et al. (2014)	Basic life support knowledge, self-eported skills and fears in Danish high school students and effect of a single 45-min training session run by junior doctors; a prospective cohort study	Knowledge of key areas of BLS is poor among high school students. One 45-minute BLS training session including theoretical aspects and hands-on training with mannequins run by junior doctors seems to be efficient to empower the students to be first responders to Out of Hospital Cardiac Arrest (OHCA).[16]
10.	Allan, K. S., et al. (2013)	The benefits of a simplified method for CPR training of medical professionals: A randomized controlled study	A simplified 2 hours training method using audiovisual feedback combined with quantitative review of Cardiopulmonary Resuscitation performance improved CPR quality and retention of these skills.[17]

CONCLUSION

Based on the reviews mentioned above, simulation based training plays role in increasing respondents' skills and performance in conducting basic life support. Some journals also state that basic life support increases survival rate of out of hospital cardiac arrest (OHCA).

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THE USE OF GAMMA KNIFE AS LATEST INNOVATION IN NUCLEAR MEDICINE FOR TREATING ARTERIOVENOUS MALFORMATION

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ABSTRACT

Arteriovenous malformation (AVM) is a congenital disorder that usually occurs in the brain where arteries and veins are connected without capillaries causing arterial and venous blood mixed. Differences in the structure of arteries and veins that cause the pressure difference anyway. When this happens the intracranial cavity, will cause an increase in intracranial pressure will cause many complications. Need to management in the form of surgery used to treat AVM radiosurgery. The action taken is radiosurgery Gamma Knife using electromagnetic waves generated by a radio isotope 60 Co With appropriate therapy procedures, AVM can be treated and pose a lower risk than other management procedures. Methods of this study were taken from the literature review that has conducted previous research using the gamma knife for the treatment of AVM. More than 500,000 patients worldwide with vascular disorders of the brain, one of which is the AVM can be cured using Gamma Knife radiosurgery so get a better quality of life for getting help from innovation in the field of nuclear medicine in the form of Gamma Knife radiosurgery. The use of Gamma Knife highly effective and efficient treatment of AVM with a lower risk than the use of the treatment of other techniques.

Keywords: *Arteriovenous Malformation, Gamma Knife, Radiosurgery*.

INTRODUCTION

Arteriovenous malformation (AVM) are vascular abnormalities consisting of fistulous connections of arteries and veins without a normal intervening capillary bed. In other definition AVM is an abnormal connection between arteries and veins, by passing the capillary system. This vascular anomaly is widely known because of its occurrence in the central nervous system.[1]

AVM is a congenital disorder that can be contained the brain and spinal cord, formed from abnormal webbing between the arteries and veins are connected by one or more fistula. In brain, AVM can lead to the development

of serious neurological disease such as epilepsy and others.[2] AVM is not a genetic disorder that is commonly though at least in the context of a specific hereditary syndrome. AVM of the brain or brain arteriovenous malformation (BAVM) are congenital lesions developing during the late somite stages between the 4th and 8th weeks of life.[3]

Nowadays, modern medical diagnose by using the latest technologies that utilize radioactive whereabouts unknown can effectively act as supporting the patient's recovery process especially in AVM therapy. There are four methods that can be used[(4)]. Such methods include the use of X-Rays, Gamma Rays, Sound Wave, and Magneticsm. This method is growing rapidly according to the function and use. The most effective for AVM therapy is using gamma rays such as gamma knife.[4]

The accuracy of Gamma Knife surgery is able to avoid damage to the cells that are adjacent to a target irradiation, and in some cases only cause fewer side effects than with regular radiation treatments.[3]

METHOD

The research method in this paper uses the literature review by gathering data from seven journals, 1 review articles, 4 books, 3 issues and newsletters. With all these sources, the data are analyzed and interpreted to the purpose of this paper is achieved. The data included information about arterivenous malformation in the central nervous system, the components of the gamma knife, and the use of gamma knife in nuclear medicine to solve the problems of the AVM, as well as some of the reasons to choose Gamma Knife compared with many therapeutic methods.

RESULT AND DISCUSSION

Gamma Knife was created in 1968 by a Swedish Neurosurgeon, Professor Lars Leksell. The tool is a tool gamma ray therapy (radiosurgery) used for the treatment of tumors and other abnormalities in the brain without opening the skull. Gamma radiation is used in Gamma knife to destroy diseased cells while keeping the other cells that are still healthy.[1,5].

The accuracy of Gamma Knife surgery is able to avoid damage to the cells that are adjacent to a target irradiation, and in some cases only cause fewer side effects than with regular radiation treatment.[3] Since Gamma Knife is used 40 years ago, more than 500,000 patients worldwide have gained a better

quality of life for these super-sophisticated tool support. Gamma Knife has become an important tool for any leading Neurosurgery clinic in the world and so on its known as gamma-knife surgery (GKS).[3]

Gamma-knife surgery (GKS) is an safe and effective modality for treatment of patient with AVM especially the brain arteriovenous malformation (BAVM). In younger patient and BAVM at 2 years with low risk of repeated hemorrhage and has minimal morbidity can be treatened by GKS may be up to 80-85%.[6]

According to the research, the outcomes of 10 patients with different grade shows the significant result. The data shows below. There were six patient with BAVM of grade 3, and the other group is consist of grade 2 and 4. This grade is based on the spetzler-martin proposal.

Patient/sex/age (y)	Clinical manifestations	BAVM grading (Spetzler–Martin)	Location of BAVM/fistula	Type of AVF	Embolic agent(s)	Outcome	Clinical/imaging follow-up (mo)
1/M/6	Headache, dizziness	2	FP/ACA	Peripheral	Coils	Cure	67/56
2/F/44	Seizure, ICH	2	FP/ACA	Central	Coils, NBCA	Cure	52/39
3/F/27	Headache, dizziness	3	F/ACA	Central	Coils, NBCA	Cure	74/53
4/M/50	Neurologic deficit, headache	3	F/ACA	Central	Coils, NBCA	Small residual BAVM	45/35
5/M/37	Seizure, headache	4	T, BG/MCA	Peripheral	Coils	Cure	65/43
6/F/32	Seizure, headache, ICH	3	F/ACA	Central	Coils	Await the effect of GKS	23/15
7/M/53	Seizure	3	TP/MCA	Central	Coils	Cure	54/42
8/F/39	Neurologic deficit, headache, ICH	3	FP/ACA	Central	Coils	Cure	58/35
9/M/56	Headache	3	F/ACA	Central	Coils	Await the effect of GKS	26/17
10/M/25	Seizure, headache	4	T, BG/MCA	Peripheral	Failure to access	Await the effect of GKS	14/12

Table 1. Demographic and clinical outcomes in 10 patients with AVF components of BAVMs after embolization and GKS.[6]

The outcomes of BAVM after GKS through MRI were total obliteration in six patient, and the leftover of patients are still awaiting for the effect of GKS. In eight patients shows that it have complete and partial regression of the symptoms result, beside that the two patients are still in medication.[6]

In other result to shows the effectiveness of GKS is display in imaging evaluation by MIR. In this image is shows the BAVM from 27-years-old woman had grade 3 BAVM in the left frontal lobe and 6-years-old boy with grade 2 BAVM in right frontoparietal lobes.[6]

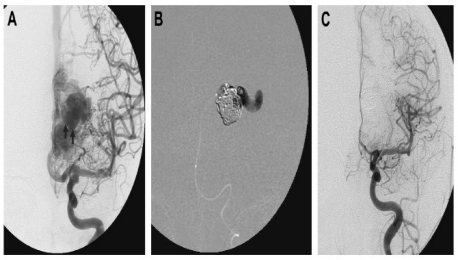

Image 1

BAVM in 27-years-old woman with grade 3.[6]

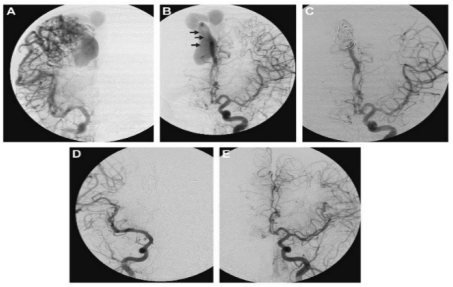

Image 2

BAVM in 6-years-old boy with grade 2.[6]

(A)Left frontal carotid angiogram depicted BAVM with a high-flow central-type AVF (arrows) in the left anterior cerebral artery. (B) The patient underwent endovascular embolization of the AVF by employing the detachable coils and NBCA mixtures, leading to total fistula occlusion (C). Follow-up carotid angiogram at 49 months after GKS showed total occlusion of the BAVM and AVF.[6] Arteriovenous fistula (AVF); Brain arteriovenous malformation (BAVM); GKS=gamma-knife surgery; NBCA=N-butyl-cynoacrylate.[6]

Bilateral frontal carotid angiograms demonstrated BAVM with an obvious high-flow AVF with dilated venous drains (arrows) (A and B). (C) Postembolization carotid angiogram revealed total fistula occlusion by employing detachable coils (D and E). Follow-up carotid angiogram at 56 months after GKS showed total obliteration of the BAVM and AVF. AVF=arteriovenous fistula; BAVM=brain arteriovenous malformation; GKS=gamma-knife surgery.[6]

At this time, embolizations of BAVM have been conducted prior to GKS that have positive effect and promising result. Previously embolized BAVM can be irregular shape which lead difficult repaired.[6] Therefore, repeat GKS was required. Furthemore, the embolization of BAVM can lead hemorrhage or ischemic stroke if the embolization carries inherent periprocedural risk.[6]

CONCLUSION

Gamma Knife is a method of treatment of brain tumors are far more than superior conventional surgery and radiotherapy methods usual. Risk during surgery and post-operative complications after surgery could be removed with the Gamma Knife. With a 0% mortality risk of post-procedure, Gamma Knife deserve strived so available to all people of Indonesia for the treatment of arteriovenous malformation.

Conflict of interest

We have no conflict of interest toward this literature review.

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PREDICTIVE FACTORS OF MORTALITY IN PATIENTS WITH ISCHEMIC STROKE IN BETHESDA HOSPITAL YOGYAKARTA

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ABSTRACT

Introduction: There are many factors that influence the cause of mortality in patients with ischemic stroke. Most of these factors are factors that can be modified either by a physician or by the patient before the stroke. By knowing these factors physicians are expected easier in evaluating the patient's initial condition, giving quick and accurate assessment of the deficit on the stroke and for society are expected in order to maintain the pattern of his life to be more healthy.

Methods: This study is a retrospective cohort study. Samples were obtained from medical records of patients at the neurology clinic at Bethesda Hospital in Yogyakarta in 2011 to 2015. Data were tested using bivariate analysis.

Results: from 206 medical records of patients with ischemic stroke, consist of 119 patients male (57.8 %) and 87 female patients (42,2%). Results of bivariate analysis showed that the variables significantly associated with mortality of ischemic stroke patients are: diabetes mellitus (RR: 1,808, 95%CI: 1,393-2,347, *p*: 0,006) , dyslipidemia (RR: 0,637, 95%CI: 0,446-0,912, *p*:0,007) , the use of ventilator (RR: 2,051, 95%CI: 1,780-2,363, *p*:0,024) , metabolic encephalopathy (RR: 1,470, 95%CI: 1,100-1,965, *p*: 0,028) and complications (RR: 1,537, 95%CI: 1,137-2,077, *p*: 0,003).

Conclusion: Diabetes mellitus, dyslipidemia, ventilator, metabolic encephalopathy and complication are associated with mortality of patients with ischemic stroke.

Keywords: ischemic stroke, the causes of mortality, predictive factors

INTRODUCTION

Ischemic stroke is an episode of neurological dysfunction which are due to focal cerebral infarction, spinal , or retina.[1] stroke patients each year are increasing. The

prevalence of stroke (with criteria diagnosed by health personnel) from 8.3 per 1000 in Riskesdas 2007 increased to 12.1 per 100 in (for the stroke of respondents 15 years and above).[2] From Stroke Registry of Bethesda Hospital in 2014 , it s show that each 3 month, stroke patients were increased. From 2460 stroke patients in Bethesda hospital in 2011-2013, 9.47 % of patients are deceased.[3]

There are many factors which influenced the cause of mortality of Ischemic Stroke Patients. Previous research states that hypertension, smoking and alcohol consumption is not related to the cause of death of stroke.[4] Another research declares that the younger age group and hypertension are associated with lower mortality in patient with acute ischemic stroke and intracerebral hemorrhage.[5] Other research show that Treatment Immediately of antihypertensive drugs after acute ischemic attack can increasing stroke mortality.[6] Other research stated that patient with history of diabetes, muscle strength and decrease in level of consciousness is a predictor factor affecting the mortality rate of stroke patients.[7] Predictors factor of mortality in patient with ischemic stroke still varied and still be debate. The purpose of this research is to review determine factors affecting the predictors of mortality in ischemic stroke patients.

METHODS

This study is a retrospective cohort study that uses secondary data, patient medical record from 2011 until 2015 period. The research sample is ischemic stroke patients at Bethesda Hospital in Yogyakarta were diagnosed with CT (computed tomography) scan of the head and not the discharge against medical advice. The independent variables were predictors of mortality factors and the dependent variable was the clinical outcome of patients with ischemic stroke. The data is processed through descriptive analysis through bivariate analysis.

RESULT

Based on the results of the bivariate analysis in Table 1 it showed that the variables which have a relationship with mortality of patients with ischemic stroke were diabetes mellitus , dyslipidemia , ventilator, metabolic encephalopathy and complications.

Table 1. Bivariate Analysis of Independent Variables and Dependents Variable

Variable	Survivors (n=103)	Deceased (n=103)	RR	95% CI	p-value
Sex					
Male	60 (50,4%)	59 (49,6%)	1,020	0,773-1,346	0,888
Female	43 (49,4%)	44 (50,6%)			
Systolic blood pressure (mmHg)					
High (>120)	81 (47,9%)	88 (52,1%)	0,806	0,591- 1,097	0,204
Normal (≤120)	22 (59,9%)	15 (40,5%)			
Diastolic blood pressure (mmHg)					
High (>80)	59 (46,1%)	69 (53,9%)	0,817	0,623-1,070	0,151
Normal (≤80)	44 (56,4%)	34 (43,6%)			
Random blood pressure (mg/dL)					
High (≥200)	32 (60,4%)	21 (39,6%)	1,301	0,986-1,715	0,080
Normal (<200)	71 (46,4%)	82 (53,6%)			
Total leukocytes *(x10⁶/L)*			1,289	0,986- 1,689	0,067
High (>11)	51 (57,3%)	38 (42,7%)			
Normal (≤11)	52 (44.4%)	65 (55,6%)			
Hipertension					
Yes	51 (47,7%)	56 (52,3%)	0,907	0,690-1,192	0,486
No	52 (52,5%)	47 (47,5%)			
Diabetes Melitus					
Yes	12 (85,7%)	2 (14,2%)	1,808	1,393-2,347	0,006
No	91 (47,4%)	101 (52,6%)			
Atrial Fibrilation					
Yes	4 (44,4%)	5 (55,6%)	0,898	0,426-1,890	0,733
No	99 (50,3%)	98 (49,7%)			
Dyslipidemia					
Yes	23 (35,9%)	41 (64,1%)	0,637	0,446-0,912	0,007
No	80(56,3%)	62 (43,7%)			
Smoking					
Yes	4 (33,3%)	8 (66,7%)	0,653	0,290-1,471	0,234
No	99 ((51,0%)	95 (49,0%)			
Ventilator					
Yes	5 (100%)	0 (0%)	2,051	1,780-2,363	0,024
No	98 (48,8%)	103 (51,2%)			

Metabolic Encephalopathy					
Yes	20 (69,0%)	9 (31,0%)	1,470	1,100-1,965	0,028
No	(83 (46,9%)	94 (53,1%)			
Complication					
Yes	68 (59,1%)	47(40,9%)	1,537	1,137-2,077	0,003
No	35 (38,5%)	56(61,5%)			

DISCUSSION

Results of the analysis showed there are factors related to the mortality of patients with ischemic stroke , so that the results are consistent with the hypothesis (diabetes mellitus (RR: 1,808, 95%CI: 1,393-2,347, *p*: 0,006), dyslipidemia (RR: 0,637, 95%CI: 0,446-0,912, *p*:0,007) , the use of ventilator (RR: 2,051, 95%CI: 1,780-2,363, *p*:0,024), metabolic encephalopathy (RR: 1,470, 95%CI: 1,100-1,965, *p*: 0,028) and complications (RR: 1,537, 95%CI: 1,137-2,077, *p*: 0,003).

Hyperglycemia persistent with glucose levels ≥155 mg/dL are associated with poor outcomes and higher mortality in ischemic stroke patients.[8] In experimental studies focal ischemia is known that hyperglycemia can increase infarct size and reduce therapeutic window of recanalization.[9] Diabetes mellitus type II contributed to the abnormalities of plasma proteins involved in the activation of platelets and blood clotting.[10] Disorders that can occur in people with diabetes mellitus is endothelial damage due to blood vessel inflammation (increased interleukin (IL)-6 , tumor necrosis factor (TNF) and TNF - α) and stress oxidative increases reactive oxygen, oxidation of lipoproteins, impaired platelet aggregation, inhibition of fibrinolysis and hypercoagulability.[11]

High level of triglyceride and low-density lipoprotein (LDL) with low level of high -density lipoprotein (HDL) Low score are associated with high score of modified Rankin Scale (mRS), a severe stroke and poor clinical outcomes.[12] Hypertriglyceride effect on thrombosis through thrombogenic alterations in coagulation system and increasing the viscosity of plasma may contribute to cerebrovascular disease.[13] Complications that occur after a stroke can affect the prognosis of patients . Gastrointestinal bleeding after stroke onset can cause hemoglobin levels go down, neurological damage, poor

functional outcome and mortality.[14] Urinary tract infections that occur during hospitalization of stroke patients had a distinctive impact that the length of hospitalization, increased maintenance costs, exposure to antibiotics intarvena, the risk of having bacteremia, a decrease in neurologic status, poor clinical outcomes and increased risk of death.[15]

CONCLUSION

There are factors that have a relationship with the mortality of patients with ischemic stroke namely diabetes mellitus, dyslipidemia, ventilator use, metabolic encephalopathy and complications.

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DEVELOPMENT OF HUMAN ENDOTHELIAL CELL CULTURE METHOD (HUMAN UMBILICAL VEIN ENDOTHELIAL CELLS) FOR RESEARCH ANTI-AGING CARDIOVASCULAR

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ABSTRACT

Introduction: The morbidity and mortality of cardiovascular disease in Indonesia is high. Endothelial cells play a key role in the pathogenesis of cardiovascular disease. Endothelial role as regulator in hemostasis and coagulation, vasomotor regulation, angiogenesis and permeability. Until now, the development of culture methods of human umbilical vein endothelial cells (HUVEC) are based on foreign protocols that are often not in accordance with the laboratory conditions in Indonesia. Need to develop a model of HUVEC cell cultures for testing the mechanism of pathogenesis of cardiovascular disease in cultured endothelial cells. Culture of human endothelial cells from umbilical vein would be presented as a model of anti-aging research cardiovascular endothelial cells in vitro.

Method: The study was conducted with the design of experiments in vitro on HUVEC cells. Vein endothelial cells isolated from the placenta. Activity in the research is to pilot plant umbilical vein endothelial cells (HUVEC) and standardize the incubation time and the concentration of collagenase as a determinant factor of growth and cell morphology. Incubation time and the concentration of collagenase is able to foster the culture of HUVEC best set as a standard method in HUVEC culture protocol.

Results: The study has gained HUVEC culture method in accordance with the conditions Gadjah Mada University LPPT laboratory. Growth and development of collagenase HUVEC optimal incubation takes 1hour. Collagenase concentration which produces optimal HUVEC culture is 12 micrograms / ml.

Conclusion: The study has been able to determine the incubation time and the concentration of collagenase that produces optimal HUVEC culture

Keywords: HUVEC; incubation time; concentration of collagenase

INTRODUCTION

Morbidity and mortality of cardiovascular disease is high. Cardiovascular disease is the leading cause of death in the group of degenerative diseases. The disease is expected to increase. Endothelial cells play an important role in the process of cardiovascular disease due to the degenerative process. Prevention or control of cardiovascular disease and its complications requires understanding the mechanisms of the pathogenesis of cardiovascular disease and its complications in bio molecular, both in vivo and in vitro. Endothelium is a component of the natural defense system that interacts directly with the reactive radicals in the body. As the wrapping layer organ in the body, the endothelium has a system of antioxidants as a natural defense system against reactive radicals. Endogenous antioxidant system involves a transcription factor Nrf2, iNOS as enzymes and TLR-4 receptors (Cines et al., 1998). Exposure on endothelial reactive radicals leads to atherogenesis by involving platelets and various inflammatory and proinflammatory cytokine modulators.

Human umbilical vein endothelial cells (HUVEC) proposed as a model of endothelial cells in vitro. Atherogenesis mechanism is still unclear needs studied more seriously to get a new drug discovery as a deterrent and to tackle atherosclerosis. Model maternal placental cells are as more profitable, compared with endothelial cells from human aorta, aortic cows and mice. The reason is placental of human origin, and easily obtained, and a larger lumen size. Since culture HUVEC successfully carried out in the early 70s, up to the 21st century had been published about 50,000 publications associated with endothelial cells (Nachman & Jaffe, 2004).

Exposure reactive radicals in the blood vessels activate endothelial cells in the process of atherogenesis through the mechanism of chronic inflammation induced by oxidative stress. Antioxidant and immunomodulatory agent may inhibit the pathogenesis of cardiovascular disease. Consumption of herbal antioxidants

including nigella sativa seed oil are expected to inhibit the degenerative processes in the body. As immunomodulators, bioactive nigella sativa has broad activity. Bioactive nigella sativa can reduce inflammatory reactions in response to asthma and toxic materials (Plumbum) (Massadeh et al., 2007), decrease the secretion of histamine by mast cells (El-Dakhakhny et al., 2000), increasing the phagocytic activity of macrophages in vivo in hamsters induced by streptozotocin (Fararh et al., 2004). Timokuinon proven to reduce reaction inflammatory on bronchus of mice, lowered Ig E and specific Ig G-OVA, lowering IL-5, IL-4, IL-13 and increase IFN γ in mice induced by ovalbumin (El-Gazzar et al., 2006), whereas the same researchers have studied about the effect of yawning nigella sativa oil can decrease leukotriene (El-Gazzar et al., 2006). The main active substance does not evaporate MBJH oil is unsaturated fatty oil being linoleic and linolenic. The main active substance is oil evaporate timokuinon, nigelon and nigelin (Farrah et al, 2004; Nickavar et.al., 2003). Unsaturated fatty acids and is a powerful antioxidant timokuinon (El sayed & Fukuhima, 2003; Mousa et al, 2004; Randhawa et al., 2002).

Thus the need to develop a test model in vitro endothelial cells for testing the mechanism of atherogenesis and nigella sativa, which is expected to provide enough specific information about the function and endothelial response. Culture of human endothelial cells from human umbilical vein or umbilical vein endothelial cells (HUVEC) will be proposed as a model of endothelial cells in vitro (Randhawa et al., 2002).

METHODS

1. Collection of cord

   Collection of umbilical cord is to obtain pieces of cord that can be used for the isolation of endothelial cells. Terms cord for culturing endothelial cells (HUVEC) are in good condition (not broken) and long enough (at least 15 cm).

2. Isolation of endothelial cells

   Isolation of endothelial cells aims to remove the endothelial cells from umbilical vein wall enzymatically in sufficient quantity and condition in life. With the isolation so that it can be done planting a primary culture of human umbilical vein endothelial cell culture (HUVEC). Implementation is done aseptically in a biosafety

cabinet (BSC) level 2 with enzymatic method.

Materials needed are the umbilical cord in the buffer solution and antibiotics, medium complete DMEM glucose, FBS aliquot (FBS A) qualified, Pen Strep aliquot, Fungizone aliquot, PBS Pen Strep 1%, Collagenase sterile (7 mg / 7ml sterile PBS (without Ca Cl and Mg Cl), Glutamine 100x, Alcohol 70%.

Tools needed are incubators, BSC level2, centrifugation, hot plate, clamps, tweezers network, needle cannula, syringes 10cc, glass bottle, funnel glass, tube conical 15cc, sterile gauze, a petri dish of glass, flask, blue yellow white tips, micropipette,

The procedure of research carried on in the research laboratory LPPT UGM.

a. Umbilicus cleared from the network and the rest of the blood with sterile gauze moistened with 70% alcohol. Each end of the umbilicus is cut transversely so that the visible presence of two arteries and veins, characteristic veins has thicker walls, large and elastic.
b. A cannula is inserted into the end of the vein approximately 1 cm, then tied tightly with string. Veins washed with FBS A through a cannula that is preinstalled with using a 20cc syringe. The foregoing is done 2-3 times. Once clean, the end of the umbilicus using either tied with strong ties and clamped.
c. Collagenase solution is inserted, while the syringe should be left attached to the cannula. Umbilicus subsequently warmed to room temperature for 60 minutes or incubated.
d. Collagenase solution containing endothelial then removed from the umbilicus a way to suck through a syringe mounted on the tip of the cannula. Collagenase is then inserted into a 18cc sterile centrifuge tube. Umbilicus 8cc rinsed with a solution of FBS A to rinse the remaining endothelial cells, for subsequent inclusion in a centrifuge tube, which already contains collagenase solution. The solution containing the endothelial centrifuged at a speed of 1000rpm for 8 minutes,

in order to obtain pellets that contain endothelial cells. The supernatant was discarded, then added 4 ml culture medium in pellet and suspended with pipetting methods are used so that the endothelial cells can be separated. The solution was transferred into a flash 25cm² that has been coated with 0.2% gelatin solution, then flash is inserted into the 5% CO2 incubator at a temperature of 37^{0}C for 30 minutes.

3. Culture of endothelial cells, stimulation and activation of endothelial cells.

   The purpose of this phase is to grow endothelial cells that have been isolated in the growing medium. Preparations containing flash endothelial cells then added to the culture medium (RPMI + 20% FBS), and incubated for 3-4 days until confluent endothelial cell cultures and monolayer.

4. Standardization of incubation time and concentrations of collagenase.

   The goal of getting collagenase incubation time that is able to produce best cell culture result.

RESULTS AND DISCUSSION

1. The result of the isolation and culture of HUVEC early

   HUVEC culture results are shown in Figure 1.

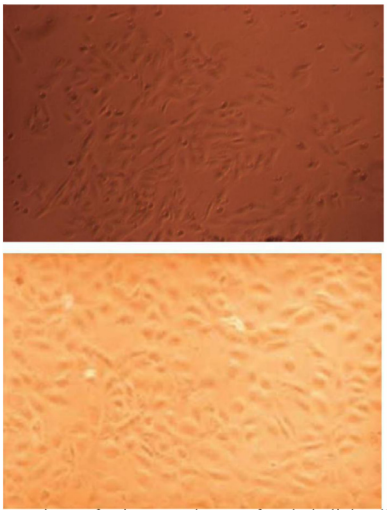

Figure 1. Comparison of primary cultures of endothelial cells (HUVEC) day 4 (A) and the cultures HUVEC day 0 (Khodarev et al., 2002) (B). 40x magnification.

The survey results revealed that HUVEC can grow well. The results showed the cell culture HUVEC grow quickly shown on the fourth day changes in cell morphology, migration and elongation in cultured cells. Cell culture results also showed only slightly contaminants. On day six of culture seemed to confluent and ready to be harvested for use in vitro testing.

2. The results of HUVEC culture with the incubation time variations collagenase

The results of cell culture based on the time of induction of collagenase are presented in Figure 2.

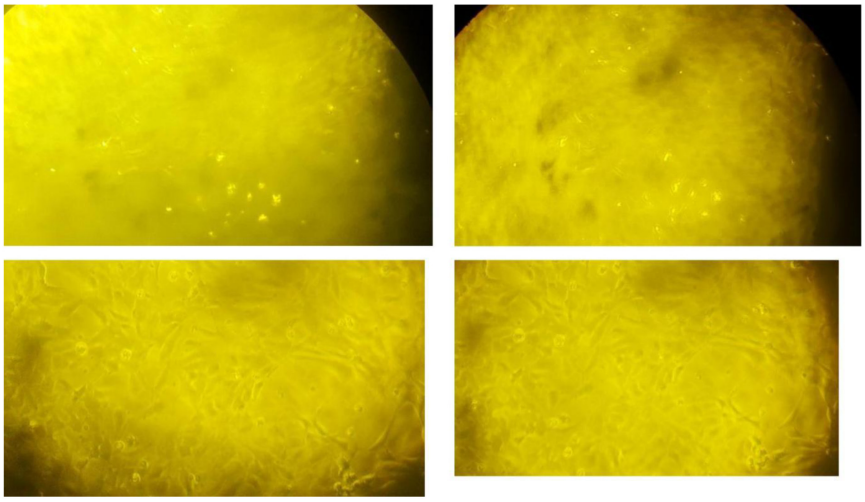

Figure 2. Pictures Photo HUVEC cell culture according to incubation time variations collagenase

Based on the results of standardization known that the incubation time long incubation collagenase are on the best culture results are for 60 minutes. The concentration of collagenase, which provides optimal culture results, was 0.5%. The results provide a new protocol modify protocol of HUVEC.com. Which according huvec.com collagenase incubation only performed for 10 minutes. The results showed that incubation of collagenase for 10 minutes yet provide optimal culture results as presented in Figure 2 c and d. This research result is slightly different from previous studies (Galley & Webster, 2004).

CONCLUSION

The results showed that it had been successfully cultured HUVEC with the addition of collagenase incubation time and concentration. HUVEC culture can be used to test antiaging.

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MEDICATION-RELATED PROBLEMS IN PATIENTS WITH TYPE 2 DIABETES: A QUALITATIVE INTERVIEWS

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ABSTRACT

Background: The general categories of medication-related problems (MRPs) were drug therapy needed, dose too low, drug monitoring needed, suboptimal drug, dose to high, adverse drug event present, more affordable alternative available, suboptimal regimen, and nonadherence. The aim of the overall study was to create a module for improving the quality of counseling practice in pharmacies. The research question directed in this sub-study was to describe medication-related problems (MRPs) in terms of type in people with type 2 diabetes, as the problems were identified through medication reviews and semi-structured interviews.

Method: Observational study with cross-sectional design. The survey contained open-ended and close-ended questions about patient's drug-related experiences, diabetes knowledge, perceptions, problems and actions. Data were collected from thirteen patients with type 2 diabetes attending internal clinic at Sleman General Hospital.

Results: A medication review was filled out by qualitative interviews with the patients. "Nonadherence" (such as limited knowledge of the disease, unwillingness to adapt lifestyle, lack of information) and the subcategory "other problems" (such as emotional well-being) were the two most common MRP primary domains identified in all patients.

Conclusions: Pharmacist needs to notice this type of MRP in type 2 diabetes when dispensing medicines to and advising patients, especially when explaining how to manage psychosocial issues.

Keywords: Type 2 diabetes; Medication-related problems; Qualitative interview; Patient interview, Pharmacist

INTRODUCTION

Type 2 diabetes is one of the non-contagious chronic diseases, characterized by the increased level of blood glucose. Epidemiological studies indicate a trend toward the

increasing incidence and prevalence of type 2 diabetes throughout the world. Data on the Diabetes Atlas 2011 released by the International Diabetes Federation (IDF) mentioned that there were 366 million people (2011) in the world who suffered from diabetes and it was expected to rise to 552 million people (2030). IDF estimated that the Indonesian population of 20-79 years old with diabetes in 2030 will rise from 7.3 to 11.8 million people (2011).[1] According to the results of the Basic Health Research, the prevalence of diabetes in Indonesia increased from 1.1% (2007) to 2.1% (2013). The Special Region of Yogyakarta is the province with the highest prevalence of diabetes (2.6%) based on the diagnosis by doctors in Indonesia.[2] Based on demographic characteristics, patients with diabetes in Indonesia are largely 55–64 years old, female, with high education, live in urban areas, entrepreneurs, and have a high ownership index quintile.[2]

The treatment of type 2 diabetes is not only used oral antidiabetic drugs or insulin, but also non-pharmacological therapy by self-regulating diet and exercise. The use of drugs or insulin is not necessarily able to control blood glucose level well. If therapeutic target is not met, it is necessary to observe therapeutic problems in a comprehensive disease management.

This study explored the problems associated with medicine use through interview. We assume that interviewing patients was an appropriate method to collect data used to complement treatment review with respect to the identification of DRPs. The patients can describe and elaborate any experiences related to drugs and disease, thoughts, actions, and their problems.[3] Several other studies identify DRPs retrospectively.[4,5] The objective of the study is to identify the type of drug-related problems among outpatients with type 2 diabetes.

MATERIALS AND METHODS

Research design

The diabetes medication-related problems were known from literatures. Theories on the self-regulation of drugs,[6] coping with illness,[2,3] user perspectives,[9] and drug-related problems[10] were used as a framework to develop interview guide. Data were collected by a semi-structured interview. The interview guide were designed to get a deep insight of the patients' perception associated with experiences, considerations, actions, and problems related to the treatment and their disease. The

study was conducted in the Sleman General Hospital. It is located in regency with most population in Yogyakarta. Patients were recruited by a purposive sampling technique. Although there was no standard guideline to determine an appropriate sample size for interviews in a qualitative study, there was a general agreement that the sample size should be determined when the saturation of data was theoretically reached.[11]

Recruitment of participant

The inclusion criteria were patients were diagnosed with type 2 diabetes, prescribed with oral antidiabetic or insulin or both, and agreed an informed consent. Senile patients were excluded from the study. Initial screening of patients was based on communication capability. 14 patients signed the informed consent, 12 of which completed the interview and 8 of which were successfully examined for HbA1c.

Data collection

Primary data were collected through semi-structured interviews and the examination of HbA1c as an indicator of glycemic control. Secondary data, such as the results of the examination of blood pressure and lipid levels, were tracked from medical records. The interviews were done when the patients were waiting for examination in polyclinic. If an interview was not completed, it was continued when they were waiting for a prescription service or in other times by appointment. The examination of HbA1c was conducted by cooperating with Prodia Clinical Laboratory. The method used was the ion exchange of HPLC that was standardized by the National Glycohemoglobin Standardization Program (NGSP). The financial incentive of IDR25,000,00 was given to each participants.

The data were collected by the first author, RW. 14 patients were recruited to involve in the interviews and signed the informed consent. Two patients were drop out from the study due to rush home after handing over the prescription and did not contact the researcher for the interview's appointment. Four participants refused taking for HbA1c testing.

Qualitative interview with patients

The interview guideline were used to frame the interview's topic. Combined open-ended and close-ended questions were applied to focus on the domains of knowledge on drugs and disease, medication adherence, social support,

psychological conditions, use of alcohol and tobacco, as well as the symptoms of other diseases. The interviews were conducted in the hospital while the participants visited their doctor, followed up when they were waiting for drug preparation, or selecting any times not distressing for the patients. One hour was allocated for an interview, but the duration of interview was dependent on the depth of each patient's response, in average of 25 to 40 minutes, not including the process to get the informed consent. The interview guideline describes in Table 3.

Before signing the consent, all participants were informed about the objective of the study as well as the risks and benefits of participation. They were informed about recording during interview and reminded that the participation was voluntary. Additional pre-interview procedures applied included notification about personal discretion and no relationship of interview and medical treatment in the next medication. Furthermore, the patients were asked to answer some questions based on all medications received.

Detection of Drug-related Problems (DRPs)

An evidence-based checklist [10] was used to identify the issues in the treatment and management of patients with type 2 diabetes systematically. In addition, the checklist aimed to intervene early as the effort to increase patients' metabolic control. The content of the checklist consisted of six parts: lifestyle management, glycemic control, blood pressure control, lipid control, platelet control, and medication adherence.

Data analysis

The characteristics of participants were analyzed by a descriptive technique. Data for numeric variables were presented in mean and standard deviation, while those for categorical variables were presented in frequency and percentage. The interviews were recorded with patient's permission, being then transcribed in verbatim or by words of investigator. Both primary data from the interviews and secondary data were analyzed based on the checklist and the conclusion was drawn based on data of drug-related problems potentially faced by the patients.

Ethical approval

The study was approved by the Medical and Health Research Ethics Committee (MHREC) Faculty of Medicine Gadjah Mada University

– Dr. Sardjito Public Hospital (Ref: KE/FK/576/EC/2015).

RESULTS

Sample description

A total of 14 patients with type 2 diabetes agreed to participate in the study, 13 were interviewed. The minimum and maximum age of participants was 35.4 and 67 years old, respectively, being mostly distributed at 51 – 60 years old. The demographic and clinical characteristics of participant were shown in Tables 1 and 2. Type 2 diabetes was more common among female patients.

Drug-related problems (DRPs)

The results of identification based on the checklist show that 21% of the patients used oral antidiabetic agents and insulin, while 67% had poor glycemic control (HbA1c > 7.0%). The number of patients with either oral antidiabetic drug or insulin was the same, i.e. 36% of the total participants. Blood glucose was controlled well in 40% of the patients who used oral antidiabetic agents only and in 20% of those who used insulin only.

Interviews with patients

Several different themes were identified. The majority of participants informed that they were adhere to medication prescribed. The participants newly diagnosed stated that they did not know much about the disease and how to manage it. Those who have long suffered from diabetes recognized that they have been able to control their lifestyle, including diet, exercise, and smoking. Some patients were less successful in treating the diabetes, particularly due to stress.

Knowledge and behavior associated with diabetes and medicines

In general, the patients recognized the function, dosage, frequency, time and method of medicine use, and storage of drugs but most did not understand the name, side effects, the ways to handle adverse effects, and drug interactions. Some patients taken herbal medicines in decoction to help lower blood glucose, but most did not believe that the traditional medicines can lower blood glucose significantly compared with either insulin or oral antidiabetic agents.

Medication adherence

The patients were averagely adhere to take drugs or inject their insulin. There was patient who has ever tried to stop taking the drugs because she felt his condition improved. During withdrawal, she did not control the diet and even

exercise. This made her realizes that diabetes cannot be cured but can be controlled, among others, by taking the drugs. Such experience was valuable for the patients. The desire to cure was motivation for most patients to always take the drugs or inject insulin despite boredom. They believed that the medicines taken given benefits more than the risk of adverse effects. They assumed that those prescribed by physicians have actually passed through various stages of both pharmacological and toxicological assays, making them safe to use.

Knowledge on disease

Only few patients claimed that they suffered diabetes due to hereditary factor, but most reported that it was due to unhealthy lifestyle, i.e. uncontrolled diet and lack of physical activity, triggering the diabetes suffered. Related to this, they are currently practicing a healthy lifestyle by regulating diet and trying to be more active in their daily lives. In general, patients have learned that discipline in managing lifestyle and using medication can improve their glycemic control. They considered that physical activity was effective to do so, among others, by gardening in the afternoon every day, playing badminton, cycling, walking, and gymnastics, particularly those with physical ability. However, some patients were not able to do such extra physical activities due to non-conducive health condition. Reasons such as no time for exercise and laziness were particular consents for the researchers.

In comparison, HbA1c in active and inactive patients were different, although the results of assessment indicate that the level of knowledge about disease and its drugs as well as medication adherence were similarly good. In addition to lack of discipline in exercise and diet, the triggers of uncontrolled blood glucose were the psychological factors. It is reported that excessive job stress and anxiety can increase blood glucose level. The more severe the responsibility, the higher the stress underwent by patients. Most patients stated that they could not overcome it. In interacting with the family, all patients expressed satisfaction over the support and understanding given by the family. There were some patients who embarrassed if others knew that they were taking a lot of drugs. They were also afraid of the judgment by others about the severity of disease suffered. However, there were also the patients who are not burdened when injecting insulin before their relatives and friends. The patients' awareness to visit the doctor regularly and perform blood test

was very high, because the desire to be healthy was so great. There was a patient reporting that when at a pilgrimage, he never injected insulin but the blood glucose was well-regulated. This is interesting for the investigator to ask more questions. When in the holy land, his physical activities increased with the routines of worship performed. In view of foods consumed, the menu was controlled for meeting nutritional balance. It can be said that his psychological condition was stable because he performed more spiritual activities, so that he was away from the depressed feeling. Arriving home, he was busy in work, had uncontrolled diet, and no time to exercise, thus causing his blood glucose uncontrolled despite adherence to the injection of insulin.

The patient's knowledge about diabetes was closely related to stress management. The more the knowledge about diabetes, the higher the understanding on stress management in terms of controlling the blood glucose. Stress can increase their blood glucose level. The results of interview showed that those who work outside their home had a higher level of stress than those who were retired or did not work outside their home. Their comprehensive knowledge and experience in diabetes was very influential in stress management. Education will lead to the effect on patients' mindset and perception on the disease management. Meanwhile, education level determines whether or not a patient is easy to understand the knowledge he or she acquired. The patients' time-consuming works will affect them to think a way to control their stress. Meanwhile, those who did not work or were retired had more time to think how to cope with stress.

The support of family or friends can also help reduce stress. By the support, they can more regularly take medicine and manage their lifestyle. Most patients stated that the role of family member to remind and pay attention made them motivated to live a healthy lifestyle and minimize the psychological stress. They know that complications can arise if their blood glucose levels were uncontrolled, and some suffered the diabetes disease and its complication. Thus, it is necessary to build a strategy to prevent the disease from being complicated.

Daily life with diabetes

The daily life lived by the patients with diabetes was as usual as those without diabetes in doing their activities. A housewife

that must cook for meeting the family's need can still prepare a specific menu for herself that is different from that for other family members. Almost all the patients did not perform a blood glucose test at home, but some perform a trial and error by themselves to find out the effect of a food on the increase of blood glucose. The results of the trial and error were used to regulate a safe portion of diet. A number of patients experienced some symptoms that led to the side effect of drugs or symptoms of other diseases, but based on their reports, the symptoms did not disturbed and only appeared occasionally. The symptoms lost when commercially available drugs or drugs prescribed by doctors were given. The patients of productive age asserted that there was no problem in their sexual lives, but some reported their sexual ability decreased.

CONCLUSION

The problems due to the target of type 2 diabetes treatment could not be met were not only caused by the inappropriate use of drugs, but also by patients' lifestyle and psychosocial problems.

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Table 1. The demographic characteristics of participants in the study

	Mean	*SD*
Age (years) (*N* = 13)	55.3	10.8
	N	%
Gender		
Male	4	24
Female	9	64
Absent	1	7
Stay		
Alone	0	-
With someone	13	93
Absent	1	7
Recent education		
Elementary school	2	15
Intermediate junior-senior high schools	8	57
Diploma	1	7
Bachelor	2	14
Postgraduate	0	0
Doctor/medical/legal	0	0
Absent	1	7
Job status		
Retired	3	21
Working	5	36
Not working	5	36
Absent	1	7

Table 2. Diabetes history of participants

	Mean	*SD*
Years of their diagnosis (*N* = 13)	9.7	9.4
HbA1c (*N* = 8)	8.3	2
IMT (kg/m^2) (*N* = 11)	25.4	4.6
	N	%
Current management		
No drug	0	-
Oral antidiabetic drugs only	5	36
Insulin only	5	36
Insulin and oral antidiabetic drugs	3	21
Absent	1	7
Blood glucose monitoring at home		
Yes	1	7
No	12	86
Absent	1	7
Self-reported history of complication and co-morbid of diabetes		
High blood pressure	11	84.6
Chronic heart disease	1	7.7
Neuropathy	1	7.7
High cholesterol	7	53.8
Eye problems	1	7.7
Kidney problems	1	7.7
Foot problems	1	7.7

Table 3. The content of interview guides

Main topic	Sub-topic
Knowledge and behavior in relation to diabetes and drugs	The use of drugs (drug name, dosage, knowledge of the effect of the drug), perception of the use of drugs, side effects (experience, consent about adverse effects, speculation about side effects), other problems with medicine use, self-regulation of medicine use (rational reasons, explanation)
Perception of disease and health as well as strategy	Causes of diabetes (smoking, lack of physical activity, food, genetics, and others), strategies to stay healthy, the ways to deal with disease and drug-related problems, the current state of health, social interaction
Daily life with diabetes and drugs	Course of the disease, symptoms, living with diabetes (family, work, daily life)

ANALYSIS OF CALCIUM CHANNEL BLOCKER USAGE FOR INFARCT MYOCARDIUM TREATMENT: A REVIEW ARTICLE

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ABSTRACT

Hypertension brought a higher risk of acute myocardial infarction. Reducing myocardial oxygen demand may decrease blood pressure and prevent ischemia or adverse cardiovascular events in coronary heart disease (CHD). Beta blocker was known to be the first line therapy for hypertensive with CHD, but its benefit and tolerability for elderly still uncertain. Calcium channel blocker (CCBs) has a good effect for a hypertensive patient with CHD and arrhythmia. Hence, we analyzed CCBs usage in managing a hypertensive patient with myocardial infarction treatment. We conducted literature study from journals and textbooks. Search engines that we used were Pubmed, google scholar, Clinicalkey, Sciencedirect, British Medical Journal (BMJ), and Journal of American College of Cardiology (JACC). Search terms were "calcium channel blocker", "calcium antagonist", "myocardial infarction", "infarct myocardium", and "management infarct". Data analysis was explained by narrative-descriptive. The result showed CCBs are generally safe and give better result in lowering blood pressure and reduction myocardial infarction risk. However, CCBs are not typically used for initial treatment. In conclusion, CCBs usually are used for some medical conditions, such angina, heart rhythm abnormalities, or as a second drug for hypertension alone.

Keywords: *calcium channel blocker, calcium antagonist, treatment infarct myocardium*

INTRODUCTION

Myocardial infarction (MI) is one of the most dangerous cardiovascular problems in these current time. MI can be together with high blood pressure condition or hypertension. It takes an important role for developing complications in cardiovascular disease, especially in myocardial infarction case. Various studies stated that hypertension's coexistence in coronary heart disease (CHD) showed a negative impact on patient's clinical manifestation and prognosis. Hypertension brought a higher risk

of acute myocardial infarction.[1] In 2007, The American Heart Association Scientific Statement recommended lower blood pressure treatment goal of less than 130/80 mmHg for a patient with coronary artery disease, stable or unstable angina, and STEMI or NSTEMI.[2]

There are many drugs for lowering blood pressure, such as diuretic, ACE inhibitors, angiotensin receptor blocker (ARBs), beta blockers, calcium channel blocker (CCBs), and so on. Beta blocker was well-known to be effective in hypertension treatment. Along with diuretics, it became the first line drugs for hypertensive CHD patients. [3] However, there was uncertainty about beta blocker benefit and tolerability for elderly patients with CHD.[4] Another antihypertensive agent, CCBs, was thought to be as, even more effective than beta blocker and diuretic[3]. In addition, CCBs are effective in controlling certain heart rhythm problems, such as supraventricular tachycardia.[5] Many guideline committees take into consideration that CCBs are a potential option for hypertension first line therapy.[6] Thus, this study was conducted to analyze CCBs usage for myocardial infarction. Our objective is to acknowledge that is CCB really safe and can be used for any condition in patients with CHD.

MATERIAL AND METHODS

A literature review was identified by books and journals online (Pubmed, google scholar, Clinicalkey, Sciencedirect, British Medical Journal, and Journal of American College of Cardiology). Identified the keywords such as "calcium channel blocker", "calcium antagonist", "myocardial infarction", "infarct myocardium", and "management infarct". Analyzed baseline Ca Channel Blocker in relation to Infarct Myocard treatment. The data was included from 2011 through 2016. Then reading the abstracts to review the full texts were in cases which were not sufficient. We found 1.010 full-text articles in a first search based on the related keywords. Then considering inclusion and exclusion criteria in the title and abstract review, about 537 qualified studies were selected, only 11 studies which authors choose.

RESULT AND DISCUSSION

Calcium channel blockers (CCB) are drugs that work by inhibiting the influx of extracellular calcium flow into the cells through specific ion channels throughout the cell wall. Generally CCBs can be divided into two broad categories, namely dihydropyridine (nifedipine, nikarpidin, amiodipin) and non-dihydropyridine (diltiazem

and verapamil). These two groups of drugs work by binding different sites on L-type calcium channel. When calcium influx is blocked, vascular smooth muscle becoming recess thus vasodilation happened and decreasing blood pressure.[6] The pharmacokinetic of CCBs drug is presented in Figure 1.

Dihydropyridine (DHP) has vasodilatory effects stronger than non-dihydropyridine. It has shorter half-lives which led reflex tachycardia. The non-DHP group work by improving survival and reducing infarct. This class also led to reduced heart rate and reduced afterload. Both groups selectively work towards the canal Ca^{2+} by 90-100%. Meanwhile, another group that is difenilpiperazin (cinnarizine, flunarizine) and diarilaminopropilamin either (bepridil) work by blocking the canal Ca^{2+} 50-70%.[6,7,8]

TABLE. Calcium Channel Blockers

Name	Half-life, h	Dose, mg·times per day	Affect LV function	Market, %	Approved by FDA for	Major Outcome-Based Hypertension Trials
Verapamil (Calan, Isoptin) (Calan SR, Isoptin SR, Verelan) (Covera-HS, Verelan-PM)	6–8 12–24 24	80–120·2 80–480·1 180–300·1	–	~8%	Hypertension, angina, atrial dysrhythmias	VHAS, CONVINCE,[10] INVEST[11]
Diltiazem (Cardizem) (Cardizem SR, Tiazac, others) (Cardizem CD, Cartia XT, others) (Cardizem LA)	6–8 8–12 18–24 24	30–90·3 120–180·2 120–480·1 120–540·1	–	~11%	Hypertension, angina, atrial dysrhythmias	NORDIL[12]
Nifedipine (Procardia, Adalat) (Procardia XL, Adalat CC)	0.2–1 2 (?) 24	10–20·3–4 30–120·1	–	~5%	Hypertension, angina	INSIGHT,[13] STONE
Nicardipine (Cardene) (Cardene SR)	6–8 8–12	20–30·3 30–60·2	–	Small	Hypertension	NICS-EH
Isradipine (DynaCirc) (DynaCirc CR)	8–12 12–18	2.5–5·2 5–10·1	– or ↓	Small Small	Hypertension	MIDAS
Felodipine (Plendil)	11–16	2.5–10·1	↓ or 0	~3%	Hypertension	HOT,[14] FEVER[15] (second-line)
Amlodipine (Norvasc)	44+	2.5–10·1	↓ or 0	~7.1%	Hypertension, angina	AASK,[5] IDNT,[9] ALLHAT,[5] VALUE, ASCOT,[16] CASE-J, ACCOMPLISH[17] (combination)
Nisoldipine (Sular)	7–12	10–40·1	– or ↓	~2%	Hypertension	ABCD
Clevidipine (Cleviprex)	0.25	1–2 mg·h, intravenously only	↓	Tiny	Hypertensive emergencies	Reflex tachycardia, hypotension
Nimodipine (generic only in United States after October 2009)	1–2	30·4–6	–	Tiny	Subarachnoid hemorrhage	Metabolized by CYP3A4; not approved for hypertension!

Abbreviations: AASK, African American Study of Kidney Diseases and Hypertension; ABCD, Appropriate Blood Pressure Control in Diabetes; ACCOMPLISH, Avoiding Cardiovascular Events Through Combination Therapy in Patients Living With Systolic Hypertension; ALLHAT, Antihypertensive and Lipid Lowering to Prevent Heart Attack Trial; ASCOT, Anglo-Scandinavian Cardiac Outcomes Trial; CASE-J, candesartan antihypertensive survival evaluation in Japan; CONVINCE, Controlled Onset Verapamil Investigation of Cardiovascular Endpoints; CYP3A4, 3A4 Isoform Of Hepatic Cytochrome P_{450} Enzyme System; FEVER, Felodipine Event Reduction; HOT, Hypertension Optimal Treatment Study; IDNT, Irbesartan Diabetic Nephropathy Trial; INSIGHT, International Nifedipine (Gastrointestinal Therapeutic System) Study: Intervention as a Goal in Hypertension Treatment; INVEST, International Verapamil-Sustained Release–Trandolapril Trial; MIDAS, Multicenter Isradipine Diuretic Atherosclerosis Study; NICS-EH, National Intervention Cooperative Study in Elderly Hypertensives; NORDIL, Nordic Diltiazem; STONE, Shanghai Trial of Nifedipine in the Elderly; VALUE, Valsartan Antihypertensive Long-Term Use Evaluation; VHAS, Verapamil Hypertension Atherosclerosis Study.

Figure 1: The resume of CCBs pharmacokinetics.[6]

CCB's Mechanism of Action

All CCBs take a role as peripheral arterial dilator agents.[6] Calcium channel blockers work by blocking the entry of the calcium into the cell. This makes relaxation of vascular smooth muscle, heart muscle contraction decreased, and the decrease in the speed of the SA node. Calcium channel blocker has a receptor on the cell membrane. Each type of calcium channel blockers has different receptors on the cell membrane. The types of calcium channel blocker that are dihidropin, verapamil and diltiazem have different effects on the physiology of calcium channels. Calcium channel blocker affects less preload load because calcium channel blocker causes relaxation of the arterial smooth muscle, but less effect on the veins. Calcium channel blockers can increase myocardial oxygen supply in a way by causing coronary dilation and causes a decrease in blood pressure and heart rate so that resulted in improved subendocardial perfusion.[8]

Influx natrium caused depolarization in early upstroke phase, then open voltage-gated Ca2+ channel (VOCC). It enables calcium ions to go into a cell and caused prolonged potential action of the heart. CCBs inhibit calcium ions influx into myocardium thus cut down calcium level to induce contraction. Cardiac myocytes have L-type Ca^{2+} channels (LTCCs) that were designated "L" due tu their long-lasting depolarization. LTCCs is having high sensitivity for CCBs (figure 2).[4,9]

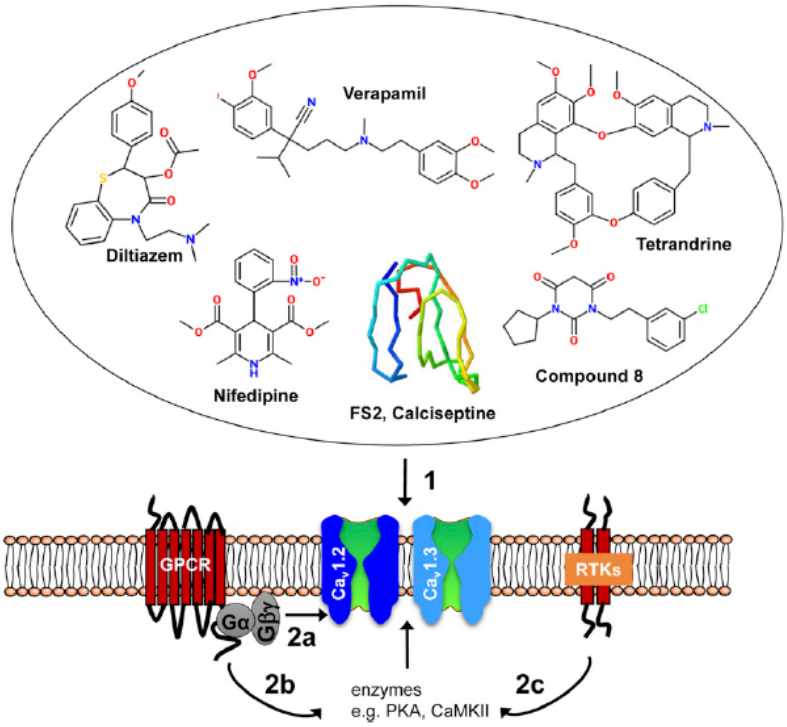

Figure 2: CCBs mechanism of action in LTCCs.[4]

Besides vasodilator effect, CCB (benidipine) was reported may inhibit aldosterone production so it will give natrium diuretic effect. A thiazide may cut down arterial pressure by reducing plasma volume and cardiac output. Afterward, combination benidipine and thiazide will give a better result to reducing blood pressure than combination benipidine and the beta blocker.[10]

At the time of diastole, the levels of calcium extracellular are 10,000 times higher than the levels of intracellular calcium. Besides, the negatively charged intracellular space. This causes calcium influx from the extracellular space into the intracellular space. The increasing levels of calcium in the cytosol stimulate sarcoplasmic reticulum release calcium in large numbers to make the sarcomere work. This reaction will cause the contraction. Calcium channels are not inhibited by tetrodotoxin. Calcium channel blockers inhibit L-type channel in humans. At the heart muscle and vascular, calcium influx has an important role in the occurrence of contractions, but this does not occur

in skeletal muscle sarcoplasmic reticulum because the system has progressed well. This is the reason why the contraction of the heart muscle can be inhibited by calcium channel blocker but not for the skeletal muscle.[8]

CCBs are metabolized by CYP3A4 enzymes. It is the same enzymes that activated clopidogrel. Clopidogrel is widely used as an antithrombotic drug for patients myocardial infarction. It has been suggested that CCBs may disrupt the efficacy of clopidogrel. However, a nationwide cohort study that conducted in Denmark by Olesen et al. showed that the clinical efficacy of clopidogrel in patients with a recent myocardial infarction is not disrupted by simultaneously using CCB treatment. This potential drug interaction is unlikely to have clinical significance.[11]

Current treatment for infarct myocardium

The aim of myocardial infarction treatment is reducing acute myocardial ischemic and limiting infarction size. For patients presenting with a STEMI, the most effective and well-established therapeutic strategy for reducing acute myocardial ischemic injury and limiting MI size is timely myocardial reperfusion using either thrombolytic therapy or PPCI. The duration of acute myocardial ischemia is a critical determinant of MI size, and as such, minimizing the time from chest pain onset to PPCI is the treatment priority.[12]

Side Effect of Long-Term Usage of Ca Channel Blocker

Calcium channel blockers can inhibit paraoxonase activity because human serum paraoxonase is calcium dependent enzyme. It needs calcium to be synthesized in the liver and it has the protective effect on free radical that can cause a formation of oxidized phospholipids in oxidized LDL and neutralizes the effects of atherogenic lipid peroxides.[13] CCB especially nifedipine can induce gingival enlargement if used too long and the dose is not maintained well. CCB can induce the enlargement by reducing the calcium ion influx, leading to a reduction in the uptake of folic acid, thus limiting the production of active collagenase. It increases the amount of connective tissue matrix dominated by collagen fibers.[14]

CONCLUSION

In patients with myocardial infarction, we found CCBs were generally safe and give better result in lowering blood pressure and

reduction myocardial infarction risk. However, CCBs were not typically used for initial treatment. In conclusion, CCBs usually used for some medical conditions, such angina, heart rhythm abnormalities, or as a second drug for hypertension alone.

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THE EFFECT OF ALOE VERA JUICE AND GLIBENCLAMIDE COMBINATION IN DECREASING LDL AND INCREASING HDL BLOOD LEVELS ON THE DIABETIC DYSLIPIDEMIA RAT MODELS INDUCED BY STREPTOZOTOCINE AND NIKOTINAMIDE

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ABSTRACT

Background: Dyslipidemia may occur in Diabetes Mellitus (DM) type 2 due to the activation of *sensitive lipase* hormone. Antidiabetic glibenclamide has been unable to repair the condition of dyslipidemia yet, especially regarding the level of LDL and HDL as the effect of diabetes. One of the attempts to solve the problem is by combining glibenclamide with *Aloe vera* which is proven to have many benefits as antidiabetic and anti-dyslipidemia.

Research Objective:To evaluate the effect of combining *Aloe vera* juice and Glibenclamide towards the changing of LDL and HDL level in dyslipidemia diabetic rats induced by streptozotocine (STZ) and nikotinamide (NA).

Research Method: Twenty male white rats Strain Wistar were randomly divided into four groups. Out of those twenty rats, five of them were made into the control group, and fifteen of them were rendered diabetes by NA(230 mg/kg, intraperitoneally) and STZ(65 mg/kg, intraperitoneally). After the induction, the rats were divided into 3 groups, namely negative control group (only given 0,5% p.o. of CMC), diabetic group which were given glibenclamide (0,18mg/200g p.o.) and diabetic group which were given glibenclamide (0,18mg/200g p.o.) and *Aloe vera* (3,6ml/200g p.o.) within 28 days. The level of LDL and HDL was evaluated during the pre-induction, post-induction, and post-treatment by using *spectrophotometer*.

Result:The combination of *Aloe vera* juice and Glibenclamide can reduce the level of LDL and increase the level of HDL in the rats models with dyslipidemia diabetes (p=0,000; 95%*CI*).

Conclusion: The findings suggest that the combination of *Aloe vera* juice and Glibenclamide has the highest LDL level reduction effect and HDL level raise in the dyslipidemia diabetic rat models.

Keywords : Streptozotocine, Nikotinamide, LDL, HDL, *Aloe vera*, Glibenklamide, Dyslipidemia, and Diabetes Mellitus

INTRODUCTION:

Diabetes mellitus (DM) is a metabolic syndrome disorder in the insulin secretion, insulin activity, or both and the chronical hyperglycemia in this DM is related with the prolonged damage, dysfunction or the failure of some body organs, especially the eyes, kidney, nerve, heart and blood vessels[1,2]. DM has four classification, i.e. Type 1, Type 2, Gestational, and others[3]. Within DM cases, there are three classical symptoms present, namely polyuri, polydipsi, and polifagi[4].

Indonesia is now in the 4th rank of the countries in the world which have the highest number of diabetic cases following the United States of America, China, and India[5]. Whereas, International Diabetic Federation (IDF) estimated in 2009 that the number of people with diabetes mellitus will increase from 7,0million in 2009 into 12,0 million in 2030[6]. Out of all DM cases present, 90% of them are DM type 2[7].

Metabolic syndrome disorder in DM patients not only disturbs the metabolism of carbohydrate but also disrupts the lipid metabolism. Consequently, the diabetic patient will experience the changing level of lipid protein plasma. One of which is the increase of LDL level and the decrease of HDL level[8]. Someone who suffers from DM has a high risk of experiencing dyslipidemia which is in accordance with a *cross-sectional* research conducted by Harris in 2005 which states that among DM type 2 patients, 55% of them will experience dyslipidemia in ≤2 years whereas 66% of them will experience it after around ≥15 years[9]. This lipid metabolism change will worsen the prognosis of the patients suffering from DM. Therefore, controlling must be conducted to the patients who suffer from diabetes mellitus so their lipoprotein plasma level is consistently within the normal limit[10]. In this term, a medication which is able to repair not only the glucose level but also the lipid protein plasma level in blood is required.

One of the chemical medicines used for DM patients is glibenclamide. This medicine is one of the synthetic medicines belongs to sulfonylurea groups which works in reducing the glucose level in the blood by stimulating beta Langerhans pancreatic cells to produce insulin[11].

Mughal's research shows that giving glibenclamide for 12 weeks to the patients with DM type 2 can increase their HDL level significantly although it has

not reached the normal level yet, whereas the total cholesterol level, triglyceride, LDL, and VLDL, does not decrease significantly. This improvement of HDL which is still under the normal value is possibly caused by the increase of Apo lipoprotein A-I (apo A-I) catabolism in dyslipidemia diabetikum[12].

One of the attempts conducted to improve both of the blood's glucose level and LDL as well as HDL level is by adding a substance which comes from herbal plants. The herbal plant which is known to be able to influence the lipid metabolism in DM type 2 is *Aloe vera*. Consuming *Aloe vera* can decrease the amount of free fatty acid by way of impeding *hormone sensitive lipase*enzyme[13]. Moreover, the utilizing of *Aloe vera* is proven to be more effective than the utilizing of glibenclamide towards the decrease of LDL level and the increase of HDL level in rats with DM type 2[14]. The research regarding the influence of combining glibenclamide with *Aloe vera* towards the decrease of LDL level and the increase of HDL level has not been conducted yet. Thus, it is expected that this research by way of combining *Aloe vera* and glibenclamide can decrease the LDL level and increase the HDL level. Subsequently, glibenclamide's role lies in controlling the blood's glucose level of DM patients, whereas *Aloe vera* is expected to be able to control lipid plasma's profile.

Research Method:

This research is an experimental research using *Pretest Posttest with Control Group Design* approach by the usage of animal experiments. The inclusion criteria of this research is male rats at the age of 2-3 months, with body weight (BW) of 180-250g, healthy rats and have been through acclimation period of 7 days, and the rats which experience dyslipidemia diabetes. This research used 20 white male Strain Wistar rats which were arbitrarily divided into 4 groups. Out of those 20 rats, 5 of them were made as the normal control group (K1) and 15 of them were induced with diabetes with 230 mg/kg of NA and 65 mg/kg of STZ in an intraperitoneal way. After the induction, the rats were divided into 3 groups, namely K2 which became the negative control group (only given 0,5% p.o. of CMC), K3 which was the diabetic group which were given glibenclamide (0,18mg/200g p.o.) and K4 which was the diabetic group which were given glibenclamide (0,18mg/200g p.o.) and *Aloe vera* (3,6ml/200g p.o.) during 28 days. The level of LDL and HDL was examined during the

pre-induction, post-induction, and post-treatment period by utilizing spektrofotometer.

This research was conducted for 41 days, with the details of 7 days of acclimatization, 5 days of induction, 28 days of treatment and one day was allocated for the final calculation of the LDL and HDL level. This research was conducted in the Centre of Food and Nutrition Studies (CFNS) or Pusat Studi Pangan dan Gizi (PAU) Gadjah Mada University laboratory. The data analysis was conducted by implementing paired sample T-Test for the pre-induction and post-induction period, One Way anova test for the decrease of LDL level and kruskall-wallis test for the increase of HDL level.

Result:

After acclimatization was conducted for seven days, and 5 days of post-induction GDP level, LDL and HDL (table 1) experienced changes and it was proven to be in accordance with the statistical test that is paired sample T-Test (table 2). However, K1 group also experienced changes despite the fact that induction was not conducted to it of which reason for this to happen was due to the fodder factor being fed. The result of the examinationcan be seen in table 5. According to the laboratory examination of Faculty of Animal Science UGM, the result shows that Br2 comfeed fodder contains 5,7% of fat and 77,293 mg/100gr of cholesterol. However, the exact effect of this Br2 fodder had not been found in any journal by the researcher yet, she only acquired the laboratory test result from UGM.

Table 1. Level of GDP, LDL and HDL (mg/dl)

		N	Average level (X±SD)			increase of GDP, decrease of LDL, increase of HDL (X±SD)
			Pre-Induksi	Post-Induksi	Post-Perlakuan	
GDP	K1	5	74,97 ± 1,7	75,47 ± 1,80	-	0,60 ± 0,50
	K2	5	73,60 ± 1,36	221,26 ± 9,08	-	147,66 ± 8,90
	K3	5	75,86 ± 1,96	223,39 ± 8,95	-	147,52 ± 7,80
	K4	5	73,92 ± 1,82	233,05 ± 11,09	-	159,13 ± 11,10
LDL	K1	5	68,55 ± 3,96	69,64 ± 3,93	62,93 ± 1,57	6,71 ± 3,06
	K2	5	63,72 ± 2,32	95,13 ± 4,41	94,75 ± 4,95	0.38 ± 0,56
	K3	5	68,66 ± 2,92	91,49 ± 2,87	78,74 ± 4,95	12,75 ± 3,96
	K4	5	66,46 ± 2,76	93,43 ± 3,89	78,74 ± 3,65	27,13 ± 4,84
HDL	K1	5	48,93 ± 2.14	48,04 ± 2,10	47,04 ± 2,36	-1.01 ± 0,50
	K2	5	47,63 ±6,38	25,95 ± 2,48	24,17 ± 2,49	-1.78 ± 0,32
	K3	5	48,81 ± 2,50	25,21 ± 1,90	38,34 ± 2,05	13,14 ± 3,31
	K4	5	47,73 ± 3,05	24,66 ± 1,81	46,49 ± 2,91	21,83 ± 2,00

Tabel 2. T-Test Dependent

	Variabel	P-Value	Information
GDP	K1	0.218	zero difference presents
	K2	0,000	difference presents
	K3	0,000	difference presents
	K4	0,000	difference presents
LDL	K1	0,008	difference presents
	K2	0,000	difference presents
	K3	0,000	difference presents
	K4	0,001	difference presents
HDL	K1	0,185	zero difference presents
	K2	0,003	difference presents
	K3	0,000	difference presents
	K4	0,000	difference presents

Furthermore, after the treatment was given during the 28 days period, it could be observed that the decrease of LDL level (table 1) was acquired from the post-induction LDL level minus post-treatment LDL level. When being compared among the groups, it turned out to be significantly different statistically with the one implementing One Way Anova and Pos Hoctests (Table 3).

Table 3. ***One Way Anova*** **LDL**

	Average different	IK 95%		P
		Min	Max	
K1 vs K2	6.32600	1.6402	11.0118	.011
K1 vs K3	-6.04000	-10.7258	-1.3542	.015
K1 vs K4	-20.42800	-25.1138	-15.7422	.000
K2 vs K3	-12.36600	-17.0518	-7.6802	.000
K2 vs K4	-26.75400	-31.4398	-22.0682	.000
K3 vs K4	-14.38800	-19.0738	-9.7022	.000

The increase of HDL level (table 1) was acquired from post-treatment HDL level minus the post-induction HDL level, and the result was also significantly different based on Kruskall-Wallis and Post Hoc Mann-Whitney tests (table 4).

Table 4. ***Kruskal-Wallis*** **HDL**

		N	Median	P
			Min-max	
Groups	K1	5	-1.04 (-1.74 – (-0.34))	< 0,001
	K2	5	-1.91 (-1.95 – (-1.20))	
	K3	5	13.28 (8.46 – 17.4)	
	K4	5	20.88 (20.16 – 24.32)	

Uji *Kruskal-Wallis*. *Uji post-hoc Mann-Whitney*: K1 vs K2 p = 0.016; K1 vs K3 p = 0.009 ; K1 vs K4 p = 0.009 ; K2 vs K3 p = 0.009 ; K2 vs K4 p = 0.009 ; K3 vs K4 p = 0.009

Discussion:

The changes in GDP level on the fifth day after the induction happened because STZ has an action mechanism which can dilapidate β cells, which results in insulin excretion disorder to happen. In which STZ which is cytotoxic in nature can cause the fragmentation of DNA cells, the alkylation of DNA by STZ through the nitro urea cluster, the transference of methyl cluster from STZ to the DNA molecule causes the dilapidation of pancreatic β cells [15]. Whereas NA is protective in nature so there is no further dilapidation of pancreatic β cells [16].

The changes of blood's lipid profile level in the inducted rats is caused by the insulin resistance in the condition of diabetes and fat lipolisis, in which the insulin resistance will cause the activation of *hormone sensitive lipase*in the adipose tissue which then causes the occurrence of hidrolisis trigliserida to become free fatty acid and gliserol. This will cause the increase of free fatty acid within the blood flow, within which the increase of this free faty acid will

cause the increase of LDL and the decrease of HDL level[17].

The level of lipid profile being examined are the LDL and HDL. The normal LDL level is > 7-27, 7 mg/dl[18]. In this research, the pre-induction LDL level detected in each group was > 27,7 mg/dl (table 2). It indicates that before the induction, the rats had already experienced the increase of LDL. The normal HDL level for rats is 35-85 mg/dl[19]. In this research, the pre-induction HDL level for each group was included in the normal HDL level (table 3). With the induction of DM, LDL level become increased and HDL decreased into below the limit of the normal level. It shows that DM can indeed influence lipid level, especially HDL level which is included as good fat [20].

After undergone treatments for 28 days, it was perceived that within K3 and K4 groups, the LDL level decreased although it had not been under the normal level yet and the HDL level increased until it reached the normal level. The changes of level in group K3 and K4 was due to the fact that glibenclamide is an anti diabetes medicine included in sulfonilurea category which will bond with the receptors in beta pancreatic cells and will cause the obstruction of sensitive ATP potassium canal which causes depolarization and the opening of calcium belvotase canal. The opening of calcium canal causes the extrication of granular insulin which will, in the end, indirectly influence the LDL and HDL level[11]. However,0, 5% of CMC did not give any effect towards the reduction of glucose level and total cholesterol in the blood since it was not digested and resobrtial[21].

Group K4 had the decrease level of LDL and the highest HDL increase compared with the other groups because

Aloe vera was given to this particular group and there are three active substances contained in *Aloe vera* which can reduce the LDL level and increase the HDL level, i.e. Niacin, Vitamin C, and Magnesium.

Niacin contained in *Aloe vera* can impede *hormone sensitive lipase* (HSL) enzyme, in the condition of insulin deficiency HSL activation can happen which causes the extrication of free fatty acid from adipose tissue[8]. Free fatty acid itself is a substance which forms VLDL in the liver, consequently, if the synthesis of VLDL is blocked or impeded, there will be a decrease on bad fat level specifically LDL and the increase of good fat explicitly HDL. Besides playing a significant role in blocking HSL,

niacin can also improve the activity of *lipoprotein lipase* enzyme which causes the decrease on triglyceride and chylomicron level[13]. Vitamin C or ascorbic acid in *Aloe vera* functions as one of the natural antioxidants which helps in the process of hydroxylation in the formation of bile salt so it increases the excretion of cholesterol and reduces the cholesterol level in the blood. Moreover, by giving vitamin C, it can increase the mineral selenium level in the blood, in which this mineral will play a part in preventing LDL's oxidation[22]. While magnesium is one of the minerals included in *Aloe vera* which has an action mechanism similar with simvastatin medicine namely preventing enzyme HMG Ko-A reductase so there is an increase in the total cholesterol excretion[23]. In addition to those substances, *Aloe vera* also contains some other substances which also help in the reduction of the rats' LDL level and the increase of their HDL level such as selenium which helps in preventing hyperlipidemia to happen.

Anthraquinonoid can also give a *laxative* effect by way of creating a gel which can accelerate the food transport in the intestine and prevent the absorption of fat, as well as producing polyphenol which can increase the excretion of cholesterol and bile acid towards the feces[24]. Moreover, glibenclamide also has the nature of not influencing the fat metabolism for diabetic patients[25].

The research that we have conducted is by way of combining *Aloe vera* and glibenclamide which can cause significant decrease of LDL and increase of HDL. This result is in accordance with the research conducted by Bunyapraphatsara *et al*. However, in their research, the lipid level being examined was specifically towards cholesterol and triglyceride and not towards LDL and HDL as conducted in our research. In addition, another difference being found is the giving of glibenclamide. In Bunyapraphatsara *et.al* research result, there was no changes in lipid level[26], whereas in our research, the giving of glibenclamide can change the lipid level although the result is lower than the ones given the combination (glibenclamide and *Aloe vera*). It might happen due to the different samples being used. Our research used rats as the sample whereas Bunyapraphatsara *et.al* research used humans or DM patients as the sample.

The researches to observe the effects of combining *Aloe vera* and glibenclamide are still limited in number. Most of them

only compared both substances without food factor. Hence, the process of the observing the effects when they were research can run optimally an in given at the same time. Accordingly, new accordance with the pre-arranged plan. In researches are required to observe whether addition, the time of the research can be the effects of giving both substances at the extended so the prolonged effect can be same time can be improved further or not.

Conclusion

The combination of glibenclamide 0,18 mg/200g BW of a rat and *Aloe vera* with the amount of 3,6 ml/200g rat BW of a / day can reduce LDL level and increase HDL level for model rats which suffer for Dyslipidemia Diabetes. The decrease of LDL level and increase of HDL level for the combined gilbenklamid and *Aloe vera* group can be bigger if compared with other groups. This reduction of LDL level and increase of HDL level is proven to be statistically significant.

Suggestion:

Further researches should better adjust the fodder provided for to the animal with the desired condition in accordance with the objectives of the research to ensure that the treatments implemented are not influenced by the implemented are not influenced by the food factor. Hence, the process of the research can run optimally and in accordance with the pre-arranged plan. In addition, the time of the research can be extended so the prolonged effect can be observed whether it will give a better result or vice versa.

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